HARVARD UNIVERSITY

BUSSEY INSTITUTION OF APPLIED BIOLOGY

CONTRIBUTIONS

FROM THE

LABORATORY OF PLANT GENETICS

VOL. I.

BOSTON
1909–1913

CONTENTS.

The Transmission of Variations in the Potato in Asexual Reproduction

BY

EDWARD M. EAST

PUBLICATION OF THE CONNECTICUT AGRICULTURAL EXPERIMENT STATION

CONTRIBUTION FROM THE LABORATORY OF GENETICS
BUSSEY INSTITUTION OF HARVARD UNIVERSITY
NO. 3

[REPRINTED FROM CONNECTICUT EXPERIMENT STATION REPORT, 1909-1910.]

THE TRANSMISSION OF VARIATIONS IN THE POTATO IN ASEXUAL REPRODUCTION.*

By Edward M. East.

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* The experimental work reported in this paper was conducted as a project under the federal appropriation for agricultural experiment stations, known as the Adams Fund. The field and laboratory work were done at the Connecticut Agricultural Experiment Station; the results were assembled at the Bussey Institution of Applied Biology of Harvard University.

INTRODUCTION.

In a previous paper (3), two series of data were presented, each giving the results from one year's selection of the extremes of a highly variable but accurately determined fluctuating character of the common potato, *Solanum tuberosum* L., when these extremes were reproduced asexually. The character under consideration was the total content of nitrogen determined by the Kjeldahl process. In one series, extremes averaging 14.07 per cent. and 8.75 per cent. in total nitrogenous matters when calculated to water-free basis, yielded crops averaging 14.70 per cent. and 12.59 per cent., respectively; in the other series, extremes averaging 18.06 per cent. and 13.37 per cent. in total nitrogenous matters, yielded crops averaging 10.90 per cent. and 9.29 per cent., respectively. These figures were due apparently to a correlation between mother and daughter tubers—if one may be allowed to use such terms—and yet the evidence even at that time did not appear to the writer to justify a conclusion that asexual fluctuations were inherited.

In the first place, the material at our disposal was a commercial stock, with nothing to guarantee its purity but our own judgment. Furthermore, it was slightly affected with scab and was treated with formalin for this disease. This treatment, poor soil and weather conditions, and the use of Paris green were so unfavorable to development that but few normal tubers resulted. This loss limited the data of the first series to one year, besides increasing the experimental error by making it impossible to compare tubers of the same state of maturity. The crop from the extremes of the second series, though much better than that of the first series, was small and could not be regarded as a normal crop of tubers. It was also a commercial stock. This experiment was terminated by a loss of the crop while it was in storage.

These statements show that it is impossible to conclude that our own observations included cases of definite inherited change among nitrogen fluctuations; on the other hand, the authenticity of certain cases reported by others, where permanent changes such as loss of color have occurred in somatic tissue, cannot be doubted. Furthermore, in an exhaustive review of the work of previous investigators (3), the writer also found evidence that in rare instances heritable change in shape, size and chemical composition *may have occurred;* yet it was quite evident that in most of these cases the already complex question had been rendered more complex by refusing to distinguish between factors that belong essentially to the study of heredity, and factors that are primarily those of the physiology of development. In spite of the unconvincing nature of the published records concerning these other changes, there is no *a priori* reason to believe that the color variations noticed in commercial practice are the only transmissible variations that occur; and in any case the nature, frequency and causes of variations that do occur are entirely undetermined.

These reasons were of themselves sufficient to make it desirable to have the work repeated, with the experimental errors eliminated as far as possible; but there was still another reason for its repetition. Since the previous experimental work had been completed, Johannsen had published his classic monograph on "Erblichkeit in Populationen und in reinen Linien" (5), in which he concluded, first, that in a pure family line arising from a single self-fertilized seed, fluctuations are not inherited; second, that gametic changes may take place within a pure line, and when such a change occurs in an individual, its progeny at once forms a separate pure line or biotype. This piece of work was so excellent in all of its features that many biologists were at once convinced that his conclusions were correct. Other investigators have waited for corroborative evidence. Such evidence has recently been forthcoming. Jennings' (4) beautiful investigations concerning inheritance in protozoa lead him to exactly the same conclusions that Johannsen had obtained with beans as a material. Johannsen left asexual fluctuations out of consideration, although no essential distinction has yet been shown between them and fluctuations of a pure line propagated by sexual reproduction. Jennings, by his work on unicellular organisms, has proved the great similarity between inheritance in sexual pure lines and inheritance in asexual reproduction. In this paper is shown the similarity between the inheritance of fluctuations in asexual reproduction in multicellular organisms and that in the classes treated by Johannsen and Jennings. Although the data are comparatively few, still they are sufficient, I think, to give a relatively high probability to this conclusion.

THE MATERIAL AND ITS TREATMENT.

In 1906 a number of both foreign and domestic varieties of potatoes was gathered together to observe the frequency with which differentiations occurred within a variety reproduced by tubers. In 1907 other varieties were added until they included over seven hundred named commercial varieties as well as a large number of seedlings. The sources of the varieties in Europe were Vilmorin, Andrieux & Cie. for France and Germany, and Sutton & Co. and Thos. Scarlett for England and Scotland. In the United States practically all of the introducers of commercial varieties contributed to the collection. Observations were made upon this stock in 1906, 1907, 1908 and 1909, although a number of varieties were discarded in 1908 because their characteristics were the same as those of other varieties. Among our unnamed seedlings was one of Early Rose, the seed from which it was grown being in all probability self-fertilized. It had no color either in the sprouts or in the skin; its shape was short oval round; its eyes deep. There were several excellent reasons why it would make a good subject for an investigation of the correlation between mother and daughter tubers when extreme fluctuations were selected. Having been raised from the seed in 1901, selected from a single hill in 1902, and carefully propagated by tubers for four years, there was no question but that we were dealing with a single variety wherein any variations present were produced asexually. In addition, there is reason to believe that the characters that it possessed are all recessive, as follows: white tubers, recessive to colored tubers; white flowers, recessive to colored flowers; deep eyes, recessive to normal eyes; roundness, recessive to elongation. Our reasons for this belief will be published in a separate paper. That the exact characters possessed by the variety are of some importance will be seen later when the evidence tending to show that bud variations are always losses of dominant characters, is produced.

Our material in 1906 for this portion of the investigation, then, consisted of about four bushels of tubers that had been produced from a single tuber in 1902. From this stock one hundred and seventeen representative tubers with the following weight distribution were selected for analysis.

TABLE I.

Distribution of Weights of Tubers of Original Stock, Crop of 1906.

A. = 137 ± 2.06 S. D. = 32.97 ± 1.45 C. V. = 24.07% ± 1.21%

Class centers in grams	85	105	125	145	165	185	205	225	245	265	285
Frequency	2	25	40	28	11	4	2	3	0	1	1

These tubers were sampled with a cork borer twelve millimeters in diameter, lengthwise but slightly to each side of the center—a method that previous experience had shown to be accurate (3). One of these samples was cut up and dried to constant weight at a temperature of 104° C., in a glycerol oven through which a current of hydrogen was passing. This gave the dry matter determination. The total nitrogen was determined by the regular Kjeldahl method as used by the Association of Official Agricultural Chemists. The total nitrogen was multiplied by the factor 5.5 and called total nitrogenous matters, although it is recognized that this factor may vary in a vegetable like the potato where the different nitrogen compounds are numerous. Yet the very fact that there are various compounds of nitrogen should give us a greater chance to determine whether variations in ability to assimilate nitrogen, constant in succeeding seasons, are produced with frequency in somatic cell division.

In discussing the data obtained, the simplest biometrical constants have been calculated, using the formulæ given by Davenport (1). The computations were all checked, but not in every case by two individuals. In each instance all of the figures have been used, although there are a few cases where strict mathematical treatment would call for the rejection of one or two extreme variates.

I believe that in cases where individual analysis is impossible, this modern statistical treatment is a great aid in concentrating the attention on the meaning of the figures *en masse,* yet there is no desire to argue that this treatment brings out more than can be seen by careful examination of the original data.

Inheritance of Fluctuations in Composition.

Results from the Analysis of the 1906 Crop.

The results of the analysis of the crop of 1906, from which were made our first selections for planting, are given in tables four to seven.

The classes in the frequency distribution of dry matter are centered on the even per cents. The mean is 18.15 ± .118 per cent., which is close to the average in water-free substance that analyses of a number of American varieties have shown. They are moderately variable, the coefficient of variation being 10.47 ± .463 per cent. Owing largely to the fact that in the highly variable character, weight, one or two individuals were found with a low per cent. of dry matter, the correlation between weight and dry matter is —.275 + .058. It is probable that the moderately wet period, during the time these tubers were maturing, is the cause of this, although it may very well be that there is generally a slight minus correlation between dry matter and weight.

The nitrogenous matters calculated to fresh material show a regular distribution with a mean of 1.92 ± .015 per cent. The standard deviation is .248 ± .011, and the coefficient of variation is 12.95 ± .580 per cent. When calculated to dry basis the mean is 10.75±.129 per cent.; the standard deviation is 2.08 ± .091; the coefficient of variation is 19.35 ± .740 per cent. There is no correlation between the weight of the tubers and the per cent. of nitrogenous matters, fresh basis; when calculated to dry basis, however, there is a slight correlation with weight, namely, .121 ± .061. As this correlation is only twice the probable error, we cannot be certain that it is a true value.

There is no doubt concerning the negative relationship between dry matter and nitrogenous matter, as the correlation coefficient is —.346 ± .055 when dealing with nitrogenous matters, fresh basis. The constant is naturally greatly increased when we use the dry matter as a basis for reducing the nitrogenous matters to comparable form. It then becomes —.758 ± .026. This relationship between dry matter and nitrogenous matters is not unexpected, for it is pretty definitely established that in all starch-storing plant parts, relatively more nitrogenous compounds are formed in the early stages of growth, and relatively more starchy compounds in the final stages. This fact should be remembered when we consider correlation between mother and daughter tubers; for it follows that the relative percentage of dry matter is a better criterion of maturity than is the weight (although actual dry matter content varies widely in different seasons), and we should try as far as is possible to compare tubers of the same degree of maturity.

Results from Growing the Extreme Nitrogen Fluctuations of the 1906 Crop.

From the one hundred and seventeen tubers of the 1906 crop that had been analyzed, ten tubers high in nitrogenous matters and ten tubers low in nitrogenous matters were selected for planting in 1907. The basis upon which they were selected was their per cent. of nitrogen, calculated to fresh basis, for the determination of dry matter took about seven days and it was thought best to plant the tubers before its completion. Table two, however, shows that the calculation to dry basis in no case affected the classification of the individual.

TABLE II.

EXTREME NITROGEN FLUCTUATIONS PLANTED IN 1907.

High Nitrogen Extremes.				Low Nitrogen Extremes.			
Wt. grams.	Dry matter.	Nit. mat. fresh b.	Nit. mat. dry b.	Wt. grams.	Dry matter.	Nit. mat. fresh b.	Nit. mat. dry b.
119	18.02	2.48	13.76	136	17.32	1.53	8.83
165	16.40	2.34	14.27	158	18.11	1.45	8.01
145	14.75	2.36	16.00	203	16.01	1.42	8.87
118	17.98	2.37	13.18	129	18.72	1.30	6.95
137	16.40	2.49	15.12	187	18.30	1.63	8.91
146	16.13	2.42	15.00	151	19.09	1.52	7.96
138	16.83	2.38	14.14	106	18.90	1.50	7.94
106	18.81	2.36	12.50	114	19.85	1.61	8.11
110	15.35	2.36	15.37	103	19.26	1.61	8.36
113	17.74	2.28	12.85	120	21.66	1.43	6.60
Ave 130	16.84	2.38	14.22	141	18.72	1.50	8.05

Each of the ten tubers of each selection was cut in four pieces of as equal weight as possible. It has been found by many experiments, that if other conditions are equal, the yield increases directly with the size of the seed piece; therefore, we endeavored to have our selected tubers and the planted pieces as uniform in size as possible. Notwithstanding the desire to obtain a normal crop, a sufficient number of tubers could not be analyzed to have the extremes especially uniform, nor were the planted pieces large enough to yield very good crops. References to table nineteen, however, will show that a fairly normal crop was obtained.

The tubers were planted on uniform soil in contiguous rows. They were allowed to develop naturally, the larvæ of the potato beetle being removed by hand and no sprays of any kind used. The season was somewhat dry during July and the first part of August, but there was a normal rainfall in the spring and in the latter part of the summer.

The chemical determinations upon the crop were made in the same manner as before. Where possible three tubers were analyzed from each hill. This would give twelve analyses of daughter tubers from each mother tuber. In a few cases, however, only one tuber was large enough for the analysis to have any value. The complete data are shown in tables twenty and twenty-one.

Table nine shows the correlation between weighted mothers and their daughter tubers in nitrogenous matters, dry basis, from the high nitrogen plot. The coefficient of correlation — .387 ± .054 shows that there was no positive correlation between the deviations of mothers and daughters of this one selection; nor was there any positive correlation between mothers and daughters in the low nitrogen plot. For the fact that there is a minus correlation as high as — .510 ± .051, I have no explanation. It is undoubtedly a physiological phenomenon, connected with the various states of maturity of different individuals. It illustrates how easily data from one season might show a positive correlation that would be mistaken for a proof of the inheritance of fluctuations. (See paper No. 3.)

Table eleven shows to better advantage that there was absolutely no inheritance of fluctuations. The weighted arithmetic means of the weighted mothers were 2.23 per cent. and 1.55 per cent. for the high and the low nitrogen plots, respectively, yet the frequency distribution of the daughters is so nearly the same as to be quite remarkable when the small number of individuals is considered. Both have the same mode at 2.5 per cent.

The means are 2.51 ± .028 per cent. and 2.51 ± .027 per cent. respectively. Two aberrant individuals raise the coefficient of variation in the high nitrogen plot to 17.41 ± .800, while in the low nitrogen plot it is 15.62 ± .771; but if these two individuals are discarded the variability is about the same.

The similarity of the progeny of the two plots is just as great when we examine the nitrogenous matter figures calculated to water-free material. The weighted arithmetic means of the weighted mothers of the high and of the low nitrogen plots are 14.11 ± .073 per cent. and 8.12 ± .015 per cent. respectively, yet the means of the daughters are 12.74 ± .171 per cent. and 12.81 ± .170 per cent. respectively.

In tables thirteen, fourteen and fifteen we have considered a possible inheritance of fluctuations in dry matter. Taking the high and low nitrogen plots separately, the coefficients of correlation between mothers and daughters are — .140 ± .062 and — .286 ± .063, respectively. There is no reason why we should not add the data from these two plots together, however, for they were grown side by side on the level land, and had the same treatment in each case. When this is done the coefficient of correlation is — .194 ± .045, which shows that there is no inheritance of fluctuations of dry matter.

Results from Growing the High Nitrogen Extremes from the High Nitrogen Plot of 1907 and the Low Extremes from the Low Nitrogen Plot of 1907.

From the analyzed tubers from the high nitrogen plot, the ten highest in nitrogenous matters, fresh basis, of those of normal development, were selected for planting in the high nitrogen plot of 1908. In like manner the ten tubers lowest in nitrogenous matters, fresh basis, of those of normal development in the low nitrogen plot, were selected for planting in the low nitrogen plot of 1908. Each tuber was divided into four nearly equal parts and planted as in 1907. The two selections were also planted in contiguous rows as before, although not on the soil formerly used.

A six weeks' drought during the growing season of this year killed some of the plants before tubers were set, and reduced the yield of all the plants. We were able to do some irrigation after the first two weeks of drought, but even this lack of water limited the setting of the tubers, so that we were able to find but sixty-seven tubers large enough to be worth analyzing.

The complete data resulting from these analyses are recorded in tables twenty-two and twenty-three. In table sixteen we have the correlation between weighted mothers and daughters from both plots in per cent. dry matter, *viz.,* .228 ± .078. This result is the first positive correlation coefficient that we have found, and as it is small and the number of individuals is small, it can have but little weight in our general conclusions. The mean of the weighted mothers being 19.01 ± .100 per cent., and that of the daughters being only 12.19 ± .129 per cent., shows that the tubers have not obtained their normal development and from this their normal dry matter. This fact is shown also by their small size (tables twenty-two and twenty-three). Since we have seen before that much more of the total amount of nitrogenous matters contained in each tuber is formed early in the season than is stored later, we can place much more dependence in the total nitrogen determination than in the dry matter determination. The abnormality of the dry matter, however, affects the reduction of the percentage of the nitrogenous matters to water-free basis.

TABLE III.

Extreme Nitrogen Fluctuations Planted in 1908.

High Nitrogen Extremes.				Low Nitrogen Extremes.			
Wt., grams.	Dry matter.	Nit. mat. fresh b.	Nit. mat. dry b.	Wt., grams.	Dry matter.	Nit. mat. fresh b.	Nit. mat. dry b.
90	16.60	3.29	19.82	77	19.57	2.03	10.37
114	17.60	2.91	16.53	109	19.08	1.87	9.80
68	20.50	3.10	15.12	88	21.43	2.02	9.43
94	18.70	2.87	15.35	88	20.62	1.97	9.55
79	19.30	3.19	16.53	105	18.28	2.06	11.27
74	18.80	3.30	17.55	114	19.81	2.14	10.80
67	21.10	3.17	15.02	118	18.55	2.14	11.53
99	17.20	3.14	18.25	168	19.80	2.08	10.50
72	18.20	2.97	16.32	92	19.28	2.18	11.30
89	16.90	3.29	19.47	92	18.79	2.21	11.76
Ave. 85	18.49	3.12	17 00	105	19.52	2.07	10.63

Although the number of individuals analyzed in 1908 was small, the resulting distribution of nitrogenous matters, when considered in the form in which they were brought together in tables seventeen and eighteen, should carry considerable weight. We are dealing, in the crop from each plot, with extremes that have been selected for two years to try to force them apart in their composition. We are really dealing, then, with a very large population from which the middle, *i. e.,* the mediocre individuals have been dropped out. If there is the slightest amount of inheritance of these deviations, the weighted arithmetic mean of the high nitrogen plot should be higher than the weighted arithmetic mean of the low nitrogen plot. But this is not the case: the means are 2.32 ± .028 per cent, and 2.48 ± .033 per cent., respectively, the nitrogenous matters, fresh basis, even averaging somewhat higher in the low nitrogen plot. The percentage of variation is also about the same in each plot, 10.26 ± .860 per cent. and 11.24 ± .942 per cent. When reduced to the basis of water-free material by use of the aberrant dry matters, the mean of the high nitrogen plot, 20.6 ± .459 per cent., is slightly higher than that of the low nitrogen plot, 19.3 ± .358 per cent., but this difference is within the limit of probable errors.

Table nineteen may be mentioned here. It was added to show the total crops obtained from each tuber planted. It sustains former conclusions by several investigators, that there is a positive correlation between weight of seed tubers and size of crop.

We may conclude from this part of the investigation that neither the relative content of dry matter nor that of the nitrogenous matters of the potato can be changed by the selection of fluctuations and their subsequent asexual reproduction.

Note.—The crop of 1908 was so much reduced by the unfavorable season that it seemed scarcely worth while to continue the experiment with such seed; the selections were made as usual, however, and were planted in 1909. As was expected, the plants had no vitality and the crop of 1909 was discarded.

Inheritance of Fluctuations in Yielding Power.

Many experiments have been conducted to find out whether the yielding power of a variety of potatoes can be increased by selection from the highest yielding plants. The writer previously has reviewed the evidence (3) and shown that it is inconclusive. In a more recent investigation, that of Waid (6 and

7) of the Ohio Agricultural Experiment Station, a positive conclusion is reached. This conclusion seems to have enlisted the support of as eminent a student of genetics as Webber (8). There are a number of points, however, upon which Waid's work needs explanation. In the first place, he seems to have taken no account of the well-known fact, that if other conditions are equal, the yield increases directly with the size of the seed piece, owing to the greater amount of stored food upon which the young plant may draw. In the second place, Waid apparently used a commercial stock, and was not absolutely certain that he was dealing with a single variety. Many varieties now upon the market are exactly alike in external characteristics and are often mixed by seedsmen, although from having originated from separate seedlings and different parents, their yielding powers may be quite different. Furthermore, we know that stock of the same asexually produced variety may be quite different in its yielding power when grown under different soil and climatic conditions. This fact, which may account for differences in Waid's own results in the second and third seasons, as well as the differences in yield in his first selections in his commercial stock, is purely a physiological phenomenon of development and should be separated from the question of inheritance which is under discussion. If actual permanent differences in yielding power are produced by asexual variation in normal, mature, healthy tubers, the yielding power of a variety could be raised by selection. We know that deterioration takes place when immature or diseased tubers are used for reproduction; this is not due to the inheritance of a variation, but to the effect upon the vitality of the plant of starting it from a diseased tuber, or from a tuber which from immaturity has not a sufficient amount of nutriment stored up for the use of the young plant. There is no question here of selection in the ordinary sense, but of selection similar to the separation of heavy from light tobacco seed, because the former gives healthier plants although their hereditary characters are the same as those of the plants from the light seed. In the same way, with potatoes it is a question of having external conditions favorable for a mature crop free from disease, and not of selection of high yielding plants to increase the yield through a variation actually transmitted to the descendants of a varying somatic cell.

In 1906 we had in stock a supply of the well-known variety Rural New Yorker No. 2, which had been grown from a single hill in 1904. A selection of tubers from the five best yielding hills was planted in 1907, and compared with five normal hills producing only one-half as much. The five best yielding hills averaged 1,200 grams of tubers per hill, with an average set of eight tubers. The check hills averaged 600 grams, with a set of four tubers each. Ten hills were planted in each case, two tubers being planted from each hill. In every case pieces of about the same weight were planted. The yield from the high yielding selections was at the rate of 101 bushels per acre; while the yield from the check hills was at the rate of 128 bushels per acre.

In 1908, four tubers from each of the best two hills were again planted from the progeny of Selection A, the high yielding hills of 1906. These were checked with four tubers from two normal, but not high yielding hills of Selection B. This year the selected hills of Selection A average 1,000 grams per hill, while the selected hills from Selection B averaged 600 grams per hill. As above, tubers of the same size were planted in each case. Selection A yielded at the rate of 96 bushels per acre; Selection B yielded at the rate of 90 bushels per acre.

In 1909 the best two hills, averaging 1,100 grams, from Selection A, were again planted, and compared with the two normal hills of Selection B, averaging 700 grams per hill. The yield from Selection A was at the rate of 115 bushels per acre; while the yield from Selection B was at the rate of 120 bushels per acre, although the four hills were planted with equal-sized tubers as before.

It is admitted that these plots were very small and that the thirty hills of the 1906 crop forming the basis of selection do not allow the selection of wide extremes, yet I have succeeded a number of times in isolating strains of maize of different yielding power with about the same number of individuals as a basis. In maize, where there is a constant state of hybridation between natural biotypes, such isolation is possible; in asexual reproduction in the potato, I do not believe it to be a common possibility. I admit, however, that in rare cases it is not without the bounds of probability that changes occur in the somatic cells whereby real differences in inherent yielding ability are produced. It may be that the results of Waid mentioned above were from one of these instances. Attention has been called to other possible explanations of them, because it seems desirable that great caution should be used in recommending asexual selection to commercial growers as a means of actual improvement of the crop, in view of the facts, first, that out of many investigations on the point no indisputable evidence of improvement has been reported, and second, that even the questionable instances of positive results are extremely rare.

INHERITANCE OF POWER OF RESISTING INHOSPITABLE ENVIRONMENT.

A number of investigators have noticed individual plants within a variety that have remained green after the majority had died. In several cases these instances have been cited as due to disease resistance in the plants in question, the disease that they were supposed to have resisted being the leaf spot disease caused by the fungus *Alternaria solani,* commonly known as early blight. In no case have I found it recorded that the dead plants were given a *post-mortem* and the cause of death actually determined as early blight. Since they have died and in the process have had spotted leaves, it has merely been taken for granted that early blight was the cause of death. Several investigators have selected seed tubers from the dead plants and also from those remaining alive, and from a comparison of their respective progenies, have made the claim that the latter transmitted the power of disease resistance.

The writer kept over 700 varieties under observation in 1907 with the idea of obtaining data upon the question. There was very little injury due to fungi during the year. Dr. G. P. Clinton did not find late blight, *Phytophthora infestans,* until just before the first frost, and then found only a few affected plants. It is possible that early blight was present during the summer, but the important immediate cause of death was the physiological trouble, tip-burn. This trouble is present every season, although more in evidence whenever there is a long drought. It starts at the large water-pore at the end of the leaf and continues over the surface until the leaf gradually dries up and dies. I examined a large number of plants of different varieties and found that the rate at which the plant succumbed to tip-burn under given weather conditions depended almost entirely upon the stage of growth of the plant. Plants that had not begun to set tubers withstood long periods of drought. The resistance to drought decreased slowly until the tubers reached about 75 grams in weight, and then decreased very rapidly, so that plants with tubers two-thirds their normal size could withstand very little dry weather. When the largest of the tubers were removed from a plant where they were one-quarter normal size, the plants continued to withstand the drought better than other plants of the same variety where all the metabolic activities were being used to store up food in tubers. Of course mutilations such as punctures of flea beetles contributed to the susceptibility of the plants in question and only varieties with about the same amount of injury from this source were taken into consideration.

In the following varieties plants were found that died at least one month before others: Magnum, Warrior, Gov. Folk, Up-to-date, Table Talk, Solief and Gem of Aroostook. From each of these varieties, tubers of the short-lived and the long-lived plants were selected as seed tubers for the next season. Since the tubers from the short-lived plants were not mature, it would not have been a strictly fair comparison to have selected large tubers from the long-lived plants, even though the latter were cut to the same sized seed pieces as the former. For this reason whole tubers of the same size were selected in each case. In the resulting crop there were two cases in which plants among the progeny of the long-lived plants were themselves long-lived, and two cases where plants from the short-lived plants were long-lived; but in each case the remaining plants of both selections died at about the same time.

It must be mentioned, however, that another variety, Mills Banner, was among those from which tubers of long-lived and short-lived plants were grown. In this case all of the progeny of Plant A, the long-lived plant, were themselves long-lived, while all of the progeny of Plant B, the short-lived plant, were short-lived. Owing to this fact, a very careful examination of the progeny of each plant was made, and led to the discovery that all of the progeny of Plant A had pink sprouts, while all of the crop from Plant B had white sprouts. Either a bud variation had taken place or an accidental mixing of two varieties had occurred.

As a result of these experiments I would not go so far as to say that variations in power of resisting physiological or fungous diseases *do not occur* in asexual reproduction, but I do believe that the relative probability that the commercial grower will obtain disease-resisting varieties by this means is negligible. In most cases of so-called disease resistance within the variety, non-infection would be the proper term; and when there is apparently a marked difference in vitality, it is due to differences in maturity at the time the plants are called upon to withstand inhospitable environmental conditions. The fact that seed pieces planted at the same time produce plants that are at different stages of maturity at the same time, has nothing to do with the question under discussion. It may be due to any of several causes, such as differences in size of seed piece, varying amounts of soil fertility, injury of the buds of the subterranean stem, or removal of young tubers by rodents.

Inheritance of Variations Generally Classed as Bud Variations.

In a former paper (2), the following statement was made in regard to bud variation in the potato:

"In the potato bud variation has almost always been confined to color variations. Mr. A. W. Sutton (in a personal communication) makes this statement: 'I have no hesitation in affirming that there is no potato in commerce to-day in England, and I might say in Europe, which owes its origin as a distinct potato to bud variation in any form whatever.' After a wide inquiry in the United States, I believe that the statement is also true for this country. Mr. Sutton cites 'Forty-fold,' 'Beauty of Hebron,' and 'Paulsen's Blue Giant,' as having varied from colored skins to white. Mr. P. deVilmorin, also, has kindly given me two instances of the same sort. In this country I have seen three color variations, and collected evidence concerning five others—apparently authentic—that are losses of color characters. But two exceptions to the rule have appeared. Mr. Sutton reports that 'Rector of Woodstock,' which was originally white, has produced a purple variegated skin. In this case, however, it is unknown whether the parent tuber possessed the power of forming the purple dye, as is indicated in some of our white varieties by their purple sprouts.

"There has appeared a similar variation also in the 'White Pearl,' reported to me by Prof. E. R. Bennett. This potato, white in color with pink sprouts, appears to have produced a purple spotted variety. Old potato growers, however, say that the parent of the 'White Pearl' had a purple skin."

The authentic evidence, that we had at that time, included but one case where a colorless variety had sported to a colored variety—the Rector of Woodstock—and even this case is doubtful from our meager knowledge of the parent variety. Although my personal knowledge has increased materially since the above was published, I have no reason to change the opinion then expressed, that practically all, if not quite all bud variations are losses of a dominant or an epistatic character allowing the appearance of a recessive or a hypostatic character. A detailed statement of the cases that have come under my observation follows.

Changes in Color.

La Bretonne, Early Sunrise, Bole Zoegling and Seedling No. 60, all pink or red varieties, have produced white variations that were constant the next season. The change affected only the color of the tuber. Seedling No. 853 in its third year from seed produced one white tuber, the remaining tubers being the normal purple. This variation came true the next year.

Several apparent changes from white to colored tubers appeared, but the changes were not constant. Arabella, Solief and Crown Jewel, all varieties in which the sprouts have a slight pink color, produced individual tubers with a marked pink coloration. The next season, however, the tubers produced by them were exactly like those of the normal variety. In 1907 tubers of the varieties Prizetaker and Aradaras appeared with deep red protuberances (Plate III). Unfortunately, neither of these protuberances produced plants. Even had the variation been constant, however, it could not have been said that color was produced by an absolutely colorless variety, for in each case the normal tubers have some pink in their sprouts.

Changes in Shape.

Four marked changes in shape from long to round tubers have been observed. In the varieties Silver Hill and Early Ohio these changes were permanent. A close observer could recognize other characteristics of the parent varieties in the round tubers, but in each case the change from a long-tubered variety to a round-tubered variety was quite distinct. Two other long-tubered varieties, White Beauty and Seedling No. 842, also produced round variations, but in these two cases the progeny were all long like their parent varieties. Two other variations were noted where the change was not permanent. State of Maine and Orphan, two oval varieties, produced single plants upon which all of the tubers were very much elongated, approaching the Lady Finger in shape. These tubers were grown the next year in comparison with normal tubers of the same variety. In both cases the crops were exactly like those regularly produced by the varieties.

Changes from Shallow to Deep Eyes.

Four permanent bud variations were observed in another chapter which, though similar to change in form, evidently affects a separately inherited factor. Potato varieties to have a commercial value must have rather shallow eyes, otherwise too much substance will be lost in peeling; but sometimes a bud variation occurs in which the tuber appears as if it had had wires attached to all of its eyes whereby the latter were drawn in toward the center of the tuber. This gives the tuber an irregular appearance very well illustrated in Plate II. Such variations appeared in varieties State of Maine, Early Ohio, Endurance and Seedling No. 843. In each case they were permanent.

Change in Habit of Growth of Tubers.

A peculiar change appeared in the variety called Pennsylvania, which is illustrated in Plate III. It consists of a prolongation of the so-called bud end of the tuber into a stem which bears another tuber at its extremity. On occasional tubers in other varieties this phenomenon has occurred, but only in this case has it been permanent and characteristic of the variety, although these abnormal tubers have been planted a number of times. I have heard the matter mentioned by old potato growers, not as a bud variation but as an indication that the variety has become reduced in vigor. The plants produced from these abnormal tubers in the Pennsylvania variety were all abnormal. Of course some tubers were formed in the regular manner on each plant, but each plant had at least one tuber showing the aberrant form. We have no evidence concerning the inheritance of this last variation in sexual reproduction, but the changes affecting the other three characters are changes from dominant to recessive. This does not prove that the reverse changes never occur, but if no such changes have been recorded by all of the numerous observers of this widely grown crop, they must be rare indeed.

Attempts to Produce Variations through Grafts.

A brief account of some experiments with grafted tubers is given here because it is thought that they have some interest in connection with asexual variation, even though the results are negative. The material used was taken from fourteen different varieties, giving several chances to find varieties that would graft upon each other. Five varieties with white skin were utilized, *viz.,* Early Carman, Moneymaker, Thorburn (white), Snowball and Clyde; in addition, Prizetaker, a variety with a white skin but with light pink sprouts, was included. The red-skinned varieties in use were Early Sunrise, Old Hemlock and Stray Beauty; while Sturtevant, Venezuela and S. Commersonii violet (Labergerie) comprised the purple-skinned varieties.

Over one hundred attempts were made to make grafts between buds of colored and white tubers. In each case we endeavored to cut the bud in half at its apical point and to graft half buds from colored and colorless tubers. Various methods of technique were tried, winding with soft twine being about as successful as the use of grafting wax. The tubers were placed where they could grow in the laboratory, and were carefully examined each day. In every grafted sprout the growing tissue of either one or the other of the two plants gained the ascendancy and in three or four days the upper point of the other sprout was left far behind. This portion of the sprout, while it did not dry up and die, did not increase in size, and was soon almost surrounded by the growing tissue of the other part of the sprout. It was quite evident that the new plant was always formed from the growing tissue of *one* variety.

The second experiment consisted in inserting a bud cut from a tuber of a colorless variety, with a brass cork borer, into a hole of the same size in a colored tuber, or *vice versa*. The other buds of the tuber were cut away and their places filled with grafting wax. Sixty of these various combinations were made and planted. When the young stems were two or three inches above the ground they were all dug up and examined. Seventy-five per cent. of the tubers had developed adventitious buds from the surrounding tuber. These sprouts were removed and the tubers replanted.

Twenty-three plants produced tubers, but absolutely no influence of the stock could be seen. It seems reasonable to suppose that a migrating character like color, when so placed that it could be utilized by the young stem, would be carried into the growing tissue and again appear in the progeny, but this was not the case. There appeared to be no influence on the scion either as a direct addition, or as a loss of a color character, or as a separate variation produced by influence of the stock. From this experience we suspect that many of the so-called graft-hybrids in the potato were merely plants exactly like the stock, produced from it by unnoted adventitious buds.

General Conclusions.

The investigations reported in the foregoing pages suggest very forcibly that the behavior of variations reproduced by budding is in many ways essentially like that of variations coming from seed. The inherited variations that were found have all but one concerned characters that mendelize in sexual reproduction, and as there is no evidence upon the inheritance of the one exception, it may be left out of consideration. Five cases of loss of color, two changes from long to round shape and four changes from shallow to deep eyes have come under our observation, all of which are common differences between seedlings out of a selfed fruit from our much crossed commercial varieties. If the causes and mechanism of production of both sexual and asexual variations are not similar or identical, it is very peculiar that no distinction between the two classes has been found. It is true that all of the asexual variations have been losses of characters, while in sexual reproduction the formation of new characters occurs. If no progressive variations occur in asexual reproduction, it must mean that there is a mechanical drawback to their production that sexual reproduction does not share; but the fact that they have not come under our observation simply proves them to be much rarer than the retrogressive variations. The last explanation parallels sexual variation, where retrogressive changes are very much more common than progressive changes, yet it seems important to call attention to a possible difference in the mechanical origin of sexual and asexual variations.

Any discussion regarding the mechanism of the production of variations is of course pure speculation, yet one cannot help feeling that the production asexually of changes that mendelize, throws some light on the subject. It follows that segregation may occur in the somatic cells, and that by division of one of these cells a line of descendants originates lacking a Mendelian character that is present in the parent cell. This fact does not develop anything new in regard to the relative importance of the chromosomes and the cytoplasm as bearers of hereditary characters, but it certainly seems to show that Mendelian segregation *is not limited to the reduction division in the maturation of sexual cells.*

Considering the amount of material under observation, the occurrence of twelve inherited variations is an unexpectedly high rate of frequency. Of course the careful scrutiny of the crop in every hill would partially account for a higher frequency than is observed by commercial growers. It might also be accounted for by the large number of colored varieties in our collection, as colored varieties form a very small proportion of the crop in the United States. Compared with an anemophilous crop like maize the per cent. of varying plants is very small, but compared with a close-fertilized crop like tobacco the difference in numbers, while still considerably less, is not remarkable. But should unobserved changes have occurred in ten times the quantity observed, there is no reason to recommend asexual selection as a commercial means of actual improvement. No changes that are of commercial value have yet been found, and, if they occur, they are rare indeed. The commercial grower would be swamped in trying out the numerous fluctuating variations that are not inherited, with scant possibility of ever finding a favorable inherited variation. Mature tubers free from disease should always be planted, but this precaution is made simply to give the young plants a normal start in life, and does not change their hereditary characters.

The classification of all of the permanent bud variations as losses of characters shows the investigation of the possible inheritance of fluctuations in composition in a new light. Since the variety used in the investigation was recessive in all the characters whose behavior in sexual reproduction is known, less probability exists that an inherited change might take place that would obscure the results on the class of variations immediately concerned. This being the case, we may feel some confidence in a conclusion that fluctuations (variations due to surrounding conditions) are not inherited. Furthermore, there is little doubt but that the cases of so-called disease resistance should be classed as noninherited fluctuations due to various physiological causes.

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TABLE IV.

CORRELATION BETWEEN PER CENT. DRY MATTER AND WEIGHT OF TUBERS IN GRAMS, CROP OF 1906.

	Per cent. Dry Matter										
Weight in Grams	14	15	16	17	18	19	20	21	22	23	
85						1				1	2
105	2	2	1	5	4	4	6	1			25
125		1		5	11	10	6	4	3		40
145	1	1	4	6	6	5	3	2			28
165		1	3	3	3		1				11
185			1	1	1	1					4
205			1	1							2
225	1			1					1		3
245											0
265					1						1
285	1										1
	5	5	10	22	26	21	16	7	4	1	117

Per cent. Dry Matter.

A. = 18.15 ± .118
S. D. = 1.90 ± .083
C. V. = 10.47 ± .463

Weight in Grams.

A. = 137 ± 2.06
S. D. = 32.97 ± 1.453
C. V. = 24.07 ± 1.21

Coef. Cor. —.275 ± .058

TABLE V.

CORRELATION BETWEEN WEIGHT IN GRAMS AND PER CENT. NITROGENOUS MATTERS, FRESH BASIS, CROP OF 1906.

	Weight in Grams.											
Nitrogenous Matters, fresh basis.	85	105	125	145	165	185	205	225	245	265	285	
1.3			1									1
1.5		1	1	2	1		1					6
1.7	1	4	17	7	5	2		1				37
1.9	1	5	10	9	4	2	1	1		1		34
2.1		10	6	4								20
2.3		5	4	4	1			1			1	16
2.5			1	2								3
	2	25	40	28	11	4	2	3	0	1	1	117

Weight in Grams.

A. = 137 ± 2.06
S. D. = 32.97 ± 1.453
C. V. = 24.07 ± 1.21

Nitrogenous Matters,
fresh basis.

A. = 1.915 ± .015
S. D. = .248 ± .011
C. V. = 12.95 ± .580

Coef. Cor. = —.056 ± .062

TABLE VI.

Correlation between Weight in Grams and per cent. Nitrogenous Matters, dry Basis, Crop of 1906.

Weight in Grams.

Nitrogenous Matters, dry basis.	85	105	125	145	165	185	205	225	245	265	285	
7	1		2									3
8		4	3	1	1							9
9		1	10	5	1	1	1	1				20
10	1	6	14	8	5	2				1		37
11		2	3	5	1			1				12
12		3	4		2	1	1					11
13		6	2	3								11
14			1	3	1							5
15		2	1	2							1	6
16		1		1				1				3
	2	25	40	28	11	4	2	3	0	1	1	117

Weight in Grams.

A. = 137 ± 2.06
S. D. = 32.97 ± 1.453
C. V. = 24.07 ± 1.21

Nitrogenous Matters, dry basis.

A. = 10.75 ± .129
S. D. = 2.08 ± .091
C. V. = 19.35 ± .740

Coef. Cor. = .121 ± .061

TABLE VII.

Correlation between per cent. Dry Matter and per cent. Nitrogenous Matters, fresh Basis, Crop of 1906.

Dry Matter.

Nitrogenous Matters, fresh basis.	14	15	16	17	18	19	20	21	22	23	
1.3						1					1
1.5			1	1	1	2			1		6
1.7				12	10	5	6	2	1	1	37
1.9	1	1	3	1	8	7	7	5	1		34
2.1	1	1	2	6	2	4	3		1		20
2.3	3	3	2	2	4	2					16
2.5			2		1						3
	5	5	10	22	26	21	16	7	4	1	117

Per cent. Dry Matter.

A. = 18.15 ± .118
S. D. = 1.90 ± .083
C. V. = 10.47 ± .463

Per cent. Nitrogenous Matters, fresh basis.

A. = 1.915 ± .015
S. D. = .248 ± .011
C. V. = 12.95 ± .580

Coef. Cor. —.346 ± .055

TABLE VIII.

CORRELATION BETWEEN PER CENT. DRY MATTER AND PER CENT. NITROGENOUS MATTERS, DRY BASIS, CROP OF 1906.

Nitrogenous Matters dry basis. \ Dry Matter.	14	15	16	17	18	19	20	21	22	23	
7						1			1	1	3
8					1	3	2	2	1		9
9			1	1	3	4	5	4	2		20
10				10	11	7	8	1			37
11			1	2	4	4	1				12
12		1	2	3	3	2					11
13		1	2	5	3						11
14	1		2	1	1						5
15	2	2	2								6
16	2	1									3
	5	5	10	22	26	21	16	7	4	1	117

Per cent. Dry Matter.

A. = 18.15 ± .118
S. D. = 1.90 ± .083
C. V. = 10.47 ± .463

Per cent. Nitrogenous Matters, dry basis.

A. = 10.75 ± .129
S. D. = 2.08 ± .091
C. V. = 19.35 ± .740

Coef. Cor. –.758 ± .026

TABLE IX.

CORRELATION IN PER CENT. NITROGENOUS MATTERS, DRY BASIS, BETWEEN MOTHERS SELECTED FOR HIGH NITROGEN, AND DAUGHTERS CROP OF 1907.

Per cent. Daughters. \ Per cent. Mothers.	12	13	14	15	16	
8			2			2
9			4	5		9
10		1	9	3	1	14
11			3	7	4	14
12		2	6	7	4	19
13	4	3	7	5	2	21
14	3	2	1	1		7
15	5	3	1	1		10
16		2	1	2		5
17		2		2	1	5
18		2	1			3
19		1				1
20		2				2
21						0
22		1				1
	12	21	35	33	12	113

Per cent. Nitrogenous Matters, dry basis, Mothers.

A. = 14.11 ± .073
S. D. = 1.147 ± .051
C. V. = 8.13 ± .365

Per cent. Nitrogenous Matters, dry basis, Daughters.

A. = 12.74 ± .171
S. D. = 2.698 ± .121
C. V. = 21.18 ± .992

Coef. Cor. – .387 ± .054

TABLE X.

Correlation in per cent. Nitrogenous Matters, dry Basis, between Mothers selected for Low Nitrogen, and Daughters Crop of 1907.

	Per cent. Mothers.			
Per cent. Daughters.	7	8	9	
7			1	1
8	1		1	2
9	1		3	4
10	1	3	2	6
11	3	8	6	17
12	5	12	5	22
13	1	5	4	10
14	3	5	6	14
15		8	2	10
16	2	2	1	5
17	1	2		3
18		1		1
19				0
20	1	2		3
	19	48	31	98

Per cent Nitrogenous Matters, dry basis, Mothers.

A. = 8.122 ± .015
S. D. = .222 ± .011
C. V. = 2.733 ± .131

Per cent. Nitrogenous Matters, dry basis, Daughters.

A. = 12.81 ± .170
S. D. = 2.493 ± .120
C. V. = 19.46 ± .972

Coef. Cor. −.510 ± .051

In the tables on the next page note the wide difference between the selected mother tubers. There are two classes vacant between the highest of the low nitrogen mother tubers and the lowest of the high nitrogen tubers, yet the progeny of each selection show frequency distributions so nearly the same that they might be interchanged. There is evidently no inheritance of these fluctuations.

TABLE XI.

FREQUENCY DISTRIBUTION OF NITROGENOUS MATTERS, FRESH BASIS, IN HIGH AND IN LOW NITROGEN PLOTS OF 1907, COMPARED WITH WEIGHTED MOTHERS.

CLASSES IN PER CENT.	1.3	1.5	1.7	1.9	2.1	2.3	2.5	2.7	2.9	3.1	3.3	3.5	3.7	3.9	4.1	A.	S. D.	C. V.
Daughters High Nit.			1	14	17	14	25	14	12	10	4		1		1	2.51 ± .028	.437 ± .020	17.41 ± .800
Weighted Mothers High Nit.						78	35									2.23		
Weighted Mothers Low Nit.	10	56	32													1.55		
Daughters Low Nit.		1	3	4	10	18	27	16	6	8	3	2				2.51 ± .027	.392 ± .019	15.62 ± .771

TABLE XII.

FREQUENCY DISTRIBUTION OF NITROGENOUS MATTERS, DRY BASIS, IN HIGH AND IN LOW NITROGEN PLOTS OF 1907, COMPARED WITH WEIGHTED MOTHERS.

CLASSES IN PER CENT.	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	A.	S. D.	C. V.
Daughters High Nit.		2	9	14	14	19	21	7	10	5	5	3	1	2	0	1	12.74 ± .171	2.698 ± .121	21.18 ± .992
Weighted Mothers High Nit.						12	21	35	33	12							14.11 ± .073	1.147 ± .051	8.13 ± .365
Weighted Mothers Low Nit.	19	48	31														8.12 ± .015	.222 ± .011	2.73 ± .131
Daughters Low Nit.	1	2	4	6	17	22	10	14	10	5	3	1	0	1			12.81 ± .170	2.493 ± .120	19.46 ± .972

TABLE XIII.

Correlation in per cent. Dry Matter between Mothers selected for High Nitrogen, and Daughters Crop of 1907.

Per cent.
Mothers.

Per cent. Daughters.	15	16	17	18	19	
16	1		1	1		3
17		1	2	3		6
18		3		6	1	10
19	7	6	1	9	3	26
20	8	7	3	4	7	29
21	2	10	1	3		16
22	1	7	2	4		14
23	1	2	1	2	1	7
24	1					1
25			1			1
	21	36	12	32	12	113

Per cent. Dry Matter,
Mothers.

A. = 16.81 ± .084
S. D. = 1.316 ± .059
C. V. = 7.829 ± .351

Per cent. Dry Matter,
Daughters.

A. = 19.98 ± .111
S. D. = 1.745 ± .078
C. V. = 8.734 ± .395

Coef. Cor. = —.140 ± .062

TABLE XIV.

Correlation in per cent. Dry Matter between Mothers selected for Low Nitrogen, and Daughters Crop of 1907.

Per cent.
Mothers.

Per cent. Daughters.	16	17	18	19	20	22	
15					1		1
16			1	1	1	1	4
17			3	1	1		5
18			4	4	2	2	12
19	1		5	4	2	2	14
20	4	1	5	11	1	4	26
21	5	1	1	9	1		17
22	1	3		7			11
23	1	1		4			6
24				1			1
25							0
26		1					1
	12	7	19	42	9	9	98

Per cent. Dry Matter,
Mothers.

A. = 18.66 ± .105
S. D. = 1.545 + .074
C. V. = 8.28 ± .4.02

Per cent. Dry Matter,
Daughters.

A. = 19.93 ± .131
S. D. = 1.923 ± .093
C. V. = 9.649 ± .469

Coef. Cor. —.286 ± .063

TABLE XV.

Correlation in per cent. Dry Matter between Mothers, and Daughters Crop of 1907, Considering both High and Low Nitrogen Plots.

Per cent. Mothers.

Per cent. Daughters.	15	16	17	18	19	20	21	22	
15						1			1
16	1		1	2	1	1		1	7
17		1	2	6	1	1			11
18		3		10	5	2		2	22
19	7	7	1	14	7	2		2	40
20	8	11	4	9	18	1		4	55
21	2	15	2	4	9	1			33
22	1	8	5	4	7				25
23	1	3	2	2	5				13
24	1				1				2
25			1						1
26			1						1
	21	48	19	51	54	9	0	9	211

Per cent. Dry Matter, Mothers.	Per cent. Dry Matter, Daughters.
A. = 17.67 ± .079	A. = 19.96 ± .085
S. D. = 1.70 ± .056	S. D. = 1.83 ± .060
C. V. = 9.62 ± .316	C. V. = 9.17 ± .301

Coef. Cor. —.194 ± .045

TABLE XVI.

Correlation in per cent. Dry Matter between Mothers, and Daughters Crop of 1908, Considering both High and Low Nitrogen Plots.

Per cent. Mothers.

Per cent. Daughters.	17	18	19	20	21	
8			1			1
9	1		1		1	3
10	2	1	1	1		5
11	4	3	3	1	1	12
12	4		4	5	2	15
13		1	8	8	1	18
14	1		4	4	1	10
15		1		1		2
16		1				1
	12	7	22	20	6	67

Per cent. Dry Matter, Mothers.	Per cent. Dry Matter, Daughters.
A. = 19.01 ± .100	A. = 12.19 ± .129
S. D. = 1.21 ± .071	S. D. = 1.56 ± .091
C. V. = 6.37 ± .371	C. V. = 12.79 ± .757

Coef. Cor. .228 ± .078

TABLE XVII.

Frequency Distribution of Nitrogenous Matters, fresh Basis, in High and in Low Nitrogen Plots of 1908, Compared with Weighted Mothers.

Classes in per cent.	1.9	2.1	2.3	2.5	2.7	2.9	3.1	3.3	A.	S. D.	C. V.
Daughters High Nit.	2	10	9	8	3	1			2.32 ± .028	.238 ± .020	10.26 ± .860
Weighted Mothers High Nit.						6	16	11	3.13		
Weighted Mothers Low Nit.		26	7						2.14		
Daughters Low Nit.	2	3	6	10	8	3	1		2.48 ± .033	.280 ± .023	11.24 ± .942

TABLE XVIII.

Frequency Distribution of Nitrogenous Matters, dry Basis, in High and in Low Nitrogen Plots of 1908, Compared with Weighted Mothers.

Classes in per cent.	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25*	27†	33	A.	S. D.	C. V.
Daughters High Nit.							2	3	3	2	1	8	5		4		1	3	1	20.6 ± .459	3.91 ± 3.25	18.98 ± 1.63
Weighted Mothers H. Nit.							6	4	9	5	4	5								17.36		
Weighted Mothers L. Nit.	2	14	10	7																10.67		
Daughters Low Nit.				1		1	1	5	2	4	2	1	7	4	3	2				19.3 ± .358	3.05 ± .253	15.80 ± 1.35

* One class vacant and omitted.

† Five classes vacant and omitted.

TABLE XIX.

The Relation between Weight of Tuber and Yield in Selections for Low Nitrogen.

Weight Seed tuber 1906.	Yield 1907 (grams).	Hills planted 1908.	Weight seed tuber 1908.	Yield 1908 (grams).
136	644			
158	1472	53–56 41–44	77 109	380 75
203	1560	49–52 45–48	88 88	220 30
129	1522			
187	1741	57–60	105	354
151	1797	65–68	114	543
106	1665	69–72	118	100
114	1776	61–64 73–76	168 69	1635 423
103	1530	77–80	92	964
120	1149			

TABLE XX.

High Nitrogen Extremes of 1906 and their Progeny of 1907

	From Hill No.	Dry Matter.	Nit. Mat. fresh basis.	Nit. Mat. dry basis.	Weight, grams.
Mother tuber, No. 19		18.02	2.48	13.76	119
Daughter tuber	1	19.4	2.09	10.77	114
“	1	19.1	2.81	14.71	122
“	1	19.4	2.00	10.31	103
Daughter tuber	2	22.4	1.97	8.79	66
“	2	22.9	2.17	9.47	55
“	2	21.6	2.11	9.77	37
Daughter tuber	3	23.0	1.98	10.65	115
“	3	22.2	2.18	9.82	66
“	3	21.3	1.74	8.17	49
Daughter tuber	4	19.3	1.86	9.63	272
“	4	19.2	1.98	10.31	123
“	4	----	----	----	----

TABLE XX—*Continued.*

HIGH NITROGEN EXTREMES OF 1906 AND THEIR PROGENY OF 1907.

	From Hill No.	Dry Matter.	Nit. Mat. fresh basis.	Nit. Mat. dry basis.	Weight, grams.
Mother tuber, No. 33		16.40	2.34	14.27	165
Daughter tuber	5	21.3	2.49	11.69	94
“	5	21.8	2.52	11.56	74
“	5	21.5	1.99	9.25	59
Daughter tuber	6	17.6	2.20	12.50	90
“	6	19.8	2.52	12.72	59
“	6	19.8	2.53	12.77	49
Daughter tuber	7	20.5	2.43	11.85	70
“	7	20.0	1.97	9.85	53
“	7	21.7	2.14	9.86	41
Daughter tuber	8	21.6	2.77	12.82	49
“	8	22.8	2.92	12.80	43
“	8	21.5	2.20	10.23	30
Mother tuber, No. 35		14.75	2.36	16.00	145
Daughter tuber	9	19.4	2.17	11.18	60
“	9	21.0	2.64	12.57	60
“	9	23.8	2.94	12.35	29
Daughter tuber	10	19.3	2.11	10.93	219
“	10	19.0	2.49	13.10	138
“	10	23.0	2.69	11.70	68
Daughter tuber	11	19.1	2.16	11.31	150
“	11	19.8	2.30	11.61	95
“	11	15.8	2.66	16.83	58
Daughter tuber	12	18.8	1.95	10.37	149
“	12	19.2	2.32	12.08	109
“	12	20.1	2.24	11.14	81
Mother tuber, No. 49		17.98	2.37	13.18	118
Daughter tuber	13	18.4	3.69	20.05	37
“	13	18.8	4.11	21.86	21
“	13	21.3	2.52	11.83	17
Daughter tuber	14	Did not grow			
Daughter tuber	15	16.6	3.29	19.82	90
“ [1]	15	17.9	3.00	16.76	59
“	15	16.4	2.27	13.84	41
Daughter tuber	16	17.9	2.26	12.62	116
“	16	22.1	3.19	14.43	39
“	16	19.9	1.97	9.90	33

[1] Planted in hills 21, 22, 23, 24 (1908).

TABLE XX—*Continued.*

HIGH NITROGEN EXTREMES OF 1906 AND THEIR PROGENY OF 1907.

	From Hill Ho.	Dry Matter.	Nit Mat. fresh basis.	Nit. Mat. dry basis.	Weight, grams.
Mother tuber, No. 55		16.40	2.49	15.12	137
Daughter tuber	17	17.1	2.64	15.44	114
"	17	20.5	2.42	11.80	85
"	17	18.6	2.44	13.12	69
Daughter tuber	18	19.2	2.17	11.30	83
"	18	19.9	2.61	13.11	75
"	18	19.5	3.09	15.84	43
Daughter tuber	19	21.5	2.42	11.25	185
" [2]	19	17.6	2.91	16.53	114
"	19	21.6	2.11	9.77	59
Daughter tuber	20	21.2	2.44	11.51	75
"	20	20.9	2.70	12.92	67
"	20	21.6	2.03	9.39	43
Mother tuber, No. 57		16.13	2.42	15.00	146
Daughter tuber	21	21.5	1.94	9.02	111
"	21	22.8	2.27	9.95	82
"	21	18.7	2.91	15.56	48
Daughter tuber	22	20.1	2.33	11.59	97
"	22	19.4	2.23	11.49	76
"	22	21.5	2.46	11.44	59
Daughter tuber	23	18.9	2.45	12.96	190
"	23	21.2	1.96	9.24	116
"	23	22.5	1.97	8.75	69
Daughter tuber	24	20.6	2.29	11.11	85
"	24	18.5	2.51	13.56	80
"	24	22.1	1.99	9.00	45
Mother tuber, No. 107		16.83	2.38	14.14	138
Daughter tuber	25	22.5	2.45	10.89	120
"	25	24.7	1.92	7.77	93
"	25	22.8	2.06	9.03	63
Daughter tuber	26	16.0	2.53	15.81	113
"	26	21.8	2.71	12.43	79
"	26	20.2	2.79	13.81	55
Daughter tuber	27	19.8	2.48	12.52	104
"	27	17.4	2.28	13.10	62
"	27	21.0	2.19	10.43	52
Daughter tuber	28	19.4	2.35	12.11	110
"	28	17.5	3.21	18.34	49
"	28	20.5	2.70	13.17	38

[2] Planted in hills 33, 34, 35, 36 (1908).

TABLE XX—*Concluded.*

HIGH NITROGEN EXTREMES OF 1906 AND THEIR PROGENY OF 1907.

	From Hill No.	Dry Matter.	Nit. Mat. fresh basis.	Nit. Mat. dry basis.	Weight, grams.
Mother tuber, No. 138		18.81	2.36	12.50	106
Daughter tuber,[1]	29	20.5	3.10	15.12	68
“	29	20.3	3.11	15.32	60
“	29	22.7	3.04	13.39	60
Daughter tuber	30	19.8	2.81	14.19	63
“	30	18.7	2.51	13.42	62
“	30	20.5	2.62	12.78	61
Daughter tuber,[2]	31	18.7	2.87	15.35	94
“	31	19.9	2.93	14.72	67
“	31	20.2	3.02	14.95	54
Daughter tuber	32	19.4	2.46	12.68	75
“	32	18.3	2.56	13.99	70
“	32	20.1	2.82	14.03	56
Mother tuber, No. 144		15.35	2.36	15.37	110
Daughter tuber	33	20.0	2.40	12.00	90
“	33	----	----	----	----
“	33	----	----	----	----
Daughter tuber	34	20.2	2.44	12.08	99
“	34	21.2	2.56	12.07	98
“	34	----	----	----	----
Daughter tuber	35	20.1	2.39	11.89	120
“	35	20.4	2.18	10.68	88
“	35	22.1	2.52	11.40	69
Daughter tuber	36	19.7	2.07	10.50	118
“ [3]	36	19.3	3.19	16.53	79
“	36	19.9	2.56	12.86	76
Mother tuber, No. 155		17.74	2.28	12.85	113
Daughter tuber,[4]	37	18.8	3.30	17.55	74
“	37	18.1	3.00	16.57	69
“ [5]	37	21.1	3.17	15.02	67
Daughter tuber	38	18.6	2.76	14.84	114
“	38	20.1	2.65	13.18	76
“	38	17.9	2.62	14.64	54
Daughter tuber,[6]	39	17.2	3.14	18.25	99
“	39	19.4	2.31	11.90	91
“ [7]	39	18.2	2.97	16.32	72
Daughter tuber,[8]	40	16.9	3.29	19.47	89
“	40	19.7	2.56	12.99	78
“	40	20.2	3.19	15.79	58

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[1] Planted in hills 1, 2, 3, 4 (1908).

[2] Planted in hills 29, 30, 31, 32 (1908).

[3] Planted in hills 13, 14, 15, 16 (1908).

[4] Planted in hills 25, 26, 27, 28 (1908).

[5] Planted in hills 9, 10, 11, 12 (1908).

[6] Planted in hills 5, 6, 7, 8 (1908).

[7] Planted in hills 37, 38, 39, 40 (1908).

[8] Planted in hills 17, 18, 19, 20 (1908).

TABLE XXI.

LOW NITROGEN EXTREMES OF 1906 AND THEIR PROGENY OF 1907.

	From Hill No.	Dry Matter.	Nit. Mat. fresh basis.	Nit. Mat. dry basis.	Weight, grams.
Mother tuber, No. 4		17.32	1.53	8.83	136
Daughter tuber	41	21.88	2.30	10.51	60
"	41	----	----	----	----
"	41	----	----	----	----
Daughter tuber	42	25.81	2.20	8.52	70
"	42	21.06	2.68	12.73	55
"	42	22.52	1.79	7.95	53
Daughter tuber	43	22.41	2.42	10.80	44
"	43	21.63	3.10	14.33	39
"	43	----	----	----	----
Daughter tuber	44	19.56	2.68	13.70	111
"	44	----	----	----	----
"	44	----	----	----	----
Mother tuber, No. 10		18.11	1.45	8.01	158
Daughter tuber, 1	45	19.57	2.03	10.37	77
"	45	19.01	2.24	11.78	83
"	45	20.35	2.60	12.78	67
Daughter tuber	46	19.60	2.88	14.70	61
"	46	----	----	----	----
"	46	----	----	----	----
Daughter tuber, 2	47	19.08	1.87	9.80	109
"	47	20.08	2.45	12.20	109
"	47	16.51	2.43	14.72	118
Daughter tuber	48	Did not grow			
Mother tuber, No. 13		16.01	1.42	8.87	203
Daughter tuber	49	20.06	2.10	10.47	65
"	49	21.08	2.27	10.77	55
"	49	19.12	2.78	14.54	70
Daughter tuber	50	21.02	2.46	11.70	103
"	50	20.04	2.28	11.38	74
" 3	50	21.43	2.02	9.43	88
Daughter tuber	51	23.06	1.69	7.33	56
"	51	20.69	3.08	14.89	88
"	51	20.23	2.36	11.67	116
Daughter tuber	52	21.52	2.41	11.20	78
"	52	20.05	2.48	12.37	100
" 4	52	20.62	1.97	9.55	88

1. Planted in hills 53, 54, 55, 56 (1908).
2. Planted in hills 41, 42, 43, 44 (1908).
3. Planted in hills 49, 50, 51, 52 (1908).
4. Planted in hills 45, 46, 47, 48 (1908).

TABLE XXI—*Continued.*

LOW NITROGEN EXTREMES OF 1906 AND THEIR PROGENY OF 1907.

	From Hill No.	Dry Matter.	Nit. Mat. fresh basis.	Nit. Mat. dry basis.	Weight, grams.
Mother tuber, No. 52		18.72	1.30	6.95	129
Daughter tuber	53	20.01	2.30	11.50	78
“	53	19.98	2.73	13.66	120
“	53	----	----	----	----
Daughter tuber	54	17.74	2.57	14.49	312
“	54	18.94	1.87	9.87	206
“	54	----	----	----	----
Daughter tuber	55	20.86	2.36	11.31	76
“	55	21.24	2.37	11.16	50
“	55	15.94	2.57	16.12	68
Daughter tuber	56	19.87	2.37	11.92	93
“	56	20.85	1.79	8.58	48
“	56	18.18	2.43	13.36	45
Mother tuber, No. 101		18.30	1.63	8.91	187
Daughter tuber	57	16.75	2.15	12.83	215
“	57	18.79	2.66	14.15	96
“	57	20.53	1.94	9.45	66
Daughter tuber	58	18.33	2.22	12.11	147
“	58	17.34	2.46	14.18	117
“ [5]	58	18.28	2.06	11.27	105
Daughter tuber	59	17.97	2.28	12.68	105
“	59	17.94	2.48	13.82	99
“	59	15.80	2.53	16.01	88
Daughter tuber	60	18.91	2.54	13.43	97
“	60	19.71	2.41	12.22	92
“	60	18.56	2.60	14.01	83
Mother tuber, No. 132		19.09	1.52	7.96	151
Daughter tuber, [1]	61	19.81	2.14	10.80	114
“	61	19.81	2.39	12.06	127
“	61	20.18	2.31	11.44	100
Daughter tuber	62	19.91	2.47	12.40	111
“	62	20.10	2.38	11.84	73
“	62	20.73	3.44	16.60	65
Daughter tuber	63	21.71	2.60	11.97	93
“	63	22.52	2.45	10.88	84
“	63	23.86	2.94	12.32	85
Daughter tuber	64	20.79	2.91	14.00	129
“	64	18.90	3.04	16.08	74
“	64	----	----	----	----

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[5] Planted in hills 57, 58, 59, 60 (1908).

[1] Planted in hills 65, 66, 67, 68 (1908).

TABLE XXI—*Continued.*

Low Nitrogen Extremes of 1906 and their Progeny of 1907.

	From Hill No.	Dry Matter.	Nit. Mat. fresh basis	Nit. Mat. dry basis.	Weight, grams.
Mother tuber, No. 146		18.90	1.50	7.94	106
Daughter tuber	65	22.79	2.54	11.14	158
"	65	23.21	2.73	11.76	104
"	65	23.23	2.62	11.28	104
Daughter tuber	66	21.36	2.53	11.84	88
"	66	22.18	2.67	12.04	70
"	66	22.31	2.54	11.38	69
Daughter tuber	67	17.43	2.35	13.48	96
"	67	20.26	2.56	12.63	60
"	67	----	----	----	----
Daughter tuber, 2	68	18.55	2.14	11.53	118
"	68	21.43	2.66	12.41	68
"	68	----	----	----	----
Mother tuber, No. 148		19.85	1.61	8.11	114
Daughter tuber, 3	69	19.80	2.08	10.50	168
" 4	69	19.28	2.18	11.30	92
"	69	14.97	2.63	17.57	69
Daughter tuber	70	Did not grow			
Daughter tuber	71	21.28	3.26	15.32	95
"	71	15.60	3.15	20.19	72
"	71	19.19	2.66	13.86	60
Daughter tuber	72	18.08	2.49	13.77	173
"	72	16.84	2.51	14.90	124
"	72	18.46	2.80	15.17	83
Mother tuber, No. 153		19.26	1.61	8.36	103
Daughter tuber	73	21.53	2.71	12.58	99
"	73	18.81	3.15	16.74	91
"	73	20 57	2.33	11.33	73
Daughter tuber	74	20.01	2.90	14.49	88
"	74	21.63	2.78	12.85	78
"	74	19.57	2.85	14.56	85
Daughter tuber	75	21.86	3.20	14.64	46
"	75	21.21	3.29	15.51	44
"	75	----	----	----	----
Daughter tuber	76	17.85	2.76	15.46	87
"	76	17.58	3.53	20.08	87
"	76	22.46	3.04	13.53	58

2. Planted in hills 69, 70, 71, 72 (1908).
3. Planted in hills 61, 62, 63, 64 (1908).
4. Planted in hills 73, 74, 75, 76 (1908).

TABLE XXI—*Concluded.*

LOW NITROGEN EXTREMES OF 1906 AND THEIR PROGENY OF 1907.

	From Hill No.	Dry Matter.	Nit. Mat. fresh basis.	Nit. Mat. dry basis.	Weight, grams.
Mother tuber, No. 157		21.66	1.43	6.60	120
Daughter tuber	77	Did not grow			
Daughter tuber	78	19.65	2.44	12.41	157
" 5	78	18.79	2.21	11.76	92
"	78	18.84	2.95	15.66	91
Daughter tuber	79	19.95	2.74	13.73	141
"	79	19.89	1.59	7.99	63
"	79	19.63	2.43	12.38	58
Daughter tuber	80	17.83	3.09	17.33	75
"	80	18.28	2.26	12.36	75
"	80	16.28	3.33	20.45	57

5. Planted in hills 77, 78, 79, 80 (1908).

TABLE XXII.

HIGH EXTREMES FROM THE HIGH NITROGEN PLOT OF 1907 AND THEIR PROGENY OF 1908.

	From Hill No.	Dry Matter.	Nit. Mat. fresh basis.	Nit. Mat. dry basis.	Weight.
Mother tuber (from tuber No. 49 in 1906)	15*a*	16.60	3.29	19.82	90
Daughter tuber	22	11.99	2.44	20.35	90
“	22	12.04	1.88	15.61	58
“	23	10.77	2.16	20.05	102
“	24	10.69	2.28	21.33	86
“	24	8.96	2.06	22.99	84
Mother tuber (from tuber No. 55 in 1906)	19*b*	17.60	2.91	16.53	114
Daughter tubers all too small to analyze.					
Mother tuber (from tuber No. 138 in 1906)	29*a*	20.50	3.10	15.12	68
Daughter tubers all too small to analyze.					
Mother tuber (from tuber No. 138 in 1906)	31*a*	18.70	2.87	15.35	94
Daughter tuber	29	9.24	2.51	27.16	62
“	32	12.63	2.66	21.06	50
Mother tuber (from tuber No. 144 in 1906)	36*b*	19.30	3.19	16.53	79
Daughter tuber	13	9.94	2.28	22.93	126
“	13	11.01	2.14	19.43	74
“	14	13.60	2.05	15.07	92
“	14	11.49	2.41	20.97	78
“	14	12.74	2.29	17.97	61
“	15	14.24	2.11	14.81	102
“	15	12.02	2.45	20.38	72
“	16	12.64	2.11	16.69	84
“	16	14.03	2.21	15.75	50
Mother tuber (from tuber No. 155 in 1906)	37*a*	18.80	3.30	17.55	74
Daughter tuber	25	10.57	2.26	21.38	125
“	27	8.47	2.79	32.94	61
Mother tuber (from tuber No. 155 in 1906)	37*c*	21.10	3.17	15.02	67
Daughter tuber	9	12.07	2.18	18.06	71
“	10	11.10	2.50	22.52	60
“	11	9.37	2.56	27.32	56
“	12	11.88	2.05	17.25	52

TABLE XXII –*Concluded.*

HIGH EXTREMES FROM THE HIGH NITROGEN PLOT OF 1907 AND THEIR PROGENY OF 1908.

	From Hill No.	Dry Matter.	Nit. Mat. fresh basis.	Nit. Mat. dry basis.	Weight.
Mother tuber (from tuber No. 155 in 1906)-------	39*a*	17.20	3.14	18.25	99
Daughter tuber---------	5	10.19	2.35	23.06	72
“ ---------	6	10.29	2.79	27.11	85
“ ---------	8	10.88	1.87	17.18	80
Mother tuber (from tuber No. 155 in 1906)-------	39*c*	18.20	2.97	16.32	72
Daughter tuber---------	37	10.67	2.12	19.86	66
“ ---------	38	10.48	2.11	20.13	62
“ ---------	39	11.05	2.30	20.81	60
“ ---------	40	13.12	2.58	19.66	50
Mother tuber (from tuber No. 155 in 1906)-------	40*a*	16.90	3.29	19.47	89
Daughter tuber---------	17	11.98	2.44	20.36	69
“ ---------	17	13.88	2.27	16.35	50
“ ---------	20	11.24	2.29	20.37	82
“ ---------	20	11.59	2.88	24.85	51

TABLE XXIII.

LOW EXTREMES FROM THE LOW NITROGEN PLOT OF 1907 AND THEIR PROGENY OF 1908.

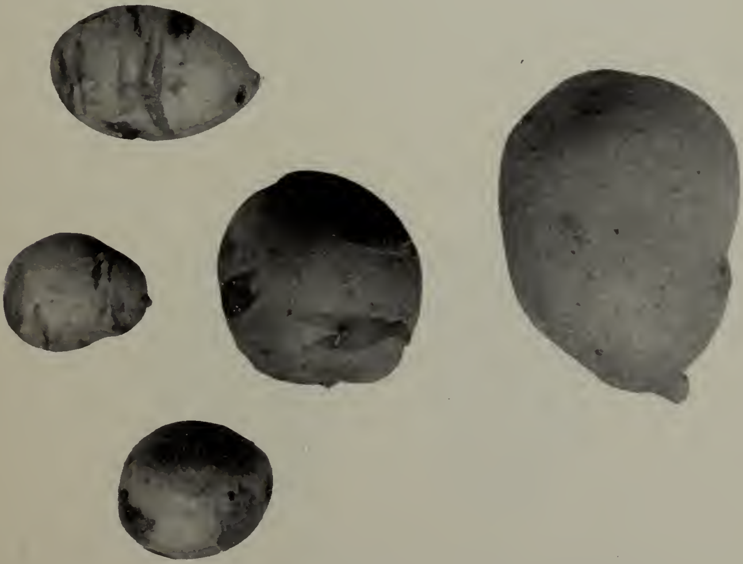
	From Hill No.	Dry Matter.	Nit. Mat. fresh basis.	Nit. Mat. dry basis.	Weight.
Mother tuber (from tuber No. 10 in 1906)--------	45*a*	19.57	2.03	10.37	77
Daughter tuber---------	53	10.39	2.38	22.91	70
“ ---------	56	12.51	2.81	22.46	88
Mother tuber (from tuber No. 10 in 1906)--------	47*a*	19.08	1.87	9.80	109
Daughter tubers all too small to analyze.					
Mother tuber (from tuber No. 13 in 1906)--------	50*c*	21.43	2.02	9.43	88
Daughter tuber---------	51	13.61	2.36	17.34	64
“ ---------	52	12.87	3.13	24.32	120
Mother tuber (from tuber No. 13 in 1906)--------	52*c*	20.62	1.97	9.55	88
Daughter tubers all too small to analyze.					
Mother tuber (from tuber No. 101 in 1906)-------	58*c*	18.28	2.06	11.27	105
Daughter tuber---------	57	14.97	2.43	16.23	121
“ ---------	58	16.36	2.70	16.50	56
“ ---------	60	11.48	2.74	23.87	116
Mother tuber (from tuber 132 in 1906)-----------	61*a*	19.81	2.14	10.80	114
Daughter tuber---------	65	12.68	2.61	20.58	69
“ ---------	66	10.80	2.41	22.31	153
“ ---------	67	12.76	2.66	20.85	113
“ ---------	67	13.06	2.77	21.21	71
“ ---------	68	11.60	2.58	22.24	85
“ ---------	68	11.58	2.57	22.19	57
Mother tuber (from tuber No. 146 in 1906)-------	68*a*	18.55	2.14	11.53	118
Daughter tubers all too small to analyze.					

TABLE XXIII—*Concluded.*

LOW EXTREMES FROM THE LOW NITROGEN PLOT OF 1907 AND THEIR PROGENY OF 1908.

	From Hill No.	Dry Matter.	Nit. Mat. fresh basis.	Nit. Mat. dry basis.	Weight.
Mother tuber (from tuber No. 148 in 1906)-------	69*a*	19.80	2.08	10.50	168
Daughter tuber---------	61	13.85	2.49	17.97	77
" ---------	61	13.39	2.33	17.40	59
" ---------	61	12.47	2.92	23.41	60
" ---------	62	13.10	2.70	20.61	101
" ---------	62	14.18	2.95	20.80	81
" ---------	62	12.77	2.32	18.17	66
" ---------	63	12.12	2.43	20.05	167
" ---------	63	11.85	2.69	22.70	126
" ---------	63	13.21	2.01	15.21	69
" ---------	64	14.11	2.03	14.38	71
" ---------	64	13.76	2.41	17.51	76
" ---------	64	15.10	1.82	12.05	69
Mother tuber (from tuber No. 148 in 1906)-------	69*b*	19.28	2.18	11.30	92
Daughter tuber---------	74	13.18	2.37	17.98	54
Mother tuber (from tuber No. 157 in 1906)-------	78*b*	18.79	2.21	11.76	92
Daughter tuber---------	77	13.04	2.49	19.10	70
" ---------	77	13.13	2.47	18.81	60
" ---------	79	13.15	2.08	15.81	63
" ---------	79	11.93	2.55	21.37	102
" ---------	79	12.32	1.93	15.66	84
" ---------	80	12.61	2.62	20.78	89
" ---------	80	12.42	----	----	109
" ---------	80	13.78	2.25	16.32	103

a. Rector of Woodstock. The only instance known where bud variation has produced color. It is unknown whether mother variety possessed purple sprouts.



b. La Bretonne. The white-skinned variation at the right was constant.

BUD VARIATION IN COLOR.

PLATE II.

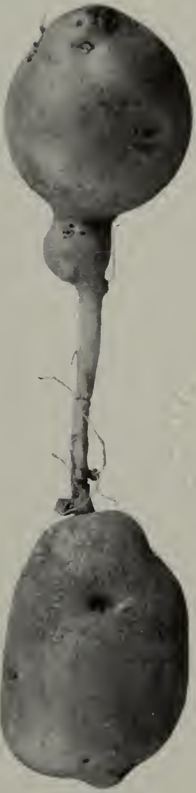
a. Early Sunrise. White variation at the right was constant.

b. Bole Zoegling. White variation at the right was constant. The apparent color in the reproduction is due to the corky layer having been rubbed off in places.

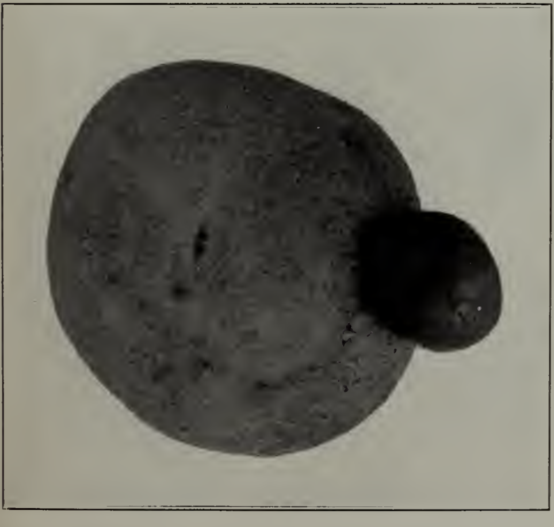
c. Early Ohio. Deep-eyed variation at the right was constant.

BUD VARIATION IN COLOR AND DEPTH OF EYE.

a. Aradaras. Apparent pink variation at right. Not constant.



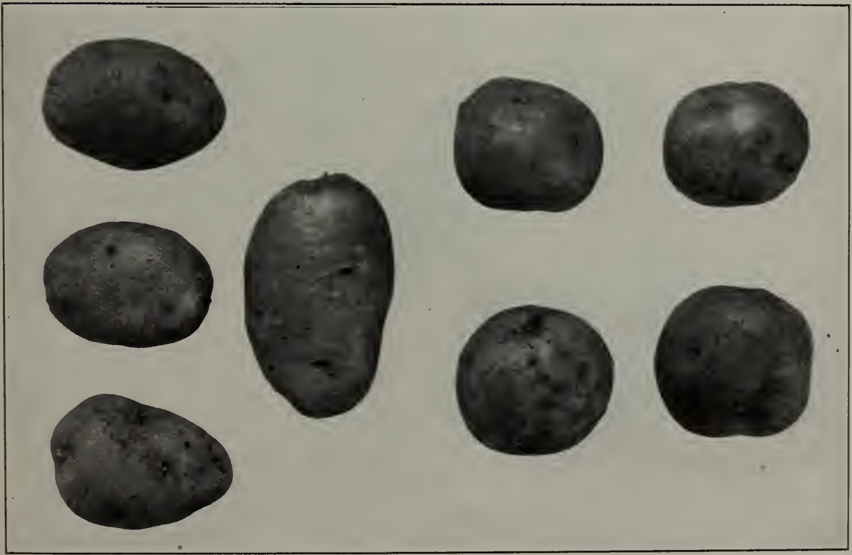
c. Peculiar variation in habit of tuber formation. Constant.



b. Prizetaker. Apparent pink variation at bud end. Not constant.

ODD BUD VARIATIONS.

a. Early Ohio. Round variation at the right was constant.



b. Silver Hill. Round variation at the right was constant.

BUD VARIATIONS IN SHAPE.

a. Normal State of Maine.

b. An elongated variation of above. It was evidently due to some attending condition during growth, for it did not reproduce itself.

BUD VARIATIONS NOT INHERITED.

GENETICAL STUDIES ON ŒNOTHERA. II

Some Hybrids of *Œnothera biennis* and *O. grandiflora* that resemble *O. Lamarckiana*

DR. BRADLEY MOORE DAVIS

NEW YORK
1911

[Reprinted, without change of paging, from the American Naturalist, 1911.]

[Reprinted from THE AMERICAN NATURALIST, Vol. XLV., April, 1911.]

GENETICAL STUDIES ON ŒNOTHERA. II

SOME HYBRIDS OF *Œnothera biennis* AND *O. grandiflora* THAT RESEMBLE *O. Lamarckiana*[1]

DR. BRADLEY MOORE DAVIS

THE status of *Œnothera Lamarckiana* is a matter which must be given serious consideration in any attempt to judge the value of De Vries's mutation theory, for the reason that the behavior of this form in throwing off marked variants (mutants) from the type has been regarded by De Vries as direct experimental proof of mutation. Indeed, the theory of De Vries may fairly be said to rest chiefly upon the behavior of this interesting plant, the account of which forms so large a part of his work, "Die Mutationstheorie," 1901–1903.

Aside from his claim of direct proof of the origin of mutations from *Œnothera Lamarckiana,* De Vries offers a considerable body of indirect evidence of the sort presented in Darwin's "Variation of Animals and Plants under Domestication," and in that extensive and very carefully sifted account of Bateson, "Materials for the Study of Variations," 1894. However, much of this indirect evidence of De Vries deals with the origin of "sports" from domesticated forms or with the origin

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[1] Contribution from the Laboratory of Genetics, Bussey Institution of Harvard University No. 7. An investigation conducted with aid from the Elizabeth Thompson Science Fund for which the author desires to express his indebtedness.

of new forms under conditions that are not typical of those of nature in the wild. For these reasons such evidence could never appeal with so much force as would direct experimental proof that a wild species is in the habit of producing suddenly new types sufficiently distinct from the parent form to rank as new species or even as strongly marked varieties.

In "Die Mutationstheorie" of De Vries the behavior of *Œnothera Lamarckiana* in giving rise to the so-called mutants is presented as evidence that new species have come into existence without intermediate steps from a form which is assumed to be typical of a species in nature. *Œnothera Lamarckiana* is made to bear the weight of an elaborate hypothesis, treating of fundamental problems, very much as the apex might be made to bear the weight of an inverted pyramid. As the equilibrium of the inverted pyramid depends upon the stability of its apex, so the value for the mutation theory of the evidence from the behavior of *Lamarckiana* must rest with the status of this plant as a form truly representative of a typical species.

De Vries from the beginning took it for granted that *Œnothera Lamarckiana* was a native American species introduced into Europe, an assumption that was perhaps not unnatural, although dangerous when the responsibility of direct proof of the origin of species by mutation was laid upon its behavior. As far as the writer is aware, *O. Lamarckiana,* as a wild American species, is unknown. No American locality can be cited where it may be found as a clear component of the native flora. There are certain records of its presence under conditions that indicate the possibility of its being sometimes a garden escape, and there is some herbarium material, referred to *Lamarckiana,* which, however, has not been tested by culture and was collected at times when the importance of the most critical judgment in identification was not appreciated. It cannot be said that American botanists are not alive to the importance of the status of

Lamarckiana, for it is well known that a certain group would follow with persistence any clue that might give evidence of its being or having been an American native species.

Critics of the evidence for De Vries's mutation theory have been aware of the point of weakness that lay in the uncertain status of *Œnothera Lamarckiana* and the suggestions of Bateson and Saunders ('02, p. 153), East ('07, p. 34), Boulenger ('07, p. 363), Leclerc du Sablon ('10, p. 266), Tower ('10, p. 322), and others have probably occurred to many, namely, that this plant is of hybrid origin and that the appearance of its "mutations" is due to the continued splitting off of variants after the manner of hybrids. This view is held by a number of American botanists with whom the writer is acquainted and represents the attitude of those who are sceptical of the importance of mutation as a factor of organic evolution in nature. If *Lamarckiana* is of hybrid origin it should be possible to obtain evidence of its probable parentage, and the present paper offers a hypothesis with a considerable body of evidence in its favor. After the evidence has been presented the hypothesis will be discussed in the concluding section entitled "The Possible Origin of *Œnothera Lamarckiana* as a Hybrid of *O. biennis* and *O. grandiflora.*"

None of the hybrids of *biennis* and *grandiflora* described in the following pages are identical with *Lamarckiana.* There are important differences, chiefly of foliage and stem markings, which distinguish the hybrids at a glance, but on the other hand these characters in taxonomy would be considered of minor importance and the hybrids, if their origin were unknown, could not be placed elsewhere than next to *Lamarckiana.* Furthermore, these differences are of a sort that are likely to be much less apparent when the results of crosses made this summer (1910) between certain types recently differentiated become known in succeeding cultures. In an investigation of this character the results, as every student of genetics knows, come slowly, and the writer feels no necessity of offering an apology in publishing preliminary data, since they are based on three seasons' study, even though he hopes to present more conclusive evidence in the future.

The cultures of the past season (1910) were grown partly at the Bussey Institution and partly in the Botanic Garden of Harvard University, where facilities were offered for which the writer is deeply indebted.

METHODS

To break the biennial habit and obtain flowering plants in one season it is only necessary to sow seeds in the hot house during the winter, where rosettes will develop, which may be set out in the open in the early spring. The cultures of 1910 were sown early in January and had developed large rosettes by May 5, when the plants were transferred to the gardens. It is best to sow the seeds thinly in large seed pans (with, of course, sterilized soil) from which each individual seedling may be potted.

Cross pollination was always performed on flowers from which the unopened anthers had been removed. The best results are generally obtained when a castrated flower is left bagged for twelve to twenty-four hours before the pollen is applied, in order that the stigma may have time to mature fully which will be evident from the moist exudation on its surface. Strong manila bags tied firmly over the pollinated flowers have proved more satisfactory than special bags of paraffin paper, which appear to hold the flowers in an atmosphere too moist for the best results. It is my practise to dip the forceps in a bottle of alcohol before each transfer of pollen and also to rinse the hands in alcohol. The pollen of *Œnothera* is so sticky that under ordinary conditions there is no danger from wind blown pollen, and by following the practise outlined above there is little or no probability of impure pollination.

In the future my plan will be to sow the seed capsule by capsule, which is the safest method to obtain quantitative results. In the past I have sown from bulk collections of seed and my studies so far must be regarded as primarily qualitative in character. This practise was followed under the impression that probably only a small proportion of the seed from a cross, apparently as extreme as that between *biennis* and *grandiflora,* would germinate. The results, however, have shown that the seed of this cross is fertile to a very high degree. In consequence my cultures of this season gave three or four times more seedlings than it was practicable to bring to maturity. A process of selection became at once necessary, which was followed with the end in view of obtaining a variety of types, but it soon became evident, with the later development of the cultures, that it is impossible to select with accuracy among the young seedlings. Thus plants which showed certain tendencies as young rosettes or seedlings of five to nine leaves often changed very materially in later growth. For these reasons my cultures of this year even as qualitative studies are undoubtedly not fully representative.

Œnothera biennis L.

My first crosses between *biennis* and *grandiflora* were made at Woods Hole, Mass., in 1908. This locality was carefully searched for rosettes of *biennis* with broad leaves and two plants of this character were transplanted from waste ground to the garden. The rosettes were similar and at maturity the plants proved to be the same form of *biennis* and were practically indistinguishable. These two plants were the starting points of two strains of *biennis,* designated A and B, which have been cultivated through two and three generations, respectively, and have proved constant.

It seems quite certain that under the name *Œnothera biennis* is included a number of races with well-marked peculiarities. These races are probably very pure, for the reason that close pollination is certainly usual, if

FIG. 1. Mature rosette of *Œnothera biennis*, B (10.21a).

not universal, among the forms of the species. The lower portions of the stigma lobes in the bud lie below the tips of the anthers which discharge their pollen before the bud opens. As a result the stigma is not only well pollinated in the bud, but cytological studies on my strains A and B have shown that the pollen tubes reach the embryo sacs before there is any opportunity for cross pollination. Hybridization in nature could hardly occur in these forms unless their own pollen should be insufficient for the number of ovules in the ovary or was much slower than foreign pollen in effecting fertilization; alternatives that are very unlikely. These conditions, similar physiologically to those of cleistogamous flowers, are present in all of the forms of *biennis* known to the writer, and have been noted by De Vries for the European types. As a result a strain once established is certain to remain pure throughout at least the great mass of its seeds.

The chief characteristics of the *biennis* strains A and

FIG. 2. Mature plant of *Œnothera biennis*, B (10.21a).

B, employed in the hybrid studies of the present paper, when under good cultivation are as follows:

1. *Rosettes.*—The mature rosettes (Fig. 1), from 3 to 4 dm. broad, have about 40 closely clustered leaves, spatulate, irregularly toothed at their base, and green except for occasional reddish spots. The rosettes are persistent and conspicuous during the early development of the main stem.

2. *Mature Plants.*—The mature plants, 1–1.5 m. high, have a spreading habit (Fig. 2) with long side branches. Stems chiefly green (brownish below), the papillate glands at the base of long hairs also green. Basal leaves on the main stem narrowly elliptical, about 16 cm. long (Fig. 3), leaves above lanceolate.

3. *Inflorescence.*—Bracts lanceolate, less than half the length of the buds (Fig. 4).

4. *Buds.*—About 5.5 cm. long, the cone 4-angled (Fig. 4). Sepals clear green, their tips not markedly attenuate.

5. *Flowers.*—Small (Figs. 3 and 5). Petals about 1.3 cm. long. Lower halves of stigma lobes (when closed) below the tips of the anthers.

6. *Capsules.*—Gradually narrowing from the base, 2–2.5 cm. long.

7. *Seeds.*—Light brown. Those developed in the ovary of *biennis* after pollination by *grandiflora* are similar to the female parent.

Fig. 3. Flowering side branch of *Œnothera biennis*, B (10.21a), with a leaf from lower portion of main stem.

The most striking peculiarities of strains A and B in comparison with certain other American types of *biennis* are:—the small flowers, green stems, and the absence of that red coloration in the papillate glands which is seen in some forms of the species and is conspicuous in *Lamarckiana.*

Since differentiating the strains A and B in 1908, I have had an opportunity of observing somewhat extensively various forms of *biennis* in the vicinity of Boston, where the prevailing type has larger flowers, frequently a broader leaf, and usually stems punctate with red tinted glands. There is considerable variation in the characters above mentioned and I have this year selected certain plants growing wild on the grounds of the Bussey Institution that in some respects are likely to prove much more satisfactory for my purposes than the strains A and B. These

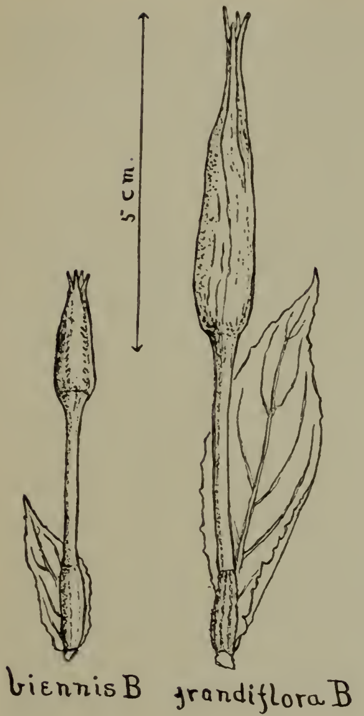

FIG. 4. Buds of *biennis* B and *grandiflora* B.

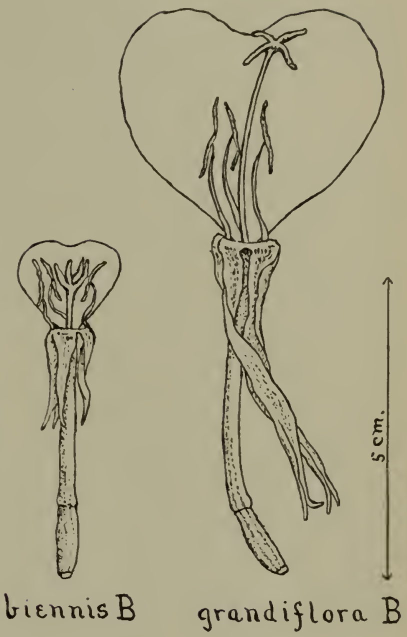

FIG. 5. Flowers of *biennis* B and *grandiflora* B.

plants (strains C and D) have been crossed this year with the best strains of *grandiflora* (B and D) and I expect to grow the hybrids in another season, when the strains will also be tested for their stability. The testing for purity is of course a necessary precaution, although, as explained above, the habit of self-pollination in *biennis* makes it very unlikely that any of these plants are tainted with foreign blood. These strains will not be further described until the prospective cultures have been grown.

Sowings from the wild seed collected as *Œnothera grandiflora,* described below, have given several plants of a southern form of *biennis* (strain S) which may prove of considerable interest since some of its characters (stem coloration, form of buds, size of flowers, etc.) are very favorable for combination with *grandiflora* with certain ends in view. This southern strain was crossed this season with *grandiflora,* and the hybrids and the parent *biennis* will be studied through further cultures.

Œnothera grandiflora Ait.

We are indebted to Dr. MacDougal ('05, p. 7) and to Miss Vail ('05, p. 9) for accounts of the rediscovery of this remarkable species of the southern United States and for a clear analysis of its probable history. Aiton's original description (1789), from material grown at Kew, states that *Œnothera grandiflora* was introduced by John Fothergill, M.D., in 1778. An expedition of William Bartram in 1776, undertaken at the request of Dr. Fothergill for the purposes of botanical discovery, records the finding of a large-flowered *Œnothera* near Tensaw (Taensa), Ala. Bartram's picturesque and excellent description of this new plant (see MacDougal, '05, p. 7), together with a herbarium specimen in the British Museum from "Hort. Fothergill, 1778," makes it evident that Bartram must have sent seed to Fothergill, through whom the plant was introduced into England in 1778.

Professor S. M. Tracy in 1904 visited the original locality and found the species growing in considerable quantity along the east bank of the Alabama river in the vicinity of Dixie Landing, which is only a few miles from Tensaw. His material agrees with the descriptions of Bartram and Aiton and, according to Miss Vail, with the herbarium specimen of Fothergill, and there seems to be no doubt that *Œnothera grandiflora,* so widely cultivated, has been rediscovered growing wild in its original locality.

My seed of *Œnothera grandiflora* was collected by Professor Tracy at Dixie Landing in September, 1907. During the past three seasons I have sampled the collection to the extent of bringing to maturity from the wild seed thirty-four plants, and in addition some seventy rosettes and young plants have passed under my inspection. Besides the above I have selected and cultivated from this material three strains (*grandiflora* A, B, and D), which have been carried through a second generation represented by twenty-four mature plants.

It became apparent, as my cultures progressed, that the *Œnothera grandiflora* growing near Tensaw is far from uniform in character. I have so far selected four distinct types of plants, only one of which, however, has been cultivated in later generations. The fact that the stigma of *grandiflora* is generally well above the tips of the anthers prevents pollination in the bud and in the opening flower, and offers a very much greater opportunity for cross pollination than is possible in *biennis*. This condition is probably responsible for the heterogeneity of the species.

The type which I have under cultivation and which has proved stable is characterized by a somewhat broader leaf than is common to the species. From it have been differentiated the following three strains (A, B, and D), which have been used in the crosses with *Œnothera biennis*.

1. Strain D came directly from a wild plant.
2. Strains A and B are the result of a cross between D and a similar plant, F, in the garden at Woods Hole in 1908. The cross was made to fix thoroughly the characters of a broad leaf and red coloration of sepals present in both parents. The two strains (A and B) are essentially similar, differing chiefly in the degree of red coloration present in the sepals, a character that is variable and probably cannot be depended upon as a fixed factor. As a matter of fact, strain D in a second generation has proved very uniform and it is not likely that I shall make further use of strains A and B. All three strains are representative of the larger proportion of the plants of *grandiflora* that have passed under my observation, being, however, what a gardener would select as luxuriant forms with broader and larger leaves than the average.

FIG. 6. Mature rosette of *Œnothera grandiflora*, B (10.4a).

In addition to the type represented by the strains A, B, and D (which is described in the paragraph below) there have appeared in the cultures from the wild seed the following three types markedly different from one another and from the general run of wild *grandifloras*.

1. A type represented by a large plant (2 m. high) peculiar for its light green broadly elliptical leaves, generally green stem, green sepals, and a close rosette of crinkled leaves strongly resembling a half-grown rosette of *Lamarckiana*. This type, represented by a single plant (*grandiflora* I) appeared this year, 1910, and is likely to prove of great interest. It will not, however, be described until its behavior in later generations has been noted and its crosses with *biennis* have been grown.

2. A type represented by a relatively small plant (1.2 m. high) with stiff, broadly lanceolate, revolute leaves, and sepals deeply blotched with red. This peculiar form

FIG. 7. Mature plant of *Œnothera grandiflora*, B (10.4c).

(*grandiflora* R) is too far from the general type of *grandiflora* to be of value in the present study.

3. A type represented by a plant (*grandiflora* Z) with narrow lanceolate leaves, much too narrow to give results, if crossed with *biennis*, that would approach *Lamarckiana*.

The chief characteristics of the *grandiflora* strains A, B, and D, employed in the hybrid studies of the present paper, when under good cultivation are as follows:

1. *Rosettes*.—The mature rosettes (Fig. 6), 2–2.5 dm. broad, have about 20 loosely clustered leaves, broadly elliptical, irregularly and sometimes deeply cut at the base, slightly crinkled, and mottled with reddish brown blotches. The rosettes are transitory, the leaves withering during the development of the main stem.

Fig. 8. Flowering side branch of *Œnothera grandiflora*, B (10.4a), with a leaf from the lower portion of the main stem.

2. *Mature Plants.*—The mature plants (Fig. 7), 1.5–2 m. high, have a more upright habit than *biennis*. Frequently the branching is profuse, the main stem and long side branches being covered with short shoots. Stems reddish, green only towards their tips, the papillate glands following the color of the stem. Basal leaves on the main stem ovate or broadly elliptical, about 15 cm. long (Fig. 8); leaves above broadly lanceolate.

3. *Inflorescence.*—Bracts early in the season leaf-like and more than half the length of the buds (Fig. 4); later becoming very much smaller.

FIG. 9. Rosette of a hybrid (10.30 La), *grandiflora* B × *biennis* A, F_1 generation.

4. *Buds.*—From 9–10 cm. long, the cone scarcely angled (Fig. 4). Sepals marked with reddish brown blotches, sometimes dull and faint; their tips attenuate.

5. *Flowers.*—Large (Figs. 5 and 8). Petals about 3.3 cm. long. Stigma lobes generally 2–5 mm. above the tips of the anthers.

6. *Capsules.*—Tapering rather sharply from the large base, 2.5–3 cm. long.

7. *Seeds.*—Dark brown. Those developed in the ovary of *grandiflora,* after pollination by *biennis,* have the same color as the female parent.

The above description is so worded as to emphasize the characters of the *grandiflora* strains A, B, and D in contrast to the *biennis* strains A and B. It should be compared with the description of the latter forms to understand clearly the conditions that appear in the hybrids, which will now be described.

FIG. 10. Mature plant of a hybrid (10.30 La), *grandiflora* B × *biennis* A, F_1 generation.

HYBRIDS IN THE F_1 GENERATION

The cultures of 1910 in the F_1 generation were hybrids of three different combinations of parents, as follows:

1. *grandiflora* B × *biennis* A (10.30), grown at the Bussey Institution.

2. *grandiflora* B × *biennis* B (10.18) and the reciprocal cross (10.19), grown at the Bussey Institution.

3. *grandiflora* A × *biennis* B (10.17) and its reciprocal cross (10.20), grown at the Botanic Garden of Harvard University.

Bearing in mind that the *biennis* strains A and B are practically indistinguishable and that the *grandiflora* strains A and B are essentially similar, the cultures as a whole would not be expected to differ markedly, which was the case.

The figures of hybrids published with this paper are from two plants in the first culture (*grandiflora* B ×

FIG. 11. Flowering side branch of a hybrid (10.30 La), *grandiflora* B × *biennis* A, F_1 generation. At the left is a leaf from the lower portion of the main stem.

biennis A). They were selected as likely to prove the most interesting for further cultures in the F_2 and later generations. These two plants 10.30 La and 10.30 Lb will be described in some detail, together with the general features of the cultures.

1. *grandiflora* B × *biennis* A (10.30). This culture gave about 400 seedlings from which 57 were selected for the breadth of the cotyledons and the shortness of their petioles. These were brought to maturity. The characters of the parents were blended in the rosettes which had long, broadly elliptical leaves, toothed below, and col-

Fig. 12. Rosette of a hybrid (10.30 Lb), *grandiflora* B × *biennis* A, F_1 generation.

ored with large reddish spots and blotches. The mature plants likewise presented the characters of both parents blended in the habit, foliage, and flowers, all the characteristics of form and measurements being clearly intermediate. It was possible to distinguish certain rosettes as being more *biennis*-like or more *grandiflora*-like than the culture in general and the mature plants from these rosettes also exhibited similar differences. The plants of the culture therefore presented a certain range, the extremes being readily distinguished as more like one parent than the other although never approaching closely to either. Between the extremes were numerous transitions.

Two rosettes of this culture were selected for their resemblance in certain particulars to *Œnothera Lamarckiana* and the mature plants from these proved to be among the most interesting in the gardens. These hybrids, 10.30 La and 10.30 Lb, were representative of a

FIG. 13. Mature plant of a hybrid (10.30 Lb), *grandiflora* B × *biennis* A, F_1 generation.

type of hybrid of *biennis* and *grandiflora* that has proved not uncommon in my cultures. The description of these two plants will now follow, arranged to bring out the salient features in comparison with the parent species and with *Lamarckiana*.

Hybrid 10.30 La. 1. *Rosette.*—The mature rosette (Fig. 9), about 3.3 dm. broad, was persistent. Older leaves more elliptical and less spatulate than those of *biennis* (Fig. 1), deeply cut at the base as in *grandiflora* (Fig. 6), spotted with red. The younger leaves were markedly crinkled as in *Lamarckiana* (Fig. 15), but narrower and more pointed.

2. *Mature Plant.*—The mature plant (Fig. 10), 1.8 m. high, had a straggling habit with long branches arising from the base, short clustered shoots above (*grandiflora*-like), a habit very different from the symmetry of *La-*

FIG. 14. Flowering side branch of a hybrid (10.30 Lb), *grandiflora* B × *biennis* A, F_1 generation. At the left is a leaf from the lower portion of the main stem.

marckiana (Fig. 16). Stem green above, mottled red below, occasional regions where red-tinted papillate glands lay on green portions of the stem as in *Lamarckiana*. Basal leaves on the main stem (Fig. 11) elliptical, about 22 cm. long, only slightly crinkled and not so long as the much-crinkled basal leaves of *Lamarckiana* (Fig. 17). Leaves on the upper portion of the plant broadly elliptical, slightly larger than those of *Lamarckiana*.

3. *Inflorescence.*—The inflorescence (Fig. 11) was strikingly similar to that of *Lamarckiana* (Fig. 17), the bracts being of about the same size and shape.

FIG. 15. Rosette of *Œnothera Lamarckiana* (10.23c).

4. *Buds.*—From 6 to 6.5 cm. long, about the same size as *Lamarckiana* and similar in form (compare Fig. 11 with Fig. 17), sepals green. Intermediate in size between those of parents.

5. *Flowers.*—Scarcely distinguishable from those of *Lamarckiana* (compare Fig. 11 with Fig. 17) and with the form and proportions of the parents blended. Petals about 2.2 cm. long. Base of stigma lobes slightly below the tips of the anthers.

6. *Capsules.*—About 2.3 cm. long, intermediate in size between those of the parents.

7. *Seeds.*—A shade of color clearly intermediate between the light and dark brown of the parents.

Hybrid 10.30 Lb. 1. *Rosette.*—The mature rosette (Fig. 12), about 3 dm. broad, was persistent. Older leaves much broader than those of *biennis* (Fig. 1), cut at the base as in *grandiflora* (Fig. 6), a lighter green than the average of the culture, and with relatively few red spots as in *Lamarckiana*. The younger leaves were

FIG. 16. Mature plant of *Œnothera Lamarckiana* (10.23c).

strongly crinkled, almost as broad as those of *Lamarckiana* (Fig. 15), but more pointed.

2. *Mature Plant.*—The mature plant (Fig. 13), 1.7 m. high, with long branches from the base, had a habit more spreading than that of *Lamarckiana* (Fig. 16), but was otherwise very similar. Stems green above, mottled red and brown below, the papillate glands of the same color as the portions of the stem upon which they lay. Basal leaves on the main stem (Fig. 14) broadly elliptical, about 20 cm. long, without crinkles and not so long as the

FIG. 17. Flowering side branch of *Œnothera Lamarckiana* (10.23zb), with a leaf from the lower portion of the main stem.

much crinkled basal leaves of *Lamarckiana* (Fig. 17). Leaves on the upper portion of the plant broadly elliptical, slightly crinkled, similar to those of *Lamarckiana.*

3. *Inflorescence.*—The inflorescence (Fig. 14) had bracts longer than those of *Lamarckiana* (Fig. 17), somewhat crinkled and similar to the bracts in figures of *Œnothera scintillans* (De Vries's "mutant" from *Lamarckiana*).

4. *Buds.*—From 6–6.5 cm. long, about the same size as those of *Lamarckiana,* but with a more pointed cone and attenuated sepal tips (contrasted in Fig. 18), sepals green. Intermediate in size between those of the parents.

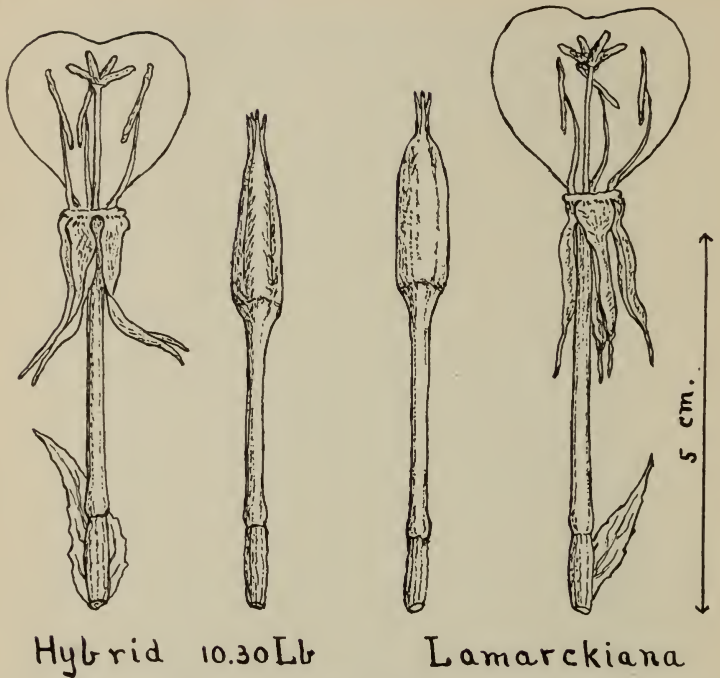

Fig. 18. Buds and flowers of a hybrid (10.30 Lb), *grandiflora* B × *biennis* A, F_1 generation, in comparison with those of *Œnothera Lamarckiana.*

5. *Flowers.*—Scarcely distinguishable from those of *Lamarckiana* (contrasted in Fig. 18), and with the form and proportions of the parents blended. Petals about 2.2 cm. long. Base of the stigma lobes slightly below the tips of the anthers.

6. Capsules.—About 2.5 cm. long, intermediate in size between those of the parents.

7. *Seeds.*—A shade of color clearly intermediate between the light and dark brown of the parents.

An examination of these two hybrid plants with respect to the contributions by their respective parents may be readily made by comparing the illustrations and descriptions of rosettes, mature plants, inflorescence, and flowers, bearing in mind that the parent *biennis* A is essentially indistinguishable from the strain *biennis* B which is here figured. It will be found that the hybrids present the characters of the parents in a blend. By a blend it must not be inferred that the characters of the hybrids are a perfect mean as to the measurement and proportions of their organs. This is certainly not the case for all of the characters of the hybrids and it would be a difficult matter to determine a perfect blend for a single character because of the fluctuating variations in the parent strains. All that I desire to demonstrate in the present account is the fact of blended conditions throughout these hybrids of the first generation, and the total absence of clear dominance of one parent over the other with respect to any character.

It would be very difficult and probably impossible to support satisfactorily a claim that either of the two parent plants exhibited its influence to a measurably greater degree than the other. To illustrate this point let us examine hybrid 10.30 La. The rosette (Fig. 9) of this plant might be said to be more like that of *biennis* than of *grandiflora,* but the habit of the mature plant (Fig. 10) with respect to its short clustered shoots is more like the latter parent than the former, and thus two characteristic stages of the plant suggest opposite conclusions. This general balance of the influence of one parent over the other was manifest throughout the greater part of the culture, but, as previously noted, a small proportion of the hybrids was readily distinguishable as being more like *biennis* or *grandiflora* although never approaching closely to either parent form.

The chief points of resemblance between the two hybrid plants just described and *Œnothera Lamarckiana* may be briefly summarized as follows:

1. The inflorescence was very similar to *Lamarckiana,* especially in the case of hybrid 10.30 La from which many branches might have been picked that as herbarium specimens could not have been separated from a mixed and varied collection of *Lamarckiana* branches similarly prepared.

2. The only essential difference between the buds lay

in the slightly greater attenuation of the sepal tips especially in the case of the hybrid 10.30 Lb.

3. The flowers of the hybrids were scarcely distinguishable from those of *Lamarckiana,* the small differences in the measurement of parts being no greater than might be expected in any reasonably large and varied culture of *Lamarckiana.* The chief difference with respect to the flower lay in the clear green color of the ovaries of the hybrids, the glands of which were not tinted red, as is characteristic of all material of *Lamarckiana* that the writer has seen.

4. Although the capsules were somewhat longer and more pointed than in the forms of *Lamarckiana* grown by the writer, they were not so long as in certain "long-fruited races" described by De Vries, who states that the fruits of *Lamarckiana* are highly variable and figures capsules as pointed as those of my hybrids (De Vries '09, p. 528, et seq., Fig. 114).

The points of difference concern chiefly the vegetative portions of the plants under discussion.

1. The rosette of the hybrids consisted of mixed forms of leaves, only the younger resembling the markedly crinkled leaves of *Lamarckiana.*

2. The habit of the mature hybrid plants was more straggling, lacking the symmetry characteristic of *Lamarckiana.* The basal leaves were not so large and were but slightly crinkled; the upper leaves, especially in the case of hybrid 10.30 Lb, were similar to *Lamarckiana.*

3. The coloration of the stem was green above and mottled red and brown below, in contrast to the green stems of *Lamarckiana* punctate with red-tinted papillate glands. Similar glands were present in the hybrids, but their color (portions of 10.30 La excepted) was that of the regions of the stem upon which they lay.

The hybrids, therefore, resembled *Lamarckiana* as to the inflorescence, floral parts, and fruits; they differed chiefly in certain vegetative characters and in the coloration of the stem. It remains to be seen through further cultures which of the two, the resemblances or the differences, are more stable in inheritance and variation.

The type of *Lamarckiana* which has been compared in this paper with the hybrids of *biennis* and *grandiflora* is one with which I have been familiar for the past five years. It has been represented in my cultures by strains from seed that has come to me through three different sources, all of the seed, however, originally being derived from the cultures of De Vries. These strains have not differed materially from one another, and as grown in my small cultures have not exhibited marked variation. The rosettes and mature plants have agreed in habit and foliage with the descriptions of *Lamarckiana* in "Die Mutationstheorie." The flowers have, however, been uniformly smaller than the measurements and figures of De Vries, the petals being about 2.5 cm. long instead of measuring 3 cm. or more. In the flower structure the position of the stigma has proved more variable than one would be led to suppose by the figures and descriptions of *Lamarckiana,* the stigma generally being but slightly above the tips of the anthers or about at their level, and in some plants distinctly below.

2. *grandiflora* B × *biennis* B (10.18), and the reciprocal cross (10.19). From about 200 seedlings of the first culture and about 250 seedlings of the second culture, 66 and 70 plants, respectively, were brought to maturity, being selected for the breadth of the cotyledon and the shortness of its petiole. As the rosettes formed there appeared much variation in the amount of anthocyan developed in the leaves, the larger proportion being marked with dull red spots and blotches, only a small number having few spots as in *Lamarckiana.* The form of the leaves likewise varied and certain of the rosettes were readily separated as being more *biennis*-like or *grandiflora*-like than the average. The plants that developed from these extreme forms of rosettes were also somewhat more like the respective parents than the mass of the culture which presented the characters of

the parents thoroughly blended in the form and proportions of habit, foliage, and flowers. Considering the cultures as a whole, there seemed to be no marked difference between the first cross and its reciprocal.

The average types of hybrids in both crosses were essentially similar and a number of types were very close to the hybrids 10.30 La and 10.30 Lb of the previously described culture. Six plants in culture 10.19 were selected for special peculiarities, but these will not be described unless their behavior in the F_2 generation should justify a detailed account.

3. *grandiflora* A × *biennis* B (10.17), and the reciprocal cross (10.20). From about 200 seedlings of the first culture and about 150 seedlings of the second culture, 49 and 60 plants, respectively, were brought to maturity, being selected for the breadth of the cotyledon and shortness of its petiole. These cultures were grown in a stiff clay at the Botanic Garden and presented an interesting contrast to the cultures previously described which were grown in a somewhat sandy well-fertilized soil. The plants were smaller and less vigorous vegetatively, although they flowered very freely. The rosettes and mature plants presented the characteristics of the parents well blended as in the other cultures. There were also a few extreme types that resembled one or the other of the parents more closely than the average. There appeared to be no significant differences between the first cross and its reciprocal. Two plants with marked peculiarities were selected from culture 10.17 and will be carried through an F_2 generation; they will be described if their further cultivation proves of interest.

Although the evidence, not being quantitative in character, is incomplete, nevertheless the following points may be noted, at least provisionally, from these observations on F_1 generations. (1) There was no indication from these cultures of a marked preponderance of either paternal or maternal influence upon the hybrids.

(2) No character of either parent was observed to be dominant. (3) It is doubtful whether there would be any material difference between a cross and its reciprocal if each were equally vigorous. (4) Although the extreme types in the culture, approaching somewhat the respective parents, could be readily distinguished, they were connected by transitional forms and a sharp line could not be drawn between two sets of hybrids, such as have been described by De Vries ('07, '08) as "twin hybrids" and reported for crosses between the *Onagra* group and *Lamarckiana*. From observations on small cultures during the season of 1909 (Davis '10, p. 113) the writer was led to believe that "twin hybrids" might be present in this cross, but he no longer regards this as probable.

HYBRIDS IN THE F_2 GENERATION

In a recent paper (Davis '10) I described two small cultures of hybrids between *biennis* B and *grandiflora* D that were grown at the Botanic Garden of Harvard University in the season of 1909. Four of the plants of these cultures were of special interest as presenting flowers and inflorescences very similar to *Lamarckiana*, although differing markedly in foliage. I unfortunately was unable to observe the early development of these plants and for this reason they were not very good forms on which to base studies of their progeny in the F_2 generation. Their seed was, however, sown this season and the cultures, described below, were of interest as indicating the probable behavior of hybrid plants of *biennis* and *grandiflora*. These four lines will not be cultivated further, since I have in the hybrids 10.30 La and 10.30 Lb material better suited to the purposes of a quantitative study for the reason that the records of their life history have been kept in detail. The seeds of these hybrids proved fertile to a very high degree, but it was practicable to grow only a small proportion of the seedlings to maturity.

1. Progeny from hybrid 9ba, *biennis* B × *grandiflora*

D. This hybrid plant (Davis, '10, pp. 112 and 113), an excellent blend of the parent forms, was similar to *Lamarckiana* in habit and floral structure, but differed in having smaller, uncrinkled leaves on the lower portions of the plant and larger bracts upon the inflorescence.

From about 600 seedlings 73 plants were carried through the rosette stage and set in the ground, being selected to represent various types. The seedlings were strikingly diverse, some having long cotyledons similar to those of *grandiflora,* others having shorter and broader ones, and a large proportion with small light yellow, etiolated cotyledons. Many of the latter seedlings died before the appearance of the second leaf, the others developed very slowly, forming rosettes one-fourth or one-third the size of the normal with more or less etiolated leaves. Twenty-three of the dwarf rosettes were set out in the garden and of these seven finally grew to be large plants similar to the average of the culture, but with a somewhat etiolated foliage; of the remainder several died and the others developed into dwarf plants from 2–8 dm. high, small leaved, sparsely branched, and with flowers smaller than the average but larger than the *biennis* parent. The behavior of these etiolated dwarfs resembled De Vries's account of the appearance of the form *albida* in his cultures of *Lamarckiana.*

As the normal rosettes approached maturity it was possible to distinguish certain ones as somewhat more *biennis*-like or more *grandiflora*-like than the average, and the mature plants which developed from these showed similar points of resemblance to the respective parents of the cross. Nevertheless, the culture as a whole presented these parental characters well blended, although exhibiting a much wider range of variation than the F_1 generation of this cross. This variation appeared to indicate a relative segregation of the parental characters deserving of detail studies upon larger cultures. There were a number of plants similar to the parent hybrid, but none markedly nearer to *Lamarckiana.*

2. Progeny from hybrids 9ba, 9bb, and 9bc, *grandiflora* D × *biennis* B. These three hybrids (Davis, '10, p. 114) presented the parental characters well blended. They were essentially similar to *Lamarckiana* in flower structure and inflorescence, but differed in foliage and habit, the leaves on the lower portion of the stem being but half the length of those similarly placed on *Lamarckiana* and with only slight traces of crinkles; the habit was *biennis*-like.

From about 350 seedlings of hybrid 9ba, 76 plants were brought to maturity, being selected as representative types of the rosette stages. A small proportion of the rosettes was dwarfed and the nine selected representatives of this type developed small plants 1–2.5 dm. high, generally without side branches; these did not flower. The normal rosettes varied greatly in the forms of leaves and extent of the red coloration, certain ones being distinctly more like the respective parents of the cross than the average; these differences were maintained in the mature plants, but to a less marked degree. The culture in general presented a habit more *grandiflora*-like than *biennis*-like, but all of the characters remained blended, although there was a considerable range of variation in flower structure and foliage. While a number of the plants were similar to the hybrid parents, none proved to be appreciably nearer to *Lamarckiana*.

There were about 550 seedlings of hybrid 9bb, from which 93 plants, selected as representative rosettes, were brought to maturity. Relatively few dwarf rosettes were present in this culture; eleven of these being selected grew into plants 1–4 dm. high, small leaved and without prominent side branches, the larger of the dwarfs developing small *biennis*-like flowers. The culture in general was more uniform than the preceding, but certain rosettes and mature plants were noticeably more like one or the other of the original parents than the average, which presented these parental characters well blended. The foliage of the culture was distinctly crinkled so that the plants resembled *Lamarckiana* more closely than those of the other cultures in the F_2 generation, differing chiefly in the smaller size of the basal leaves and in the absence of red tinted papillate glands on a green stem, the stem being mottled with red. One plant of this culture (10.12 Lz) was selected for marked peculiarities, but will not be described unless its behavior in an F_3 generation proves of sufficient interest.

About 800 seedlings of hybrid 9bc appeared in the culture, from which 95 plants were selected as representative types of rosettes. A few dwarf rosettes were present, six of which set in the ground developed into unbranched plants about 1 dm. high, that failed to flower. The culture in general exhibited considerable variation, the most interesting types of plants being several with light green, smooth, obtusely pointed leaves, similar in shape to *Lamarckiana,* but without crinkles. There was shown the same previously described tendency on the part of a few rosettes and mature plants to depart from the average of the culture towards the characteristics of the respective parents of the cross, maintaining, however, a blended structure of their parts.

Considering these cultures of F_2 generations in comparison with the F_1 generations that have been grown, the most striking feature is the greater range of variation exhibited not only by the F_2 plants as a whole, but by their different parts. Since the studies were not quantitative in character, because such a large proportion of the seedlings were necessarily discarded, it has not seemed best to describe the variations in detail and such an investigation is deferred for the present. However, in this increased variation is clearly indicated at least a relative segregation of the parental characters in the F_2 generation.[2]

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[2] Extensive cultures from the seed of the two hybrids 10.30 La and 10.30 Lb, described in this paper, are now (February, 1911) seedlings with 4–5 leaves which already show marked segregation in this F_2 generation, with the extreme types closely resembling seedlings of the parents of the cross and between these a large range of intermediates.

THE POSSIBLE ORIGIN OF *Œnothera Lamarckiana* AS A HYBRID OF *O. biennis* AND *O. grandiflora*

We have shown that hybrids between certain strains of *Œnothera biennis* and *O. grandiflora* may be synthesized, which approach somewhat closely to *Œnothera Lamarckiana,* and there is good reason to believe that further experimentation will result in the production of forms with a more perfect resemblance. It is now important to ascertain, as far as this is possible, whether there are any historical reasons why *Lamarckiana* may not have arisen either accidentally or intentionally from such a cross.

Œnothera Lamarckiana appears to have been under cultivation in the gardens of the Muséum d'Histoire Naturelle at Paris in 1797, being described by Lamarck[3] under the name *grandiflora.* Shortly afterwards Seringe[4] renamed the form *Lamarckiana,* recognizing it to be distinct from the *grandiflora* of Aiton.

As previously noted from the investigations of MacDougal ('05) and Vail ('05), the evidence is very clear that *grandiflora* was introduced into England in 1778 and was at that time under cultivation at Kew. Forms of *Œnothera biennis* had of course been in European gardens for many years previous to 1778. There was therefore a period of about eighteen years (1778–1797) during which hybrids between *biennis* and *grandiflora* might have arisen in Europe before the earliest known record of the cultivation of *Œnothera Lamarckiana* in Paris. So striking an American novelty as *Œnothera grandiflora* would almost certainly have been passed on from Kew to other botanical gardens and in the interval between 1778 and 1797 is likely to have become widely distributed and cultivated. On historical grounds then there seems to be no reason with respect to the date of the first recorded recognition of *Œnothera Lamarckiana* why this form might not have arisen in Europe as a hybrid of *biennis* and *grandiflora.*

[3] "Encyclopédie Méthodique Botanique," Vol. IV, p. 554, 1797.

[4] De Candolle's "Prodomus," Vol. III, p. 47, 1828.

Let us suppose that it should be shown that *Œnothera Lamarckiana* was in existence previous to the date 1778, what effect would such evidence have on the hypothesis that the form is a hybrid of *biennis* and *grandiflora?* It would not in the writer's opinion have weight against experimental proof that *Lamarckiana* or forms closely resembling this plant may be synthesized as hybrids of these wild American species. It would not prove that Lamarck's plant in Paris (1797) was not a hybrid. It would merely indicate that *Lamarckiana,* having arisen as a hybrid in America, was introduced as such into Europe. As already pointed out, the position in *grandiflora* of the stigma well above the anthers gives ample opportunity for chance hybridization in nature. Indeed. the diverse forms that have appeared in my cultures from seed of *grandiflora* collected in the field clearly show that the species is far from homogeneous in character, a condition that is probably due to a large amount of cross pollination. It may be expected that careful search, especially in the southern United States, will bring to light occasional plants with characters intermediate between *grandiflora* and other species, such as, for example, southern types of *biennis,* but it is also probable that the behavior of such plants in culture will show them to be heterozygous in character, *i. e.,* hybrids.

There have been two attempts to establish the presence of *Lamarckiana* in Europe previous to 1778 when *grandiflora* was introduced at Kew. MacDougal ('07, pp. 5, 6) refers to *Lamarckiana* a description and figure of an *Œnothera* by Miller. Plate 189. Fig. 2, for the "Gardener's Dictionary," 1760. This figure, published in 1757, is of a large-flowered *Œnothera* with petals 2 or 2.2 cm. long and by its side (Fig. 1) is a smaller-flowered form. With respect to the point under discussion, the most important features of these figures, clearly shown by the drawing, is the position and form of the stigmas, *well below* the tips of the anthers and with the lobes *unexpanded* in open flowers. These are peculiarities of *biennis* and, in the writer's experience, are not characteristic of *Lamarckiana* where the stigma lobes are usually expanded in the open flower and generally above or about on the level with the tips of the anthers. Furthermore the size of the petals in the illustration of the large-flowered type (Fig. 2) is no greater and indeed not so great as in some forms of *biennis*. Both of the figures show the essential characteristics of the flower of *biennis* to which they have generally been referred in taxonomic accounts. For these reasons the view of MacDougal that the illustration of the large-flowered type (Fig. 2) is of *Lamarckiana* and establishes its presence in Europe previous to 1757, is to the writer not convincing.

The second attempt to establish the presence of *Lamarckiana* in Europe previous to 1778 is the announcement of Gates ('10) that certain marginal notes in a copy of Bauhin's "Pinax," 1623, give in Latin an accurate description of this plant although differing in one or two minor characters. Gates presents an outline of the points which indicate to him that the description refers to *Lamarckiana,* but the notes themselves are not published. A full account is promised, in which we may expect to see these Latin notes and judge of them for ourselves, and comments on this announcement will be reserved for the present.

Finally we must return to the question of whether or not it appears probable that *Œnothera Lamarckiana* is at present a component of the American native flora. De Vries ('05, p. 368) refers to *Lamarckiana* certain herbarium material at the New York Botanical Garden and Missouri Botanical Garden, both collected by A. W. Chapman in Florida (1860 or earlier), and also material in the Philadelphia Academy of Science collected by C. W. Short at Lexington, Ky. A thorough search (MacDougal, '05, p. 6) by several botanists in the vicinity of Lexington Ky., Nashville Tenn., Knoxville Tenn., and Courtney Mo., in the endeavor to find living plants that might be identified as *Lamarckiana,* was unsuccessful. Later, Miss Vail (MacDougal, '07, p. 67) came to the conclusion that the plant from Lexington, Ky., is *grandiflora,* and a possible escape from cultivation. I have not seen the herbarium material mentioned above, but in the light of the fact that many dried specimens could be prepared from my hybrids which as such would be considered *Lamarckiana,* it is clearly necessary that evidence from herbarium material should be weighed with much caution. The average herbarium material of the *Œnotheras* is generally not sufficient to show the peculiarities of the earlier phases of development (rosettes and basal foliage) which in the case of *Lamarckiana* furnish diagnostic characters that are necessary for a full identification. Unless the evidence of field collections is followed up by garden cultures, there is the possibility of numerous errors of interpretation.

A specimen in the Gray Herbarium of Harvard University is stated by MacDougal ('05, p. 5) to agree perfectly with *Œnothera Lamarckiana,* but in this view the writer can not accord. This plant was apparently grown in the Cambridge Botanical Garden, Massachusetts and bears the date 1862. The specimens are accompanied by the significant notes in the hand writing of Dr. Asa Gray "from seed of Thompson, Ipswich," and "said by English horticulturists to come from Texas." The flowers are large, with petals about 4.5 cm. long and sepals about 5 cm. long, very attenuate, the tips projecting 1 cm. beyond the folded petals in the manner characteristic of *grandiflora.* The stigma lobes are also *grandiflora*-like in their length, about 8 mm., and in their position, about 5 mm. above the tips of the anthers. A large detached leaf, about 18.5 cm. long, with some evidence of former crinkles, suggests by its form (although rather small) the basal leaves of *Lamarckiana.* The flowers and upper foliage of this specimen, however, agree very closely with broad-leaved types in my cultures of *grandiflora* and do not resemble the *Lamarckiana* that I have grown from seeds of De Vries, or with his figures and descriptions in "Die Mutationstheorie.'' If this plant could be established as derived from *Œnotheras* introduced into England by Messrs. Carter and Co. at about 1860 from seeds said to come from Texas, it would be a point of great importance, as will appear in the following paragraphs.

De Vries ('05, pp. 384–385) offers strong evidence that the strains of *Lamarckiana* at present cultivated in Europe have a genetic relation to seed of Messrs. Carter and Co., of London about 1860. This seed is stated to have been received unnamed from Texas and plants grown from it were pronounced by Dr. Lindley to be *Lamarckiana.* A specimen from one of these plants is figured in ''The Floral Magazine,'' Vol. II, Plate 78, 1862, this plate being reproduced in ''L'Illustration Horticole,'' Vol. IX, Plate 318, 1862. This plate shows an *Œnothera* with flowers about 10 cm. (4 inches) in diameter and with a large amount of red coloration on the sepals and ovaries; the stigma is figured both above and below the tips of the anthers. The flowers of this illustration are larger than those of *Lamarckiana,* as known to the writer, and would do for *grandiflora* except for the position of the stigma which is much closer to the anthers than is typical for this species. The red coloration of the sepals and ovaries is much too deep for typical *Lamarckiana* and not unlike some forms of *grandiflora,* but the sepal tips, as drawn, are not so long or so pointed as in the latter form. Indeed the identification of this plate with any probable *Œnothera* is very difficult and the reasons why it should be called *Lamarckiana* are to the writer far from convincing, although it would perhaps be as easy to argue for this identification as for any other.

It is, however, possible that new light may be thrown on the composition of the cultures of Carter and Co. through the plant in the Gray Herbarium described above. The date of this specimen, 1862, together with the very suggestive notes of Dr. Gray '' from seed of Thompson, Ipswich,'' and '' said by English horticulturists to come from Texas," make it appear possible that this plant was derived from the cultures of Carter and Co. If this could be established it would indicate that forms very close to *grandiflora* were present in the cultures or seeds of this firm. It is not at all improbable that Texas with its immense area and very great range of climatic conditions may harbor *grandiflora* or related types especially since it is known to be rich in species of *Œnothera* and to have a number of large flowered representatives.

There may have been thus a second introduction into England of *grandiflora*-like types through Carter and Co. at about the year 1860. While there is of course no means of knowing whether their cultures were uniform, it is altogether probable that the results of their sowings gave a diverse progeny, since that has been my experience with seed from Alabama. There seems to be no reason why chance hybrids may not have been present or why *grandiflora*-like strains might not have shortly hybridized with European forms of *biennis*. These possibilities are mere matters of speculation to which little assistance is given by the puzzling plate in "The Floral Magazine" and in "L'Illustration Horticole" referred to above. A search among the English herbaria might, however, result in the discovery of specimens which would materially assist in the solution of a very interesting question—the identity of the plants grown by Carter and Co. At present the specimen in the Gray Herbarium appears to offer the most important evidence bearing upon the question. The contention that *Lamarckiana* was introduced in the form of a native American species at this date, 1860, seems to the writer to be without sufficient foundation.

The American botanist will ask himself why, if *Lamarckiana* was present in America as a native species in 1860, no localities are known where it may be observed in the field. It will be hard for him to believe that so strong and vigorous a plant, if a wild species, has become so recently extinct when, as he well knows, the *Œnotheras* are established as remarkably successful forms in our flora. The fact that *Lamarckiana* is not known as a component of the native American flora stands as the most serious obstacle to the view that this plant is representative of a wild species. The writer believes it very probable that plants more or less resembling *Lamarckiana* will occasionally, or perhaps rarely, be found in parts of America and under circumstances indicating that they are not garden escapes, but it seems to him equally probable that these forms when tested in culture will give evidence of a heterozygous, or hybrid nature. The mere records of such plants as handed down by the average type of herbarium specimen, unaccompanied by experimental cultures, will have little or no value for the present problem—the origin of *Œnothera Lamarckiana.*

Summary

This paper offers a body of evidence which shows that hybrids resembling *Œnothera Lamarckiana* may be synthesized from certain strains of the American native species *O. biennis* and *O. grandiflora.* The resemblances of the hybrids to this plant are strongest with respect to the inflorescence, buds and flowers. The differences are chiefly manifest in the basal foliage of the mature plant, in the coloration of the stem, and in the more straggling habit of the hybrids. The rosettes of the hybrids present mixed forms of leaves, the younger with points of similarity to *Lamarckiana.* Bearing in mind that other strains of *biennis* have characteristics more *Lamarkiana*-like than those of strains A and B, herein described, it is more than probable that the hybrids from certain crosses made this season (1910), when grown in future cultures, will come closer to the desired end—the synthesis of a hybrid so similar to *Lamarckiana* as to be practically indistinguishable by the usual taxonomic tests.

Exception is taken to the claim of MacDougal ('07.

pp. 5, 6) that Miller's Plate 189, Fig. 2, for the "Gardener's Dictionary," 1760, establishes the presence of *Lamarckiana* in Europe previous to the date, 1778, when *grandiflora* is known to have been introduced into England. The view of De Vries that strains of *Lamarckiana* were introduced into England about 1860, through seed of Messrs. Carter and Co. said to come from Texas, is discussed with reference to certain specimens in the Gray Herbarium of Harvard University and in the light of the author's experience with seed from Alabama, indicating that Carter and Co. probably had *grandiflora*-like types in their cultures which were also likely to have been of a mixed character. The absence, so far as is known, of *Lamarckiana* as a component of the native American flora is emphasized as a point of great importance against the claim that *Lamarckiana* was introduced into Europe as an American wild species.

A working hypothesis is presented as a result of the writer's experimental studies and in relation to such historical evidence as is available, to the effect that *Œnothera Lamarckiana* arose as a hybrid between certain types of *biennis* and *grandiflora,* recognizing that under these names must, for the present at least, be included a number of races which can only be clearly defined by laborious genetical investigations. The precise time and place of such an origin for *Lamarckiana* is a matter of mere speculation, but there seems to the writer no good reason why hybridization between *biennis* and *grandiflora* might not have taken place in Europe between 1778 and 1797 (when *Lamarckiana* was first recognized at Paris) and also at later dates, as for example about 1860. It is also possible that *Lamarckiana* may have been introduced as a chance hybrid from America, but the probability of such an origin is naturally rather remote.

The bearing of the possible hybrid nature of *Œnothera Lamarckiana* upon the claim of De Vries that the behavior of this plant demonstrates the origin of new species by mutation from a form representative of a typical wild species will be sufficiently evident to require no comment at this time. A discussion of the matter will therefore be reserved until the writer has proceeded further with his studies.

CAMBRIDGE, MASS.,
November, 1910.

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[*Reprinted from* SCIENCE, *N. S.*, *Vol. XXXII.*, *No. 811*, *Pages 93–95*, *July 15*, *1910*]

NOTES ON AN EXPERIMENT CONCERNING THE NATURE OF UNIT CHARACTERS

SOME time ago the writer planned[1] a series of experiments designed to throw some light on the nature of unit characters. Only one part, of which the following is a brief extract, has been completed. If an apology is necessary for daring to present negative results, I might say that even if proof of a negative is logically impossible, such evidence does give an idea of the relative frequency of the occurrence of the event in question. It is sometimes forgotten that a small probable error is as desirable in this case as when the results are positive. In addition to this fact, however, it is a pleasure to call attention to a line of experimentation which, though familiar to all biologists, has not had the serious consideration that it deserves. I mean the work of MacDougal in trying to produce mutations or transmissible variations by artificial means. Even if one does not accept as fact that the definite and transmissible changes which have occurred in Dr. MacDougal's injection experiments were caused directly by the introduction of semi-toxic solutions into the mother plant's ovary, he should admit that the method proposed is well worth his earnest attention. It is capable of several modifications and extensions—two of which I shall describe—which if given sufficient trial might yield results with important bearings somewhat apart from the original scope of MacDougal's investigations. Even if many experiments on limited popula-

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[1] At the Connecticut Agricultural Experiment Station, under the federal appropriation known as the Adams fund.

tions should give no positive results, it should be remembered that progressive variation occurs but rarely in nature, possibly but one variant in millions of individuals. One ought then to expect to increase this proportion only if he can multiply artificially the effectiveness of nature's causes; and it seems hardly reasonable to be disappointed if positive results are not obtained from experiments with only 1,200 or 1,500 plants.

One method which in spirit is an extension of the injection work was suggested by Osborne's investigations on plant proteids. Work on the ultimate composition of pure proteids has only been touched, but the fundamental researches that Osborne and his associates have carried on for the past twenty years have shown, even with the crude methods of our general analytic chemistry, that the proteids of different species of plants are very different in composition, the differences becoming more definite as the plants are further apart in the natural system. These facts immediately suggest the possibility that if the plant of one species could in its first life stages utilize the stored proteids of the endosperm or cotyledons of a very different type, changes would probably be induced in it, some of which might be heritable. This treatment is quite different from that where plants are fed different quantities of inorganic compounds in the form of the so-called essential elements of soil fertility. Food compounds and enzymes or producers of enzymes of a different kind from those ordinarily produced and used by the plant, are ready for its use in the very early formative period of ontogeny. If any changes can be expected to come about indirectly through changes in nutrition, they should be expected to occur under such treatment. It is possibly not a phenomenon that could occur naturally, yet since variations are caused by some cell activity different from the normal, they might very well be caused by the production of a different proteid or part of a proteid molecule, different from that normally produced but similar to what is produced by other plants. Then again similar conditions are probably produced when severe changes in temperature occur during the maturation of the seed. In fact, abnormal temperature conditions seem to have similar effects on somatic cells, for Webber[2] once stated that after the last great Florida "freeze" bud variations were very numerous in the trees that were severely injured. It is likely, too, that specialized parasites may have had changes in their own structure brought about in this way. Of course one must recognize the fact that a great many data are at hand on the effect of the stock upon an engrafted scion, all of which are negative or questionable. In this case, however, conditions are not similar. The bud or branch used as a scion is not at all in the same ontological stage as are the young seedlings in the experiments proposed. I have mentioned this matter at greater length than I had intended, but I have had the chance to try only some preliminary experiments with grafted cotyledons, and hope the plans might find favor with others who could work on a larger scale.

The second experiment which included observations on 1,200 plants is somewhat different. There seemed a possibility at least that if certain characters are due to the presence of an enzyme, this enzyme might actually be added to a plant which had not inherited either the enzyme itself or the ability to produce it, if the proper time of addition and the proper method could be found. This is pure speculation with no analogies, although it is likely that the mosaic disease of tobacco is an enzyme effect that can be communicated by infection. This disease, however, is not in-

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[2] In a personal communication.

herited through the seed, and may prove to be bacterial.

The subjects selected for experiment were two varieties of tomatoes, Sutton's Best of All, a variety with red flesh, and Golden Queen, a variety with yellow flesh. Hurst had already shown that all tomatoes possessed yellow flesh and that presence and absence of red flesh acts as a simple Mendelian pair. In order to be certain that we are dealing with the same characters, however, the two varieties were crossed and Hurst's work corroborated. Golden Queen, the yellow variety, was grown to flowering in sterilized soil. Flowers were castrated and bagged. When the stigmas were receptive they were pollinated with pollen from the same plants, which had been kept pure by bagging. At intervals of one hour after pollinating—up to fifteen hours—different ovaries were injected with one of three solutions. Solution one was obtained by macerating one part by volume of seeds of the red variety in four parts water. Solution two was made by macerating the flesh of ripening fruit of the red variety and adding 50 per cent. water. Solution three was made by macerating pollen from the red variety in about nine times its bulk of water and filtering.

Seed was obtained from most of the injected ovaries, but the resulting fruits gave absolutely no trace of red coloration. The seeds from the treated ovaries were again planted in sterilized soil and gave nothing but normal Golden Queen fruit.

I have no doubt but that an experiment of this kind seems utter foolishness; most experiments yielding negative results do. Biologists, however, have generally accepted the suggestion of physiological chemists that life processes are in the nature of enzyme processes. Perhaps this is because one is behind a safe barrier of ignorance when he speaks of enzymes. But in the case of plant sap colors and animal pigments there certainly is reason to believe that their production is accelerated by enzyme action. If this is true, color-producing enzymes should show action comparable to that of other enzymes. As to the general properties of enzymes, however, little is known. Perhaps they can be stated in the following definition. Enzymes are catalysts that have thus far been produced only by living organisms. Two of their properties may be mentioned that especially interest us here: one, which they hold in common with inorganic catalysts, that of changing the rapidity of progress of a reaction already initiated, but not appearing in the final product; the other, that of possessing colloidal nature and a large molecule. The size of the molecules of all known enzymes and their colloidal nature makes it improbable that any extract containing a color-producing enzyme should reach the ovules of a treated ovary; it is not at all impossible, however, that such an extract might come in contact with the male nucleus as it is journeying from the stigma to the micropyle. Further, if one may argue from the work that has been done on artificial digestion, enzymes should be able to do their work after extraction. This work, then, simply shows failure under the conditions described. It may be that failure should always be expected, yet with proper analysis of some of the attendant physical and chemical processes, some valuable results might be obtained.

E. M. East

Harvard University

[Reprinted from THE AMERICAN NATURALIST, Vol. XLIV., July, 1910.]

INHERITANCE IN POTATOES

PROFESSOR EDWARD M. EAST

HARVARD UNIVERSITY.

A STUDY of the behavior of certain plant characters in inheritance formed part of an investigation into the factors connected with the improvement of the common potato as a commercial crop, begun at the Connecticut Agricultural Experiment Station in 1906. This work was really a continuation of investigations made by the writer at the University of Illinois from 1902 to 1905, along broader and somewhat different lines. In 1908[1] the many disheartening difficulties attending hybridization were discussed, but it was shown to be possible to overcome several of the obstacles by proper treatment. The conclusions drawn at that time have not been changed by further experience, but the hindrances caused by external conditions not under control have been so great that the work has been discontinued. For example, in 1908 a prolonged drought at the time the fruits were forming, caused one hundred and fifty cross- and self-pollinated seed-berries to drop off while yet too immature for the seed to germinate. Not a single hand-pollinated flower matured its fruit.

Recently, a part of the pedigree records were lost in a fire which destroyed one of the buildings of the Connecticut Agricultural Experiment Station. For these reasons the data reported here do not represent fairly the amount of work done upon the subject, for the actual number of plants under observation was considerably larger than the figures reported. The complete figures had been studied with some care before the loss of the records, and it is thought that the remaining records are a fair sample of the whole.

The records contain observations on only one genera-

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[1] Some essential points in potato breeding. Biennial Report, Conn. Agr. Exp. Station. 1907–1908, 429–447, 1908.

tion of plants, together with the characters possessed by their parents. I have endeavored to find what characters were possessed by the parents of the varieties used in crossing but have found no trustworthy data. The following conclusions, therefore, are tentative. Nothing is known about the behavior of the characters when extracted. The data show that *certain characters segregate,* they give some evidence as to dominance and recessiveness, but they do not show the exact behavior of the Mendelian factors concerned, under different combinations.

COLOR IN THE PLANT STEM

Many varieties have a purple anthocyan sap color which gives the plant stem a dark appearance quite distinct from the clear green stems of the varieties in which it is absent. The color is variable in amount in different varieties. In some it extends throughout the petioles and petiolules; in others it can only be detected on the stems of the young seedling. My counts were made on seedlings about four inches high.

The color is evidently of the same nature as that found in many other cultivated plants. Its widespread occurrence and seeming uselessness in the plant's economy would place it in the category of typical varietal characters in the sense used by De Vries. It forms a single allelomorphic pair with its absence.

One purple-stemmed variety selfed gave all purple-stemmed progeny. Four purple varieties selfed, each showed segregation into two distinct classes, purple and non-purple. Fifty-four purple plants and seventeen non-purple plants were obtained. (These figures as well as those that follow are the records saved from the fire.) In each of these cases we may take it that the parent plants were heterozygous for the purple color, and approximated the simple three to one Mendelian ratio when self-pollinated. Four green-stemmed varieties were also selfed, and produced nothing but green-stemmed progeny.

One of these pure green-stemmed varieties was crossed on one of the heterozygous purples, and thirteen seedlings were obtained. Six plants were purple-stemmed and seven were green-stemmed. This result is what would be expected when crossing DR × R.

Color in the Flowers

All potato flowers have a ray of yellow extending from the limb of the corolla toward the apex of each lobe. The remainder of the corolla is either white or purple. There is wide zygotic (in potatoes, therefore, varietal) variation in the intensity of the purple sap color, but the flowers should probably be classed as either purple or white. The fact that the variety color, whether light or dark, remains true when propagated asexually, does not necessitate more than one Mendelian pair. It is undoubtedly a quantitative difference in the same pigment which is kept constant by the asexual method. Why somatic cell divisions should reproduce a color shade so exactly, while sexual reproduction gives rise to varying shades is unknown. It is the more peculiar since in animals visible division of the chromatin appears to be much more accurate in the sexual cells than in the somatic cells. The somatic cell appears to have the power of developing and of regenerating only the quantity of color originally apportioned to it, except on the rare occasions when all of the potential color activity goes to one daughter cell and the other is left without it. When this occurs, branches resulting from the descendants of the second cell are "sports" or "bud-variations" in which the original character is lost.

Only two varieties of potatoes with flowers other than purple or white have been noticed. The variety *Holland fleur de June* has blossoms which are decidedly yellow. Several attempts to self this variety and to cross it with other varieties failed. One other variety, a nameless seedling of unknown origin, possessed a true blue flower. No admixture of red which would give it a purple tinge could be detected. Even this color, however, may be of the same nature as the purple color, the difference being in the completeness of the reaction forming the blue dye. It will be remembered that litmus reacts in this manner. Several cross-pollinated and several self-pollinated fruits were obtained from this variety, but none of the seedlings had flowered in 1909, the second year of their growth.

The seedlings of the potato are very slow to flower in a New England environment, and but few flower records were obtained among several hundred plants. One selfed variety with purple flowers gave progeny all with colored flowers. Three selfed varieties with purple flowers gave both purple and white flowers: the total number of seedlings that flowered was nineteen, of which fourteen were purple-flowered and five were white-flowered. Three selfed white-flowered varieties gave nothing but white-flowered progeny.

Since three white-flowered varieties gave nothing but white flowers and three out of four colored varieties showed a hybrid condition with segregation of color, the purple is probably dominant to its absence. Color and no color is probably a single Mendelian pair, but this can not be stated with certainty from such meager data.

COLOR IN THE TUBERS

Potato tubers, when colored,[2] are either purple or red. In both cases the color may extend over the entire tuber or may be limited in extent. No definite mosaic pattern is formed when the color is limited, but the splashes of color are restricted to pretty definite areas. It is probably due to a separate Mendelian factor, for the mosaic varieties and the self-colored varieties are distinct. Tuber color varies quantitatively more than flower color. Many varieties show no color in the skin, and can be classified only by examining the young shoots when the

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[2] Colorless skins may vary from white to dark brown in different varieties. This is entirely due to their possessing corky layers of various thicknesses.

latter are about half an inch in length. The progeny of such varieties belong to the same classes as the progeny of self-colored varieties. They give fewer self-colored seedlings, however, which may be due to the action of one or more unknown heritable factors. I have not attempted to separate the self-colored from those showing color in the young shoots, but have classified both as colored varieties.

The results of selfing varieties with different color characters are as follows:

Selfed purples gave either all purples (one variety); purples, reds and colorless (two varieties); or purples and colorless (three varieties). Selfed reds gave either all reds (two varieties), or reds and colorless (two varieties). Selfed red varieties gave no purple progeny. Three colorless varieties (that is, no color in *either* the *tuber skin* or *young shoots*) were selfed, giving all colorless progeny.

Without considering factors for limiting color, these results seem to show that purple and red are separate Mendelian units, each dominant to its absence, and that purple is epistatic to red.

It is an interesting fact that although the purple varieties and the red varieties are distinct color types without intermediates and that mosaic varieties of each are known, yet in no case has a mosaic variety appeared in which splashes of the two colors are found. We may conclude therefore that the two colors are formed by the action of other factors upon the same chemical constituent. If we assume that the red color is a lower form of oxidation than the purple color and that they are produced by different oxidases R and P acting upon the same substance C, the results obtained are explained, for the presence of P would oxidize all of the stubstrate to the purple color.

Shape of Tuber

Potato tubers vary in shape from a length six times the median diameter to a length about the same as the median diameter. The varieties, the length of whose tubers is not over one and one-quarter times the medium diameter, I have called round. Two selfed round varieties gave only round progeny. Twelve varieties with oval tubers when selfed gave elongated, oval and round progeny. The ratio of other types to round was about nine to one. Either there is a series of factors for shape with the round type as the final subtraction form, or the oval types are heterozygotes of elongated and round. The latter interpretation is more likely to be correct, because oval types have been the popular market types for many years and therefore been used as parents in crosses.

Depth of Eyes

Shallow buds or eyes are required for profitable commercial varieties, yet from one fifteenth to one fourth of the progeny of ten selfed varieties were deep-eyed forms. Three selfed varieties gave no deep-eyed progeny. No progeny of deep-eyed seedlings were obtained, but it seems probable that this character is recessive to shallow eye.

The writer is fully aware that these few observations do not prove that the characters in which potato varieties differ all segregate in Mendelian proportions after crossing. A long series of crosses is necessary to analyze correctly the behavior in inheritance of such characters as shape. On the other hand, the color characters in stem, blossom and tuber are definite and discontinuous, and are alternative in inheritance. The chaotic appearance of the progeny of our commercial potatoes is only apparent. They readily fall into a simple classification and their exact behavior in inheritance could be readily determined if it were not for the difficulties attending successful crossing.

As the writer has previously stated,[3] certain characters pair with their own absence in crossing and these

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[3] "The Transmission of Variations in the Potato in Asexual Reproduction," Biennial Report, Conn Agr. Exp. Sta. 1909–1910, 119–161, 1910.

character pairs are the ones affected when a somatic mutation or bud variation occurs in asexual reproduction. Simple loss of the factor takes place. Segregation, therefore, takes place at other times than the reduction of the chromosomes.

In a previous paper,[4] the writer analyzed the data then extant concerning the hypothesis of degeneration or "running out" of potato varieties. The conclusion was that no degeneration due to continued asexual propagation occurs. No data have been obtained which refute this view, but the study of progeny of selfed potato varieties has suggested an explanation of a certain amount of diminution in yield after long-continued asexual propagation. All commercial potato varieties which have been selfed and their progeny grown, have proved to be heterozygous in at least two characters. It has been shown[5] that when maize biotypes are crossed, the F_1 generation has greater vigor and gives larger yields than the parents. It is a condition apart from inheritance, and is probably due to the heterozygous condition of certain characters in the germ cells. It may be correlated with the actual mechanical operation of segregation. Since potato varieties are retained in cultivation on the basis of yield and since those on the market have been found to be heterozygous in many of their characters, probably the same phenomenon is the cause. May there not be a gradual loss of the stimulus due to crossing through continued bud propagation, so that the variety has only the vigor of one homozygous in the same characters? The variety of course remains heterozygous for those characters in which it was originally heterozygous, yet there may be a gradual decline of the stimulus to cell division than it once possessed.

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[4] A "Study of the Factors influencing the Improvement of the Potato," Bull. Ill. Agr. Exp. Sta., No. 127, 375–456, 1908.

[5] East, E. M., "The Distinction between Development and Heredity in Inbreeding," AMER. NAT., 43: 173–181, 1909.

A MENDELIAN INTERPRETATION OF VARIATION THAT IS APPARENTLY CONTINUOUS

PROFESSOR EDWARD M. EAST

NEW YORK
1910

[Reprinted, without change of paging, from the AMERICAN NATURALIST 1910.]

[Reprinted from THE AMERICAN NATURALIST, Vol. XLIV., Feb., 1910.]

A MENDELIAN INTERPRETATION OF VARIATION THAT IS APPARENTLY CONTINUOUS[1]

PROFESSOR EDWARD M. EAST

HARVARD UNIVERSITY

THERE are two objects in writing this paper. One is to present some new facts of inheritance obtained from pedigree cultures of maize; the other is to discuss the hypotheses to which an extension of this class of facts naturally leads. This discussion is to be regarded simply as a suggestion toward a working hypothesis, for the facts are not sufficient to support a theory. They do, however, impose certain limitations upon speculation which should receive careful consideration.

The facts which are submitted have to do with independent allelomorphic pairs which cause the formation of like or similar characters in the zygote. Nilsson-Ehle[2] has just published facts of the same character obtained from cultures of oats and of wheat. My own work is largely supplementary to his, but it had been given these interpretations previous to the publication of his paper.

In brief, Nilsson-Ehle's results are as follows: He found that while in most varieties of oats with black

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[1] Contributions from the Laboratory of Genetics, Bussey Institution, Harvard University, No. 4. Read before the annual meeting of the American Society of Naturalists, Boston, December 29, 1909.

[2] Nilsson-Ehle, H. Kreuzungsuntersuchungen an Hafer und Weizen. Lunds Universitets Årsskrift, N. F. Afd. 2., Bd. 5, No. 2, 1909.

glumes blackness behaved as a simple Mendelian monohybrid, yet in one case there were two definite independent Mendelian unit characters, each of which was allelomorphic to its absence. Furthermore, in most varieties of oats having a ligule, the character behaved as a mono-hybrid dominant to absence of ligule, but in one case no less than four independent characters for presence of ligule, each being dominant to its absence, were found. In wheat a similar phenomenon occurred. Many crosses were made between varieties having red seeds and those having white seeds. In every case but one the F_2 generation gave the ordinary ratio of three red to one white. In the one exception—a very old red variety from the north of Sweden—the ratio in the F_2 generation was 63 red to 1 white. The reds of the F_2 generation gave in the F_3 generation a very close approximation to the theoretical expectation, which is 37 constant red, 8 red and white separating in the ratio of 63:1, 12 red and white separating in the ratio of 15:1, 6 red and white separating in the ratio of 3:1, and one constant white. He did not happen to obtain the expected constant white, but in the total progeny of 78 F_2 plants his other results are so close to the theoretical calculation that they quite convince one that he was really dealing with three indistinguishable but independent red characters, each allelomorphic to its absence. Nor can the experimental proof of the two colors of the oat glumes be doubted. The evidence of four characters for presence of ligule in the oat is not so conclusive.

In my own work there is sufficient proof to show that in certain cases the endosperm of maize contains two indistinguishable, independent yellow colors, although in most yellow races only one color is present. There is also some evidence that there are three and possibly four independent red colors in the pericarp, and two colors in the aleurone cells. The colors in the aleurone cells when pure are easily distinguished, but when they are together they grade into each other very gradually.

Fully fifteen different yellow varieties of maize have been crossed with various white varieties, in which the crosses have all given a simple mono-hybrid ratio. In the other cases that follow it is seen that there is a di-hybrid ratio.

No. 5–20, a pure white eight-rowed flint, was pollinated by No. 6, a dent pure for yellow endosperm. An eight-rowed ear was obtained containing 159 medium yellow kernels and 145 light yellow kernels. The pollen parent was evidently a hybrid homozygous for one yellow which we will call Y_1 and heterozygous for another yellow Y_2. The gametes Y_1Y_2 and Y_1 fertilized the white in equal quantities, giving a ratio of approximately one medium yellow to one light yellow. The F_2 kernels from the dark yellow were as follows:

TABLE I.[3]

F_2 SEEDS FROM CROSS OF No. 5–20, WHITE FLINT × No. 6 YELLOW DENT, HOMOZYGOUS FOR Y_1 AND HETEROZYGOUS FOR Y_2

Dark Seeds Heterozygous for Both Yellows Planted

Ear No.	Dark Y.	Light Y.	Total Y.	No Y.
1	270	56	326	29
2	101	215	316	27
3	261	52	313	28
5	273	284	557	35
10	358	117	475	25
12	296	72	368	19
13	207	156	363	35
14	387	102	489	29
Total	2153	1054	3207	227
Ratio			14.1	1

The ratios of light yellows to dark yellows is very arbitrary, for there was a fine gradation of shades. The ratio of total yellows to white, however, is unmistakably 15:1.

In the next table (Table II) are given the results of F_2 kernels from the light yellows of F_1. Only ear No. 8, which was really planted with the dark yellows, showed yellows dark enough to be mistaken for kernels containing

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[3] In these tables only hand pollinated ears are given.

both Y_1 and Y_2. The remaining ears are clearly monohybrids with reference to yellow endosperm.

TABLE II.

F_2 SEEDS FROM SAME CROSS AS SHOWN IN TABLE I

Light Yellow Seeds Heterozygous for Y_1 Planted

Ear No.	Dark Y.	Light Y.	No Y.
1		359	117
2		144	54
3		173	63
4		433	136
6		316	120
8	331		109
8a		229	86
9		325	115
10		227	87
11[4]		4	434
12		318	118
13		256	93
Total		3111	1098
Ratio		2.8	1

In a second case the female parent possessed the yellow endosperm. No. 11, a twelve-rowed yellow flint, was crossed with No. 8, a white dent. The F_2 kernels in part showed clearly a mono-hybrid ratio, and in part blended gradually into white. Two of these indefinite ears proved in the F_3 generation to have had the 15:1 ratio in the F_2 generation. Ear 7 of the F_2 generation calculated from the results of the entire F_3 crop must have had about 547 yellow to 52 white kernels, the theoretical number being 561 to 31. The hand-pollinated ears of the F_3 generation (yellow seeds) gave the results shown in Table III.

The F_3 generation grown from the other ear, Ear No. 8, showed that the ratio of yellows to whites in the F_2 generation was about 227 to 47. As the theoretical ratio is 257 to 17, the ratio obtained is somewhat inconclusive. A classification of the open field crop could not be made accurately on account of the light color of the yellows and

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[4] Discarded from average. This ear evidently grew from one kernel of the original white mother that was accidentally self-pollinated. The four yellow kernels all show zenia from accidental pollination in the next generation.

TABLE III.

NO. 11 YELLOW × NO. 8 WHITE

F_3 Generation from Yellow Seeds of F_2 Generation

Ear No.	Dark Y.	Light Y.	Total Y.	No Y.	Ratio They Approximate.
1	116	95	211	19	15Y : 1 no Y
14			88	5	15Y : 1 no Y
5	181	122			$3Y_1Y_2 : 1\ Y_{1 \text{ or } 2}$
4		253		68	3Y : 1 no Y
6		193		73	"
8		163		79	"
11		108		35	"
9		456			Constant $Y_{1 \text{ or } 2}$

the presence of many kernels showing zenia. Table IV, however, showing the hand-pollinated kernels of the interbred yellows of the F_2 generation, settles beyond a doubt the fact that the two yellows were present.

TABLE IV.

PROGENY OF EAR NO. 8 OF THE SAME CROSS AS SHOWN IN TABLE III

F_3 Generation from Yellow Seeds of F_2 Generation

Ear No.	Dark Y.	Light Y.	Total Y.	No Y.	Ratio They Approximate.
10	101	188	289	25	15Y : 1 no Y
11	89	219	308	23	15Y : 1 no Y
3		233			constant light Y
9	dark and light		331		3 dark : 1 light Y
13	dark and light		350		3 dark : 1 light Y
8		294		108	3 light : 1 no Y
15		221		87	3 light : 1 no Y
1[5]		197		203	

In a third case an eight-rowed yellow flint, No. 22, was crossed with a white dent, No. 8. Only four selfed ears were obtained in the F_2 generation. Ear 1 had 72 yellow to 37 white kernels. This ear was poorly developed and undoubtedly had some yellow kernels which were classed as whites. Ear 4 had 158 yellow and 42 white kernels. It is very likely that both of these ears were mono-hybrids, but the F_3 generation was not grown. Ear 5 had 148 yellow and 15 white kernels. Ear 7 had 78 yellow and 5 white kernels. It seems probable that both of these ears

[5] Kernel from which this ear grew was evidently pollinated by no Y.

were di-hybrids, but only Ear 5 was grown another generation. The kernels classed as white proved to be pure; the open field crop from the yellow kernels gave 14 pure yellow ears and 14 hybrid yellow. Theoretically the ratio should be 7 pure yellows (that is, pure for either one or both yellows) and 8 hybrid yellows (4 giving 15 yellows to 1 white and 4 giving 3 yellows to 1 white). Five hand-pollinated selfed ears were obtained. Three of these gave mono-hybrid ratios, with a total of 607 yellows to 185 white kernels. One ear was a pure dark yellow (probably $Y_1Y_1Y_2Y_2$). The other ear was poorly filled, but had 27 dark yellows (probably Y_1Y_2) and 7 light yellow kernels (Y_1 or Y_2). Unfortunately no 15:1 ratio was obtained in this generation, but this is quite likely to happen when only five selfed ears are counted. The gradation of colors and the general appearance of the open field crop, however, lead me to believe that we were again dealing with a di-hybrid.

Two yellows appeared in still another case, that of white sweet No. 40 ♀ × yellow dent No. 3 ♂. Only one selfed ear was obtained in the F_2 generation giving 599 yellow to 43 white kernels. Of these kernels 486 were starchy and 156 sweet, which complicated matters in the F_3 generation because it was very difficult to separate the light yellow sweet from the white sweet kernels. Among the selfed ears were three pure to the starchy character, and in these ears the dark yellows, the light yellows and whites stood out very distinctly. Ear 12 had 156 dark yellow; 47 light yellow; 14 white kernels. Ear 13 had 347 dark yellow; 93 light yellow; 25 white kernels. The third starchy ear, No. 6, had 320 light yellow; 97 white kernels. Two ears, therefore, were di-hybrids, and one ear a mono-hybrid.

The ears which were heterozygous for starch and no starch and those homozygous for no starch, could not all be classified accurately, but it is certain that some pure dark yellows, some pure light yellows, some showing segregation of yellows and whites at the ratio 15:1, and some showing segregation of yellows and whites at the ratio of 3:1, were obtained,

One other case should be mentioned. One ear of a dent variety of unknown parentage obtained for another purpose was found to have some apparently heterozygous yellow kernels. Seven selfed ears were obtained from them, of which two were pure yellow. The other five ears each gave the di-hybrid ratio. There was a total of 1906 yellow seeds to 181 white seeds, which is reasonably close to the expected ratio, 1956 yellow to 131 white.

It is to be regretted that I can present no other case of this class that has been fully worked out, although several other characters which I have under observation in both maize and tobacco seem likely to be included ultimately. Nevertheless, the fact that we have to deal with conditions of this kind in studying inheritance is established; granting only that they will be somewhat numerous, it opens up an entirely new outlook in the field of genetics.

In certain cases it would appear that we may have several allelomorphic pairs each of which is inherited independently of the others, and each of which is separately capable of forming the same character. When present in different numbers in different individuals, these units simply form quantitative differences. It may be objected that we do not know that two colors that appear the same physically are exactly the same chemically. That is true; but Nilsson-Ehle's case of several unit characters for presence of ligule in oats is certainly one where each of several Mendelian units forms exactly the same character. It may be that there is a kind of biological isomerism, in which, instead of molecules of the same formula having different physical properties, there are isomers capable of forming the same character, although, through difference in construction, they are not allelomorphic to each other. At least it is quite a probable supposition that through imperfections in the mechanism of heredity an individual possessing a certain character should give rise to different lines of descent so that in the F_n generation when individuals of these different lines are crossed, the character behaves as a di-hybrid instead of as a mono-hybrid. In other words, it is more probable that these units arise through variation in different individuals and are combined by hybridization, than that actually different structures for forming the same character arise in the same individual.

On the other hand, there is a possibility of an action just the opposite of this. Several of these quantitative units which produce the same character may become attached like a chemical radical and again behave as a single pair. Nilsson-Ehle gives one case which he does not attempt to explain, where the same cross gave a 4:1 ratio in one instance and 8.4:1 ratio in another instance. In his other work characters always behaved the same way; that is, either as one pair, two pairs, three pairs, etc. In my work, the yellow endosperm of maize has behaved differently in the same strain, but it is probably because the yellow parent is homozygous for one yellow and heterozygous for the other. They were known to be pure for one yellow, but it would take a long series of crosses to prove purity in two yellows.

Let us now consider what is the concrete result of the inter-action of several cumulative units affecting the same character. Where there is simple presence dominant to absence of a number n of such factors, in a cross where all are present in one parent and all absent in the other parent, there must be 4^n individuals to run an even chance of obtaining a single F_2 individual in which the character is absent. When four such units, $A_1A_2A_3A_4$ are crossed with $a_1a_2a_3a_4$, their absence, only one pure recessive is expected in 256 individuals. And 256 individuals is a larger number than is usually reported in genetic publications. When a smaller population is considered, it will appear to be a blend of the two parents with a fluctuating variability on each side of its mode. Of course if there is absolute dominance and each unit appears to affect the zygote in the same manner that they do when combined, the F_2 generation will appear like the dominant parent unless a very large number of progeny are under observation and pure recessives are obtained. This may be an explanation of the results obtained by Millardet; it is certainly as probable as the hypothesis of the non-formation of homozygotes. Ordinarily, however, there is not perfect dominance, and variation due to heterozygosis combined with fluctuating variation makes it almost impossible to classify the individuals except by breeding. The two yellows in the endosperm of maize is an example of how few characters are necessary to make classification difficult. First, there is a small amount of fluctuation in different ears due to varying light conditions owing to differences in thickness of the husk; second, all the classes having different gametic formulæ differ in the intensity of their yellow in the following order, $Y_1Y_1Y_2Y_2$, $Y_1y_1Y_2Y_2$ or $Y_1Y_1Y_2y_2$, Y_1Y_1, Y_2Y_2, Y_1y_1, Y_2y_2, y_1y_2. As dominance becomes less and less evident, the Mendelian classes vary more and more from the formula $(3+1)^n$, and approach the normal curve, with a regular gradation of individuals on each side of the mode. When there is no dominance and open fertilization, a state is reached in which the curve of variation simulates the fluctuation curve, with the difference that the gradations are heritable.

One other important feature of this class of genetic facts must be considered. If units $A_1A_2A_3a_4$ meet units $a_1a_2a_3A_4$, in the F_2 generation there will be one pure recessive, $a_1a_2a_3a_4$, in every 256 individuals. This explains an apparent paradox. Two individuals are crossed, both seemingly pure for presence of the same character, yet one individual out of 256 is a pure recessive. When we consider the rarity with which pure dominants or pure recessives (for all characters) are obtained when there are more than three factors, we can hardly avoid the suspicion that here is a perfectly logical way of accounting for many cases of so-called atavism. Furthermore, many apparently new characters may be formed by the gradual dropping of these cumulative factors without any additional hypothesis. For example, in *Nicotiana tabacum* varieties there is every gradation[6] of loss of leaf surface near the base of the sessile leaf, until in *N. tabacum fruticosa* the leaf is only one step removed from a petioled condition. If this step should occur the new plant would almost certainly be called a new species; yet it is only one degree further in a definite series of loss gradations that have already taken place. If it should be assumed that in other instances slight qualitative as well as quantitative changes take place as units are added, then it becomes very easy, theoretically, to account for quite different characters in the individual homozygous for presence of all dominant units, and in the individual in which they are all absent.

Unfortunately for these conceptions, although I feel it extremely probable that variations in *some* characters that seem to be continuous will prove to be combinations of segregating characters, it is exceedingly difficult to demonstrate the matter beyond a reasonable doubt. As an illustration of the difficulties involved in the analysis of pedigree cultures embracing such characters, I wish to discuss some data regarding the inheritance of the number of rows of kernels on the maize cob.

The maize ear may be regarded as a fusion of four or more spikes, each joint of the rachis bearing two spikelets. The rows are, therefore, distinctly paired, and no case is known where one of the pair has been aborted. This is a peculiar fact when we consider the great number of odd kinds of variations that occur in nature. The number of rows per cob has been considered to belong to continuous variations by DeVries, and a glance at the progeny from the seeds of a single selfed ear as shown in Table V seems to confirm this view.

There is considerable evidence, however, that this character is made up of a series of cumulative units, inde-

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[6] It is not known at present how this character behaves in inheritance.

TABLE V.

PROGENY OF A SELFED EAR OF LEAMING MAIZE HAVING 20 ROWS

Classes of rows	12	14	16	18	20	22	24	26	28	30
No. of ears	1	0	5	4	53	35	19	5	2	1

pendent in their inheritance. There is no reason why it should not be considered to be of the same nature as various other size characters in which variation seems to be continuous, but in which relatively constant gradations may be isolated, each fluctuating around a particular mode. But this particular case possesses an advantage not held by most phenomena of its class, in that there is a definite discontinuous series of numbers by which each individual may be classified.

Previous to analyzing the data from pedigree cultures, however, it is necessary to take into consideration several facts. In the first place, what limits are to be placed on fluctuations?[7] From the variability of the progeny of single ears of dent varieties that have been inbred for several generations, it might be concluded that the deviations are very large. But this is not necessarily the case; these deviations may be due largely to gametic structure in spite of the inbreeding, since no conscious selection of homozygotes has been made. There is no such variation in eight-rowed varieties, which may be considered as the last subtraction form in which maize appears and therefore an extreme homozygous recessive. In a count of the population of an isolated maize field where Longfellow, an eight-rowed flint, had been grown for many years, 4 four-rowed, 993 eight-rowed, 2 ten-rowed and 1 twelve-rowed ears were found. Only seven aberrant ears out of a thousand had been produced, and some of these may have been due to vicinism.

On the other hand a large number of counts of the number of rows of both ears on stalks that bore two ears has shown that it is very rare that there is a change

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[7] The word fluctuation is used to designate the somatic changes due to immediate environment, and which *are not inherited*.

greater than ± 2 rows. If conditions are more favorable at the time when the upper ear is laid down it will have two more rows than the second ear; if conditions are favorable all through the season, the ears generally have the same number of rows; while if conditions are unfavorable when the upper ear is laid down, the lower ear may have two more rows than the upper ear. Furthermore, seeds from the same ear have several times been grown on different soils and in different seasons, and in each case the frequency distribution has been the same. Hence it may be concluded that in the great majority of cases fluctuation is not greater than in ± 2 rows, although fluctuations of ± 4 rows have been found.

A second question worthy of consideration is: Do somatic variations due to varying conditions during development take place with equal frequency in individuals with a large number of rows and in individuals with a small number of rows? From the fact that several of my inbred strains that have been selected for three generations for a constant number of rows, increase directly in variability as the number of rows increases, the question should probably be answered in the negative. This answer is reasonable upon other grounds. The eight-rowed ear may vary in any one of four spikes, the sixteen-rowed ear may vary in any one of eight spikes; therefore the sixteen-rowed ear may vary twice as often as the eight-rowed ear. By the same reasoning, the sixteen-rowed ear may sometimes throw fluctuations twice as wide as the eight-rowed ear.

A third consideration is the possibility of increased fluctuation due to hybridization. Shull[8] and East[9] have shown that there is an increased stimulus to cell division when maize biotypes are crossed—a phenomenon apart from inheritance. There is no evidence, however, that

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[8] Shull, G. H., "A Pure-line Method in Corn Breeding," *Rept. Amer. Breeders' Assn.*, 5, 51–59, 1909.

[9] East, E. M., "The Distinction between Development and Heredity in Inbreeding," Amer. Nat., 43, 173–181, 1909.

increased gametic variability results. Johannsen[10] has shown that there is no such increase in fluctuation when close-pollinated plants are crossed. I have crossed several distinct varieties of maize where the modal number of rows of each parent was twelve, and in every instance the F_1 progeny had the same mode and about the same variability.

Finally, a possibility of gametic coupling should be considered. Our common races of flint maize all have a low number of rows, usually eight but sometimes twelve; dent races have various modes running from twelve to twenty-four rows. When crosses between the two subspecies are made, the tendency is to separate in the same manner.

Attention is not called to these obscuring factors with the idea that they are universally applicable in the study of supposed continuous variation. But there are similar conditions always present that make analysis of these variations difficult, and the facts given here should serve to prevent premature decision that they do not show segregation in their inheritance.

Table VI shows the results from several crosses between maize races with different modal values for number of rows. Several interesting points are noticeable. The modal number is always divisible by four. This is also the case with some twenty-five other races that I have examined but which are not shown in the table. I suspect that through the presence of pure units zygotes having a multiple of four rows are formed, while heterozygous units cause the dropping of two rows. The eight-rowed races are pure for that character, the twelve-rowed races vary but little, but the races having a higher number of rows are exceedingly variable.

When twelve-rowed races are crossed with those having eight rows, the resulting F_1 generation always—or nearly

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[10] Johannsen, W., "Does Hybridization Increase Fluctuating Variability?" *Rept. Third Inter. Con. on Genetics*, 98–113, London, Spottiswoode, 1907.

TABLE VI.

CROSSES BETWEEN MAIZE STRAINS WITH DIFFERENT NUMBERS OF ROWS

Parents. (Female Given First.)	Gen.	Row Classes.						
		8	10	12	14	16	18	20
Flint No. 5		100						
Flint No. 11		1	4	387	7	1		
Flint No. 24		100						
Flint No. 15		100						
Dent No. 6				6	31	51	18	4
Dent No. 8			3	54	36	12	2	
Sweet No. 53[11]		1	5	25	4			
Sweet No. 54[11]		25	2	1				
No. 5 × No. 53	F_1	1	7	13				
No. 5 × No. 6	F_1	11	18	27	3			
No. 11 × No. 5	F_1	2	4	18				
No. 11 × No. 53	F_1	2	5	17				
No. 24 × No. 53	F_1	57	8	3				
No. 15 × No. 8	F_1	1	14	26	3	1		
No. 15 × No. 8 (from 10-row ear)	F_2	14	15	28	9	1		
No. 15 × No. 8 (from 12-row ear)	F_2	4	13	25	6	3		
No. 8 × No. 54	F_1	1	6	14				
No. 8 × No. 54 (from 12-row ear)	F_2	11	25	38	2	1		

always—has the mode at twelve rows. In one case cited in Table VI, No. 24 × No. 53, nearly all the F_1 progeny were eight-rowed. It might appear from this, either that the low number of rows was in this case dominant, or that the female parent has more influence on the resulting progeny than the male parent. I prefer to believe, however, that the individual of No. 53 which furnished the pollen was due to produce eight-rowed progeny. Unfortunately no record was kept of the ear borne by this plant, but No. 53 sometimes does produce eight-rowed ears.

When a race with a mode higher than twelve is crossed with an eight-rowed race, the F_1 generation is always intermediate, although it tends to be nearer the high-rowed parent. Only one example is given in the table, but it is indicative of the class. These results are rather confusing, for there seems to be a tendency to dominance in the twelve-rowed form that is not found in the forms with a higher number of rows. I have seen cultures of other investigators where 12-row × 8-row resulted in a

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[11] Approximately.

ten-rowed F_1 generation, so the complication need not worry us at present.

The results of the F_2 generation show a definite tendency toward segregation and reproduction of the parent types. I might add that in at least two cases I have planted extracted eight-rowed ears and have immediately obtained an eight-rowed race which showed only slight departures from the type. Selection from those ears having a high number of rows has also given races like the high-rowed parent without recrossing with it. It is regretted that commercial problems were on hand at the time and no exact data were recorded. It can be stated with confidence, however, that ears like each parent are obtained in the F_2 generation, from which with care *races* like each parent may be produced. *Segregation seems to be the best interpretation of the matter.*

These various items may seem disconnected and uninteresting, but they have been given to show the tangible basis for the following theoretical interpretation. No hard and fast conclusion is attempted, but I feel that this interpretation with possibly slight modifications will be found to aid the explanation of many cases where variation is apparently continuous.

Suppose a basal unit to be present in the gametes of all maize races, this unit to account for the production of eight rows. Let additional independent interchangeable units, each allelomorphic to its own absence, account for each additional four rows; and let the heterozygous condition of any unit represent only half of the homozygous condition, or two rows. Then the gametic condition of a homozygous twenty-rowed race would be $8 + AABBCC$, each letter actually representing two rows. When crossed with an eight-rowed race, the F_2 generation will show ears of from eight to twenty rows, each class being represented by the number of units in the coefficients in the binomial expansion where the exponent is twice the number of characters, or in this case $(a + b)^6$.

The result appears to be a blend between the characters of the two parents with a normal frequency distribution of the deviants. Only one twenty-rowed individual occurs in 64 instead of the 27 expected by the interaction of three dominant factors in the usual Mendelian ratios. The remainder of the 27 will have different numbers of rows, and, by their gametic formulæ, different expectations in future breeding as follows:

1 $AABBCC = 20$ rows.
2 $AaBBCC = 18$ rows.
2 $AABbCC = 18$ rows.
2 $AABBCc = 18$ rows.
4 $AaBbCC = 16$ rows.
4 $AaBBCc = 16$ rows.
4 $AABbCc = 16$ rows.
8 $AaBbCc = 14$ rows.

There are four visibly different classes and eight gametically different classes. It must also be remembered that the probability that the original twenty-rowed ear in actual practise may have had more than three units in its gametes has not been considered. This point is illustrated clearly if we work out the complete ratio for the three characters, and note the number of gametically different classes which compose the modal class of fourteen rows in Table VII. It actually contains seven gametically different classes and not a single homozygote. If this conception of independent allelomorphic pairs affecting the same character proves true, it will sadly upset the biometric belief that the modal class is *the type* around which the variants converge, for there is actually less chance of these individuals breeding true than those from *any other* class.

TABLE VII

THEORETICAL EXPECTATION IN F_2 WHEN A HOMOZYGOUS TWENTY-ROWED MAIZE EAR IS CROSSED WITH AN EIGHT-ROWED EAR

Classes	8	10	12	14	16	18	20
No. ears	1	6	15	20	15	6	1

The conception is simple and is capable theoretically of bringing in order many complicated facts, although the presence of fluctuating variation will be a great factor in preventing analysis of data. I have thought of only one fact that is difficult to bring into line. If $8AA$, $8BB$ and $8CC$ all represent homozygous twelve-rowed ears—to continue the maize illustration—and none of these factors are allelomorphic to each other, sixteen-rowed ears should sometimes be obtained when crossing two twelve-rowed ears. I am not sure but that this would happen if we were to extract all the homozygous twelve-rowed strains after a cross between sixteen-row and eight-row, and after proving their purity cross them. In some cases the additional four-row units would probably be allelomorphic to each other and in other cases independent of each other. On the other hand, this is only an hypothesis, and while I have faith in its foundation facts, the details may need change.

Castle has raised the point that greater variation should be expected in the F_1 generation than in the P_1 generations when crossing widely deviating individuals showing variation apparently continuous. If the parents are strictly pure for a definite number of units, say for size, a greater variation should certainly be expected in the F_1 generation after crossing. But considering the difficulties that arise when even five independent units are considered, can it be said that anything has heretofore been known concerning the actual gametic status of parents which it is known do vary in the character in question and in which the variations are inherited, for the race can be changed by selection within it. It may be, too, that the correct criterion has not been used in size measurements, for, as others have suggested, solids vary as the cube root of their mass, whereas the sum of the weights of the body cells has usually been measured and compared directly with similar sums.

Attention should be called to one further point. Many characters in all probability are truly blending in their inheritance, but there is another interpretation which may apply in certain cases. I have repeatedly tried to cross Giant Missouri Cob Pipe maize (14 feet high) and Tom Thumb pop maize (2 feet high), but have always failed. They both cross readily with varieties intermediate in size, but are sterile between themselves. We may imagine that the gametes of each race, though varying in structure, are all so dissimilar that none of them can unite to form zygotes. Other races may be found where only part of the gametes of varying structure are so unlike that they will not develop after fusion. The zygotes that do develop will be from those more alike in construction. An apparent blend results, and although segregation may take place, no progeny as extreme as either of the parents will ever occur.

I may say in conclusion that the effect of the truth of this hypothesis would be to add another link to the increasing chain of evidence that the word mutation may properly be applied to any inherited variation, however small; and the word fluctuation should be restricted to those variations due to immediate environment which do not affect the germ cells, and which—it has been shown—are not inherited. In addition it gives a rational basis for the origin of *new* characters, which has hitherto been somewhat of a Mendelian stumbling-block; and also gives the term unit-character less of an irrevocably-fixed-entity conception, which is more in accord with other biological beliefs.

THE GENOTYPE HYPOTHESIS AND HYBRIDIZATION

PROFESSOR E. M. EAST

Reprinted from The American Naturalist, Vol. XLV., March, 1911.

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THE GENOTYPE HYPOTHESIS AND HYBRIDIZATION[1]

PROFESSOR E. M. EAST
HARVARD UNIVERSITY

It sometimes seems as if the hypercritical attitude had become an obsession among biologists. A proper judicial spirit is of course essential to science, but do not biologists often require a large amount of affirmative data before assenting to a proposition which is in reality a simple corollary of one already accepted?

For example, Darwin emphasized small quantitative variations as the method of evolution, although he recognized the occurrence of larger changes both quantitative and qualitative. De Vries, on the other hand, emphasized large variations—especially qualitative variations—as the real basis of evolution, although he too admitted the existence of lesser changes. He distinctly states that a mutation or new basis for fluctuating variation, may be so small that it is obscured by the fluctuations themselves.

If relative frequency of occurrence is a criterion of the value of variations in organic evolution, which is not necessarily so, Darwin's point of view is probably the nearer correct. If one could find a unit basis for describing variations in terms of the physiological economy of the organism concerned, *i. e.,* if one knew exactly what was a large change and what was a small change, he would probably find that a random sample of inherited variations followed the normal curve of error. By this I

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[1] Read at the symposium on the "Genotype Hypothesis" at the meeting of the American Society of Naturalists, Ithaca, N. Y., December 28, 1910.

Contribution from the Laboratory of Genetics, Bussey Institution of Harvard University.

The experimental results are from cooperative work between the Connecticut Agricultural Experiment Station and the Bussey Institution of Harvard University.

mean that small variations would center closely around a mode, and large variations would occur with a relative frequency inversely proportional to their size. The point that I wish to emphasize, however, is that neither Darwin nor De Vries recognized the proper distinction between a mutation and a fluctuation. Darwin made no distinction. De Vries, however, considered fluctuations to be linear; that is, to be limited to increase and decrease in characters already present. He thought that selection of such variations brought about changes in the selected population due to the inheritance of the fluctuations, but that the selected populations returned to the mean of the general population after selection ceased. Mutations, on the other hand, were gains or losses of entire characters—qualitative changes—which were transmitted completely, *i. e.,* were constant, from the beginning. De Vries did indeed state that mutations could take place in any direction, which would involve the idea of linear change or quantitative mutations; yet it seems quite evident from his general attitude in "Die Mutationstheorie" that to his mind qualitative and quantitative variations were quite distinct.

Many practical breeders had long known, however, that the selection of linear variations often produced new races which were as constant as any races, provided they were not exposed to crossing with individuals of the general population from which the selected race had come. Why this was true was unknown. It was felt that there was a real distinction between certain variations, to which Darwin had not called attention; yet it was felt that the De Vriesian idea was not wholly correct. It has been in making this distinction clear-cut and definite that Johannsen has rendered his great service. His elaborate extensions of the genotype conception of heredity have cleared up many debated points, and corroborative evidence has been received from so many lines that it can hardly be doubted that the main points of the hypothesis are correct. It may seem, therefore, as if the superstructure of this conception were too elaborate to rest upon a simple foundation; yet I can not see but that the basis of the entire hypothesis is the fact that a fluctuation is a non-inherited variation produced upon the soma by environmental conditions, while the inherited variation, the mutation if you will, is any variation qualitative or quantitative, that is germinal in character. This being so, it seems scarcely necessary for an elaborate proof of the proposition, for it is nothing but a corollary to that part of Weismannism which was already generally accepted.

Of course it is recognized that pure Lamarckism still has followers to whom neither Weismannism in any form nor the genotype conception of heredity could appeal. But to thorough Weismannians and to those who believe in occasional germinal response to environmental conditions, it seems as if both propositions must be acceptable and their interdependence apparent.

Let us follow this line of reasoning to its logical conclusion in regard to the physiology of heredity. The Mendelian notation has been generally accepted as a convenient way of accounting for the facts of heredity in certain markedly discontinuous characters. It has been questioned by many, however, whether the Mendelian conception is not rather an apparent interpretation of a relatively small number of facts than a general law. De Vries has even suggested that there are definite physiological reasons why certain characters should Mendelize and others should not. His idea is that Mendelian segregation occurs when a germinal determinant for a character (*Anlage*) meets an opposing determinant, and when no such opposition exists the character in the cross-bred organism breeds true. Now the universal tendency of the facts of breeding is towards an interpretation the opposite of this. When a determinant from one parent meets with no such determinant from the other parent (presence and absence hypothesis), Mendelian segregation appears. When the same determinant is received from both parents, segregation can not be proved, for the character breeds true.

In fact the many results of experimental breeding during the past few years have convinced me that De Vries's general conception of this matter is incorrect. There may be finally a considerable modification of our ideas regarding the ultimate nature of Mendelian unit characters and the exact meaning of segregation, yet the universal applicability of a strict Mendelian system to interpret the facts of heredity becomes more and more apparent every day. And the point that I wish to emphasize is that Mendelian inheritance is a genuine corollary of the genotype hypothesis if the latter is applicable to a population in a state of natural hybridity. In my work with maize where free intercrossing does occur I am convinced of the existence of genotypes in a state of natural hybridization. Furthermore, these genotypes can be isolated by inbreeding. If it were true, then, that only certain markedly discontinuous characters such as color Mendelize, how could genotypes which differ from each other in size characters be isolated? It is not expected, however, that the statement that Mendelian inheritance and the genotype hypothesis are interdependent will be received without proof. Data that are believed to furnish such proof are submitted here.

When Mendelism was a new idea it was natural that the behavior of many hybrids should be regarded as irreconcilable to such a system of interpretation. The earlier criticisms arose largely through the misconception that dominance instead of segregation was its essential feature. Later, when so many complex results from pedigree cultures were fitted into a strict and simple Mendelian notation, it was objected that the investigators could by expert juggling of a sufficient number of factors interpret according to their system any experimental results they might obtain. Perhaps a few biologists regarded as a personal affront the gradual growth of the idea that the facts of heredity were complex, but it is hardly likely that many could regard this complexity as an invention of Mendelians. The latter would only too gladly have the facts as simple as possible.

There have remained, however, several instances in which hybrids apparently did not segregate in the F_2 generation. Mendel himself investigated one such case, the genus *Hieracium*. The investigation of Ostenfeld[2] made this case perfectly clear by showing that the hybrids reproduced apogamously. Such asexual reproduction may also explain the behavior of hybrids between species of brambles which are also said to breed true in all their characters. These cases, however, and others among animals of which human skin color is the example *par excellence,* may be left out of consideration because no exact data concerning them have been forthcoming. There remain the experiments of two careful investigators who observed no segregation in the F_2 generations of their hybrids, those of Lock[3] upon heights of maize plants and those of Castle[4] upon weights and ear lengths of rabbits. Lock expected that if segregation occurred it would be into two classes, *i. e.,* simple mono-hybridism. For this reason he made no measurements which would show whether he obtained the kind of segregation which as is shown later in this paper, does occur in maize hybrids. Castle[5] has recently admitted the possibility that his numbers were not large enough to prove definitely that segregation involving several small unit characters does not occur in the ear length of rabbits.

The difficulty attending this earlier work was that there was no way of explaining different manifestations of the same character. Segregating characters could always be interpreted either as the presence and absence of a unit

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[2] Ostenfeld, C. H., 1904, ''Zur Kenntnis der Apogamie in der Gattung *Hieracium,*'' *Ber. Deutsch. Bot. Ges.,* 22: 7.

[3] Lock, R. H., 1906, '' Studies in Plant Breeding in the Tropics,'' III, Experiments with Maize, *Ann. Roy. Bot. Gard. Peradeniya,* 2: 95–184.

[4] Castle, W. E., *et al.,* 1909, ''Studies of Inheritance in Rabbits,'' Carnegie Inst. Wash. Pub., 114: 5–70.

[5] In lectures at the Lowell Institute, Boston, Mass., 1910.

giving a 3:1 ratio, or as the complemental action of two different units each allelomorphic to its absence, giving 9:3:3:1 ratios or modifications of them. Nillson-Ehle[6] and the writer,[7] however, have shown that several units each allelomorphic to its own absence may be the determinants of what appears to the eye as a single character. In the above paper the writer suggested that if such ratios as 15:1 and 63:1—di-hybrid and tri-hybrid ratios, respectively—were found in considerable numbers, then higher ratios of this kind might account for the apparent constancy of hybrids in characters that seemed to be continuous. For, if—as is quite probable—the additional units increase the activity of the character in question, and if there is no dominance,[8] it is quite evident that hybrids may be intermediate between the two parents. All the pure classes in a complex character of this kind would indeed be difficult to isolate, but segregation could be absolutely proved by a comparison of the variability of the F_1 and F_2 generations.

Since writing the above paper I have obtained clear evidence of 15:1 ratios in two other cases. The first is a red pericarp color, the second is the condition of endosperm in maize which gives dented seeds as distinct from that which gives flinty seeds. There is even considerable probability that higher ratios occur which affect the latter character. In another paper[9] I have shown photographic evidence of size segregation in varieties of *Nicotiana rustica* and stated that similar evidence of segregation of size character in maize had been obtained. The following figures and tables show sufficient of the evidence from the maize crosses to demonstrate conclusively

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[6] Nillson-Ehle, H., 1909, "Kreuzungsuntersuchungen an Hafer und Weizen," *Lunds Universitets Årsskrift*, N. F., Afd. 2., Bd. 5, Nr. 2, 1–122.

[7] East, E. M., 1910, "A Mendelian Interpretation of Variation that is Apparently Continuous," Amer. Nat., 44: 65–82.

[8] One dose, *i. e.*, receiving the same gene from a single parent, would on the average increase the manifestation of the character half as much as two doses.

[9] East, E. M., 1910. "The Rôle of Hybridization in Plant Breeding," *Pop. Sci. Mon.*, Oct., 1910, pp. 342–354.

that size characters segregate. It is hoped that this evidence will make us more cautious about accepting uncorroborated statements about characters which are definite exceptions to the Law of Mendel. It is by no means certain that no such exist, but no experimental proof of hybrids non-Mendelian in character has been made.

A further proof of segregation of size characters has recently been made in a preliminary note by Emerson.[10] He states that definite segregation occurs in beans, gourds, squashes and maize. His full data are therefore awaited with great interest.

Table I shows the frequency distribution of the heights of plants in a cross between no. 5 a medium-sized flint maize and no. 6 a tall dent maize. Sufficient seed was obtained in a previous season so that the entire series could be grown in rows side by side during one summer. This procedure eliminates any possibility that the variability of the F_2 generation might have come from varying conditions of soil fertility.

It will be noticed that the F_1 generation is nearly as tall as the taller parent. This increase in size is not due to dominance. It is the increased vigor that comes from crossing in maize, and while it obscures the hereditary differences in size, it is really a problem of development and not of heredity as was shown in a previous paper.[11]

The distribution of heights in the F_2 generation is seen by simple inspection of the table to be more variable than the F_1 generation in the case of each ear planted. Reduced to simple terms by the calculation of the coefficient of variation in each case, however, the two generations can be compared more accurately. In the F_1 generation the C.V. $= 8.68 \pm .553$ while in the various F_2 generations from different ears the coefficients of variation run from $12.02 \pm .559$ to $15.75 \pm .684$.

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[10] Emerson, R. A., 1910, "Inheritance of Sizes and Shapes in Plants," Amer. Nat., 44: 739–746.

[11] East, E. M., 1909, "The Distinction between Development and Heredity in Inbreeding," Amer. Nat., 43: 173–181.

TABLE I[12]

Frequency Distribution of Heights of Maize Plants in Cross (5 × 6)

No.	Class Centers in Inches for Heights of Plants																					A.	S.D.	C.V.
	51	54	57	60	63	66	69	72	75	78	81	84	87	90	93	96	99	102	105	108	111			
5	3	2	4	5	11	17	17	18	10	6	4											68.22 ± .406	6.49 ± .324	9.51 ± .421
6													1	2	2	13	14	16	14	8	3	101.18 ± .400	5.07 ± .283	5.01 ± .279
(5 x 6)- F_1											5	4	3	9	9	6	5	4	4	7		94.53 ± .740	8.21 ± .519	8.68 ± .553
(5 x 6)- 1 F_2	2	1	3	6	3	5	10	10	22	16	10	7	11	13	6	10	8	4	4	2		81.00 ± .685	12.76 ± .486	15.75 ± .684
(5 x 6)- 2 F_2				2		2	4	2	6	5	12	8	12	6	5	8	3	10	8	2	2	88.33 ± .802	11.78 ± .567	13.34 ± .675
(5 x 6)- 8 F_2	2				4	4	12	7	9	13	12	15	10	8	8	1	1	1				79.46 ± .621	9.55 ± .438	12.02 ± .559
(5 x 6)-14 F_2		1	2	4	2	7	8	17	21	11	27	24	16	9	11	10	5	4	2			81.24 ± .513	10.28 ± .364	12.65 ± .450

TABLE II[13]

Frequency Distribution of Heights of Plants in Cross (54 × 60)

No.	Class Centers in Inches for Heights of Plants																							Total
	22	25	28	31	34	37	40	43	46	49	52	55	58	61	64	67	70	73	76	79	82	85	88	91	
No. 54													2	6	10	17	24	26	21	14	14	7	3	1	145
No. 60	←	—	—	—	—	—	→[14]																		100
(60–5 x 54) F_1														←	—	—	—	—	—	—	→[14]				55
(60–8 x 54) F_1											←	—	—	—	—	—	—	—	—	—	—	→[14]			26
(60–3 x 54) F_1												←	—	—	—	—	—	—	—	—	—	→[14]			30
(60–5 x 54) F_2[15]						2	2	1	8	18	26	44	95	117	126	92	59	31	22	3	3				649
(60–8 x 54) F_2[16]						1		1	5	6	13	13	35	29	36	27	21	20	9	6	4	1	1		228
(60–3 x 54) F_2[17]						5	15	13	32	31	68	60	72	34	47	14	13	5							409

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[12] Grown side by side in same season.

[13] Distributions giving figures grown side by side in same season.

[14] Extremes were measured and number of plants counted. All were strongly convergent around the central classes.

[15] Grown from five different ears.

[16] Grown from two different ears.

[17] Grown from three different ears.

Table II shows a similar distribution of heights in cross between no. 60, a dwarf pop maize commonly known as Tom Thumb, and no. 54, a sugar corn known as Black Mexican. The distribution of heights of no. 54 was obtained in the same season as the F_2 generation. They were both grown upon the same plot of ground in which the soil appeared to be quite uniform. Unfortunately, the exact distribution of the heights of no. 60 and of the F_1 plants which were grown in previous seasons, is unknown. The range of the variates shown by the black lines, however, is correct. Furthermore, from notes recorded at the time we know that the F_1 generation was comparatively uniform, the greater number of variates being distributed around classes 67, 70 and 73 inches. In this case, also, the effect of crossing is shown by the relatively high plants of this generation. The plants of the F_2 generation show a wide range of variation. The highest individuals are practically the height of the highest individuals of the taller parent, no. 54. The lowest plants of F_2 do not reach the lower range of no. 60. I interpret this as due to continued heterozygosis in other characters and to physiological correlation. By the latter term I mean that since the plants of no. 60 are very small, F_2 segregates of the same size could only be expected where the ears and seeds also are very small. But since the ears and seeds of these plants also show segregation in new combinations, normal growth correlation probably resulted in a somewhat larger average size. For example, little 40-inch plants were found with ears three times the length of normal ears of no. 60. It is likely that such plants might have been smaller if they had been recombined with the characters necessary for the production of smaller ears.

Table III and Figs. 1–4, show the lengths of ears in the cross just described. In making this table the best ear from each plant that bore a well-filled ear was taken. The small ears, therefore, do not represent poor, unfilled or supernumerary ears. The coefficients of variability

TABLE III[18]

FREQUENCY DISTRIBUTION OF LENGTHS OF EARS IN CROSS (60 × 54)

No.	Class Centers in Cm. for Lengths of Ears																	A.	S.D.	C.V.
	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21			
No. 60	4	21	24	8														6.6±.073	.81±.051	12.27±.783
No. 54									3	11	12	15	26	15	10	7	2	16.8±.121	1.87±.088	11.13±.531
(60–5 x 54) F_1					1	12	12	14	17	9	4							12.1±.121	1.51±.088	12.48±.722
(60–5 x 54) F_2 [19]			4	5	22	56	80	145	129	91	63	27	17	6	1			12.7±.058	1.99±.037	15.67±.296
(60–8 x 54) F_2 [20]			1	10	19	26	47	73	68	68	39	25	15	9	1			12.9±.076	2.25±.053	17.44±.413
(60–3 x 54) F_2 [21]			2	5	17	33	33	33	27	21	13	10	11	12	1	2	1	12.6±.128	2.81±.087	22.30±.744

TABLE IV

FREQUENCY DISTRIBUTION OF WEIGHTS OF SEEDS OF CROSS (60 × 54)

No.	Class Centers in Grams for Weights of 25 Seeds																				A.	S.D.	C.V.
	2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5	6.0	6.5	7.0	7.5	8.0	8.5	9.0	9.5	10.0	10.5	11.0	11.5			
60	7	22	28	4																	2.7 ± .034	.39 ± .024	14.44 ± .899
54									2	2	5	7	14	10	5	4	1	2	1	2	8.3 ± .107	1.21 ± .074	14.54 ± .963
(60–5 x 54) F_1				5	12	13	17	4	3												4.6 ± .059	.64 ± .041	13.91 ± .912
(60–5 x 54) F_2 [22]		1	7	17	49	98	113	83	69	43	27	13	8	1		1					5.3 ± .032	1.09 ± .022	20.56 ± .442
(60–8 x 54) F_2 [23]				1	3	15	16	17	23	23	18	12	8	9	1	1					6.2 ± .067	1.23 ± .047	19.84 ± .799
(60–3 x 54) F_2 [24]				4	10	24	41	53	40	52	21	19	9	8	2	3					6.0 ± .047	1.17 ± .033	19.50 ± .563

[18] No. 60 and F_1 gen. grown in 1909, No. 54 and F_2 gen. in 1910.

[19] Grown from five ears.

[20] Grown from two ears.

[21] Grown from three ears.

[22] Grown from five ears.

[23] Grown from two ears.

[24] Grown from three ears.

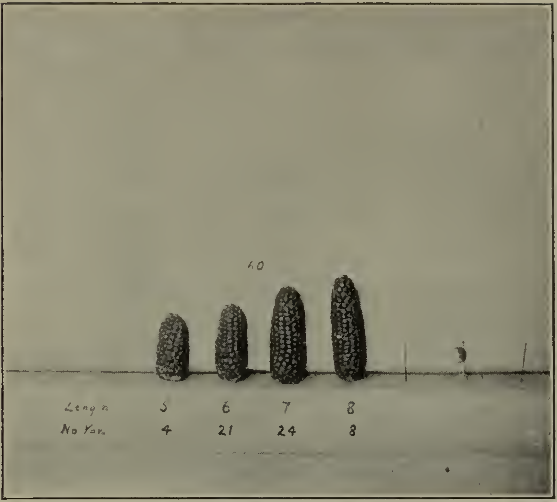

FIG. 1. No. 60, female parent, illustrating variation in length of ear (⅙).

have again been calculated, but they hardly emphasize the real segregation as well as do the photographs which were made from representative ears of the different classes found in the actual crop.

Table IV shows the segregation of weights of seeds in F_2 in this same cross. Fig. 5 shows the average size of

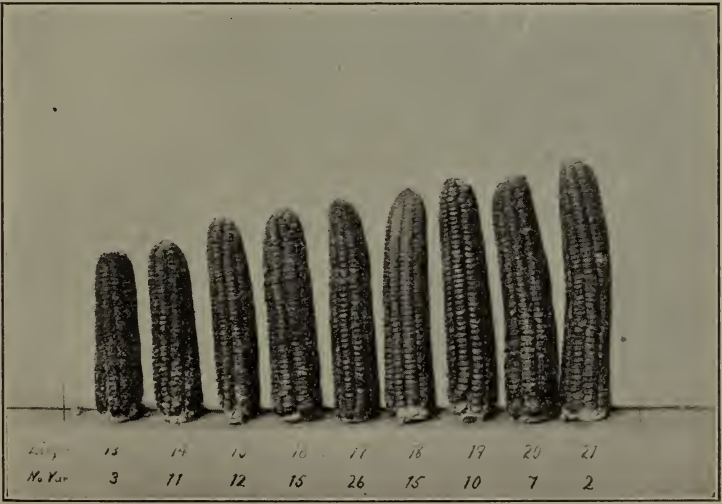

FIG. 2. No. 54, male parent, illustrating variation in length of ear (⅙).

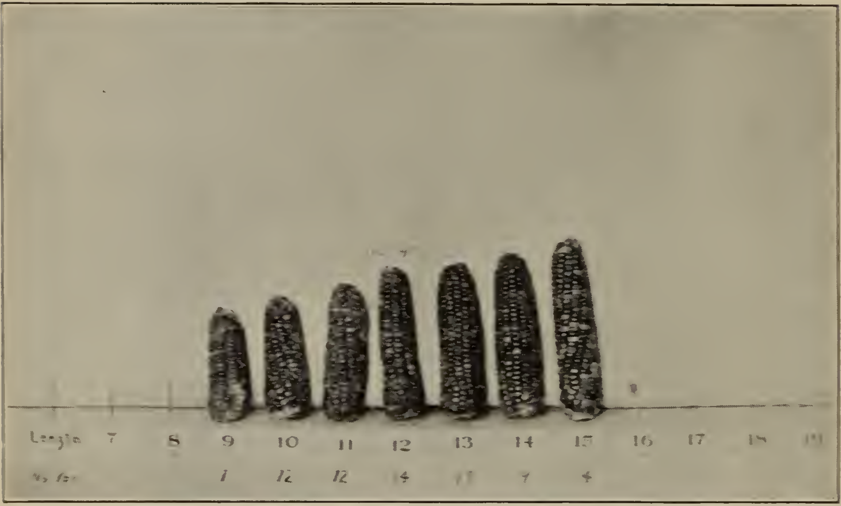

FIG. 3. Variation in length of ear of F_1 generation of cross between No. 60 and No. 54 ($\frac{1}{6}$).

the seeds of the two parents and the F_1 generation and the extremes of the F_2 generation. In making the weights for this table, it was necessary to use a scheme by which the sugary wrinkled seeds of the Black Mexican parent, no. 54 could be weighed as starchy seeds. This

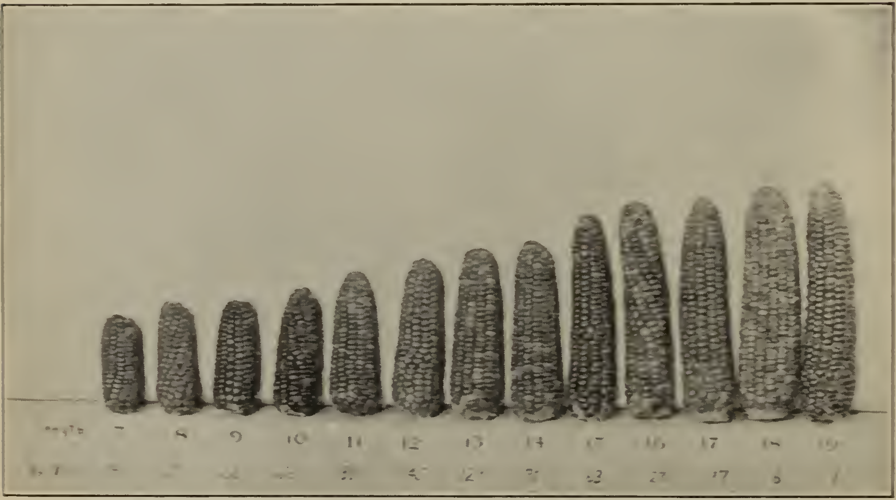

FIG. 4. Variation in length of ear of F_2 generation of cross between No. 60 and No. 54 ($\frac{1}{6}$).

was done by planting the no. 54 between rows of the hybrid. Sufficient crossed seeds which had become starchy through *Xenia* were obtained to make the weights given. Not all of the ears, however, had 25 starchy seeds, which accounts for the small number of plants measured. Furthermore, the seeds of no. 54 were a rather mixed lot, which of course resulted in a higher variability than would probably have been found if only seeds of the individual plant of no. 54 which was used as the male parent of the cross, could have been planted. Perhaps it should be noted here since the question might arise, that since the size of the seeds on an ear is governed by the development of the pericarp, the sugar corn, no. 54, was unaffected in other ways than by having the pericarp filled out with starch by the hybridization which occurred attended by the resultant *Xenia*.

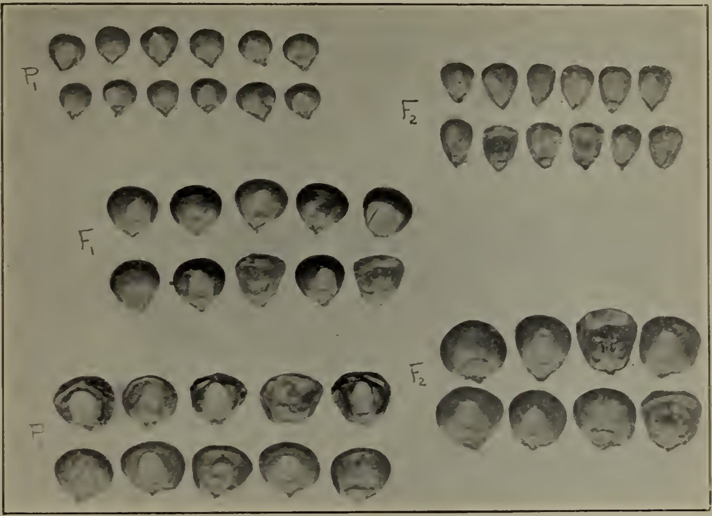

FIG. 5. Average size of seeds of No. 60 and No. 54 and the F_1 generation of the cross between them. Extremes of the F_2 generation.

In Tables III and IV the measurements and weights of the F_1 generation were recorded from only one cross, although three crosses between the two varieties were made. It might be said that one has the right to compare only the F_2 generation of cross of which the F_1 generation is given. If this were granted our conclusions in regard to segregation would be the same. It might be said, however, that sufficient records were made of the F_1 generations of the other crosses to know that they differed but little from the family of which the data were recorded. In addition, it is a fact that general populations of the two parents were studied, and their variation was undoubtedly greater than would have been that of the inbred progeny of the three parent plants of either variety.

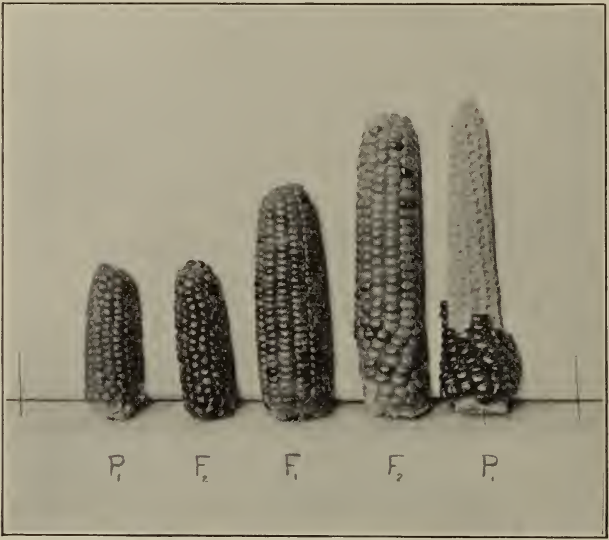

FIG. 6. Average ears of No. 60 and No. 58 and the F_1 generation of the cross between them. Extremes of the F_2 generation.

An additional cross between Tom Thumb pop maize and a small purple flint is illustrated in Fig. 6. The ears pictures show the average size of the two parents and the F_1 generation, and extremes of the F_2 generation.

In conclusion there are two points I wish to notice. Unquestionable segregation in size characters has been shown by comparison of the F_1 and F_2 generations. It can scarcely be doubted that some of these segregates will breed as true as the parent forms, yet one can scarcely do more than speculate in regard to the specific characters that are concerned in developing either organs or individuals of certain sizes. There are probably many characters that interact together in developing certain characters, although the actual determinants in the germ cells may be transmitted independently. These interdependent reactions during development obscure to us the real causes and what we regard as independent characters may be but indirect results of unknown causes. For example, the ability to evert their starch when heated has been the distinguishing character of the subspecies called *Zea mays everta,* the pop maizes. This character so called, however, is the resulting physical condition of the starch caused at least partially by the small size, the thickness and the toughness of the enveloping pericarp.

For these reasons it may not be possible—at least very soon—to point out even the number of characters concerned in size developments. From the number of extreme segregates obtained in each case I might venture to state that the size of ear in the cross shown in Fig. 5 is apparently due to not less than three characters, while the size of ear in the other cross pictured seems to be due to not less than four characters.

THE RÔLE OF SELECTION IN PLANT BREEDING

BY PROFESSOR E. M. EAST

Reprinted from the POPULAR SCIENCE MONTHLY, August, 1910.

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THE RÔLE OF SELECTION IN PLANT BREEDING[1]

BY PROFESSOR E. M. EAST
HARVARD UNIVERSITY

WHEN one attempts to give some idea of the principles and of the methods and scope of plant breeding, the matter falls naturally into two parts, the rôle of selection and the rôle of hybridization. This is not because the subject is really thus separable, but because the methods in use fall into these categories. One must, of course, use selection after hybridization, but there are a number of plants of great agricultural value, in which either the flowers are too small for artificial crossing or in which other reasons make it desirable to use simply selection in their improvement. It is of these that this paper will treat.

The particular work discussed has been selected because it will illustrate certain principles, not because it is regarded as more important than other work of like nature. The work of many quiet men who are striving for the good of mankind by their efforts toward the improvement of plants deserves to be mentioned, but unfortunately the limits of a single paper are too narrow to discuss principles and to say much about practise, and knowledge of the former should be made more widespread in order that the latter may be appreciated.

The non-botanical public can not be blamed if it receives comparatively worthless productions with greater acclaim than those of value when the former obtain all the publicity and no voice is raised in protest. Exploitations of new plant introductions of little value have certainly been numerous in the past few years. Perhaps this has been a public benefit, for it has increased the general interest in plant breeding and has stimulated many laymen to study the subject in order to be able to separate the wheat from the tares when dealing with new varieties. It is strange, perhaps, with our reputation for always looking for the dollar sign, that the new agricultural productions of greatest economic value have always received less notoriety than the production of horticultural novelties of limited use and small importance; yet such is the case. It is doubtful whether the production of a new field corn that would increase the yield in the United States by ten per cent. would obtain more than a passing notice from the press; yet such an increase would add $100,000,000 per annum to the wealth of

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[1] This paper is based on one a series of poular lectures delivered at the Bussey Institution of Harvard University April and May, 1910. A second paper will follow.

the country, and the individual who was responsible would deserve to be ranked among the greatest benefactors of the commonwealth.

This illustration serves to show something of the extent of the benefits that may be confidently expected from the improvement of cultivated plants; but the full extent of our rightful expectations is at least ten per cent. increase in both quality and quantity of all the great crops of the United States. In fact this is a very conservative forecast based upon what has been accomplished in the past. Men like Haynes with his "Blue stem" wheat and J. S. Leaming with his "Leaming" corn have perhaps made an even greater percentage increase in the value of the returns from the land upon which their productions have been grown. Their results were obtained largely in the latter half of the last century and even greater advances should be made in the future. This statement is made because, in the last quarter of the nineteenth century, experimental biology was in the same relative position in which chemistry stood in its beginning. During the century chemistry made wonderful advances; during this—the twentieth—century experimental biology will make similar progress. And one of the first and most important applications of the facts discovered will be to guide and direct man in producing new plants and animals by more direct and certain methods.

When one speaks of producing new plants, however, he should not be misunderstood. Man has not yet actually produced new variations (although the time may come when even this is possible); he simply works with the variations which have occurred through natural causes of which little is known. The isolation of a varying plant and from it the production of a variety, or the combination of desirable characters from one strain with other desirable characters from different strains, comprises the total aim and desire of the plant breeder. The idea is simple; to put the idea into practise successfully is often a tedious and difficult task.

As in hybridization the ease with which results can be obtained by selection depends largely upon flower structure. In selection, however, the relative facility with which artificial cross-pollination can be accomplished is of small importance. What one wishes to know is whether cross-pollination or self-pollination takes place naturally. Practically all plants are occasionally cross-fertilized naturally, and many of them have devices whereby they are nearly always crossed; but we are coming to see that cross-fertilization is not as essential to plant life as Darwin endeavored to prove in his "Cross- and Self-fertilization in the Vegetable Kingdom." Wheat, for example, is almost always self-fertilized; yet it has kept its vigor for thousands of years. The importance of this fact to the selectionist is easily seen. If seed from several varieties of wheat is mixed and planted, each variety remains true to its type because of self-pollination, and during the growing season the plants can be compared and any desirable type selected for future propagation. In a cross-pollinated plant like maize this is not the case. The pollen is carried by the wind through long distances and varieties planted close together are continually intercrossed. The isolation of a particular type is not simple as in the case of wheat, but may be prolonged through many generations. Each prize ear selected for future planting will have had at least a few and possibly many of its seeds fertilized by pollen from less desirable strains. When these seeds are grown they of course again fertilize the seeds of the desirable plants with a frequency proportionate to their number.

In the case just cited recourse may be had to artificial self-pollination. Several hundred seeds are thus produced at one operation and the work of isolating the new variety is made materially easier. But suppose we are dealing with red clover where the flowers are small, almost sterile with their own pollen and produce only one seed. In this crop, the long and tedious method of continuous selection just mentioned must be used, for there is no other way. This method is often called the pedigree-culture method. The main idea of the plan is that the seeds of *single* plants are grown in isolated plots, and the character of the mother plant judged by the characters of the progeny. This method has given much better results than the so-called German method, which consisted in planting a mixed lot of seeds from several of the best plants. For example, the German sugar-beet raisers have for years analyzed large numbers of sugar-beets and have grown their seed from the mother beets showing the highest percentage of sugar. No particular attention was paid to the general average of the progeny of each beet; those were bred from which appeared to be the best as shown by the polariscope sugar test. In this way the amount of sugar produced per acre was gradually increased, but progress was slow and cessation of selection immediately caused the sugar content to decline.

To see the real reason of this we must go back to the time of Darwin. The data from which Darwin proved the doctrine of descent came in large measure from domestic animals and cultivated plants. He saw that plants varied among themselves and that by selection of the variants new types were gradually produced. From these facts he argued that all evolution had taken place by the selection of minute variations and generally through the selective agency of a contest for life taking place among all living organisms. This he called the agency of natural selection. Later, however, Bateson, Korshinsky and de Vries called attention to the fact that many new types of animals and plants are known to have originated suddenly. There was no gradual evolution of the type; it simply appeared fully formed. This hypothesis, called the "mutation theory," found great favor among plant breeders for they knew that many times they had noticed and isolated plants showing new characters from their cultures, and had carefully made selections for further improvement of the new strain, but that generation after generation showed no further progress. LeCouteur, whom de Vries cites as the first known user of the pedigree culture method, had a case in point. From the heterogeneous lot of wheat plants which he was growing, he isolated a uniform type of great merit which he called "Bellevue de Talavera." For years after, this strain was subjected to selection in order to bring about further improvement, but the efforts were made in vain, for no new heritable variations were produced. Yet something was lacking from this theory. Sometimes there did appear to be a gradual improvement by selection. De Vries said that this was merely a temporary improvement made by selection of quantitative variations. He believed that when selection ceased, sooner or later the improved types would return to the original type of the variety from which it had been produced. The real interpretation of the facts and one which fitted all the parts of the puzzle together, came from the work of Johannsen and later investigators. It is an explanation that should have been thought of before, but like many other important discoveries, it was too simple for ordinary minds to grasp. Weismann had shown years before that the inheritance of characters acquired through outside influences during the development of the body was probably mythical. His investigations led him to believe that there is a continuity between the reproductive or germ cells of different generations, and that the body is nothing but a temporary house built to shelter them. Injuries to the house have no effect on the future generations unless the germ cells themselves are affected. Later Boveri and others, through their cytological studies, showed that the future germ cells are laid down at a very early stage in certain animal organisms and that very few cell divisions take place before the maturation of the reproductive organs and the production of active germ cells. The body cells he found to be built up by continuous cell division of a very different part of the original fertilized egg. Since no biologist, however, had found or is likely to find similar cytological phenomena in plants, no one seemed to grasp the idea that here was the key to the question that had been puzzling the plant breeders. Johannsen, however, brought matters straight by his experiments on beans. He found that commercial varieties of beans, though pure in grosser characters, such as color, were actually very mixed types when such characters as length or weight were studied. Several investigations were undertaken on size characters, the characters most rapidly affected by changes in environment. He found that his commercial variety fluctuated around an average size and that when seeds larger or smaller than this type were selected they responded to it in whichever direction the selection was made. The progeny of the selected beans were not so extreme, however, as their parents but regressed toward the average character of the parent race. This was nothing new. Galton had discussed the matter a decade before and had interpreted the regression as due to the "pull toward mediocrity" exerted by former ancestors that must have been on the average mediocre. Johannsen was not satisfied with this interpretation and in order to investigate the subject more thoroughly introduced the individual pedigree culture method, or pure line method as he spoke of it, into his work. All of his plants under experiment were self-fertilized for successive generations, so that all of his future bean progeny were descendants of single individuals from the original commercial variety. Each pure line he found to fluctuate around a typical size just as the commercial variety had done. Some types were exactly the same as the original mixed type, but others fluctuated around averages that would have been considered more or less extreme variations in the original. He then grew extreme variants from each of his pure lines and made the discovery that no progress at all was made by repeated selections of this kind. The progeny of the high extremes and the progeny of the low extremes each were found to fluctuate around the same pure line average. It was quite evident then that in the first place he had been dealing with a mixed race. This mixture consisted of sub-races each with a heritable difference in the character size. These heritable variations, however, were obscured by size fluctuations produced by differences in moisture, sunlight and fertilizer received by the different individual plants. There was even a difference in the size of individual beans on the same plant, due probably to location of some pods in places on the plant more desirable than others for the utilization of the plant's soluble foods waiting to be stored in the seeds. *These differences due to immediate environment were not inherited.* They behaved exactly as the acquired characters of an animal. This made the rôle of selection clear. The only improvement that selection can achieve is to isolate a substrain if such a substrain or substrains exist in the variety under experiment. When this substrain has been isolated, selection has absolutely no effect, and even if continued for countless generations will have no effect until nature produces one of the heritable changes which are so much rarer than the fluctuations produced by environment. It is also evident that the older idea that improvements made by continued selection—*i. e.,* gradual isolation of a type—are inconstant, is wrong. The explanation is that since non-inherited fluctuations obscure the heritable variations, only a pure line method can absolutely isolate a pure strain; and in the German method of mass selection with poor control against mediocre pollen, the chances were overwhelmingly in favor of the selected type recrossing with the more commonly cultivated and poorer type from which it came.

To my mind this work should clear up the strife between the critics and the adherents of evolution by mutation. It is evident that there are variations that are inherited and variations that are not inherited. If we call the one a mutation and the other a fluctuation, we have a distinction that will stand analysis. Why should a further distinction be made? De Vries believes mutations to be qualitative, fluctuations quantitative. Nevertheless, quantitative changes that are transmissible occur in much greater numbers than do qualitative changes. Opponents of mutation believe wide jumps appear too seldom to have been a factor in organic evolution, but they can not deny that they do occur. There are too many authentic cases in variation under domestication. Yet no one who has had experience in breeding plants will deny that small variations (not fluctuations) occur with much greater frequency. While it is impossible to prove it, I believe that the mathematical law of error controls the transmissible variations as well as fluctuations. If one could collect a random sample of variations that are inherited he would probably find that a great many forces act as the causes, and therefore as in ordinary probability, the extreme changes—that is, the great variations—occur with less frequency. One should remember, however, that in our present state of physiological knowledge, he can not know with much certainty which of two changes that apparently differ greatly in magnitude is really the greater in the light of the plant's economy.

It might be well before leaving this part of the subject to speak of one other point. In a strain that has been self-fertilized for several generations, gradual progress has sometimes been made by selection. This probably comes about because the parent plant is still hybrid in regard to certain characters, and it is to their recombinations that the intensification or reduction of certain apparently single characters but which are really combinations of separately heritable characters, is due. According to the law of chance with repeated self-fertilizations any strain approaches a constant condition in all of its characters when unselected, but one can not say when this state is reached unless he knows the exact number of hybrid characters in the beginning and can recognize each.

If we were to take up the crops of the United States which owe their present excellence and future prospects in large measure to the isolation of superior strains by selection, we should cover a great majority of the agricultural wealth of the country. Of course natural cross-fertilization and even occasional artificial hybridization have played important parts by causing recombinations of characters, but selection has been the main cause of improvement. Two of the important crops, tobacco and wheat, are very seldom cross-pollinated naturally; nevertheless new types are continually appearing in the fields. To make new varieties

FIG. 1. TYPES APPEARING IN A SINGLE FIELD OF MAIZE. A strain like the ear near the center has been isolated.

it simply takes an alert eye for their detection, comparative tests to prove their merit and the time needed to produce a sufficient increase for commercial use. Some of our other important grain crops like oats and rye are more often cross-pollinated, as is also our chief grass crop, timothy. But as maize is probably the most difficult crop to deal with, and is a typical cross-pollinated plant as well as our most important cereal, perhaps it will be of interest to take a short survey of some of the problems with which one has to deal when endeavoring to improve it by selection.

Maize is the only one of our cereals that is monœcious. The tassel contains the pollen or male element while the silks are the stigmas of the female flowers. In order that the pollination of the silks shall be relatively certain, each tassel produces about thirty million pollen grains; and as the ears average less than five hundred seeds apiece, there are about sixty thousand pollen grains produced for each kernel. With such a large amount of superfluous pollen floating around in the air, there is a great deal of inter-crossing between the neighboring plants. This fact has been an obstacle to the improvement of maize, but it has been offset by one advantage it possesses over the other cereals, that of producing large ears. Since each individual ear must be handled and its characters noted at husking time, it is not strange that ears with desirable variations sufficiently striking to catch the eye of the grower have become the parents of numerous distinct varieties. By selecting desirable seed ears and isolating them from other varieties, various strains have been produced that are remarkably uniform in characters such as color that have forcibly attracted the attention of the breeder. Even in these strains, however, there are many natural types growing side by side and continually crossing with each other. There are stalks

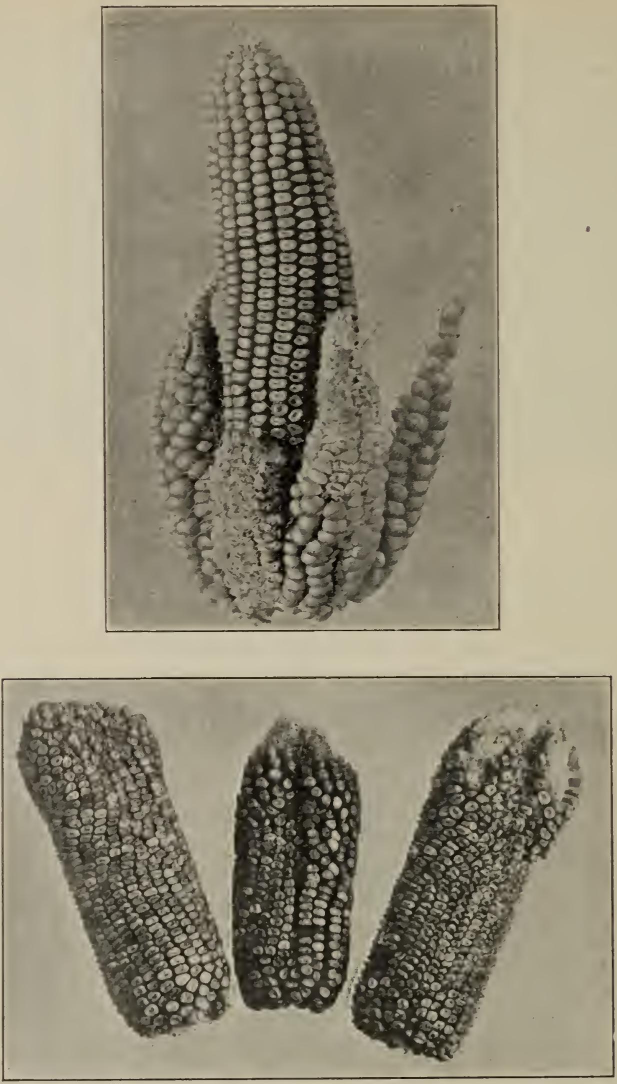

Fig. 2. Inherited Abnormalities Appearing in Maize. All pollen from these plants and from their normal sister plants must be prevented from maturing.

which bear their ears high and stalks which bear them low, stalks with long and stalks with short ear shanks, stalks with different leaf markings and with notably different tendencies to produce suckers. Differences are everywhere present even in the ears, as is shown in the accompanying photograph (Fig. 1). A large number of these differences are simply fluctuations produced by the environment and are not inherited. The obscuration of heritable variations by the fluctuations and the mixed condition of the natural types makes it a difficult task to isolate the most productive types. Many variations of technique have been proposed for the prosecution of the work, but are all based upon the idea of proving the capacity of a mother ear by the characters of the progeny produced. If a very large number of ears are included in the original stock, it is unquestionable that some of them will transmit more desirable characters than others. It only remains to test them out by growing the seed of each ear in marked plots or rows and gradually eliminating the undesirable types.

The accompanying diagrams, showing the work of the Illinois Agricultural Experiment Station in their experiments in selecting for high and low protein content, and high and low oil content, admirably illustrate the rapidity with which progress can be made by selecting only from the maternal side, even in the face of constant intercrossing. This work the writer believes has given a complete corroboration of Johannsen's conclusions on pure lines. This interpretation has been made, however, from their published data, and the Illinois station should not be held responsible. This work of breeding to change the composition of maize was started in 1896 with a hazy Darwinian idea that as corn was known to vary in composition, continuous selection of extreme variations would produce a continuous change in type. A very old type—Burr's White—furnished the foundation stock. A chemical analysis was made of parts of the individual ears each year, and the extreme ears planted. From the first, the four lines above mentioned were planted in isolated plots and were continually selected in the same direction. After ten generations the average crop of the high protein line had reached 14.26 per cent., while the low protein line was only 8.64 per cent.; the high oil strain had reached 7.37 per cent., while the low oil strain was reduced to 2.66 per cent. These facts clearly show the rapidity with which results can be obtained

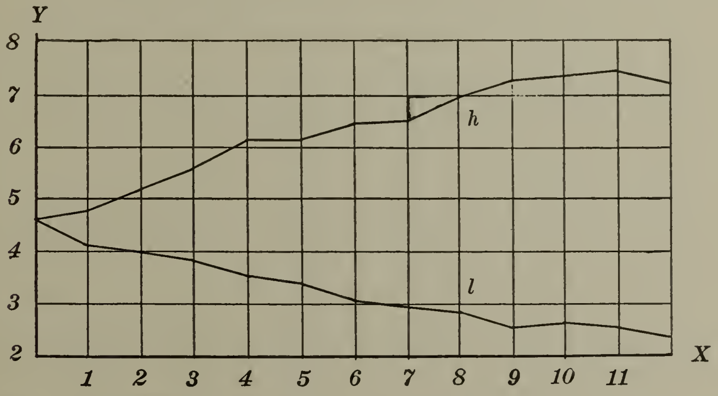

FIG. 3. DIAGRAMMATIC REPRESENTATION OF THE RESULTS OF THE ILLINOIS AGRICULTURAL EXPERIMENT STATION in selecting for high and for low ~~protein~~ content. \ oil
Y, per cent. protein in crop; X, generations; h, high protein strain; l, low protein strain.

by this method of selection even with a crop that is often cross-fertilized. But the diagrams show other facts. The published records show that the variability of the race was but little, if any, reduced by continuous selection. With extreme variants comparatively as far removed from each year's type, available for planting in each successive generation, the gain each year should have been at the same rate, if the Darwinian interpretation of the rôle of selection were correct. On the contrary, we notice that the regular curve fitted to the crop averages for ten generations, is first concave showing great progress made by selection, is later convex as progress becomes slower, and last becomes horizontal

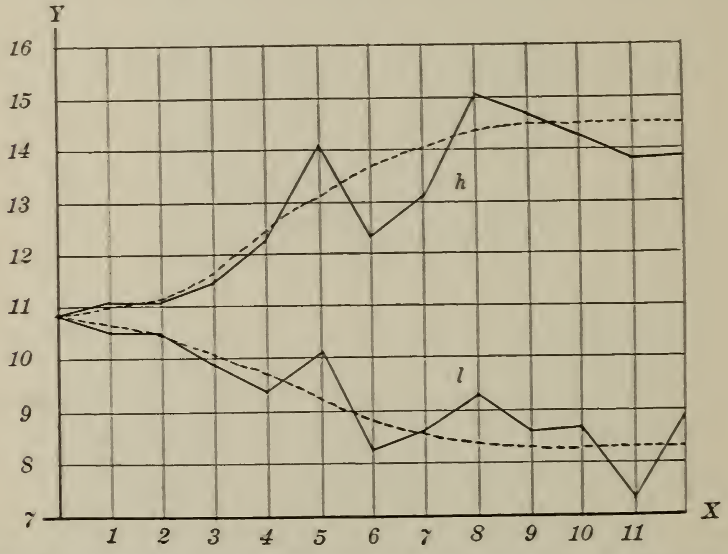

FIG. 4. DIAGRAMMATIC REPRESENTATION OF THE RESULTS OF THE ILLINOIS AGRICULTURAL EXPERIMENT STATION in selecting for high and for low ~~oil~~ content. *Y*, per cent. oil in crop; *X*, generations; *h*, high oil strain; *l*, low oil strain.

as no more progress results. It is very evident that the original stock was a mixed race containing sub-races of various composition intermingled by hybridization. Selection rapidly isolated these sub-races. The isolation was practically complete at the eighth generation in the case of the protein strains and the ninth generation in the oil strains. After this selection accomplished nothing. That the effect of selection was simply the isolation of a sub-race and not a continuous response, is further demonstrated by the fact that in 1903 another plot was started with seed from the isolated high oil strain. After four years' cessation of selection, the average composition of the crop remained the same, showing that after complete isolation of a homogeneous type no retrogression of the selected character occurs unless intercrossing with mediocre strains takes place. Fluctuation in composition still appears, but this is the non-inherited kind produced by external conditions.

It is sometimes somewhat difficult to see why selection of this kind should yield results slowly. There are indeed many points concerning which little is known. One may picture to himself, however, that where crossing is always likely to occur and where the apparent character is in reality a combination of a number of separately inherited characters, many thousands or even millions of individuals would have to be grown to run a fair chance of obtaining the most desirable combination. By growing a few individuals in which the desired character is intensified in successive generations, the combination wanted may be obtained with the use of smaller numbers.

I have stated that nothing can be accomplished by selection after a pure line or genotype as Johannsen calls them is isolated, unless a new transmissible variation is produced by nature. The questions then arise: how often may such changes be expected? and, what is their nature? Such changes are of two kinds,[2] progressive where a new character appears, or retrogressive where a character is lost. But little can be said as to their relative frequency. Undoubtedly some species are in a more unstable condition than others and give more of such variations, as de Vries has already suggested. On the other hand, certain unknown combinations of external conditions may favor germ-cell changes. They are both rare, the progressive changes being relatively much less frequent than the retrogressive changes, but they are sufficiently common for several to have come within the knowledge of every experienced breeder.

There is another type of variation much more closely related to changes occurring in "pure lines" than is generally supposed. I refer to what is commonly known as bud variation or vegetative sports. Retrogressive variations of this kind are probably no rarer than the same kind of changes occurring in pure lines. No authentic progressive variations (as distinguished from digressive) are known. In my own experience in growing eight hundred species and varieties of tuberous solanums (largely potato varieties), fifteen retrogressive variations have been noticed, and the changes that occurred were exactly like those occurring in seed-propagated strains.

The relative value of progressive and retrogressive variations is difficult to estimate. In organic evolution the former must have been far more valuable; commercially the latter are often of great worth. We may cite, for example, the great value of the bush or dwarf varieties of beans, peas and tomatoes that have originated as retrogressions.

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[2] De Vries also gives a third kind, digressive variations, such as occur when a character previously possessed by but latent in the plant appears. This class is unnecessary. Digressive characters appear either through the loss of a complementary inhibiting factor or the gain of a complementary factor necessary for it to become active.

In closing I should like to call attention to a fact both of evolutionary and of commercial importance. The first generation of crosses between nearly related types generally grows more vigorously than the pure types themselves. If the fertility is not impaired, they even fruit more freely. This is undoubtedly the explanation of Burbank's quick-growing hybrid walnuts, but if they were self-pollinated and grown for another generation a large percentage of the progeny would lose this character. In naturally self-pollinated types like tobacco, one sees the phenomenon expressed as greater vigor in a cross; in a continually inter-crossed species like maize the same thing is shown by a loss of vigor when the plants are self-pollinated. It is clear then that if pure strains of maize are gradually isolated by selection, by the same token they lose in vigor and productiveness. The original mixed strain may contain sub-strains some of which are much more productive than others. The less productive types may be discarded, but at the same time there is a loss of vigor from the fact that they are withdrawn from hybrid combinations. The logical procedure, then, is to isolate two high-yielding types, combine them by hybridization, and grow only the first generation of the cross. This is not mere theory, for by using such methods I have obtained from 100 to 200 bushels of shelled corn per acre on small plots. Unfortunately, this method can not be used to advantage on many crops, but in the case of maize the procedure is simple. There are many breeders using the isolation method of improvement. From

FIG 5. EFFECTS OF INBREEDING IN MAIZE. Outer ears inbred four generations. Middle ear the result of their crossing, first generation.

FIG. 6. RESULTS OF CROSSING TWO INBRED STRAINS OF MAIZE. At the right average of the parents after three generations inbreeding, 61 bushels per acre. At the left crop of first generation cross of the inbred strains, 101 bushels per acre.

them the grower obtains two strains and plants them in alternate rows. At flowering time all of the male flowers or tassels are removed from *one* of the plants of the varieties before they shed their pollen. All the ears that these plants produce are crossed with the other variety. It is this seed that produces the vigorous plants.

This method might be made the basis for some very valuable work in forestry. It is quite conceivable that many important timber trees might be found where nearly related species or varieties would cross readily. Experiment would show how great an increase in rapidity of growth could be expected, and whether such an increase would pay for the increased expense of hand hybridized seed.

FIG. 7. SILVER HILL. Normal at left. Bud variation isolated by selection at the right

One may summarize by saying that two important points cover the whole rôle of selection. The first point is that nature continually causes variations to appear in plants. The majority of these variations are simply accelerations or retardations of development of the whole or of certain parts of the plant due to good or bad environment at critical stages of the plant's growth. These variations are not inherited because the reproductive or germ cells are not affected. Other variations, however, are being constantly produced by nature—though much more rarely—which do affect the reproductive cells and are transmitted to the plant's progeny. These variations are the basis of selection. They are constant from the beginning and remain so unless changed by a second variation affecting the same constituent in the reproductive cells that is due to develop the character in question.

The second point to be remembered is that the whole aim and action of selection is to detect the desired heritable variants among the useful commercial plants and through them to isolate a race with the desired characters. When this is accomplished, selection can then do nothing until nature steps in and produces another desirable variation.

In other words, the results of selection are not continuous. Selection does not gradually perfect a character. The production of heritable variations is intermittent and the intermissions may be long. If the practical results seem to be parts of a continuous process, it is because of the imperfect methods at hand to isolate the desirable variations from their combinations with undesirable characters formed by natural hybridization.

THE RÔLE OF HYBRIDIZATION IN PLANT BREEDING

BY PROFESSOR E. M. EAST

Reprinted from the POPULAR SCIENCE MONTHLY, October, 1910.

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THE RÔLE OF HYBRIDIZATION IN PLANT BREEDING[1]

By PROFESSOR E. M. EAST
HARVARD UNIVERSITY

THE word hybridization has been used with many meanings. The term is used here to denote the crossing of any two plants that differ from each other in a heritable character, whether they are of the same or of different species.

There is intimate connection between the rôle of hybridization and the rôle played by selection. It comes about in this way. Inherited variations are produced by nature with considerable profusion. New characters appear and old characters are lost: these form the working basis of selection. But whether they are large or small they are usually inherited completely. They are the units of heredity; or, if they are sometimes transmitted in units of lesser degree, they may be compared to chemical radicals.

The main object of hybridization then is the shuffling of these units in the first hybrid generation and their recombination in the next generation. There are, however, various phenomena attending hybridization, and I will endeavor to illustrate the following as those of most importance: (*a*) Recombination of characters and their fixation, (*b*) production of desirable combinations in the first hybrid generation and their continuation by asexual propagation, (*c*) production of fixed first generation hybrids, (*d*) production of blends.

If we begin at the real beginning in this discussion, we must say a few words concerning the actual mechanical operations of crossing. The first foundation stone to be laid is a knowledge of the flowering habits and flower structure of the plants to be used. Of course a careful examination of the flowers will show the easiest and surest method of removing the stamens of the flowers that are to be pollinated and of protecting them from foreign pollen. What is not so easily determined are the precise conditions under which the cross should be made to be successful. The proper preparation of the breeding plot even before the plants are grown is necessary. One takes it for granted that some fertilizer will be used, for the plants must be normal to seed well. The three essential elements of soil fertility are nitrogen, potassium and phosphorus, and to get the best results compounds of these elements must be present in proper proportions. First, available potash must be

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[1] This paper is based on one of a series of popular lectures delivered at the Bussey Institution of Harvard University, April and May, 1910.

present in normal quantity although a certain excess will not be harmful. If nitrates are present in excess, however, vegetative growth will be over stimulated and seed production will be small. A lack of phosphates will produce the same effect upon seed production, but for a different reason. Phosphorus is an essential constituent of the proteid compounds found in large quantities in the seed. Therefore, if the plants are to be in the best condition for crossing, the soil should contain just the right amount of nitrates for normal growth and a generous supply of potash and phosphates. The exact amounts must be determined by experience for each soil and each species of plant.

Other necessary knowledge that can be obtained only from experience is which are the best flowers on the plant to serve as parents of the cross and what is the proper time for their pollination. For example, in the grasses the first flowers that appear usually form larger, healthier seed than the later blossoms. In most of the Solanaceæ, the petunias, browallias, etc., the exact opposite is true. The time when the individual flower is most receptive to pollen is even more narrowly limited. Both premature and delayed pollination is the cause of many failures and the optimum time should be accurately determined. Having exercised these precautions, it remains to study carefully the structure of the flower in order that it may be emasculated—*i. e.*, the anthers removed before the pollen is shed—with sufficient adroitness that neither the anthers shall be opened nor the parts of the pistil injured. Only a few buds upon a single flower spike should be operated upon if they are to be given the best chance of development. If the buds are very small and some pollen unavoidably reaches them, it may be washed off with comparative safety with a dental syringe if done immediately. It is often recommended that the calyx and corolla be cut away when emasculating. This should be avoided if possible and the floral envelopes left as a protection to the pistil. After emasculation the buds should be protected from foreign pollen until time for pollination, and again after pollination at least until the fruits have begun to form. This protection may be an ordinary paper bag when the crossing is done in the field. In the greenhouse I find that a square of thin celluloid rolled around the flower and caught with two rubber bands, each end being protected with absorbent cotton plugs, is a better device. It gives excellent protection and allows transpiration.

But enough of the technique of hybridizing; the phenomena attending it are of more importance. After the pollen is placed upon the stigma it begins to grow until it reaches the ovule. Down this tube comes the male cell which contains the potentialities of its parent plant. This cell fuses with the female cell in the ovule and fertilization is accomplished. From this combination the seed and later the hybrid plant results, half of its characters coming from the plant which furnished the pollen and half from the plant to which it has been applied. If one studies the characters of several such hybrids, he finds many surprising facts. It usually makes no difference which plant is the mother plant, the result is the same. Certain characters are found in the hybrid that are identical with those possessed by the male parent and other characters the same as those possessed by the female parent. Other characters appear to have resulted from the blending of those of the two parents, while still others appear to be entirely new. The plant may be sterile if the cross is between widely differing species, but if it is fertile and the flower of the hybrid is self-fertilized, the plants resulting from this seed present still more surprises. For example, if one has crossed a pear-shaped yellow tomato with a round red tomato, in the second hybrid generation he will find individual plants bearing fruit of four kinds, pear-shaped yellow and round red, as were the two parents, and in addition pear-shaped red and round yellow. In other words all possible combinations occur and in definite proportions. Stated as a principle it may be said that where either of the parent plants possesses characters absent from the other, the potential characters remain pure in the germ cells of the hybrid and recombine as if by chance. This is the most important feature of the only law of heredity of which there is any exact knowledge—the law of Mendel. Let us illustrate the action of the law. Such a character as starchiness, as shown in "flint" maize, is either present or it is not present. The flinty appearance of the seed is due to the possession of some character that causes the maturation of plump starch grains. When this character is absent, the seeds dry up without maturing their starch grains, and present the wrinkled appearance common to sweet maize. Pairs of characters such as these, affecting a certain plant structure, are called contrasted or allelomorphic pairs. When a sweet maize is crossed with a flint maize, the resulting seeds are all flint like. That is, the dominant character or the character that calls for the presence of the structure or compound in question, manifests itself in the first hybrid generation. Complete dominance, however, is not a general phenomenon in crosses and as its importance is slight as compared with the second law, that of segregation of the pure characters (potentially) in the germ cells of the hybrid, we will discuss it no further. The second law predicts that in the generations succeeding a cross, plants grown from the self-fertilized seeds of the hybrid reproduce both contrasted characters in the proportion of three of the dominant or "presence" characters to one of the recessive or absent character. Furthermore, inbred or self-fertilized plants bearing the recessive character continue ever after to breed true, while of those plants bearing the dominant character one third are pure and breed true while two thirds are hybrids and again throw the recessive character in one fourth of their offspring.

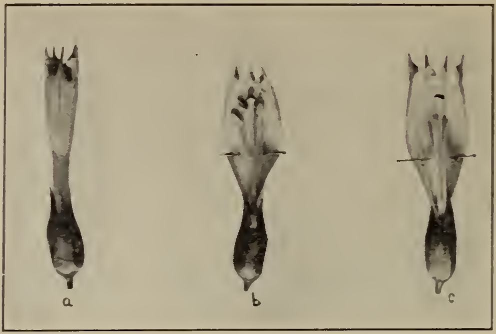

FIG. 1. CASTRATION OF BUDS OF *Nicotiana tabacum* BEFORE CROSSING. *a*, correct stage for castration; *b*, method of slitting the corolla; *c*, castrated bud.

The theory supposes that when a dominant and a recessive character meet in a cross, the germ cells which are produced in the hybrid do not blend these characters, but possess either the one or the other; and as the possession of either character is a matter of chance, on the average 50 per cent. will bear the dominant and 50 per cent. will bear the recessive character. In a plant, for example, 50 per cent. of the pollen cells would bear the dominant and the other 50 per cent. would

FIG. 2. CASTRATED BUD OF *Impatiens sultani* SHOWING METHOD OF PROTECTION IN GREENHOUSE WORK.

bear the recessive character. One half of the egg cells, likewise, contain the dominant, and one half the recessive character.

Now, if we could pick out at random any one hundred pollen or male cells to fertilize any one hundred egg or female cells, we can see that there are equal chances for four results. A dominant male cell might meet a dominant female cell, a dominant male cell a recessive female cell, a recessive male cell a dominant female cell, and a recessive male cell a recessive female cell.

We have (D + D), (D + R), (R + D), and (R + R) plants formed in equal quantities, but as the two middle terms are the same, we can reduce the formula to one (D + D) to two (D + R) to one (R + R). But wherever there is a D present in the germ cell, the dominant character shows, while the recessive character is hidden. The one part or 25 per cent. of the individuals showing the character (D + D) will appear just like the two parts or 50 per cent. of the individuals having the character (D + R). Therefore, there will be 75 per cent. of the individuals which will show the dominant or D character, while 25 per cent. will show the recessive or R character. These 25 per cent. showing the R character will ever after breed true, because they contain nothing but the recessive character; while of the 75 per cent. showing the dominant character, one third or those having the pure (D + D) character will breed true in succeeding generations, while the other two thirds having the (D + R) or hybrid character will again split in the next generation.

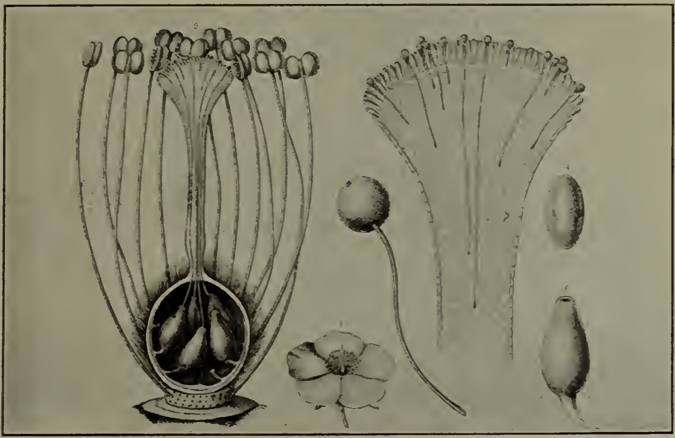

FIG. 3. THE COURSE OF THE POLLEN-TUBE IN A ROCK-ROSE (*Helianthemum marifolium*). After Kerner and Oliver. 1, single flower; 2, essential organs of flower: course of pollen tubes shown diagrammatically; 3, pollen tubes penetrating the tissue of the pistil; 4, dried pollen grain; 5, pollen grain germinating; 6, ovule.

For all practical purposes in plant breeding the mere fact of segregation is of greatest importance and the complexity of recent Mendelian interpretations need not bother us. Suffice it to say that most plant breeders have accepted the explanation that the recessive character is simply the lack or absence of the character in question, while the dominant character is its presence. This is simply a slightly different interpretation of the same facts and simplifies some of the more complex results of crossing. Instead of 50 per cent. of the germ cells bearing the flint character and 50 per cent. bearing the sweet character when sweet corn is crossed with flint corn, one should think of all of the germ cells bearing the ability to produce the wrinkled sweet corn seeds, but that 50 per cent. of them contain in addition the presence of a flint or starch producing character. In other words, the "starchy" character is superimposed upon the "sweet" character. The dominant and recessive characters in such a cross, then, are simply the presence and absence of the starchy character.

When several character pairs differentiate the two parent plants in a cross, all possible recombinations are formed, the relative frequency with which the combinations occur being simply the algebraic product of as many of the simple ratios as there are character pairs.

The importance of these Mendelian facts to the commercial plant breeder is great. In crossing plants differing in several simple characters that segregate after hybridization he may rest assured of two things. First, that with a sufficient number of progeny in the second hybrid generation, every possible recombination of the characters present will be represented by at least one *pure* specimen. Second, that these pure specimens when selfed, or pollinated with their own pollen, will breed true. It should be remembered, however, that one may have to self a number of plants to get the combination desired with all characters pure, for if any dominant characters are concerned, their purity can be ascertained only by breeding for another generation. As an illustration we may take the snap dragon, Antirrhinum. There is a long series of colors that segregate. There is also a type called the "Delilah," where the tube of the corolla is uncolored. Starting with this form in only one color, the whole color series of Delilah forms may be reproduced by crossing with the self colored strains. Or, one may combine the dwarf habit of growth of the Dwarf Champion tomato, with any of the various colors and shapes now on the market which have the ordinary tall habit of growth. Sometimes a very simple recombination is of very great commercial value. The so-called Havana type of wrapper tobacco grown in the Connecticut River valley has large leaves and a short stocky habit of growth. It produces from nineteen to twenty-one leaves. There is another type grown under cheese cloth shade which has a tall habit of growth with about twenty-six smaller leaves. The tall slender habit of growth makes it an undesirable type to grow in open fields where it is apt to be blown down. Mr.

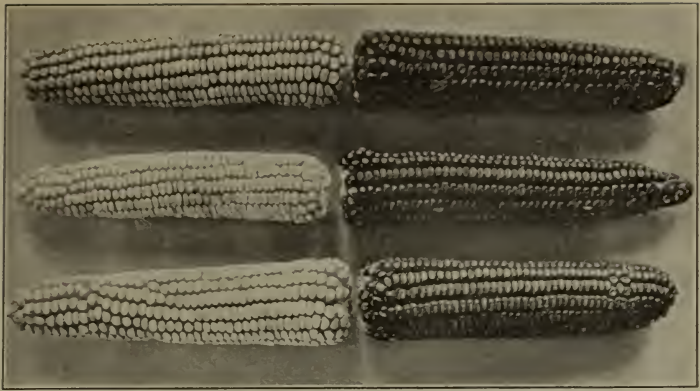

FIG. 4. SHOWING ABSOLUTE SEGREGATION IN SECOND HYBRID GENERATION. These red and white ears grew from a single self-pollinated ear of the first hybrid generation of a cross between red and white maize.

A. D. Shamel, of the United States Department of Agriculture, crossed these two types. A new type called the Halladay has been produced with the higher number of leaves of the Cuban parent and the stocky habit of growth and large leaves of the Havana parent. The first interpretation of this result was that an entirely new variation had appeared, for the Cuban type usually has but twenty-two or twenty-three leaves. The writer has been able to show, however, that the actual strain of the Cuban used as the parent of the cross has on the average twenty-six leaves, and data have now been collected that show that the new variety is a simple recombination of the characters possessed by the two parents giving an out-door type averaging thirty per cent. greater yield than the old Havana strain. In a similar way Biffen has produced a rust resistant high-yielding wheat by crossing two varieties each of which possessed but one of these desirable qualities. Orton has combined the edible quality of the watermelon with the wilt resistance of the citron, and Webber has increased the ability of the orange to resist cold by crossing with the hardy trifoliate orange.

Recent accurately controlled investigations in hybridization have shown that many apparently complex results yield to simple explana-

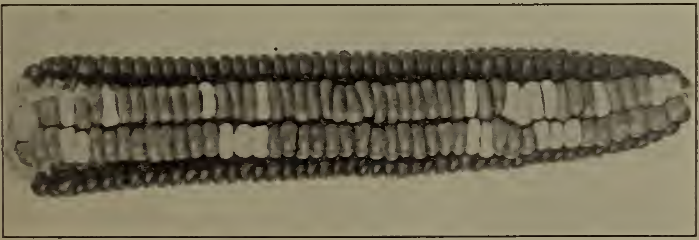

FIG. 5. RED MAIZE EAR WITH PERICARP REMOVED, showing segregation of yellow and white endosperm beneath it.

tions by use of the Mendelian theory. For example, two or more hereditary factors may be necessary for the production of an actual tangible character. If factors A and B must be present for its production, then a plant carrying only factor A and another carrying only

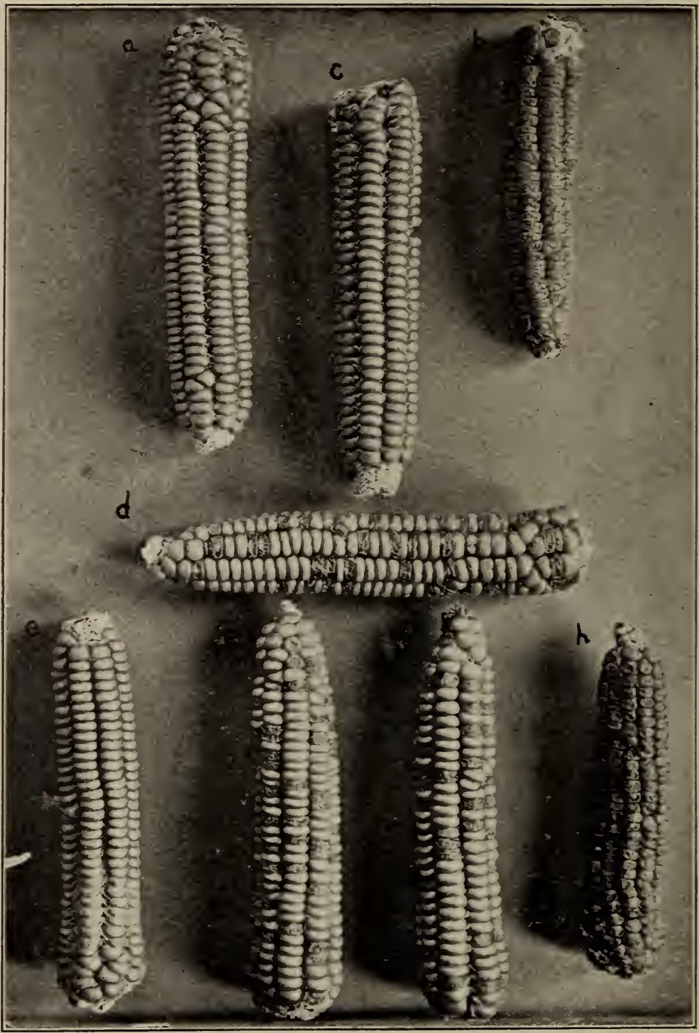

FIG. 6. MENDELIAN SEGREGATION IN MAIZE. *a* and *b*, the two parents, starchy and sweet maize; *c*, the first hybrid generation showing dominance of starchiness; *d*, the second hybrid generation showing segregation with the ratio of three starchy to one wrinkled seed. Lower row daughters of *d*. *e*, *f* and *g*, results of planting starchy seeds. One ear out of three is pure starchy. *h*, result of planting sweet seeds. Ear is pure sweet.

factor B do not possess the character. But let the two plants be crossed and the character appears. There are two white varieties of sweet peas; each, however, contains one of the two factors necessary for the pro-

FIG. 7. AT LEFT "HAVANA" PARENT, AT RIGHT "CUBAN" PARENT OF CROSS SHOWN IN FIG. 8. The "Havana" has short habit of growth, large leaves averaging 19 to 21 in number. The "Cuban" has tall habit of growth and averages 26 medium-sized leaves.

duction of a purple variety. When these two white varieties are crossed, the purple variety results. The second generation, however, produces seven whites to every nine purples. Such segregation into purples and whites may not be desirable; all purples may be wanted. This brings us to a consideration of class B of the four classes of phenomena attending hybridization, the production of desirable character combinations in the first hybrid generation and their continuation by asexual propagation. This class really includes several distinct types of occurrences. The purple sweet pea produced from the two whites will serve as an illustration of the first type. In certain plants (not meaning the sweet pea, however) it is as simple to reproduce by cuttings as by seed. The cuttings are simply parts of the plant from which they come and are identical with it in character.[2] If in a species of this kind a desirable character is formed by the union of two or more hereditary factors and one wishes to reproduce the character indefinitely, asexual reproduction by cutting serves the purpose admirably.

[2] There are certain cases like variegation that are exceptions to this rule.

There is another case of a different kind. Sometimes the hybrid character is different from the character of the parents, even though the exact parental characters are reproduced by segregation in succeeding generations. The commercial carnation form is the result of crossing the single carnation with the huge worthless doubles called "busters." Reproduced by seed the commercial carnation throws both singles and busters, showing that segregation of the parental characters takes place; but as these plants are easily reproduced by cuttings, and the cuttings are all of the commercial type, sexual reproduction is only resorted to for the sake of producing new varieties. Another common phenomenon attending hybridization is sterility. Many very beautiful flowers produce no seed at all. This is even an advantage in some cases, because the plants flower more profusely than if they were spending their energies in the production of seed. Here again, cuttings are resorted to to reproduce the hybrid, or, as in the case of seedless oranges, the cuttings are grafted into an older rootstock instead of being rooted.

I stated at the beginning that there were two other classes of hybridization phenomena, the production of fixed first-generation hybrids and the production of blend hybrids. It is probable in the last analysis that the true explanation of these cases is the same; so we will consider them together. It is believed by many that there are kinds of inheritance other than Mendelian, that is, inheritance where no segregation occurs. Far be it from me to deny this; *I simply state the fact that there are no exact data extant proving other kinds of inheritance.* Such data may be found, but it is useless to speculate upon other laws without such evidence. There are several cases in which either new characters that breed true or blended characters that breed true *appear* to have been formed, but they have not been studied with sufficient care for an analysis of their mode of inheritance to be accurate and final. It is in crosses between true species that hybrids have been formed seemingly as constant and uniform as their parent species. Janczewsky has produced several such hybrids. Perhaps the most famous, however, are the blackberry-raspberry crosses first produced by the late E. S. Carman, editor of the *Rural New-Yorker* and later by Luther Burbank and others. Several hybrids having a commercial value have been made in this genus (*Rubus*), and all of them reproduce approximately true from seed. These are the facts and show what may sometimes be expected by hybridizers when crossing true species; but I wish to point out that this does not necessarily mean that we are dealing with a new mode of inheritance. Bramble species produce seedlings that are quite variable and in which the variations are extremely difficult to describe; there is, therefore, no exact information as to the relative variability of the hybrid seedlings as compared to that of the two parents. It may be said, then, that it is yet unknown whether there is partial segregation.

FIG. 8. FIRST HYBRID GENERATION OF CROSS BETWEEN "HAVANA" AND "CUBAN" VARIETIES OF TOBACCO SHOWN IN FIG. 7. Plant is taller than either parent showing the increased vigor due to a cross. Size of leaf of "Havana" is dominant. Habit of growth of "Cuban" is dominant. Number of leaves is intermediate, but approaches the "Cuban."

But why should there not be complete segregation to the types of each parent? In the first place, because it is likely that numerous separately heritable characters are concerned, and when *n* pairs of characters are concerned it takes four to the *n*th power seedlings to run an even chance that there will be one plant like each of the parents. When we consider that with ten pairs of characters, this means over 1,000,000 individuals, we can see with what enormous numbers one has to deal. In the second place these hybrids are only partially fertile, and as I have suggested in former papers, some consideration must be given the fact that there may be selective fertilization that works against ex-

FIG. 9. RECOMBINATION OF CHARACTERS OF PLANTS SHOWN IN FIG. 7, occurring in the second hybrid generation. This is a uniform and constant type having the short habit of growth and large leaves of the "Havana" parent, combined with the high number of leaves of the "Cuban" parent. It is now grown in the Connecticut River valley and yields 40 per cent. more than the Havana type.

treme segregation. To take a hypothetical case, suppose two plants are crossed in which the flowers of one are twice as long as the flowers of the other and that this extra length is controlled by three or four separately heritable factors. If only a few of the egg cells can be fertilized on account of dissimilarity from the pollen cells, one would expect only those seeds to be formed that would come from the fusion of the germ cells nearest alike. Intermediates would therefore be more likely to be formed than extremes. There is one other possible way of accounting

FIG. 10. SEGREGATION OF SIZE CHARACTERS. At left *Nicotiana rustica brazilia*. This plant was crossed with *N. rustica scabra* shown at left in Fig. 11. At right is a segregate of the second hybrid generation which is exactly like its parent. Unfortunately it has branched at the base or the similarity would be more striking.

FIG. 11. SEGREGATION IN SIZE CHARACTERS. At left *Nicotiana rustica scabra*. This plant was crossed with *N. rustica brazilia* shown at left in Fig. 10. At right is a segregate of the second hybrid generation exactly like its parent in size of plant, leaf and flower and in habit of growth.

for constant intermediate hybrid races which I think has never before been mentioned. In crossing species of the genus Nicotiana, I have had plants develop from seed that have apparently been formed apogamously, that is, formed from an immature egg cell without fertilization. It is evident that this is induced by the extraordinary irritation of foreign pollen. The true hybrid plants that are formed are generally blends in the first generation. The question, then, arises: May not the difficulty of maturing sexual cells in a wide cross sometimes cause apogamous development and therefore a continued propagation of a constant and uniform race?

All but the last of these suggestions may also be pertinent in the case of varietal crosses where there is said to be a blending of characters that deal with size. I am not certain, however, that all the so-called blend hybrids might not show segregation if studied in large numbers. I have found such segregation in size characters in crosses of both maize varieties and of tobacco varieties.[3]

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[3] In the writer's paper "The Rôle of Selection in Plant Breeding" in the August number of this journal, the legends for figures three and four unfortunately were interchanged in printing.

INHERITANCE IN MAIZE.

By E. M. East and H. K. Hayes.

Connecticut Agricultural Experiment
Station Bulletin, No. 167,

and

Contribution from the Laboratory of Genetics
Bussey Institution of Harvard University No. 9.

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INHERITANCE IN MAIZE.

BY

E. M. East and H. K. Hayes.

INTRODUCTION.

The investigations reported in this paper were begun in the spring of the year 1906. During the first four years the work was conducted at the Connecticut Agricultural Experiment Station. Since the fall of 1909, it has been carried on both there and at the Bussey Institution of Harvard University. Strictly speaking the researches comprise more than five years' work, for several of the pure varieties used as parent stocks had been selfed for the two previous years, so that a number of crosses were made in 1905 with full assurance that as far as most of the visible characters were concerned, the parent strains were pure. There was some further advantage gained in that the writers have been interested in experimental maize breeding since 1900, for without this experience the probable error of the results would be greatly increased.

Genetic research during the past decade has been very fruitful of results; nevertheless one could scarcely say that the field has been thoroughly surveyed, much less that any part of it has been completely investigated. The rediscovery of Mendel's law in 1900 opened up a new era in the search for the principles

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*"Contribution from the Laboratory of Genetics, Bussey Institution of Harvard University, No. 9."

of heredity. Mendel's chief discovery — segregation of potential characters in the germ-cells of hybrids and their fortuitous recombination — was one of the really great achievements in biology, but even so, it may be questioned whether his chief legacy is not his method of work. As has already been stated by Bateson, previous investigators even including the biometricians dealt with facts *en masse*, and the seeming order of the mathematical formulas deduced served rather to conceal than to reveal the individual facts. Mendel's method of individual analysis by the study of simple characters in carefully controlled pedigree cultures, however, has yielded and will continue to yield results of great value to science. Still, since we are dealing, as yet, with the simplest elements of the science of genetics, the subject matter of an investigation may be expected to yield results (other things being equal) somewhat in the proportion in which it fulfills the following technical requirements.

1. The genus or species under investigation should be variable. There should be a goodly list of types which are differentiated by definite characters easy of determination. That is, the differences should be largely qualitative and not quantitative.

2. The different types should be freely fertile *inter se*, unless an investigation of the causes of sterility is contemplated.

3. The flower structure should be such that the technique of crossing and selfing is simple and accurate.

4. Since the accuracy of an analysis of the manner in which characters are inherited increases — generally speaking — as the square root of the number involved, the subjects should return a large number of seed per operation (selfing or crossing).

5. The flowering branches should be numerous. This is necessary for three reasons. If one is dealing with flower characters he must be able to determine the character of the plant from a mature flower while immature blossoms still remain for the production of the controlled seed. Obviously, it is also an advantage when dealing with plant characters,

to have more than one opportunity to secure a desired union. Further, it is advantageous to be able to make several different crosses upon one plant.

6. Seed should be viable for several years in order that different generations may be compared at the same time. This enables one to reduce to a minimum the physiological fluctuation due to varying environment which many characters undergo in a marked degree.

7. The subject material should be "workable" cytologically in order that it may be attacked from both standpoints.

It might be remarked here that some botanists consider that genetic research can throw no new light upon evolution and upon the meaning of species, unless the subject material is an uncultivated genus or species. This criticism is apparently no more pertinent than the one the chemists had to meet years ago when they were told that synthetic compounds could not possibly be the same as those produced by nature. The fact of physiological fluctuation due to varying environment is admitted, but it is *not admitted* that the *mechanism* of hereditary transmission of the character in question is affected by these fluctuations.

Some idea as to the effectiveness of an inquiry concerning inheritance in maize from the standpoint of science may be gained then by examining the degree in which the plant fulfils the above requirements.

Although the forms of maize are regarded by botanists as belonging to the one species *Zea mays* L., there is probably no species of the flowering plants — if horticultural color varieties are excepted — that appears under such varied forms. These forms are perfectly fertile *inter se*, moreover, so that the first and second of our requirements are fulfilled perfectly. The third requirement, that of an easy technique and accurate control of desired matings, is met very imperfectly. The plant is monœcious. Ordinarily, this type of flowering habit is desirable in pedigree culture work because accidental selfing is usually much more easy to prevent than it is in hermaphroditic plants. In the case of maize, however, there is such an enormous production of pollen that it is continually present in the air of the maize field. In spite of all precautions, therefore, seeds of unknown paternal ancestry do creep into the cultures. The general error due to this source has been determined in cases which are described later, but the determination of a probable error in a mass of data is not sufficient in genetic work. An actual error in which a single seed of unknown paternity becomes the ancestor of a pedigreed line, is sufficient to upset all inductions drawn from the data. For this reason the cultures have had to be larger than would otherwise have been necessary.

The requirement of a large number of seeds from one union to reduce the probable error of chance fertilization among gametes differing in character is quite satisfactory in maize as from two hundred and fifty to twelve hundred seeds are produced on the cobs of the various races. The small number of flowering branches, however, is a serious objection. In some cases there are two or even three and four ears upon each plant; but in most cases, especially in the large races, there is but one ear. And even where there is an extra ear one gains but little advantage. The ears mature about the same time and it is impossible to find out what seed characters the plant possesses before pollinating the ear which is to have its place in the controlled culture. The disadvantage of this fact to the investigator is apparent if one remembers that when studying ear abnormalities sometimes twenty to twenty-five cobs must be selfed by hand to be reasonably certain that one selfed ear with the desired characters is obtained.

Maize seed is rather delicate and must be carefully dried in a place where there is a good circulation of air. When dried until the moisture content is only about ten per cent, it remains in fairly good condition for three seasons. Seed older than this is almost worthless. In fact, there is a possibility of obtaining distorted results even in the second year. Ninety-eight to one hundred per cent of properly dried seed should germinate the next spring after harvesting, but this percentage falls to about ninety the year following. If, therefore, seeds of any particular gametic structure should lose their vitality first, incorrect results would be obtained.

The chromosomes of maize are small and difficult to study and scarcely anything is known of their behavior during the maturation divisions.

This discussion should give some idea of the advantages and disadvantages that maize presents as subject material for a genetic investigation from the standpoint of pure science. The plant, however, even if not as perfect as some others from this point of view, has another claim which ought not to be disregarded. The fact that maize is the basis of the agricultural wealth of the country makes it eminently desirable that every fact about the inheritance of its characters, should be learned as soon as possible. It is only through the application of such knowledge that the present arbitrary, and, in a way, unscientific methods of its improvement as an economic crop will be placed upon a definite and orderly basis.

PART I.

THE MATERIAL AND THE PROBLEM.

The Plant and its Origin.

Although there is no absolute information as to the origin of maize, most botanists agree that its original home is Mexico (Harshberger '93) or the region to the south of there. As to how it originated there has been much speculation, and various views are held by different writers. We think it not out of place to give here a synopsis of the most important theories, because in our opinion, the results from the pedigree culture work on the inheritance of plant characters described in Parts IV and V throw considerable light on the subject.

The Tribe Maydeæ of the order Gramineæ comprises but seven genera and only sixteen or seventeen species. The two genera which interest the maize student are Zea and Euchlæna both of which are monotypic. The especial distinctions between the two are given by Lamson-Scribner (: 00) in his key to the genera of Maydeæ as follows:

"**Euchlæna,** pistillate spikes axillary fasciculate, distinct, axis of each articulate."

"**Zea,** pistillate spikes axillary, grown together, forming a compound spike with a much thickened, continuous axis."

His complete descriptions are:

"*Euchlæna* Schrad. Ind. Sem. Hort. Gœtt. 1832. Spikelets unisexual, monœcious; the staminate 2-flowered, in pairs, one sessile the other pedicellate, arranged in terminal paniculate racemes; the pistillate 1-flowered, sessile and solitary at each joint of an obliquely articulate rhacis of a simple spike; the spikes fasciculate in the leaf axils and each more or less enveloped by a foliaceous bract. Glumes in the staminate spikelets 4, acute, the first two membranaceous, empty; flowering glumes smaller and like their paleas, hyaline. Stamens 3. Glumes of the pistillate spikelets 4, the outer one broad and boat-shaped, smooth, soon becoming hard, surrounding the inner glumes and narrow rhacis, second glume empty coriaceous, third glume hyaline with a palea but no flower; fourth or flowering glume with its palea hyaline. Styles very long, filiform, shortly bifid at the apex.

Tall annuals with long and broad leaves, closely resembling Indian corn in habit. Species one with several varieties in Mexico and Central America."

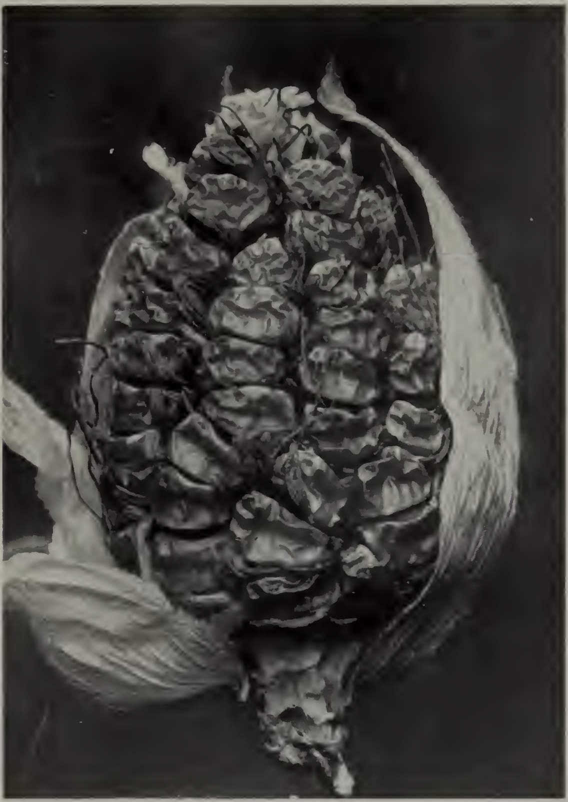
"*Zea* Linn. Sp. Pl. 971. 1753. Spikelets unisexual, monoecious; the staminate 2-flowered in pairs, one sessile the other pedicellate, along the numerous branches of a terminal panicle; the pistillate 1-flowered, sessile, crowded in several rows, along a much thickened continuous axis arising from the lower leaf-axils and closely enveloped by numerous large foliaceous bracts. Glumes four, awnless; those of the staminate spikelet acute; those of the pistillate very broad and obtuse or emarginate. Grain hard, only partially inclosed by the fruiting glumes. A well-known tall and striking annual grass with erect stems and broad leaves. The terminal staminate inflorescence forms the "spindle" [tassel], and the long projecting styles of the pistillate flowers constitute the "silk." The cob is formed by the union of the axes of several female spikes into a much thickened body. Species one or two, of American origin, presenting many varieties in cultivation known as corn, Indian corn or maize."

From these descriptions of the two monotypic genera, it is seen that *Euchlæna mexicana* Schrad., the common teosinte, is not greatly different from *Zea mays* L., our ordinary maize. Indeed to one who has grown and followed the extraordinary variability of both, it does not seem a greater step from teosinte to the maize most similar to it — the short many branched pop or flint types — than it does from the small dwarf pop maize to the giant dent forms. Teosinte is perfectly fertile with maize, which fact has led to some confusion, for Watson ('91) thinking that hybrids between the two constituted a pure wild species, named it *Zea canina* Watson. Segura (Harshberger, '96), however, by remaking the crosses and growing them near the region where the "Zea canina" was found, clearly proved the true nature of the latter. Harshberger ('93) first fell into the same error as Watson but later (Harshberger '96) discovering the true state of affairs suggested that maize originated either from (1) a cross between teosinte and some extinct but closely related plant, which by variation under a better environment finally produced a plant with larger maize-like ears; or that it came from (2) a cross between teosinte and a race of the latter that had varied under long continued cultivation. The basis of Harshberger's argument that teosinte must have been crossed by another form is his idea that only in the progeny from a cross would sufficient variability have appeared to have produced the more vigorous plant — the aboriginal maize.

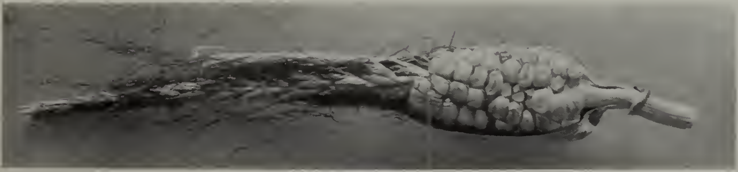
More recently Montgomery [: 06] has advanced the theory that teosinte and maize are both descended from an unknown many-branched grass whose branches terminated in a panicle of spikelets bearing hermaphrodite flowers. He says: "As evolution progressed, the central tassel came to produce only staminate flowers, these being higher and in a better position to fertilize the flowers on the lower branches. At the same time, the lateral branches came to produce only pistillate flowers, their position not being favorable as pollen producers, while, on the contrary, they were favorably placed to receive pollen. This differentiation in the flowers was accompanied by a shortening of the internodes of the lateral branches until they were entirely enclosed in the leaf sheaths [the husks]." The especial difference between the evolution of teosinte and of maize was thought to have been in the development of the ear of the first from the lateral branches of the tassel-like panicle and the ear of the second from its central spike. This argument of Montgomery is directly opposed to the old theory that the cob of modern maize is the result of a fusion of a number of two-rowed pistillate spikelets such as are found upon teosinte. His theory then, emphasizes the nature of the changes that took place; Harshberger's theory, the way they were transmitted.

In addition to these views it seems only necessary to mention that since maize is the only grass with a naked seed, the podded variety *Z. mays tunicata* Sturt. is by many considered to be an earlier stage in maize development.

Our own views on the subject have resulted from a consideration of the behavior of the characters of the various races of



a Ear with hermaphrodite flowers from the dwarf plant which appeared in Stowell's Evergreen sugar maize.



b. Mature seeds on male spike of plant heterozygous for starchiness, showing segregation. A common physiological fluctuation.

Abnormalities

maize when crossed, the data on which they are based being given later. The matter is largely speculation and should be considered as such. It is merely the simplest manner of interpreting the known facts, by connecting maize with the other Maydeæ by a short series of changes that involve characters that mendelize. On the whole it does not differ greatly from Montgomery's theory.

Since we now believe that the essential rôle of hybridization is to recombine the characters possessed by the parent plants in definite ratios without actually producing anything new, [new combinations may produce characters formerly unknown], there is no necessity of postulating hybridization of teosinte with a more maize-like variety. It is known that when teosinte is cultivated in rich soil it sometimes produces ears having an irregular development of four rows. This is only an expression of one of the commonest modes of variation, repetition of parts or meristic variation. This variation in the ear has taken place again and again in maize and is inherited although sometimes obscured by physiological fluctuation. The ear of maize then is a meristic variation produced from the central spike of the tassel of the lateral branches of teosinte or of a teosinte-like plant, and not a fusion of the lateral spikelets. Lateral spikelets still appear in maize, apparently as if variation ran in grooves or paths of least resistance. This character has been found to segregate in a manner essentially Mendelian. The podded character also mendelizes and is allelomorphic to its absence. If then progressive meristic variations occurred in the central spikes of the side branches of the teosinte-like ancestor, followed by retrogressive variations affecting both the lateral spikes of the lateral branches and the pod character, a plant would have originated bearing naked hermophroditic ears. Further change might easily have come about, as Montgomery suggests, by a shortening of the side branches producing the modern husk, and finally the origination of the monœcious character. The latter occurrence is not at all hard to picture for the change of the staminate inflorescence to an hermaphroditic or even a pistillate one, is something which is exceedingly common in all or almost all strains of maize. It is a physiological fluctuation produced by excessive rainfall and fertile soil. The appearance of stamens on the modern maize ear is much more rare but that it does occur is shown by the ear pictured in Plate I. In fact one of our sterile dwarf mutations had nothing but hermaphroditic flowers.

The Varieties of Maize.

Although all of the varied forms of maize are regarded by modern taxonomists as sub-divisions of the species *Zea mays* L., many varieties have at various times been given specific rank. The Index Kewensis gives the six following types as species. The original sources have been consulted but the descriptions have been shortened to include only essential facts.

Z. Curagua, Molina, J. I. Saggio sulla storia naturale del Chili. pp. 306, Bologna, 1810. = *Z. mays.*

This variety is distinguished by its serrate leaf-edge. It has never been cultivated in the United States, but appears to be a flint type, *Z. mays indurata.* Syn. *Z. Caragua*, Stend. Nom. ed II, ii. 797.

Z. erythrolepsis, Bonafous, M. Histoire naturelle, agricole et économique du Maïs. Folio, pp. 181, Plates 19, Paris, 1836, = *Z. mays.*

"*Glumis rubris, seminibus compressis.*" "Le Maïs à rafle rouge cultivé sur les rives du Missouri, se distinque par l'aplatissement de ses grains, et surtout par le coleur rouge des écailles et corallines de l'epi femelle. La constance de ce caractère tend à lui mériter le titre d'espèce."

This form could hardly be considered a variety as it is a common variation in all of the commonly recognized varieties.

Z. hirta, Bonafous, M. Note sur une nouvelle espèce de Maïs. Ann, Sci. Nat. Ser. I v.17; 156-158. 1829. = *Z. mays.* "*Foliis hirtis et dependentibus; spiculis masculis sessilibus, diandris triandrisve; antheris subaureis.*"

A good variety, originally sent to Bonafous from Balbis of the Jardin des Plantes de Lyon. It varies into a series of flint, pop and dent types.

Z. japonica, Van Houtte, Fl. des Serres, XVI (1865-67), 121. t. 1673-74. 1867. = *Z. mays.* Syn. *Z. vittata*, Hort. and *Z. variegata*, Hort.

A small variety with leaves variously striped with white. A small flint type is the one chiefly cultivated for ornament, but the variety occurs again and again in fields of all of our common maize strains. It could undoubtedly be isolated pure by careful selection of these individuals.

Zea macrosperma Klotzsch. Bot. Zeitung 9; 718. 1851. In der Sitzung des Ges. naturf. Freunde zu Berlin. = *Z. mays*. Seed received by Humboldt from Cuzco. It is simply a large seeded dented starchy type.

Zea rostrata, Bonafous, M. Ann. Soc. Agr. Lyon. v. (1842), 197. = *Z. mays*. Simply a hook-seeded form of pop maize, somewhat similar to our common rice pop.

Of these types *Z. mays Curagua* Molina and *Z. mays hirta* Bonafous might be considered as good varieties. The four remaining names and also the varieties of *Z. mays* listed in the Index Kewensis might well be placed under the classification porposed by Sturtevant ('99), leaving out his *Zea mays amyleasaccharata* because the latter is a type which is probably identical with the "flinty" sweet corns with which canners have so much trouble. The three ears from the San Padro Indian collection sent to Sturtevant by Palmer, and upon which he based the variety, failed to yield a mature crop in Geneva, New York. It is therefore unknown whether this type would really prove true. Sturtevant's classification follows although I have added the word *mays* and have listed them as varieties instead of species. It is not strictly correct to give him as the authority for the names, as he used them specifically, but since they have come into general use in the United States it seems more convenient to keep them. Sturtevant himself based his claim for separate species principally upon the fact that intermediates were either absent or rare. This fact comes about, as will be shown later, from the alternative manner in which the distinguishing characters are inherited. In reality many other characters are inherited in the same manner, and it is only because the chief characters of these five varieties are striking to the eye that it is advantageous to keep them in use.

Zea mays tunicata, the pod corns. Sturtevant, Bul. Torr. Bot. Club 1894, p. 355 (Also St. Hil., Ann. Sci. Nat. 16; p. 143, fide De Candolle). A form in which each kernel is enclosed in husks (usually four) besides the foliaceous bracts that enclose the ear.

This form was first described by C. Bauhin in 1623, and has been the basis of a long list of synonyms since that time. It is probable that the prototype of the species possessed this character for it would thus be linked closer to the other Maydeæ. The strains now obtainable under this name have been hybridized until ears can be found whose kernels run the whole gamut of the other four kinds. The tendency of plants to form anew characters once possessed that have been lost, is well illustrated here. We have come across several ears of our ordinary varieties in which a few of the kernels at the base have been podded. Sturtevant gives two authentic cases where fully podded ears have appeared in other varieties under such conditions that it is very improbable that it was the result of hybridization.

Zea mays everta, the pop corns. Sturtevant, Bul. Torr. Bot. Club, 1894, p. 324.

"This [species] group is characterized by the excessive proportion of the corneous [starch in the] endosperm and the small size of the kernels and ear. The best varieties have a corneous endosperm throughout. This gives the property of popping, which is the complete eversion or turning inside out of the kernel through the explosion of the contained moisture on the application of heat."

Strains of the pop maizes are the smallest of our cultivated corns, and although there are varieties that reach a height of nine feet when cultivated on fertile soil, plants comparable in size to the average dent or starchy maize are never found. There appears to be a distinct correlation between size of seed and size of plant; therefore, since one never obtains large size seeds without a development of soft starchy matter, pop kernels much larger than those now grown are not likely to be produced through selection or hybridization.

Two forms of seed are known in the pop corns; one is simply a small seed with rounded crown similar in shape to the small flints; the other, characteristic only of pop corns, is peaked at the point where the style or "silk" was attached.

Other variations such as purple colored aleurone cells, yellow endosperm, red silks, and red and variegated pericarps characterize the pop maizes in common with the flint, sweet, dent and starchy corns. The modal number of rows also varies in different varieties from eight to sixteen. In this, pop maize is similar to dent, sweet and starchy, but different from flint maize. It is doubtful whether any true flint maize exists with a mode for number of rows higher than twelve.

Zea mays indurata, the flint corns. Sturtevant, Bul. Torr. Bot. Club, 1894, p. 327.

This group is characterized by the seeds having a corneous starchy endosperm, surrounding a soft starchy center immediately behind or partially surrounding the embryo. The strains in common cultivation are considerably larger than the pop corns, but varieties do exist which form a definite series from pop to dent differing only by the amount and extent to which soft starch replaces corneous starch in the endosperm. The same color varieties that were described for pop corns occur.

Zea mays indentata, the dent corns. Sturtevant, Bul. Torr. Bot. Club, 1894, p. 329.

A group characterized by the extension of the soft starch until it completely covers the summit of the seed. Corneous starch, however remains at the sides of the kernel and acts as a frame work to support the drying seed. The soft starchy portion shrinking in drying to a much greater extent than the other forms a characteristic indentation. Dent varieties occur averaging from five feet to twenty feet (reported) in height, with from eight to twenty-four rows as the modes (extremes to thirty-six rows). The usual color varieties occur.

Zea mays amylacea, the soft or flour corns. Sturtevant, Bul. Torr. Bot. Club, 1894, p. 331.

A group characterized by entire absence of corneous starch in the endosperm. Uniform shrinkage in drying usually gives a seed with no indentation. The mummy corns of Peru, Mexico and the southern United States appear to belong to this group, but this is not absolutely certain. The specimens that we have examined belonging to the New York Botanical Garden might have been flint corns which owe their floury appearance to partial decomposition.

This group marks the final disappearance of corneous starch in the endosperm. It is the end of a series beginning with the pop corns and coming up through the flints and dents. For this reason one might expect them to possess the largest seeds, as the length of time necessary for maturing the seed undoubtedly has something to do with the amount of soft starch formed. The plants are indeed large, but seeds occur varying from the size of the smaller flints to that of the larger dents. The origin of the starchy corns is not due simply to their correlation with the general plant structure and therefore a simultaneous origin with large varieties, but is dependent upon a separate character or group of characters. The usual color varieties occur.

Zea mays saccharata, the sweet corns. Sturtevant, Bul. Torr. Bot. Club, 1894, p. 333.

"A well defined group characterized by the translucent horny appearance of the kernels and their more or less crinkled, wrinkled or shriveled condition." The sweet corns are simply pop, flint and dent varieties (East :09) that have lost their ability to mature starch normally. Some few starch grains are produced but they are generally small, angular and abortive. The reserve material of the endosperm seems to undergo a decomposition to cane sugar and the various hexoses. There is apparently something more than a simple non-development of starch, for the sweet corns in the "milk" state contain greater percentages of sugar than do the starchy varieties in a similar stage of ripeness. The same color varieties occur.

The Problem and its Treatment.

It is apparent that maize furnishes an admirable series of types which are perfectly fertile among themselves. The primary object of our work is to obtain pure forms of these diverse types by inbreeding, then to test the mechanism of inheritance of each separate character by controlled matings and an analysis of the resulting progeny. In doing this we simply follow Mendel's method of the individual analysis of pedigree cultures.

The specific questions attacked are numerous. Our principal object is to find whether the different characters under observation all obey the same law of heredity or whether separate principles are involved, and whether characters apparently inherited independently are not sometimes correlated with each other. The question of dominance of a character in the first generation of a cross has also interested us. Some characters are perfectly dominant, other characters imperfectly dominant, while still others form heterozygous combinations differing from either of the parents. There are even cases in which dominance appears to be reversible. If such a thing is possible, an explanation is desirable. The study of the phenomenon of Xenia, which has already formed the basal object of Correns' (: 01) fine monograph, throws some light on these questions.

Another object we have kept in mind is the problem of the purity of extracted homozygotes. It is a matter of common knowledge that characters that have been lost through retrogressive variations — characters that behave as mono-hybrids in inheritance — often reappear. The reverse of this phenomenon is also true. Is it because there is a phylogenetic "path" in which these changes run, so that the same variation appears again and again, or is there no absolute purity of the germ-cells but only a comparative purity as indicated by Morgan (: 10)?

The idea of prepotency has been held with great tenacity up to the present time. We hope these researches will throw some light upon this subject of so much importance to practical breeders. If there are individuals whose constitution is such that chance production of zygotes is interfered with, the fact brings many complications into the study of inheritance; but such complications must not interfere with the facts. Various other questions will be discussed in the proper places and for this reason will not be considered further here.

It may be well to mention that although these questions smack of the technical, it is maintained that in just so far as one contributes toward their solution, that far is the broad practical problem of better methods for the production of new economic maize types solved. The questions of purity of homozygotes, inheritance of size and number of ear-rows in the different sub-species are easily seen to be of practical agricultural importance. Other questions may seem of less importance or even of no importance from this point of view, but this is fallacious as is easily shown by illustrations from the science of chemistry where abstruse theoretical researches have continually proved to be the most practical in the end.

In certain quarters there has been a marked reaction against the continued Mendelian interpretation which has been given to every paper published since the year nineteen hundred in which actual experimental studies concerning the mechanism of inheritance have been reported. This reaction has taken the form of a philosophical query as to whether the characters of the organic complex of which living organisms are composed can in any sense be dissected and analyzed into the "units" of heredity which are the basis of Mendelian inheritance. Further it has been questioned whether there is any justification for the increasing complexity with which Mendelian formulæ are involved. It has been argued that with a multiplicity of "factors" any particular case can be interpreted as segregating according to the Mendelian hypothesis. For these reasons the writers wish to have their position in reporting the following investigations distinctly understood at the beginning.

It is fully understood that there is danger in improper analysis of complex ratios from pedigree cultures. This is inevitable. Yet it is not a more pertinent criticism to condemn complexity in biological facts than it is to frown upon the intricacies of modern organic chemistry because it is so different from the simple chemistry of Liebig. The answer is that the facts of heredity are complex.

In regard to the question of the ultimate nature of unit characters or the possibility of absolute segregation of characters in the germ-cells so that in the recessive there is actual absence of the character (gene) in question, we must await more results from the different points of view of the breeder, the cytologist, the physiologist and the physiological chemist. The facts reported in genetic investigations remain indelible. The interpretation of these facts may or may not be correct; they simply arbitrarily represent the facts in a convenient system of notation much as the facts of chemistry are represented by structural formulæ. This is the idea in the minds of the authors in the following report. It is thought moreover to represent the attitude of most genetic investigators and the excuse for making the above statements lies in the fact that unfortunately one often finds no appreciation of this attitude by biologists not actually engaged in genetic research.

We have, then, used the ordinary Mendelian notation, with allelomorphic pairs interpreted as presence and absence of characters not because we know that there is actual absence but because this interpretation fits our present knowledge. We have interpreted complex characters such as height which we are not able to analyze completely, as segregating characters. The conclusion that there is a segregation in the second hybrid generation very much in excess of the sum of the non-inherited fluctuation and of other variation due to the heterozygous condition of the pure (?) forms used and also of their combination in the first hybrid generation, is justified by the data. This is the essence of Mendelian theory; and, whether absolutely correct or not, it is an interpretation that cannot fail to be valuable to the commercial plant breeder. It gives him some knowledge of what may be expected in his endeavors to produce new types of maize by hybridization.

It might also be mentioned that following Johannsen, the word "gene" has been used to signify that substance present in the germ-cell which represents potentially the "unit character" or whatever it may be called that acts as an entity in heredity.

Previous Work on Inheritance in Maize.

Before describing in detail the material used in these experiments it may be well to give a short summation of the previous work in the field.

The early hybridists, Camerarius, Logan, Pontedera and Henschel, each made a desultory study of maize crosses, but obtained no results of present interest. Hardly more satisfactory are the papers of Dudley (1724), Sageret ('26), Puvis ('37), Gärtner ('49), Naudin ('63), Hildebrand ('67, '68), Vilmorin ('67) and Focke ('81), although these researches — representing work of the principal students of hybridization of the period — each gives several observations concerning the immediate effect of pollen upon the endosperm, — that phenomenon called Xenia by Focke ('81). These observations, however, can hardly be compared with those made since the cause of Xenia was discovered for the obvious reason that the facts concerning the changes in the endosperm were almost lost to sight in the search for effect of cross-pollination on the tissues of the maternal parent.

In the work of a slightly later period particularly in the United States (Kellerman and Swingle '89, '90; McCluer '92; Morrow and Gardner '92) a great improvement was made in the methods of investigation employed. The parental stock was often inbred to establish its purity, crosses were made by hand upon protected flowers, and the resulting progeny were studied with great care. Many facts of inheritance are unconsciously reported in their papers which are confirmed in the post-Mendelian work which gives them a meaning For example one finds these data in Kellerman and Swingle ('91). A *chance hybrid evidently produced by the pollination of a white maize with pollen from a variety with purple aleurone cells was inbred. A hand-pollinated ear contained 370 seeds, of which 206 were blue, 71 pink, 71 orange-yellow and 22 pure white. One wonders how the essential facts of dominance and segregation remained unnoticed in the face of such ratios as this. But even if it is interesting to reread these papers and consider them from a more modern viewpoint, it is hardly profitable to discuss them further here. The work previous to 1900 was in the wrong epoch, and since that time three valuable contributions to the subject in hand have been made. (De Vries 1899 and 1900, Correns 1899, 1900 and 1901, and Lock 1906.)

It is interesting at least, to note that in the cases of both De Vries and of Correns the studies of maize hybrids in which presence and absence of yellow and presence and absence of starch in the endosperm were concerned, contributed largely to their independent discoveries of dominance and segregation in hybrids, which they both unselfishly credited entirely to Mendel after their discovery of his previous paper. Thus *Zea mays* shares with *Pisum sativum* the honor of being the subject material in the establishment of Mendel's laws.

Correns' (:01) beautiful monograph was written with the especial idea of furnishing an explanation of the phenomenon of Xenia, but it naturally contributed a large amount of data upon the mechanism of inheritance of the characters with which he worked.

Correns' technique was as follows. The seeds were planted first in pots, allowed to attain a healthy start, and finally transplanted to the field. In the first year (1894) the plants to be used as "mothers" were planted together in his experiment field, castrated at the right time and the silks protected between

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* The immediate parents were thought to have been white, but this was probably an error.

pollinations with paper bags. The individuals that furnished the pollen were planted together in places apart from the proposed mother plants and from them pieces of the male panicles (tassels) were carried in glass bottles to the mother plants. Slight changes in the plan of planting were made in 1895 and 1896, but I cannot find in any case that either the male flowers were protected from foreign pollen during their maturation or that special care was taken to have pollen for a cross furnished by an individual plant. Furthermore in handling the hybrids individuals were not selfed but bred *inter se*. Some of the families were given to gardeners who were growing no other maize, while others were detasseled and naturally pollinated *en masse* with the pollen of a pure race. The first method predominated. We can see then that the methods in use furnished correct results only when the characters in question were simple and of such nature that the races could be kept pure by inspection. Complex ratios such as are furnished when maize with purple aleurone cells is crossed with various white maizes differing in gametic structure, could not possibly be analyzed correctly.

Correns reached conclusions regarding the mode of inheritance of the following characters but it must be borne in mind that these results came from the study of data more or less massed, and not the study of individual crosses in as precise a manner as that outlined by Mendel.

Yellow endosperm was found to be dominant to its absence, and starchiness dominant to absence of starchiness (sweet). Both of these characters behaved as Mendelian mono-hybrids. It cannot be definitely stated, however, that crosses between all races of maize where presence and absence of these characters are concerned would give the same results. Long aleurone cells also proved dominant to short aleurone cells, and red pericarp to absence of red, but Correns was not entirely satisfied that these characters behaved as simple Mendelian monohybrids although he supposed this to be the case.

Purple aleurone cells appeared to form an allellomorphic pair with absence of purple, but he found that the heterozygotes when bred *inter se* did not give the normal number of whites. What he took to be heterozygotes of the same character were either pure purple, partial purple, or pure white when the purple was used as the male parent. In the reverse cross the purple appeared to be fully dominant. Correns (: 01) endeavors to explain this phenomenon by the fact that in the formation of the hybrid endosperm two nuclei come from the female and but one from the male parent. He supposes that in some cases this may cause a dominance of the female characters. This purple character seemed to interfere with normal inheritance in still another case (Correns : 02), where a blue sweet corn was crossed with a non blue pop. Here the second generation yielded only about 15½% of sweet kernels out of a total of over 8,000. Pollinated with the recessive parent there appeared nearly 50% of sweet kernels so that the female germ-cells seemed to have segregated normally. Correns suggested that in this case the four possible combinations of characters in the germ-cells did not take place with equal facility.

Our own data shows the error in the first case to be due to the fact that white races differ in their gametic structure in characters which affect the purple color. The observations in the second case have not been confirmed, but were probably due to improper classification of the heterozygous dominants and the recessives. (See starchy and non-starchy crosses.)

The shape and size of seed and relative weight of embryo and endosperm Correns thought behaved in a non-Mendelian manner, although he was not prepared to say in exactly what manner these characters were inherited.

Lock (: 06) carried out a much more extended series of maize crosses at Peradeniya, Ceylon, from 1902 to 1906. His technique in certain cases was a considerable improvement on that of Correns in that both the male and female inflorescences were enclosed in bags and thus crosses were made between single individuals. Unfortunately his method was later changed and cross pollination was accomplished by planting the two races in alternate rows on an isolated plot of ground, and detasseling all plants of the race which it was proposed to use as the female parent. This method of course gave no chance to make a proper analysis of complex characters for could not be known just what gametic composition was possessed by the male parent. This criticism was anticipated by Lock himself but the method was used because he desired to have a large amount of data from which to establish the mathematical accuracy of Mendel's hypothesis of definite segregation and chance mating. In the cases of starchy and non-starchy, and yellow and non-yellow endosperm Lock's results were in accord with those of Correns. Furthermore he showed definitely that red pericarp behaved as a Mendelian character, allelomorphic to absence of red. Lock also crossed indented and non-indented races and remarks that in the F_2 generation a high degree of variability appeared, but without making crosses between individual plants and studying the progeny he could not decide whether or not Mendel's Law was followed. No data is reported on inheritance of height of plants but a number of crosses were made between F_1 plants and the shorter of the parental races, and he states that no segregation into short and intermediate plants took place. The plants on the contrary were remarkably uniform in height and he believed blended inheritance to be the rule for this character.

Lock's results in crossing races with purple aleurone cells with races with non-purple aleurone cells is so seriously complicated from the fact that he followed out no individual crosses that it is impossible to criticize his data. From the fact that individual ears showed such different ratios as 3 : 1, 9 : 7 and 1 : 3 we may suspect that he was dealing with white races of varying composition such as are described in our work on this character.

These short abstracts from the work of Correns and of Lock do not give an adequate idea of the large amount of painstaking investigation for which each should be credited however, and anyone interested in the subject should therefore consult the original papers.

The Material Used.

The types of maize which furnished the parental stock with which crosses were made for this series of studies were in most cases inbred by hand for at least two generations before any hybrids were actually made. When this procedure was impossible the parental ears were obtained from various commercial growers who made a specialty of the types which they furnished. From the maize obtained from them single ears were selected and planted. The plants forming the immediate progeny of these ears were used in part as the parents of crosses and in part to inbreed. When any of the seeds from the original ear were found to be heterozygous in any characters the fact is noted when the crosses are described. In this manner we were able to determine the purity of the parental stock used, for all of the grosser characters. Of course new variations were continually isolated and these were given numbers which show their origin. For example, the original stock of Longfellow corn is No. 15; if, however, new variations appeared in the Longfellow progeny they were numbered 15-1, 15-2, etc.

The following descriptions, then, comprise only original material; that is, single ears of various commercial varieties.

Zea mays tunicata. The podded corns.

21. Podded maize.

A fourteen-rowed ear with four husks around each kernel in addition to the usual paleas. The seeds looked like rice pop; they were small but showed a considerable amount of white starchy matter.

Zea mays everta. The pop corns.

20. A flint-like 8-row purple pop.

A pop with purple aleurone cells, showing a small amount of white starchy matter immediately behind the embryo, sufficient to keep the seeds from popping well. Ear 15 cm. long, 11 cm. in cir. Seeds .9 x .9 cm., white endosperm. Cob white.

60. Tom Thumb pop.

A dwarf true pop. Ear 7.5 cm. long, 8 cm. in cir., 12-rowed; pericarp colorless. Seeds rounded, true pop, .5 x .4 cm., endosperm yellow. Cob white.

23. White rice pop.

A white true pop. Ear 15.5 cm. long, 10 cm. in cir., 16-rowed. Seeds white, .9 x .5 cm., hooked. Cob white.

26. A white, flint-like pop.

A white true pop with rounded flint-like seeds. Ear 17 cm. long, 9 cm. in cir., 8-rowed. Seeds .8 x .9 cm. rounded. Cob white.

27. Red rice pop.

A true rice pop with red pericarp. Used only for inheritance of pericarp color.

28. White rice pop.

A true rice pop with white or colorless pericarp. Used only in cross with No. 27.

Zea mays indurata. The flint corns.

4. Benton maize.

An eight-rowed race intermediate between the flint and the dent corns. Ear 34 cm. long, 14 cm. in cir., 8-rowed, pericarp red becoming colorless at summit. Seeds 1.1 x 1.4 cm., some very slightly dented; endosperm yellow, slightly more starchy than a true flint. Cob white.

5. Watson flint.

A true flint with a pericarp rose red when developing in full sunlight, the seeds at the tip usually showing simply red striations beginning at point of attachment of the silk. Ear 27 cm. long, 13 cm. in cir., 8-rowed. Seeds 1.0 x 1.2 cm., endosperm colorless. Cob white.

11. Sturges' flint.

A twelve-rowed yellow flint race. Ear 20 cm. long, 14 cm. in cir., 12-rowed, pericarp colorless. Seeds 1.0 x 1.0 cm., endosperm yellow. Cob white.

13. Sanford flint.

An eight-rowed race. Ear 30 cm. long, 13 cm. in cir., 8-rowed; pericarp colorless. Seeds 1.0 x 1.3 cm.; endosperm colorless. Cob white.

15. Longfellow yellow flint.

An eight-rowed yellow race. Ear 27 cm. long, 11.5 cm. in cir.; 8-rowed; pericarp colorless. Seeds .9 x 1.2 cm.; endosperm bright yellow. Cob white.

17. Palmer's red-nosed yellow flint.

An eight-rowed yellow race. Ear 22 cm. long, 12 cm. in cir.; 8-rowed; pericarp a dirty red at the sides of seed becoming almost colorless at summit. Color not deep as in common red maize. Seeds 1.0 x 1.4 cm.; endosperm yellow. Cob white.

24. Rhode Island white cap.

An eight-rowed flint race. Ear 29 cm. long, 12 cm. in cir.; 8-rowed; pericarp colorless except for a slight pink tinge of rose similar to No. 5 but less in amount. Seeds .9 x 1.2 cm.; endosperm colorless. Cob white.

25. Brindle flint.

A common flint race not breeding true to the character from which it derives its name, — a mosaic pericarp formed by slashes of dark red extending irregularly from the point of the attachment of the silk. Eight-rowed true flint.

Zea mays indentata. The dent corns.

2. Illinois low protein dent.

A white dent selected for low proteid content at the Illinois Agricultural Experiment Station for eight generations. Protein content 8.30 per cent. Ear 19 cm. long, 18 cm. in cir.; 16-rowed; pericarp colorless. Seeds 1.5 x .8 cm.; endosperm colorless; white starchy matter largely increased in summit over usual dent type. Cob white.

8. Illinois high protein dent.

A white dent selected for high proteid content at the Illinois Agricultural Experiment Station for eight generations. Proteid content 15.46 per cent. Ear 20 cm. long, 14 cm. in cir.; 14-rowed; pericarp colorless. Seed 1.1 x .9 cm.; endosperm colorless; white starchy matter decreased from amount usual in dent types but summit still well dented. Cob white.

3. Leaming dent.

A yellow dent race. Ear 21 cm. long, 16 cm. in cir.; 20-rowed; pericarp colorless but sometimes very slightly tinted with dirty brick red at sides of seeds. Seeds 1.3 x .7 cm.; endosperm dark yellow; considerable soft starch at summit; well dented. Cob dark red.

6. Leaming dent.

Same race as No. 3 but of different ancestry. Ear, 19.5 cm. long, 18.5 cm. in cir.; 18-rowed.

7. Leaming dent.

Same race as No. 3 but of different ancestry. Ear 18 cm. long, 17 cm. in cir.; 20-rowed.

9. Leaming dent.

Same race as No. 3 but of different ancestry. Ear 18.5 cm. long, 16.5 cm. in cir.; 16-rowed.

12. Leaming dent.

Same race as No. 3 but of different ancestry. Ear 19 cm. long, 17 cm. in cir.; 18-rowed.

16. Leaming dent.

Same race as No. 3 but of different ancestry. This ear was 18-rowed and perfectly formed. It was surrounded by five lateral branches each having either four or eight rows of seeds.

1. Missouri cob pipe dent.

A very large dent race characterized by large cob. Ear 28 cm. long‘ 22.5 cm. in cir.; cob 14 cm. in cir.; 20-rowed; pericarp colorless. Seeds 1.5 x .9 cm.; endosperm white. Red cob.

22. Mosaic red dent.

A dent characterized by dark intense red pericarp. Used only for study of that character.

Zea mays amylacea. The flour corns.

10. White floury.

A thoroughly floury race, showing absolutely no corneous starch. Ear 22 cm. long, 14.5 in cir.; 14-rowed; pericarp colorless. Seeds 1.2 x 1.0 cm.; endosperm colorless. Cob white.

Zea mays saccharata. The sweet corns.

19. Stowell's evergreen.

A large-eared extremely wrinkled-seeded late sugar corn. Ear 16 cm. long, 15.5 cm. in cir.; 14-rowed; pericarp colorless. Seeds 1.4 x .7 cm.; endosperm colorless. Cob white.

18. Early Crosby.

A twelve-rowed sugar corn. Ear 14.5 cm. long, 14 cm. in cir.; 12-rowed; pericarp colorless. Seeds .9 x .9 cm.; decidedly wrinkled but thick full seeds; endosperm colorless. Cob white.

54. Black Mexican.

An eight-rowed sugar corn characterized by purple aleurone cells. Ear 13.5 cm. long, 12 cm. in cir.; 8-rowed; pericarp colorless. Seeds .9 x 1.1 cm.; aleurone cells purple; endosperm colorless.

Methods Used.

In carrying out the large amount of tedious routine work necessary in the collection of data from the crosses of the above types, a great effort was made to reduce experimental errors to a minimum. No part of the work was left to farm workmen except the preparation of the breeding plots and their cultivation. The planting, labeling of families, crossing, selfing, harvesting, filing of seed and collection and reduction of data were done by the authors. The senior author alone is responsible for the details of the work until 1909. In 1909 and 1910 the senior and junior authors both shared in the labor. Since 1907 efficient aid in harvesting, filing seed, etc., has been given by Mr. C. D. Hubbell of the Conn. Agr. Exp. Station. In 1910 Mr. D. W. Davis and Mr. O. E. White, graduate students at Harvard University, aided in selfing ears of various selections.

The ears have always been shelled and seeds classified and filed in seed envelopes. Where there has been the least question about classification the work has been duplicated by two observers. If then there has been a doubt concerning the characters borne by particular seeds, those in question have been grown for another generation. The planting has been done from the seed envelopes directly to the field. There they were planted in hills three and one-half feet apart each way, four seeds to the hill. It was not considered necessary to start the seeds in the greenhouse in sterilized soil as is done with smaller seeds. Maize seed very seldom germinates after remaining in the ground over the winter in this climate. Furthermore the corn which was not hand pollinated was not husked directly on the field so that there was but little chance that any seeds should remain upon the ground. Great care was taken not to drop seeds at planting time in other than the hills marked out. These were covered carefully and after the young plants appeared above the surface, any individuals not exactly in the hill were removed. No plants have ever given evidence that they were misplaced and there is every reason to believe that the work is accurate in this regard.

The different families were marked in the field by heavy stakes to which wired tree labels were attached. As an additional precaution against mis-labeling or misplacement of labels, however, a planting plan was always kept on file showing the exact location of every plant in the field. With this safeguard every field stake might have been removed without making the least confusion.

All crossing and selfing were done by hand. Individual plants were used as the male parent in nearly every case. If possible the male parent of a cross was also selfed with its own pollen so that selfed seed of that individual was accessible if necessary. If for any reason it was particularly desirable to have the progeny of a plant where through an accident none of its own pollen was available, it was pollinated from a sister plant. This fact was always noted, however, and the male parent selfed if possible.

Heavy manila paper bags were used to protect both male and female inflorescences from foreign pollen. These were found much more desirable than paraffined bags as the latter were likely to become inverted and filled with water during a rain storm. The manila bags stood up well in the rain, dried out quickly, and seldom failed to furnish dry viable pollen. The tassels were bagged about three days before any pollen was ripe. Of course here there was a slight chance of enclosing foreign pollen. This pollen, however, would have been three or more days older than the pollen coming from the bagged flowers, and therefore much less viable. Even disregarding this fact, however, the immense amount of pollen furnished by the bagged infloresence would so dilute any foreign pollen that the ratio would be at least 10,000 to 1 in favor of the former.

The female flowers were always bagged of course before any of the silks were showing, and any bracts or leaves showing foreign pollen were carefully removed. Here again, however, is a slight chance of enclosing foreign pollen. This error has been determined by bagging 53 ears and allowing them to remain in the bags. Forty-four ears formed no seeds, six ears formed one seed each, two ears formed two seeds each and one ear formed four seeds. There are over five chances to one then that no viable foreign pollen enters in this way.

The pollination is accomplished by removing the bag from the tassel, shaking out the empty anthers and dusting the pollen over the silks of the proposed mother plant. The bag covering the silks is not entirely removed but is held so that its opening is horizontal with the silks resting inside. The pollen is then shaken in at the opening as quickly as possible, taking care not to let the silks touch the hands or clothing of the operator or the leaves or stem of the plant. It is sometimes impossible to keep from touching the silks with the fingers, as it may be necessary to rearrange them in the bag. To guard against contamination from this source the hands are carefully cleaned with 95 per cent. alcohol after each pollination.

The silks at the base of the ear mature first; those at the tip of the ear last. For this reason, if one is to be absolutely certain of a well filled ear, it is necessary to pollinate two or three times with fresh pollen. This procedure has the disadvantages of increasing the error, however, not to speak of the difficulty of obtaining pollen, so that in this work but one pollination was made in each case. When pollinated about five days after bagging, fairly well filled ears were generally obtained, particularly with the small races.

Immediately after pollination the ear is rebagged and tagged. From this time until the ears are mature they are inspected every little while to see that the bags are not too tight for the maturing seeds. The bags remain on until the ears are harvested. They are then picked, husked, tagged with wired tree labels and dried. Boards through which wire nails have been driven are hung from the ceiling of the drying room to prevent the depredations of mice. The ears are impaled upon these nails and thus dry surrounded by a current of air.

Experimental Error.

The manipulation during pollination is undoubtedly productive of an experimental error which even the most careful work cannot entirely prevent. This error was determined as follows. Twenty-five ears were bagged and allowed to remain in this condition for five days. The bags were then opened and given the manipulation that was necessary for hand-pollination, except that no pollen was applied. The ears were then rebagged and remained so until harvest time. No seed were formed on sixteen ears; three ears produced one seed each; four ears produced two seeds each; while one ear produced four seeds and one ear produced five seeds.

There is a possibility then of an experimental error of five or six seeds out of the two hundred to eight hundred produced per ear. This is to be considered as a maximum error and not the probable error, the latter being less than one seed per ear.

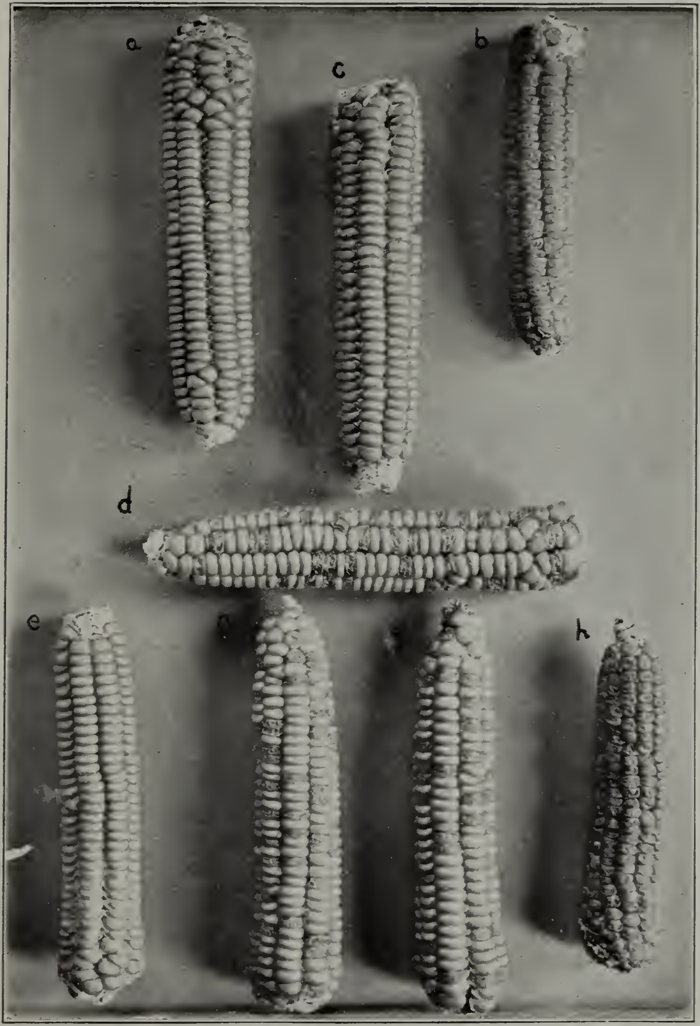
PART II.

ENDOSPERM CHARACTERS.

These hybridization studies are all reported under the headings of the different characters investigated, as this seems to be the method calculated to show the data with the least confusion. The female parent is written first, using the variety number given under the description of the material under investigation. For example 7 x 54 represents a cross of Leaming yellow dent female with Black Mexican sweet male. When this cross is grown and hand-pollinated selfed ears are obtained, they are numbered (7 x 54)-1, (7 x 54)-2, (7 x 54)-3, etc. Should ear number 2 be grown for still another generation, the crop obtained is numbered (7 x 54)-2-1, (7 x 54)-2-2, etc., thus the exact generation of a particular ear is always shown. The characters under consideration are known by letters; '*S*', for example means presence of starchy character and '*s*', absence of starchy character: '*P*', represents presence of purple aleurone cells; '*p*', its absence. An ear numbered (7 x 54)-2-1 P S represents an ear of the third or F_3 generation from which purple starchy seeds have been selected for planting.

Starchiness and Non-starchiness.

Starchiness is the condition of the endosperm of all of Sturtevant's maize varieties except *Zea mays saccharata*, regardless of the physical condition — corneous starch or soft starch — in which it appears. The starch grains are fully developed and possess a shape characteristic of the species *Zea mays*. The sugar maize does not have the ability to develop these starch grains to maturity. Some starch is formed but it remains small, angular and abortive, hence the seeds ripen from the stage of maturity called the "milk" without much change, giving the seed a wrinkled translucent appearance. The difference in size of the starch grains in the two races is shown in Table 10. This difference in the size of starch grains however,



a. No. 24 Rhode Island white cap (starchy parent); *b.* No. 53 Crosby non-starchy parent); *c.* result of cross 24x53 showing heterozygous seeds in which starchiness is completely dominant. *d,* an ear with F2 seeds showing mono-hybrid segregation. Lower row daughters of *d.* *E, f* and *g,* results from planting starchy seeds. One ear out of three is pure starchy. *h,* result from planting non-starchy seeds.

Segregation of Starchiness and Non-starchiness.

is not the whole difference between starchy and non-starchy races. As the starchy races ripen, starch formation goes on at a steady rate, while in the non-starchy races there is an actual breaking down of endosperm materials into cane sugar and various hexoses. This is shown by determinations we have made of reducing sugars in both starchy and non-starchy races when both were at the "milk" stage of maturity. The non-starchy races contained from one and one-fourth to two and one-half as much reducing sugar as the starchy races.

Correns (: 01) has already shown that starchiness behaves as a Mendelian dominant allelomorphic to its absence. Dominance was complete, and segregation generally * exact and inheritance discontinuous. It is not to confirm his work that the matter is taken up here, but to consider other questions to which the data are relevant. These questions relate chiefly to the mathematical hypothesis of Mendelism, to prepotency of individuals, and to gametic purity. The data from which the problems are discussed are not selected, but the figures

TABLE 1.

NO. 15 FLINT STARCHY X NO. 54 NON-STARCHY.

Ear No.	S	*s*	Total	Ratio per 4	Dev.	P. E.
(15 x 54)-1	135	48	183	2.9508 : 1.0492	0.0492	0.0864
(")-2	253	85	338	2.9944 : 1.0056	0.0056	0.0635
(")-3	150	42	192	3.1248 : 0.8752	0.1248	0.0843
(")-4	238	96	334	2.8504 : 1.1496	0.1496	0.0639
(")-6	190	72	262	2.9008 : 1.0992	0.0992	0.0722
(")-8	302	96	398	3.0352 : 0.9648	0.0352	0.0586
(")-11	242	105	347	2.7896 : 1.2104	0.2104	0.0627
(")-15	236	79	315	2.9968 : 1.0032	0.0032	0.0658
(15 x 54)-2-1	235	70	305	3.0820 : 0.9180	0.0820	0.0669
(")-2-2	242	79	321	3.0156 : 0.9844	0.0156	0.0652
(")-2-3	248	66	314	3.1592 : 0.8408	0.1592	0.0659
(")-2-4	227	68	295	3.0780 : 0.9220	0.0780	0.0680
(")-2-5	200	59	259	3.0888 : 0.9112	0.0888	0.0726
(")-2-6	182	74	256	2.8436 : 1.1564	0.1564	0.0730
(")-2-7	238	91	329	2.8936 : 1.1064	0.1064	0.0644
(")-2-8	195	58	253	3.0832 : 0.9168	0.0832	0.0734
(")-2-9	162	38	200	3.2400 : 0.7600	0.2400	0.0826
(")-2-10	131	53	184	2.8476 : 1.1524	0.1524	0.0861
(")-2-11	132	40	172	3.0696 : 0.9304	0.0696	0.0891
(")-2-12	101	32	133	3.0376 : 0.9624	0.0376	0.1013

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* The one exception was the pop and sugar cross mentioned later.

include only about one-fourth of the hand-pollinated ears at our disposal, belonging to the starchy and non-starchy cross. This number seemed sufficient for our purpose, and the segregating kernels were not counted on the remaining ears. It should be mentioned however, that any wide departures from the normal on any of the four hundred selfed heterozygous ears of this cross would have been noted and reported if such had occurred.

Dominance was found to be complete. In no case was there the slightest difference between the homozygous and the heterozygous seeds in either outward appearance or in the character of the starch cells when examined microscopically. Whatever it is that is brought in by the starchy parent to cause starch formation is sufficiently active to bring about complete change when present in one "dose" (that is from one parent). As in all endosperm characters, when S is the male parent the starchiness appears in the current generation so called, giving the most perfect illustration of Xenia there is known. As a matter of fact, one is not dealing with the current generation but with the F_1 generation, the endosperm being a younger generation than the plant which bears the ear. In no case, in an experience with several thousand seeds, did an F_1 seed showing Xenia fail to show a heterozygous condition; nor did extracted recessives (sugar seeds) of the F_2 generation ever show a heterozygous condition. From this, one may conclude that the second male nucleus that fertilizes the endosperm nucleus always bears the same characters as the first male nucleus that fertilizes the embryo nucleus or egg. Several heterozygous seeds have been found, however, that were not completely starchy, but had developed bilaterally into half starchy and half non-starchy. There was not a gradual change from the one condition to the other, but a distinct line of demarkation, with one side as absolutely distinct from the other as are the pure races of each kind. None of these seeds were homozygous starchy, and Correns' interpretation of similar phenomena as cases in which the second male nucleus did not fuse with the endosperm nucleus but each developed separately, seems well founded. Attention is called to the matter for this reason. It is an hypothesis generally received with quiescence if not with acquiescence, that starchiness (and other "presence" characters) is due to presence of an enzyme not possessed by the allelomorph. Now if this is true, the enzyme must be a colloid with such large molecules that there is absolutely no dialysis, otherwise it seems as if it would diffuse through the unripe seed sufficiently to act as a catalyser throughout the entire endosperm. No matter what is the correct interpretation, there is certainly a definite chain of hereditary transmission of characters from cell to cell during development, and each original cell follows an inertia of its own with little influence on others.

TABLE 2.

NO. 24, FLINT STARCHY X NO. 54, NON-STARCHY.

Ear No.	S	*s*	Total	Ratio per 4	Dev.	P. E.
(24 x 54)–1	274	94	368	2.9784 : 1.0216	0.0216	0.0609
(")–2	219	73	292	3.0000 : 1.0000	0.0000	0.0684
(")–6	256	89	345	2.9680 : 1.0320	0.0320	0.0629
(")–8	200	64	264	3.0304 : 0.9696	0.0304	0.0719
(")–9	155	69	224	2.7680 : 1.2320	0.2320	0.0781
(")–10	212	59	271	3.1292 : 0.8708	0.1292	0.0710
(")–11	213	77	290	2.9380 : 1.0620	0.0620	0.0686
(")–12	268	80	348	3.0804 : 0.9196	0.0804	0.0626
(")–13	264	106	370	2.8540 : 1.1460	0.1460	0.0607
(")–14	227	90	317	2.8644 : 1.1356	0.1356	0.0656
(24 x 54)–1–2	207	68	275	3.0108 : 0.9892	0.0108	0.0704
(")–1–6	223	75	298	2.9932 : 1.0068	0.0068	0.0677
(")–1–8	235	90	325	2.8924 : 1.1076	0.1076	0.0648
(")–1–9	106	36	142	2.9860 : 1.0140	0.0140	0.0980

We have said that dominance appears to be complete; segregation also appears to be complete. It is seldom necessary to subject extracted recessives to proof by growing them a further generation. Some strains of non-starchy maize, No. 18 for example, are much less wrinkled than others; and when such a strain is crossed with a flint type there is less difference between dominants and recessives in appearance than when certain other types are crossed. But in no case is there the least difficulty in separating the segregates correctly. Whether this apparent segregation is as complete as it appears we shall discuss presently. It should further be mentioned that the seeds can also be classified with absolute exactness by microscopical examination.

Tables 1-9 contain the proportion of the starchy and non-starchy seeds obtained as progeny when heterozygous seeds were planted; although, as was stated before, only a few ears from each family were counted. One object in view is to show the behavior of starchy and non-starchy in several races.

TABLE 3.

NO. 5, FLINT STARCHY X NO. 18, NON-STARCHY.

Ear No.	S	*s*	Total	Ratio per 4	Dev.	P. E.
(5 x 18)-4	181	68	249	2.9176 : 1.0924	0.0924	0.0740
(“)-8	172	66	238	2.9908 : 1.1092	0.1092	0.0757
(“)-10	215	68	283	3.0388 : 0.9612	0.0388	0.0694
(“)-16	225	69	294	3.0612 : 0.9388	0.0612	0.0681
(“)-18	186	61	247	3.0120 : 0.9880	0.0120	0.0743
(“)-21	136	42	178	3.0560 : 0.9440	0.0560	0.0876
(“)-25	176	50	226	3.1152 : 0.8848	0.1152	0.0777
(“)-30	218	68	286	3.0488 : 0.9512	0.0488	0.0691

There is reason to believe that different races can be identical in appearance, but may have such different gametic composition that they may affect a character possessed by a race with which they may be crossed, in very different manners. (See purple aleurone cells and non-purple.) Examination of the tables shows this not to be the case with starchy and non-starchy. All of the starchy and non-starchy races with which crosses have been made behave in exactly the same manner. There is no difference in appearance in heterozygotes from different races that is not accounted for by the different shaped seeds possessed by the parents, and wide variations in shape occur only in generations later than F_1. In the F_1 generation the shape is intermediate between that of the two parents.

TABLE 4.

NO. 11, FLINT STARCHY X NO. 18, NON-STARCHY.

Ear No.	S	*s*	Total	Ratio per 4	Dev.	P. E.
(11 x 18)-7	220	74	294	2.9932 : 1.0068	0.0068	0.0681
(“)-14	218	81	299	2.9164 : 1.0836	0.0836	0.0676
(“)-15	200	77	277	2.8880 : 1.1120	0.1120	0.0702
(“)-22	235	87	322	2.9192 : 1.0808	0.0808	0.0651

If one examines the tables carefully however, he sees at once that there is quite a difference in the ratios obtained. They vary from a ratio of 2.7896 : 1.2104 in ear (15 x 54)-11, Table 1 to a ratio of 3.2020 : 0.7980 in ear (19 x 7)-2, Table 7. This brings up an important question. Does this discrepancy represent an expected probable error in chance matings; or, is there a prepotency in certain families through which excessive numbers of dominants or of recessives tend constantly to reappear? Correns did indeed find in one family such an excess of starchy seeds, but it is not known whether this apparent prepotency was transmitted in further generations.

Tables 1-9 show several cases where ears with a ratio deviating from the expected 3 : 1 of Mendelian hypothesis have been grown for another generation. For example, ear (8 x 54)-1 of Table 6 has a ratio of 2.9420 : 1.0580 while ear (8 x 54)-5 of the same table has a ratio of 3.0780 : 0.9220; yet the progeny of these ears average just about the 3 : 1 ratio of theory. There are even more ears with an excess of recessives from the ear that had the excess of dominants and *vice versa*. Other deviants have been grown for several generations, and while the exact ratios have not been recorded it may be stated with confidence that wide deviations occurring in considerable numbers would have been noticed while making other records. It may be concluded then that no prepotency or tendency to aberrant ratios is a constant characteristic of any of our families. How then can the discrepancies from theoretical ratios be explained?

To study this question the probable errors of all of the ratios have been calculated. The method used has been that of Johannsen (: 09, p. 405), except that the mean error has been reduced to the probable error by multiplying by the factor 0.6745. The standard deviation of a Mendelian proportion is $\pm\frac{\sqrt{p \times q}}{p+q}$ where p and q are the Mendelian terms, in this case 3 and 1. Then S. D. $= \pm\frac{\sqrt{3 \times 1}}{4} = \pm\frac{1.7321}{4} = \pm 0.4330$. The probable error, $E = \pm 0.6745 \frac{\text{S. D.}}{\sqrt{n}}$ where n is the total number of variates.

To find out whether the different ratios given in Tables 1-9 are what should reasonably be expected if the Mendelian theory of chance matings of equal numbers of gametes *S* and *s* in both male and female germ-cells is correct, it should be understood just what is meant by probable error in the law of error. Plus errors and minus errors should occur with equal frequency, small errors should occur more frequently than large errors, and very large errors should not occur. Determined as above the probable error means that the chances are:

1 to 1 that the true value lies within ± E
4.5 to 1 that the true value lies within ± 2E
21 to 1 that the true value lies within ± 3E
142 to 1 that the true value lies within ± 4E

The theory of error also provides for errors of any size in their proper frequency or rather infrequency, but as a matter of fact in practice if errors greater than ± 4E occur they are probably due to experimental errors or avoidable mistakes.

We may consider each ear given in Tables 1-9 as a determination and its probable error as the probable error of a single determination. With this in mind we find that in the 94 ears tabled there are 49 plus errors and 45 minus errors. Further we find that the theoretical mode or 0 error is almost 3 : 1 being in fact very slightly greater. The errors are distributed as follows:

Within ± E 47.8% — Theory 50.0%
Within ± 2 E 83.0% — Theory 82.3%
Within ± 3 E 96.8% — Theory 95.7%
Within ± 4 E 100.0% — Theory 99.3%

The sum total of these segregates is 23529 to 7811, a ratio of 3.0031 : 0.9969 ± .0066.

It should be mentioned simply in order to suppress no data that one ear was found with a probable error somewhat in excess of ± 4 E. It was found by growing this ear for another generation however that this was due to an experimental error. There was a great excess of sugar seeds, but in the starchy seeds there proved to be about 4 heterozygotes to 1 homozygote. Since no other ear like this has ever been obtained, and since it is known that during the progress of this experiment several ears were first selfed with pollen killed by rain and afterward pollinated with pollen from a sugar plant to get material for another purpose, it seems highly probable that this ear was of a similar mixed parentage and that its explanatory label had been lost.

As to whether these data support the Mendelian hypothesis or not there may be slight grounds for a difference in opinion. Our own opinion is that when we take into consideration the chance for experimental error, the ratios are well within the limits of probable error. One thing at least is brought out clearly, the behavior of segregates in more than one generation and a variety of matings are necessary, if one is to draw conclusions as to the exact mode of inheritance of character pairs from small numbers.

One further point remains for discussion. Do the extracted homozygotes breed true? In other words, is segregation an absolute separation of a gene from its absence? or, is there only a relative segregation? Morgan (: 10) has suggested that relative segregation may explain Mendelian facts, if one presupposes that when the amount of the gene falls below a certain limit the dominant fails to develop. This idea while interpreting the facts in the F_2 generation is inadequate to explain the apparent purity of further generations of extracted recessives, for if this hypothesis were true many recessives would show the dominant character when crossed.

In Table 9 is shown the segregation of extracted dominant starchy seeds. The ratio is as nearly the expected 2 heterozygotes to 1 homozygote as could well be expected. Several

TABLE 5.

NO. 17, FLINT STARCHY X NO. 54, NON-STARCHY; NO. 18, NON-STARCHY X NO. 58, FLINT STARCHY; AND NO. 7, DENT STARCHY X NO. 54, NON-STARCHY.

Ear No.	S	*s*	Total	Ratio per 4	Dev.	P. E.
(17 x 54)–1	328	102	430	3.0512 : 0.9488	0.0512	0.0563
(18 x 58)–1	332	102	434	3.0600 : 0.9400	0.0600	0.0561
(7 x 54)–1	379	137	516	2.9380 : 1.0620	0.0620	0.0514
(")–2	493	131	624	3.1604 : 0.8396	0.1604	0.0467

thousand dominant homozygotes have been bred for further generations and these have all bred true to the starchy character. This is, in general, the case with the extracted non-starchy seeds. Furthermore, there are in commercial use many sugar corns that are extracted recessives. Golden Bantam, Late Egyptian and many others are examples of races that have originated from crosses with starchy varieties. The wrinkled seeds have been selected and have bred true. Out of the many million seeds that are annually grown for the canning factories, however, there does appear an occasional ear with semi-starchy seeds. These ears transmit the character and give the canners no end of trouble. There is no way to find out whether these ears appear only on varieties which somewhere in their ancestry had a starchy parent. One can only say that they do appear in ratios not exceeding one ear in ten thousand. By some lucky chance some of these ears made their appearance in our controlled cultures. All of our extracted recessives have proved true to non-starchiness (*) except from the progeny of ear (8 x 54)-1-6. The majority of the progeny of this ear were also non-starchy, but three ears appeared which were decidedly semi-starchy, one of which is shown in Plate III, fig. b. There was no possibility that these ears could have grown from a normal heterozygous seed. They were not plump seeds like a true heterozygote nor did they segregate into starchy and non-starchy in the next generation. The entire ear was rather uniformly semi-starchy and quite different from the true starchy ears. Microscopical examination showed definitely that starch grains had been developed normally to a size intermediate between the true starchy and the true sweet seeds of the same family. This fact is shown in Table 10.

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* There are other cases where some apparent starchiness is always developed, namely when pop races are crossed with non-starchy races. We interpret this as being due to the small size of the resulting F_2 seeds borne on intermediate F_1 ears. When the seeds are small the endosperm material more nearly fills the pericarp than when they are large. The wrinkled condition is therefore less apparent. If one has had considerable experience in classifying starchy and non-starchy seeds, such crosses are seen to show normal segregation. If, however, careful classification is not made and the seeds are not tested in further generations pop and non-starchy crosses always appear to show an excess of starchy seeds. It is suggested that this is the explanation of Corren's failure to obtain normal ratios in a similar cross. These cases are not real exceptions to the statement made above, however, for recessives extracted from pop crosses are never grown commercially as sugar corns.

a. P2, F1, and F2 seeds from cross between No. 19 Stowell's Evergreen sugar and No. 2 Illinois low protein dent maize.

b. Middle ear is a semi-starchy ear No. (8x54)-1-6, progeny of an extracted recessive (wrinkled) seed. On the left is an extracted dominant (starchy) ear of the same cross. On the right is a well wrinkled ear, sister of No. (8x54)-1-6.

Segregation of Starchiness and Non-Starchiness.

a Random sample of progeny of starchiest seeds of semi-starchy ear shown in PLATE III.

b. Random sample of progeny of most wrinkled seeds of semi-starchy ear shown in PLATE III.

GAMETIC PURITY.

TABLE 6.

NO. 8, DENT STARCHY X NO. 54, NON-STARCHY.

Ear No.	S	*s*	Total	Ratio per 4	Dev.	P. E.
(8 x 54)-1	381	137	518	2.9420 : 1.0580	0.0580	0.0513
(")-2	340	137	477	2.8512 : 1.1488	0.1488	0.0535
(")-3	400	141	541	2.9576 : 1.0424	0.0424	0.0502
(")-5	384	115	499	3.0780 : 0.9220	0.0780	0.0523
(8 x 54)-1-3PC	243	64	307	3.1660 : 0.8340	0.1660	0.0667
(")-1-4PC	302	105	407	2.9680 : 1.0320	0.0320	0.0579
(")-1-6PC	231	81	312	2.9616 : 1.0384	0.0384	0.0672
(")-1-8PC	321	117	438	2.9316 : 1.0684	0.0684	0.0558
(")-1-14PC	238	66	304	3.1316 : 0.8684	0.1316	0.0670
(")-1-6P	145	40	185	3.1356 : 0.8644	0.1356	0.0859
(")-1-11P	268	78	346	3.0984 : 0.9016	0.0984	0.0628
(")-1-13P	320	107	427	2.9976 : 1.0024	0.0024	0.0565
(")-1-15P	293	96	389	3.0128 : 0.9872	0.0128	0.0592
(")-1-1	237	88	325	2.9168 : 1.0832	0.0832	0.0648
(")-1-2	236	88	324	2.9136 : 1.0864	0.0864	0.0649
(")-1-4	176	60	236	2.9832 : 1.0168	0.0168	0.0760
(")-1-10	242	80	322	3.0064 : 0.9936	0.0064	0.0651
(")-1-29	272	93	365	2.9808 : 1.0192	0.0192	0.0611
(8 x 54)-5-2	236	90	326	2.8956 : 1.1044	0.1044	0.0647
(")-5-3	294	99	393	2.9924 : 1.0076	0.0076	0.0589
(")-5-4	147	61	208	2.8268 : 1.1732	0.1732	0.0810
(")-5-5	277	72	349	3.1748 : 0.8252	0.1748	0.0625
(")-5-6	357	123	480	2.9748 : 1.0252	0.0252	0.0533
(")-5-8	324	109	433	2.9932 : 1.0068	0.0068	0.0651
(")-5-10	306	85	391	3.1304 : 0.8696	0.1304	0.0591
(")-5-11	249	86	355	2.9732 : 1.0268	0.0268	0.0638

Seeds from the ear shown in Plate III, fig. b, were divided into two classes, those most nearly starchy and those most nearly non-starchy, and planted. A number of selfed ears were obtained from each class. Those resulting from the seeds most nearly non-starchy were in part what would immediately be classified as non-starchy and in part as starchy in appearance as the parent ear. The ears resulting from the seeds most nearly starchy were all as starchy as the parents and certain of them even more so. This fact is shown in Plate IV. Microscopical examination of the most starchy seeds of this generation showed that the starch grains were most of them developed to normal size. The ears were not uniform nor was there uniform starchiness among the seeds of a single ear. Seeds could be selected which formed a series running from true sweet to true starchy, yet those most nearly starchy had a rough appear-

TABLE 7.

NO. 19, NON-STARCHY X NO. 7, DENT STARCHY AND NO. 19, NON-STARCHY X NO. 8, DENT STARCHY.

Ear No.	S	s	Total	Ratio per 4	Dev.	P. E.
(19 x 7)–2	297	74	371	3.2020 : 0.7980	0.2020	0.0607
(")–5	486	156	642	3.0280 : 0.9720	0.0280	0.0461
(19 x 7)–5–1	304	109	413	2.9444 : 1.0556	0.0556	0.0575
(19 x 8)–1	183	64	247	2.9636 : 1.0364	0.0364	0.0743
(")–2	464	152	616	3.0128 : 0.9872	0.0128	0.0471
(")–3	449	151	600	2.9932 : 1.0068	0.0068	0.0477
(")–4	303	96	399	3.0376 : 0.9624	0.0376	0.0585
(")–5	414	139	553	2.9948 : 1.0052	0.0052	0.0507

ance very different from the well-filled pericarp of the true starchy seeds of the same family. These seeds will be selected for starchiness and if uniform ears are finally obtained, will be crossed with non-starchy again to see if their behavior is the same as normal starchy maize. Provisionally one is forced to one of two conclusions. *Either homozygous recessives (and likewise dominants) are not complete segregates, but products of a partial quantitative separation of genes allowing traces of the dominant character to remain, traces which may sometimes accumulate sufficiently to bring out the dominant character: or, progressive variations are constantly taking place in small numbers, most often along paths that have been passed before.*

TABLE 8.

NO. 60, POP STARCHY X NO. 54, NON-STARCHY.

Ear No.	S	s	Total	Ratio per 4	Dev.	P. E.
(60–5 x 54)–2	274	82	356	3.0788 : 0.9212	0.0788	0.0619
(")–6	273	102	375	2.9120 : 1.0880	0.0880	0.0603
(")–8	163	53	216	3.0184 : 0.9816	0.0184	0.0795
(")–11	191	58	249	3.0684 : 0.9316	0.0684	0.0740
(")–12	249	84	333	2.9908 : 1.0092	1.0092	0.0640
(60–3 x 54)–1	296	87	383	3.0912 : 0.9088	0.0912	0.0597
(")–5	260	107	367	2.8336 : 1.1664	0.1664	0.0610
(")–6	243	73	316	3.0760 : 0.9240	0.0760	0.0657
(60–8 x 54)–1	227	67	294	3.0884 : 0.9116	0.0884	0.0681
(")–8	224	71	295	3.0372 : 0.9628	0.0372	0.0680

It is our opinion that dominant starchiness — if it is the same dominant starchiness — has been formed anew. It occurs too rarely to support a partial segregation theory such as Morgan's (: 10). If it is asked why starchiness is the character that arises anew rather than another variation, it is suggested that the peculiar chemical structure of the germ cell of maize may be such that a molecular readjustment is much more likely to bring about starchiness than any other variation. Such a path of least resistance for variations might account for the many cases in animals and plants where the same variation has apparently occurred again and again.

Conclusion.

These starchy and non-starchy crosses represent a much larger number of individuals than have ever before been studied in accurately controlled pedigree cultures. Taking them as a whole they show that the mechanism by which the members of an allelomorphic pair are distributed among the gametes, is accurate. The aberrant ratios sometimes obtained are what should be expected by the Law of Error. They are not inherited, and we believe this to show that there is no such thing as prepotency *per se* which would cause abnormal ratios. We might extend this conclusion further and say that there is no conclusive evidence of a failure of segregation of male gametes or of selective fertilization (Lock : 06), or of partial gametic coupling that presupposes gametes bearing opposite genes to be formed in unequal numbers (Bateson and Punnett : 08). Disbelief in prepotency of the kind described above does not indicate disbelief in different "potencies" as described by Davenport (: 10). Different potencies, that is various degrees of manifestation of the same character due to its modification during development by the action of *other developing* genes possessed by the individual, is a different thing and is entirely logical. In prepotency or potency of this kind segregations are perfectly normal, and modifications which occur in characters are due to the gametic constitution of the individual.

The aberrant ratios obtained by Correns in the pop-sugar cross referred to above, may have been due to modification by other unknown characters possessed by the parents, but it seems more likely that they were due to improper classification of dominants and recessives for the reason that recessives in such crosses although hyaline and easily classified microscopically often do fill the pericarp with endosperm material owing to the small size of the seed.

If then, in cases of simple mono-hybrids where there are no complications, a ratio of 3.0031 : 0.9969 ± .0066 is obtained; are we not compelled to take the view that segregation occurs at the reduction division? Could any less exact division give the distribution of genes necessary for such exact recombinations? Of course it has long been suspected that this was the time of segregation, but Bateson (: 09 p. 271) has felt that obstacles were in the way of interpreting the chromosomes as such important bearers of hereditary qualities. These obstacles were three in number; first, it is objected that no correspondence has been shown between visible differences of type (except sex) and chromosome differences; second, that no correspondence between complexity of type and chromosome numbers has been shown; and third, that bud sports are somatic segregates. There are, it seems to us, no real obstacles here. One should expect that the quality of the chromosome and not shape or number, is the important fact. It is even likely that most of the important morphological characters are carried by all of the chromosomes, hence a doubling of chromosome number as has occurred in *Oenothera gigas* may be relatively unimportant. The case of bud sports is also fairly clear since Winkler (: 09) has shown that a graft hybrid between the black night shade and the tomato proved to have the sum of the haploid numbers of the two parents and not the sum of the diploid numbers. The somatic cell then has a regulatory apparatus of its own. What might be called the normal bud sport (other sports probably occur from abnormal cell divisions) is probably due to the fusion of two somatic cells of a heterozygote, followed by a reduction, in which one of the homozygote forms appears. It must be not understood however that because Bateson's objections are considered surmountable, we therefore believe it to be *proved* that the chromosomes are the sole bearers of hereditary characters and that the *reduction division* is the time of Mendelian segregation. Judgment must still be suspended on these matters.

TABLE 9.

EARS FROM F_2 GENERATION PLANTS OF STARCHY AND NON-STARCHY CROSSES.

Starchy Seeds Planted.

Selection	Heterozygous S	Homozygous S
(8 x 54)–1	6	4
(")–2	4	5
(")–3	30	17
(")–5	75	31
(")–1–1	67	32
(")–1–2	44	25
(")–4	71	38
(")–10	48	28
(15 x 54)–2	46	13
(")–3	25	17
(24 x 54)–1	28	14
Total	444	224
Ratio	1.93	1

TABLE 10.

RANDOM COMPARISON OF DIAMETER OF STARCH GRAINS.

Extracted Starchy Seeds from (8 x 54)–1 and Semi-Starchy and Non-Starchy from (8 x 54)–1–6.

Diam. in mm.	.009	.017	.034	.052	.069	.086	.103	.12	.138	.155	Total
No. variates from starchy seeds	..	1	9	23	34	66	36	16	12	3	198
No. variates from semi-starchy seeds	..	17	52	57	48	17	11	5	..	..	227
No. variates from non-starchy seeds	34	94	52	13	..	..	..	..	..	..	193

Yellow and Non-yellow Endosperm.

Correns (: 01) and Lock (: 06) each found a yellow color in the endosperm which behaved with its absence as a single allelomorphic pair. We have found *two* * yellow colors in the endosperm each behaving when crossed with its absence, as an independent allelomorphic pair. A part of the experiments with these characters has been described in a previous paper (East :10). In this paper some further data are presented.

Both of these yellow colors, although they behave in inheritance as separate entities are either identical or very similar in composition. They are insoluble in water, somewhat soluble in alcohol and easily soluble in ether, chloroform, benzine, benzol and carbon bisulphide. They occur in rhombic plates in the starch cells and possibly also in the chromoplasts although this is not certain. From these facts it might be supposed that they were hydrocarbons with compositions similar to carotin. They do not give the general reactions however which the fatty pigments or lipochromes — of which carotin is an example — give with sulphuric acid or iodine dissolved in aqueous potassium iodide. Independent of their solubility reactions, this would class them with the anthochlorins (Courchet '88).

Considering the importance to Mendelian theory of the discovery that two similar and possibly identical characters may each act with its own absence as an independent allelomorphic pair, further chemical investigations are being made which will be reported in a separate paper. It may simply be stated here that as far as is known these colors are indistinguishable, but as they behave differently in crosses they will be known as Y_1 and Y_2.

A number of crosses were made between yellow and non-yellow which gave only 3 : 1 ratios. The remaining crosses shown in Tables 11-16 each showed one or more ears with dihybrid ratios.

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* Lock mentioned that light yellow seeds appeared in his crosses, but he classed them as whites which vitiates his study of Mendelian numerical proportions.

TABLE 11.

F_2 SEEDS FROM CROSS OF NO. 1 WHITE DENT X NO. 7 YELLOW DENT.

Ear No.	Y	*y*	Ratio Approx.
(1 x 7)-1	587	212	3 : 1
(1 x 7)-2	127	30	3 : 1

Table 11 gives the results from two selfed ears of No. 1 white dent crossed with No. 7 yellow dent. They approximate 3 to 1 ratios although ear No. 1 has an excess of non-yellow and ear No. 2 an excess of yellow seeds. This cross proved to be too late for the Connecticut climate and the resulting F_3 seeds were immature and difficult to classify. Yellow was dominant and appeared as Xenia in the F_1 seeds but the F_2 seeds varied in different ears in a peculiar manner. Where there was sufficient soft starchy matter in the caps of the seeds the heterozygotes were considerably lighter colored at the cap than when the seeds possessed more corneous starch. The same phenomenon occurred in reciprocal crosses; so that when there was sufficient soft starchy matter the heterozygotes could be distinguished from the homozygotes either way the cross was made. (See cross of floury yellow with non-yellow.)

Ear (1 x 7)-1Y gave only one selfed ear with 126 yellows of various shades, 14 white, and 3 doubtful seeds. The mother seed was probably Y_1 Y_2 y_1 y_2. Several open field ears from yellows with white caps all proved to be heterozygous, thus proving the above statement regarding Xenia. The crop from the white seeds proved pure for non-yellow.

TABLE 11A.

F_3 SEEDS OF EAR NO. 2 OF CROSS SHOWN IN TABLE 11.

Yellow Seeds Planted.

Ear No.	Y	*y*	Ratio Approx.
(1 x 7)-2-3	423	108	3 : 1
(“)-2-7	351	108	3 : 1
(“)-2-8	375	120	3 : 1
(“)-2-12	343	111	3 : 1
(“)-2-14	577	. . .	Pure yellow
(“)-2-17	286	58	3 : 1
(“)-2-18	209	87	3 : 1
(“)-2-19	360	135	3 : 1
(“)-2-20	341	105	3 : 1
(“)-2-22	319	92	3 : 1
(“)-2-23	408	168	3 : 1
(“)-2-25	633	40	15 : 1

Table 11a shows the results from planting (1 x 7)-2 Y seeds. Ears Nos. 3 and 17 have an excess of yellow seeds. Possibly they were 15 : 1 ratios in which the yellows were very light and could only have been classified with certainty by growing the supposed whites another generation. The remaining ears all showed 3 : 1 ratios except ear No. 25. This ear was clearly a 15 : 1 ratio. The crop from (1 x 7)-2y (extracted whites) gave 12 pure white ears, showing that the classification of the F_2 seeds was correct.

A cross between No. 5 white flint and No. 6 yellow dent (Tables 12 and 12a) showed in all cases complete dominance of yellow. In the F_1 seeds which were of course flinty like the mother, there was no soft starch in the cap and the heterozygotes were exactly like pure yellow flint seeds. In the F_1 plants the F_2 homogygous and heterozygous yellow seeds were also indistinguishable. It was necessary to grow them to distinguish heterozygous yellow from homozygous yellow. In the F_2 plants with F_3 seeds, however, there was a considerable segregation of dented ears from flint ears. Here as in the cross of (1 x 7) it was fairly easy to distinguish heterozygous yellows from homozygous yellows when the seeds of the former had a well developed soft starchy zone in the cap.

Although as has been stated the F_1 seeds were all exactly like pure yellow flint seeds, they nevertheless belonged to two classes. The Xenia seeds (F_1 seeds) of the hybrid ear contained 159 seeds which were dark yellow and 145 seeds which were a considerably lighter yellow. This striking phenomenon was not understood until another generation was grown from the seeds. Table 12 showing the selfed ears resulting from the dark seeds, and Table 12a showing the selfed ears resulting from the light seeds make this matter plain. Excluding ear (5 x 6)-9 from Table 12 because it evidently came from a pure yellow seed grown in this family through an error, and ear (5 x 6)-11a from Table 12a which evidently grew from a self-pollinated seed of the mother No. 5, it is clear that the No. 6 plant furnishing the pollen for the cross was homozygous for one yellow and heterozygous for the other. The classification into light and dark yellows was somewhat arbitrary and therefore some ears in Table 12 gave ratios of 3 : 1 and some ears in Table 12a gave ratios of 15 : 1 but the fact that about one-half of the F_1 seeds had a gametic formula of $Y_1\ y_1\ Y_2\ y_2$ and one-half the formula $Y_1\ y_1$, or $Y_2\ y_2$, is certain. Ear (5 x 6)-7a is the only ratio in doubt. It is probably 15 : 1 as the yellows were very light and difficult to classify, and some were probably placed with the non-yellows.

TABLE 12.

F_2 SEEDS FROM CROSS OF NO. 5 WHITE FLINT X NO. 6 YELLOW DENT.

Dark Yellow Seeds Planted.

Ear No.	Y	*y*	Ratio Approx.
(5 x 6)–1	326	29	15 : 1
(")–2	316	27	15 : 1
(")–3	313	28	15 : 1
(")–7	354	122	3 : 1
(")–8	331	109	3 : 1
(")–9	307	. . .	Pure yellow
(")–10	475	25	15 : 1
(")–11	298	113	3 : 1
(")–12	368	19	15 : 1
(")–13	363	35	15 : 1
(")–14	489	29	15 : 1
(")–15	427	118	3 : 1

TABLE 12A.

F_2 SEEDS FROM SAME CROSS AS SHOWN IN TABLE 12.

Light Yellow Seeds Planted.

Ear No.	Y	*y*	Ratio Approx.
(5 x 6)–1a	359	117	3 : 1
(")–2a	144	54	3 : 1
(")–3a	173	63	3 : 1
(")–4a	433	136	3 : 1
(")–5a	557	35	15 : 1
(")–6a	316	120	3 : 1
(")–7a	450	49	10 : 1
(")–8a	229	86	3 : 1
(")–9a	325	115	3 : 1
(")–10a	227	87	3 : 1
(")–11a	. . .	434	Pure white
(")–12a	318	118	3 : 1
(")–13a	256	93	3 : 1

Tables 13, 13a, b, c, d, show results from an opposite cross. No. 11, yellow flint was the female parent and No. 8, white dent was the male parent. There was no effect of Xenia, as the F_1 hybrid seeds were as yellow as the pure No. 11. Table 13 shows the results from the F_1 hybrid seeds. Every ear approximates a 3 : 1 ratio except ears (11 x 8)-7 and (11 x 8)-8. Ear (11 x 8)-7 is shown afterwards by Tables 13b and c to have been in reality a 15 : 1 ratio. In other words it was a $Y_1 y_1 Y_2 y_2$

TABLE 13.

F_2 SEEDS FROM CROSS OF NO. 11, YELLOW FLINT X NO. 8, WHITE DENT

Yellow Seeds Planted.

Ear No.	Y	*y*	Ratio Approx.
(11 x 8)–1	358	154	3 : 1
(")–2	124	41	3 : 1
(")–3	389	127	3 : 1
(")–4	340	96	3 : 1
(")–6	252	83	3 : 1
(")–7	454	145*	3 : 1
(")–8	204	70**	3 : 1

---

* ** Proved to be a mixture of Y y and y, with preponderance of Y y.

ear. Ear (11 x 8)-8 probably was also of the same character as about half of the seeds classed as white proved to be heterozygous. Table 13d shows only two ears out of eight to have been other than white but an inspection of the open field crop showed such a large proportion of apparently heterozygous ears, that this ratio is probably not the real one.

Ear (11 x 8)-2 proved to be $Y_1 y_1$ or $Y_2 y_2$ as is shown in Table 13a. There is a ratio of about 2 heterozygous to 1 homozygous ears.

TABLE 13A.

F_3 SEEDS OF EAR NO. 2 OF CROSS SHOWN IN TABLE 13.

Yellow Seeds Planted.

Ear No.	Y	*y*	Ratio Approx.
(11 x 8)–2–1	275	. .	Pure yellow
(")–2–3	237	75	3 : 1
(")–2–5	244	71	3 : 1
(")–2–7	374	. .	Pure yellow
(")–2–8	344	113	3 : 1
(")–2–9	280	. .	Pure yellow
(")–2–10	99	31	3 : 1
(")–2–11	173	38	3 : 1
(")–2–15	274	75	3 : 1

Ear (11 x 8)-7 was evidently wrongly classified as is shown in Tables 13b and 13c. Ear (11 x 8)-7-1 is probably a 15 : 1 ratio. If this is true then there were 2 ears with gametic formula $Y_1 y_1 Y_2 y_2$, 2 ears with gametic formulæ $Y_1 y_1$ or $Y_2 y_2$, 1 ear with formula $Y_1 Y_1 Y_2 Y_2$ [Ear (11 x 8)-7-9], and 3 ears with formulæ $y_1 y_2$. The apparently white seeds from this ear were not all non-yellow, but partly pure and partly heterozygous light yellows. That is, they were $Y_1 Y_1$ or $Y_2 Y_2$ or $Y_1 y_1$ or $Y_2 y_2$.

TABLE 13B.

F_3 SEEDS OF EAR NO. 7 OF CROSS SHOWN IN TABLE 13.

Yellow Seeds Planted.

Ear No.	Y	*y*	Ratio Approx.
(11 x 8)–7–1	207	25	8 : 1
(")–7–4	253	68	3 : 1
(")–7–6	193	73	3 : 1
(")–7–8	163	79	3 : 1
(")–7–9	456	. .	Pure yellow
(")–7–11	108	35	3 : 1
(")–7–14	88	5	15 : 1

TABLE 13C.

F_3 SEEDS OF EAR NO. 7 OF CROSS SHOWN IN TABLE 13.

Apparently White Seeds Planted.

Ear No.	Y	*y*	Ratio Approx.
(11 x 8)–7–1a	. . .	504	Pure non-yellow
(")–7–2a	271	. . .	Pure light yellow
(")–7–3a	. . .	330	Pure non-yellow
(")–7–4a	323	117	3 : 1
(")–7–5a	. . .	300	Pure non-yellow

TABLE 13D.

F_3 SEEDS OF EAR NO. 8 OF CROSS SHOWN IN TABLE 13.

White Seeds Planted.

Ear No.	Y	*y*	Ratio Approx.
(11 x 8)–8–1	406	194	3 : 1
(")–8–3	. . .	394	Pure non-yellow
(")–8–6	. . .	560	"
(")–8–7	. . .	348	"
(")–8–9	. . .	490	"
(")–8–11	. . .	360	"
(")–8–12	. . .	360	"
(")–8–13	442	. . .	Pure yellow

TABLE 14.

F_2 SEEDS FROM CROSS OF NO. 11 STURGES' YELLOW FLINT X NO. 24 SANFORD'S WHITE FLINT.

Yellow Seeds Planted.

Ear No.	Y	y	Ratio Approx.
(11 x 24)–3	467	164	3 : 1
(")–4	320	137	3 : 1
(")–5	499	142	3 : 1
(")–6	356	116	3 : 1

Table 14 shows the results from selfing the F_1 seeds of a cross between No. 11, yellow flint and No. 24 white flint. There was no effect of Xenia. The ears gave 3 : 1 ratios and the extracted non-yellows proved to be pure in the F_3 generation.

TABLE 15.

F_2 SEEDS FROM CROSS OF NO. 15 LONGFELLOW FLINT X NO. 8 WHITE DENT.

Yellow Seeds Planted.

Ear No.	Y	y	Ratio Approx.
(15 x 8)–1	305	73	3 : 1
(")–2	166	12	15 : 1
(")–3	246	85	3 : 1
(")–4	428	142	3 : 1
(")–5	393	124	3 : 1
(")–6	353	106	3 : 1
(")–7	480	140	3 : 1

Table 15 gives the results from selfing the F_1 seeds of a cross between No. 15, Longfellow yellow flint and No. 8, white dent. There was no appearance of Xenia in the F_1 seeds. The F_2 seeds segregated in 3 : 1 ratios with the exception of ear (15 x 8)-2. This ear was originally classified as bearing 128 yellow and 50 non-yellow seeds. The F_3 seeds produced by the supposed whites, however, showed the correct ratio to have been 166 yellow and 12 non-yellow. The whites proved true in three other ears. The white seeds from ear (15 x 8)-1 were not grown, and therefore the large excess of yellow seeds cannot be explained. It is possible of course that this ear as well as one or two others that were not planted really had light yellows classified as whites. If this were true one might consider that the original mother plant was homozygous for one yellow and heterozygous for the second. It seems not improbable that this was the case, for the same results were obtained in two other instances.

TABLE 16.

F$_2$ SEEDS FROM CROSS OF NO. 19 WHITE SWEET X NO. 7 YELLOW DENT.

Yellow Seeds Planted.

Ear No.	Y	*y*	Ratio Approx.
(19 x 7)–2	277	77	3 : 1
(")–5	599	43	15 : 1

One other cross, No. 19 non-yellow sweet and No. 7 yellow dent (Table 16), gave di-hybrid ratios. The hybrid seeds were yellow starchy varying somewhat in shade. Only two selfed ears were obtained from the F$_1$ seeds. As shown in Table 16 one is a 3 : 1 ratio and one is a 15 : 1 ratio. Here again is evidence that the male parent was homozygous for one yellow and heterozygous for the second yellow. To be sure there is a slight excess of non-yellows in ear (19 x 7)-5, but this is accounted for in the F$_3$ generation. The supposed non-yellows gave one heterozygous yellow to seventeen non-yellows. The true ratio

TABLE 16A.

F$_3$ SEEDS OF EAR 5 OF CROSS SHOWN IN TABLE 16.

Dark Yellow Starchy Seeds Planted.

Ear No.	Y	*y*	Ratio Approx.
(19 x 7)–5–1	315	98	3 : 1
(")–5–6	320	97	3 : 1
(")–5–9	19	1	15 : 1
(")–5–12	203	14	15 : 1
(")–5–13	440	25	15 : 1

then is 601 : 41 which is very close to theoretical expectancy. The results from planting the yellow starchy seeds of (19 x 7)-5 are shown in Table 16a. Unfortunately the admixture of segregates with wrinkled endosperm made these a little difficult to classify, but there is scarcely a doubt that 2 ears were mono-hybrids and three ears di-hybrids, although no dependence can be placed on ear (19 x 7)-5-9 with only 20 seeds. No pure yellows were obtained from these seeds unless ear (19 x 7)-5-9 were of this class. The deficiency of these data was supplied by the crop of the yellow sweet F_2 seeds of the same ear. Twelve selfed ears were obtained. They are not given in a table because we were not able to prove the classification by growing for another generation, and it is difficult to make *exact* visible classifications of yellow and non-yellow sugar seeds. There is scarcely any doubt however that two ears were pure for both yellows (seeds all dark yellow), two pure for light yellow, (seeds all light yellow) three heterozygous for one yellow (seeds light yellow and white), one at least and probably two heterozygous for two yellows (seeds dark yellow, light yellow and white) and the rest homozyous for one yellow and heterozygous for one yellow (seeds dark yellow and light yellow).

This family gave by far the best demonstration of two yellows as far as the eye is concerned. The ears homozygous for two yellows would never have been classed as the same variety with those homozygous for one yellow. Nearly all the seeds were absolutely distinct, and yet when they were arranged in a series there would always be a number that were difficult to place.

Table 17 gives the F_2 segregates of a mono-hybrid cross between No. 10 white flour and No. 6 yellow dent. There seems to be no question of a di-hybrid ratio, but the cross is interesting for another reason. The heterozygous seeds are lighter than the homozygous so that the effect of Xenia is shown either way the cross is made; that is, Xenia is shown both where *white flour* is crossed with yellow, and where *yellow flour* is crossed with white. The effect is the same as that shown when light starchy caps are formed when a starchy yellow dent is pollinated by a non-yellow, but as in this case the whole seed is floury, therefore it is all changed to lighter yellow.

It might be mentioned that No. 60 yellow pop crossed with No. 2 white, and No. 9 yellow dent crossed with No. 10 flour also show Xenia. The hybrid seeds become so much whiter that there is no difficulty in distinguishing the greater part of them from homozygous yellows.

TABLE 17.

F_2 SEEDS FROM CROSS OF NO. 10 WHITE FLOUR AND NO. 6 YELLOW DENT.

Yellow Seeds Planted.

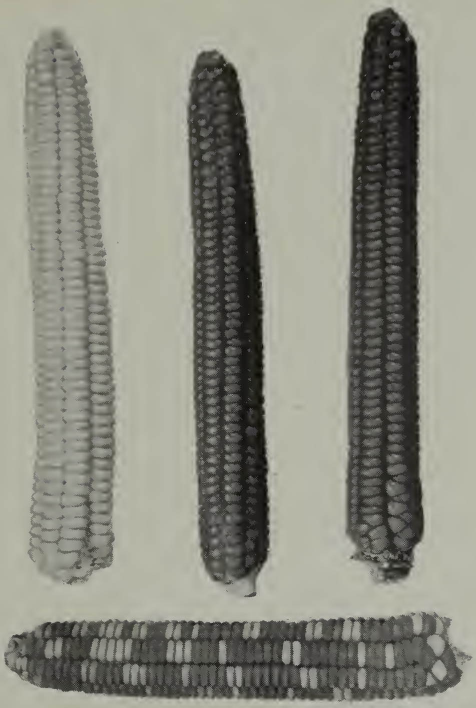
Ear No.	Dark Y	Light Y	Total Y	*y*	Ratio Approx.
(10 x 6)–1	162	357	519	187	3 : 1
(")–2	141	242	383	119	3 : 1
(")–3	175	301	476	156	3 : 1
(")–4	131	243	374	127	3 : 1

Conclusions.

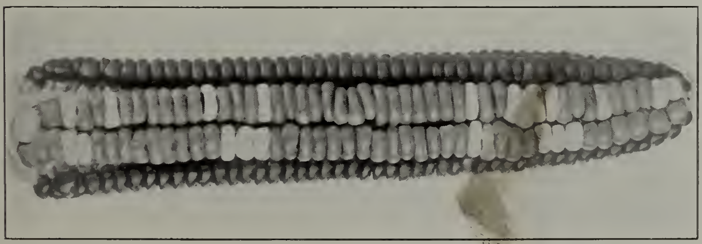
This completes the list of crosses in which new facts have been observed in regard to yellow endosperm. Other crosses might be described where simple mono-hybrid ratios were obtained, but these have already been described by Lock. The di-hybrid ratios have been described in greater detail because they belong to a class of facts having a very important theoretical bearing on the Mendelian hypothesis, which is discussed later in the paper.

It should perhaps be stated that Correns' other general facts have been corroborated. The pure extracted dominants of the F_3 generation have appeared in about the general ratio of 1 homozygote to 2 heterozygotes when dealing with mono-hybrids. There have been insufficient numbers to determine the exact ratio of extracted dominants when dealing with di-hybrids, but in both cases the F_4 generations have in every case bred true. This fact we hold to be more important than the ratio. It may look somewhat queer to say that the extracted F_2 non-yellows have always bred true, when a number of cases have been described in which the seeds that were thought to be non-yellows, proved to be heterozygous yellows. This

PLATE V.



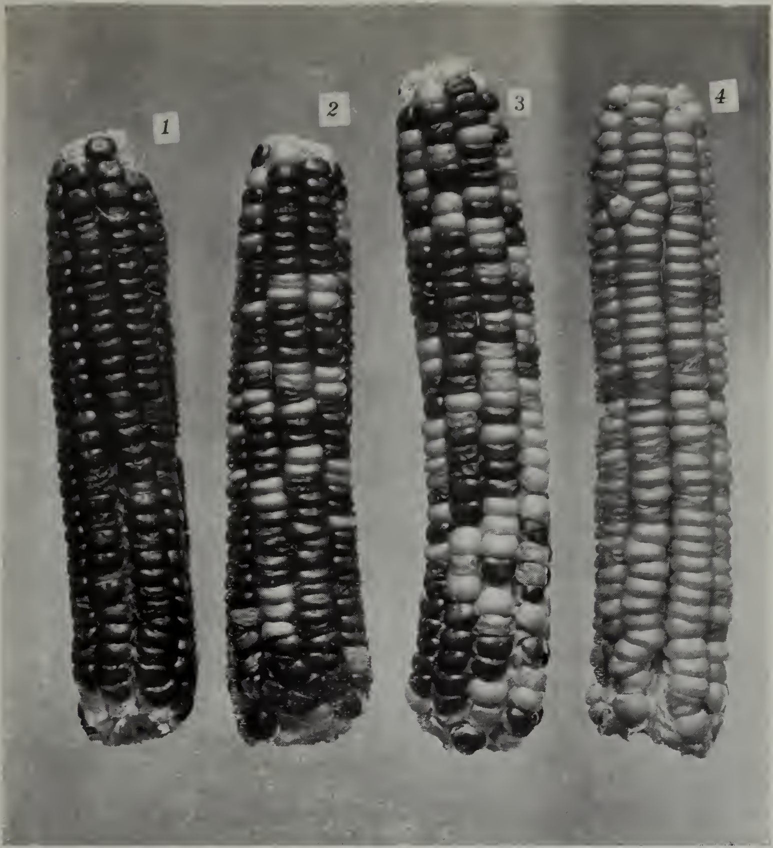
a. At left, No. 24 Rhode Island white cap (white endosperm), at right, No. 15 Longfellow (yellow endosperm). In center, hybrid showing dominance of yellow. Below, F2 seeds showing segregation.



b. An ear showing dominance of red pericarp in F_1. The pericarp has been removed from two rows of seeds, showing mono-hybrid segregation of F2 endosperms beneath it into yellow and non-yellow.

Segregation of Yellow and Non-Yellow Endosperm.

PLATE VI.



a. Cross 24x54. 1. Ear (24x54)-12-5; a pure extracted purple. 2. Ear (24x54)-12-6; purples 208, non-purples 65, a 3:1 ratio. 3. Ear (24x5b)-12-4; purples 147, non-purples 117, a 9:7 ratio. 4. Ear (24x54)-12-3; a pure extracted non-purple.

b. Purple seeds produced by random crossing of non-purple seeds of ear (24x54)-12 shown in Table 18g. 1. Ear (24x54)-12-9x12x8; ratio 1 purple: 3 non-purple. 2. Ear (24x54) 12-11x12x10; ratio 1 purple: 1 non-purple.

Inheritance of Aleurone Color.

is due to the simple fact that the seeds with the gametic formula $Y_1\ y_1$ or $Y_2\ y_2$ vary in color intensity so that it is generally impossible to *classify correctly* from 1 to 5 seeds per ear. These F_2 seeds prove their gametic structure in the F_3 generation; and those that have behaved as pure extracted non-yellows in F_3 have never given anything but pure non-yellows in the F_4 generation.

The occurrence of the two yellow colors casts a further doubt upon the correctness of Lock's work since his main object was to show the truth of Mendel's mathematical conclusions when dealing with large numbers. Our results both here and in the case of the purple aleurone cells show the futility of not making crosses between individuals and of not selfing individual F_1 plants. This is a further excuse for presenting in detail the individual crosses between starchy and non-starchy races with the same object as Lock.

Purple and Non-purple aleurone cells.

The consideration of the inheritance of this character includes also that of a hypostatic red color which appears in crosses between the various purple and non-purple families. The pigments are both fairly easily soluble in water. They are seen first in the aleurone cells of the maturing seeds a few days after fertilization. When the seed is mature the red color becomes an intense dark rose madder, and the purple becomes almost black. Several tests of each pigment were made by macerating the aleurone cells in 50% alcohol and testing the filtrate. With lead subacetate both turned green and a green precipitate separated. The precipitate from the red seeds was somewhat darker and turned greenish brown on evaporation while that from the purple seeds remained a lighter green. Ferrous sulphate added to the red pigment produced but little if any change in color although a dirty precipitate separated on shaking. When added to the blue pigment, however, a dark blue precipitate separated leaving the liquid colorless. This precipitate left a blue residue on evaporation, while the residue from the red pigment was simply a slight discoloration, dark, but with no distinct color except possibly a redness at the edges. Ferric chloride however gave markedly different reactions in the two cases. Added to the red pigment an orange color was produced which became somewhat darker on evaporation. The precipitation was slight. Added to the blue pigment the color was first greenish with blue edges. This turned dark blue and a bluish precipitate separated which later turned green and remained so on evaporation. With ferric alum there was no change except that each pigment became more intense in color. Sodium hydroxide formed brownish green precipitates, darker with the purple. Acids gave red colorations which were lighter with the red pigment. The acid and alkali tests are evidently the usual reactions with vegetable color "indicators" and differ only through the various amounts of pigment present.

It is recognized that tests such as these are arbitrary in nature and cannot form the basis of conclusions as to the chemical composition of the pigments. It seems certain however that they differ somewhat in composition, although they are probably different stages of oxidation of the same color base.

It will be seen in the following pages that purple crossed with different strains of non-purple gave different results. This is clearly due to the various gametic formulæ possessed by the different whites. It may also be that the purples differ somewhat among themselves in unseen characters even though they were pure for purple when selfed. Our analysis of the large amount of data which follows shows that there is simple Mendelian segregation and recombination of several factors and that there is really no confusion of results such as led Correns and Lock to advance various supplementary hypotheses to account for the facts. The use of the color factor C, shows how Lock obtained his purples by crossing white seeds supposedly heterozygous for purple, with white; but it is impossible to analyze his data since individual pollinations were not made. A supplementary hypothesis of Correns should also be mentioned because, if it were true it would necessitate a very different conception of the interpretation of the inheritance of all endosperm characters. Correns supposed that purple × non-purple always gave purple while non-purple × purple sometimes gave non-purple and sometimes gave purple. He accounted for this by the supposition that since the endosperm nucleus is formed by the union of *two maternal* nuclei with *one paternal* nucleus, therefore the maternal endosperm characters would often dominate the paternal characters through the effect of the greater amount of maternal nuclear material. This is *never* the case and the fact is quite important. If Correns' supposition were true and the amount of nuclear matter determined the characters to be formed, no Mendelian segregation of endosperm characters and their recombination by chance matings could be demonstrated. Since all of our data shows it to be untrue, it follows that the *quality* and not the quantity of nuclear material is the important thing. The nucleus evidently regenerates or throws off material to come to its proper adjustment for the performance of its functions, and *always* in accordance with the *quality* of its structure.

In order to facilitate a consideration of the data, it will be presented in families. Each family comprises the progeny resulting from a particular cross. They are taken up in the order of increasing complexity.

Family (24 and 54)

This family includes all of the progeny of the cross of No. 24 white flint with No. 54 Black Mexican sweet, this being the variety with purple aleurone cells. The Black Mexican which furnished the pollen for this cross had proved true to the purple color for three generations, but pollen for the crosses of the different hybrid families came from several different ears. For this reason there is no certainty that the purple aleurone parent had the same gametic structure in each family. The data for the above family are reported in the sub-divisions of Tables 18 to 20. In these tables there is no correlation of the purple and starchy characters, there being a simple 3 : 1 relationship of starchy and non-starchy seeds in both the case where purples and non-purples were obtained in F_2 in the ratio of 3 : 1 and where they are obtained in the ratio of 9 : 7. We may therefore leave this character out of consideration and consider only the purple character.

The F_1 seeds formed in the hybrid ear were all purple. Upon growing these seeds nine selfed ears were obtained with the ratios of purples to non-purples shown in Table 18. The purple color of these segregates was of full depth and covered the entire seed with one or two exceptions. These exceptions were zygotic variations due to heterozygosis and were quite different from the partial or light purples obtained in other families. In the latter case it was due to a transmissible gametic factor which will be explained later. Table 18 shows the ratio of purples to non-purples to be 3 : 1 in the case of seven ears and 9 : 7 approximately in the case of two ears. This immediately suggests the mono-hybrid ratio in the first case and a di-hybrid ratio in the second case. That this is the true state of affairs is shown by the behavior of the seeds of these ears in later generations. The progeny of the purple seeds of ear (24 x 54)-1 (Table 18a) were either pure purples or heterozygous purples segregating in the ratio of 3 : 1. The non-purple seeds of the same ear (Table 18b) produced only non-purples. The same ratio was obtained from purple seeds of ear No. (24 x 54)-11 shown in Table 18c.

The fact that F_3 extracted non-purple seeds continued to breed true is shown by the results of the F_4 generation shown in Table 18d. Extracted purple starchy seeds were also planted from Ear No. (24 x 54)-1-4 and ten selfed ears proved pure. Twenty-six ears were also obtained from the open field crop which were also pure purple, six being pure starchy and twenty heterozygous starchy.

These continued 3 : 1 ratios with purity of the extracted homozygote are what should be expected from the progeny of the mono-hybrid ears of Table 18. If the 9 : 7 ratios given by ears No. 9 and No. 12 of Table 18 are true di-hybrid ratios resulting from the interaction of two factors both of which are necessary for the production of the purple color, one should expect in the F_3 generation but one pure purple out of nine to occur and the remaining ears to be about 50% monohybrids with a 3 : 1 ratio and 50% di-hybrids with a 9 : 7 ratio. The progeny of ear No. (24 x 54)-12 (Tables 18e, 18f) shows how nearly these expectations are confirmed. Out of a total of nineteen selfed ears two were pure purple, ten were mono-hybrids and seven were di-hybrids. It must be concluded therefore that the purple color is due to the action of the factor P upon another color factor C, which is probably similar in nature to that which Bateson found in sweet peas. The gametic structure of No. 24, the non-purple variety, evidently differed in the ovules of the seeds of the original hybrid ear. Part of them lacked both P and C and gave a 9 : 7 ratio when crossed with the purple (C P), and part of them contained either P or C and therefore gave a mono-hybrid ratio when crossed with C P. If one supposes C to be contained by the non-purple in the first case then the result is as follows, Cp x C P = C C P p. The gametes formed differ only in presence or absence of P and a simple mono-hybrid ratio is obtained in the F_2 generation.

In the second case the cross is c p x C P = C c P p, and the F_2 populations have the formulae and ratios 9 C P : 3 C : 3 P : 1 c p, the first nine being purple and the last seven being white. This being the case the various non-purple seeds of F_2 should prove true non-purples when selfed but should sometimes give purples when crossed. The non-purples exist in the following ratios:

1 C C p p
2 C c p p
1 c c P P
2 c c P p
1 c c p p

When crossed at random there are 7 x 6 = 42 possible combinations of which 24 should give all non-purple and 18 some purples. Of these eighteen ears 2 should be pure purples, 8 purples and non-purples in the ratio 1 : 1, and 8 purples and non-purples in the ratio of 1 : 3. In Tables 18g and 18h besides the selfed non-purples seven combinations of different non-purples are shown, besides several reciprocal crosses. Of these one combination and its reciprocal gives a 1 : 1 ratio and one combination and its reciprocal gives a 1 : 3 ratio.

None of the F_2 seeds of the selfed ears of this cross showed any seeds with red aleurone cells. Among the open field ears containing F_2 seeds however, were noticed several seeds with aleurone cells of a peculiar blue color and several of the red color. Five selfed ears were obtained from the blue aleurone seeds (Table 19). Four of these ears gave 9 colored (P and R) seeds to 7 non-colored and one gave a simple mono-hybrid ratio in which no reds were found. The red seeds varied in shade until the darkest seemed to the eye to be purple. They could be separated accurately only by a microscopic examination of sections of the aleurone cells. The purples (the blue seeds proved to be exactly like ordinary purple seeds) occurred in greater numbers than the reds but the exact ratios were not determined in this family, because their parentage was not certain.

The red seeds found in the open field ears also proved to be heterozygous for red as shown by Table 20. They gave simple 3 : 1 ratios except ear No. 1 which proved to be pure red although heterozygous for starchiness. F_3 seeds were obtained from the red seeds of ear No. 8 as is shown in Tables 20a and 20b. It happened that in this small number five pure red ears were obtained and only three ears that were heterozygous and segregated in the ratio of 3 : 1.

Besides the ears shown in Table 20a, two ears from extracted red seeds were crossed with pure extracted non-purples (whites) of the F_3 generation of cross (24 x 54). Ear No. 1 gave 125 purples and 123 non-purples. Ear No. 2 gave 108 purples and 124 non-purples. The red ears, the maternal parents of the crosses, were evidently heterozygous and therefore a 1 : 1 ratio was obtained. The non-purple which furnished the pollen must have carried the P factor which oxidized the seeds which otherwise would have become red to the purple color. This fact proves the epistatic nature of P over R and is a further proof of the di-hybrid nature of the purple color. Another ear crossed with non-purples of the same family as above gave all purple seeds. This ear evidently was homozygous for red and all of its seeds were oxidized to purple. Two other of these red ears were crossed with extracted purples of the same cross from which came the extracted whites used above. The seeds of the resulting ears were all purple. (See Plate 8a.)

Several red non-starchy seeds from ear (24 x R)-16-8 (Table 20b) were also planted. Three selfed ears resulted in two pure for red and one giving 248 reds to 60 non-reds, a 3 : 1 ratio. One ear of this lot was crossed with the same extracted purples used in crossing the starchy red seeds resulting in an ear with all purple seeds. Another ear was crossed with one of the extracted non-purples used in crossing the red starchy seeds and resulted in an ear with 119 purple starchy and 124 non-purple starchy seeds. The results from the non-starchy seeds of this family were therefore the same as those from the starchy seeds.

The non-red seeds from (24 x R)-16-8 both starchy and non-starchy bred true to the non-red character. Four crosses between individual ears of this lot were made and the resulting seeds were all non-red. This is the result which should be expected from an ear giving a mono-hybrid ratio as did ear (24 x R)-8 and shows that the purples resulting from the crosses between the non-purples coming from the 9 : 7 ratios were not accidental.

TABLE 18.

F₂ SEEDS FROM CROSS OF NO. 24 WHITE FLINT X NO. 54 PURPLE ALEURONE NON-STARCHY.

Purple Aleurone Starchy (PS) Seeds Planted.

Ear No.	PS	Ps	pS	ps	Total P	Total p	Ratio Approx.
(24 x 54)–1	207	67	67	27	274	94	3 : 1
(")–2	170	54	49	19	224	68	3 : 1
(")–6	197	65	59	24	262	83	3 : 1
(")–9	83	44	72	25	127	97	9 : 7
(")–10	166	40	46	19	206	65	3 : 1
(")–12	153	40	115	40	193	155	9 : 7
(")–8	159	41	41	23	200	64	3 : 1
(")–11	166	55	47	22	221	84	3 : 1
*(")–13	205	81	59	25	286	84	3 : 1

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* All purple seeds were full dark purples except a few splashed purples from this ear.

TABLE 18A.

F₃ SEEDS OF EAR 1 OF SAME CROSS AS TABLE 18.

Purple Aleurone Starchy (PS) Seeds Planted.

Ear No.	PS	Ps	pS	ps	Total P	Total p	Ratio Approx.
(24 x 54)–1–3	...	..	..	..	144	..	Pure P
(")–1–4	...	..	..	..	384	..	Pure P
(")–1–5	...	..	..	..	96	..	Pure P
(")–1–11	...	..	..	..	320	..	Pure P
(24 x 54)–1–2	161	55	46	13	216	59	3 : 1
(")–1–6	171	56	52	19	227	71	3 : 1
(")–1–8	180	71	55	19	251	74	3 : 1
(")–1–9	79	29	27	7	108	34	3 : 1
(")–1–10	255	..	91	..	255	91	3 : 1
(")–1–14	195	..	80	..	195	80	3 : 1
Total	..	..	..	..	1251	410	....

TABLE 18B.

F_2 SEEDS OF EAR 1 OF SAME CROSS AS TABLE 18.

Non-Purple Aleurone Starchy (ps) Seeds Planted.

Ear No.	P	p	Ratio Approx.
(24 x 54)-1-4a	..	308	Pure white
" -1-5a	..	312	"
" -1-6a	..	362	"
" -1-7a	..	320	"
" -1-15a	..	296	"

TABLE 18C.

F_3 SEEDS OF EAR 11 (TABLE 18A) OF SAME CROSS AS TABLE 18.

Purple Aleurone Non-Starchy (Ps) Seeds Planted.

Ear No.	Ps	ps	Ratio Approx.
(24 x 54)-11-2	312	..	Pure P
" -11-3	368	..	"
" -11-4	280	..	"
" -11-1	240	82	3 : 1
" -11-5	197	78	3 : 1
" -11-6	205	52	3 : 1
" -11-11	40	12	3 : 1
Total Het.	682	224	....

TABLE 18D.

F_4 SEEDS OF EAR (24 x 54)-1-6 (EXTRACTED PS, TABLE 18B) OF SAME CROSS AS TABLE 18.

Non-Purple Starchy (ps) Seeds Planted.

Ear No.	ps	PS
(24 x 54)-1-6-1	All	
" -1-6-2	"	
" -1-6-5	"	
" -1-6-8	"	
" -1-6-1 x (24 x 54)-8-5 RS	..	All
" -1-6-6 x " -8-8 RS	..	"
" -1-6-12 x " -8-3 RS	..	"
" -1-6-9 x " -8-10 RS	..	"
" -1-6-10 x " -8-1 RS	..	"

Open-field crop all white.

a. Flint and dent segregates from F2 of cross 8x54. Flint character carried by No. 54.

b. F3 types from cross 8x54. 1. Pure extracted purple (PPCC). 2. Pure extracted parti-colored ((PPcc). 3. Pure extracted non-purple (ppCC or ppcc).

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TABLE 18E.

F_3 SEEDS OF EAR (24 x 54)–12 OF SAME CROSS AS TABLE 18.

Purple Starchy (PS) Seeds Planted.

Ear No.	PS	p	SS or Ss	Ratio Approx.
(24 x 54)–12–1	280	. .	Ss	Pure
(“)–12–2	147	40	Ss	3 : 1
(“)–12–3	190	60	Ss	3 : 1
(“)–12–4	147	117	Ss	9 : 7
(“)–12–5	288	. .	Ss	Pure
(“)–12–6	208	65	Ss	3 : 1
(“)–12–7	188	115	Ss	9 : 7
(“)–12–8	237	72	SS	3 : 1
(“)–12–8_1	212	72	Ss	3 : 1
(“)–12–9	159	120	SS	9 : 7
(“)–12–10	145	56	Ss	3 : 1
(“)–12–11	95	30	Ss	3 : 1
(“)–12–12	179	59	Ss	3 : 1

TABLE 18F.

F_3 SEEDS OF EAR (24 x 54)–12 OF SAME CROSS AS TABLE 18.

Purple Non-Starchy (Ps) Seeds Planted.

Ear No.	P	p	Ratio Approx.
(24 x 54)–12–1a	160	53	3 : 1
(“)–12–2a	186	64	3 : 1
(“)–12–3a	137	115	9 : 7
(“)–12–4a	97	65	9 : 7
(“)–12–6a	109	80	9 : 7
(“)–12–9a	123	120	9 : 7

TABLE 18G.

F_3 SEEDS OF EAR (24 x 54)–12 OF SAME CROSS AS TABLE 18.

Non-purple Starchy (pS) Seeds Planted.

Ear No.	P	p	Ratio Approx.
(24 x 54)–12–3b	. .	All	. . . .
(“)–12–4b	. .	“	. . . .
(“)–12–6b	. .	“	. . . .
(“)–12–7b	. .	“	. . . .
(“)–12–12b	. .	“	. . . .
(“)–12–2b x 12–4b	. .	“	. . . .
(“)–12–4b x 12–2b	. .	“	. . . .
(“)–12–5b x 12–1b	. .	“	. . . .
(“)–12–8b x 12–9b	13	62	1 : 3
(“)–12–9b x 12–8b	81	226	1 : 3
(“)–12–10b x 12–11b	79	86	1 : 1
(“)–12–11b x 12–10b	93	99	1 : 1

TABLE 18H.

F_2 SEEDS OF EAR (24 x 54)–12 OF SAME CROSS AS TABLE 18.

Non-Purple Non-Starchy (ps) Seeds Planted.

Ear No.	P	p
(24 x 54)–12–2c	..	All
(")–12–3c	..	"
(")–12–5c	..	"
(")–12–9c	..	"
(")–12–10c	..	"
(")–12–12c	..	"
(")–12–13c	..	"
(")–12–1c x 12–4c	..	"
(")–12–4c x 12–1c	..	"
(")–12–7c x 12–3c	..	"
(")–12–3c x 12–7c	..	"
(")–12–6c x 12–8c	..	"
(")–12–8c x 12–6c	..	"

TABLE 19.

F_2 SEEDS FROM CROSS BETWEEN NO. 24, WHITE FLINT AND NO. XP PURPLE ALEURONE.

Purple Aleurone Starchy (PS) Seeds Planted.

Ear No.	P+R	p	Ratio Approx.	Notes
(24 x P)–16–2	287	192	9 : 7	SS: some seeds red
(")–16–5	141	117	9 : 7	ss: few P's strongly colored
(")–16–6	165	115	9 : 7	ss: few P's strongly colored
(")–16–7	278	89	3 : 1	Ss: 84 P's lighter at cap
(")–16–8	253	193	9 : 7	ss: 69 P's lighter at cap

TABLE 20.

F_2 SEEDS FROM CROSS BETWEEN NO. 24 WHITE FLINT AND NO. XR RED ALEURONE.

Red Aleurone Starchy (RS) Seeds Planted.

Ear No.	RS	Rs	rS	rs	Total R	Total r	Ratio Approx.
(24 x R)–16–1	160	52	..	..	212	..	Pure red
(")–16–4	26	12	13	3	38	16	3 : 1
(")–16–6	140	43	53	22	183	75	3 : 1
(")–16–8	195	73	41	19	268	60	3 : 1

TABLE 20A.

F_3 SEEDS OF EAR (24 x R)–16–8 OF SAME CROSS AS TABLE 20.

Red Aleurone Starchy (RS) Seeds Planted.

Ear No.	R	r	Ratio Approx.
(24 x R)–16–8–3	360	..	Pure red
(")–16–8–4	161	65	3 : 1
(")–16–8–5	60	..	Pure red
(")–16–8–6	172	53	3 : 1
(")–16–8–8	320	..	Pure red

TABLE 20B.

F_3 SEEDS OF EAR (24 x R)–16–8 OF SAME CROSS AS TABLE 20.

Red Aleurone Non-Starchy (Rs) Seeds Planted.

Ear No.	R	r	Ratio Approx.
(24 x R)–16–8–1a	160	..	Pure red
(")–16–8–2a	248	60	3 : 1
(")–16–8–3a	280	..	Pure red

Family (8 x 54)

The F_1 Xenia seeds of the cross between No. 8 non-purple dent starchy and No. 54 purple non-starchy were all purple in color. Four selfed ears were obtained when these hybrid seeds were planted. The segregation of the F_2 seeds is shown in Table 21. A new phenomenon of peculiar interest appeared in this family. A certain number of seeds were solid dark purple, others were splashed dark purple, others were a very faint purple and have been called particolored, while still others were without the purple color. The splashed dark purples were seeds that had a break in the purple color; that is the purple color was dark but appeared in patches. These splashed purples are found in all of the purple-non-purple crosses except the family * just described. It seems evident then that they are due to the interaction of characters which

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* Only one or two splashed purples were ever found in family (24 x 54).

happen to be absent from the (24 x 54) family; but at the same time they are zygotic variations which are not inherited, for their progeny are exactly like the progeny of the dark purple seeds. Further, these patches are not in a regular pattern nor does the selection of seeds of this nature have the slightest tendency to fix the phenomenon as a separate character. There is reason for believing however that no homozygous purples (C C P P) are ever of this nature, and that the splashing is simply due to incomplete dominance, but caused by a factor or factors brought in by the non-purple parent.

The fact that particolored or very light purples which transmitted the character also appeared in this family made it seem probable that a new character had appeared, making the family a tri-hybrid. But this is not the simplest interpretation. We have seen in the other family that the behavior of purple is best interpreted as the interaction of two factors C and P. In this family the hypothesis that either Cp or cP seeds are not pure whites but very light purples is supported by all of the data. At first sight it seems more reasonable that they should have the formula Cp. If in accordance with older interpretations of color inheritance, the purple color is formed by an enzyme, P acting upon a chromogen C it is more reasonable to suppose that in the presence of the chromogen an exceedingly small amount of the enzyme might give rise to the particolored seeds, than it is to believe that the normal amount of enzyme would form the purple color with a trace of chromogen. The reason for this statement rests upon the well known fact that enzymes are organic catalysers and can accelerate reactions involving quantities very disproportionate to their own amount. There is an objection to this interpretation, however, for when particolored seeds are crossed with those having red aleurone cells and which therefore have the gametic formula R C, they invariably give purples. This proves that the gametic formula of the particolored seeds is c P and they are so designated in the tables.

The suggestive work of Miss Wheldale (: 09, : 09a, : 10) in correlating the results of biological chemistry with those of genetics, has made it very probable that a basic chromogen is present in all flowers which are able to form a sap color, and that the complexities of color inheritance may be referred to

PLATE VIII.

a. 1. Pure purple aleurone resulting from crossing pure extracted red aleurone with pure purple. 2. Same result from crossing pure extracted red aleurone with colorless aleurone. 3. Seeds half purple resulting from crossing heterozygous red aleurone with colorless aleurone. 4. Result from selfing the male parent of 3.

b. 1. (24x54)8-3 pure extracted red aleurone. 2. (24x54)-8-6 heterozygous red aleurone. Cut does not show color value when compared with Fig *a.*

INHERITANCE OF ALEURONE COLOR.

the dual nature of the oxydases necessary for the formation of the color compounds. It is quite likely that the color in the aleurone cells of maize is similar in nature to flower color; and, as we fully agree with Miss Whedale's conclusions, none of our factors C, R and P are to be regarded as chromogens. The argument above is in agreement with this viewpoint. If one wishes to denote a chromogen, the addition of an X to represent it, common to both families, makes no difference in the interpretation of the results.

If we are dealing with a di-hybrid ratio, one pure purple ear out of every nine should be expected in the F_2 generation. Tables 21a and 21b show that one such ear was obtained out of seventeen ears. If the total purple seeds and the sum of the particolored and white seeds is considered in Tables 21, 21a and 21b a close approximation to a 9 : 7 ratio is obtained. If the particolored seeds could in every instance be distinguished from whites the ratio of purples to particoloreds to whites should be 9 : 3 : 4. It will be noticed however that in the ears from which this ratio should be expected there is generally an excess of whites. This is explained by the fact that particoloreds especially when non-starchy are not always distinguishable from whites. The last two ears shown in Table 21d are in fact ears grown from seeds which were originally classed as whites. If this hypothesis in regard to the particolored is true, one should expect the purple F_2 seeds to give in the F_3 generation, one ear pure purple, two ears showing segregates of purple and particolored in the ratio of 3 : 1, two ears showing segregates of purple and non-purple in the ratio of 3 : 1, and four ears showing purples, particolored and non-purples in the ratio 9 : 3 : 4. Among the ears received (Tables 21a, 21b) there were one of the first class, six of the second class, three of the third class and seven of the fourth class.

Tables 21c and 21d show the results from growing the particolored seeds of the same ear, No. (8 x 54)-1. One ear should be pure particolored to two showing segregates of particolored and non-purple in the ratio of 3 : 1. Out of the fifteen ears obtained three were evidently of the first class and twelve of the second class.

In all of these tables the progeny of hybrid starchy seeds segregated normally.

Seeds classified as non-purples were also planted from this same ear No. (8 x 54)-1. The thirteen selfed ears resulting as progeny of starchy seeds all proved to be non-purple. Two particolored ears, however, appeared in the eight selfed ears resulting from planting non-purple, non-starchy seeds. This showed that there was more difficulty in classifying the non-starchy non-purples than in classifying starchy non-purples. Non-purple seeds planted from ear No. (8 x 54)-5 also gave a few particolored progeny.

Four thousand seeds from tested whites of the F_3 seeds were planted in an isolated plot the next season and were allowed to inter-cross naturally. If we were dealing with di-hybrid non-purples in this case, such inter-crossing should give some purples, such as were obtained in the (24 x 54) family. The resulting crop of this large number of plants however were all true non-purples, proving that we were dealing with non-purples with formulæ either CC, Cc or cc. Further proof of the constitution of the particolored is shown in the following facts. No particoloreds ever gave full purples. Furthermore, pure extracted particoloreds (c c P P) from ear No. (8 x 54)-1-13b of Table 21c were grown for another generation and their gametic structure tested by various crosses. Several of these ears were selfed and all proved to be pure particoloreds (ccPP). Three different ears were crossed with pure extracted purples from progeny of ear No. (24 x 54)-1-4. As would be expected all of the seeds were purple. Two of the ears however had a decided reddish purple color while one was dark purple without the reddish tint. Four ears were crossed with extracted red seeds (RRCC)· All produced purple seeds. Nine ears were crossed with plants of the progeny of the non-purples of ear No. (24 x 54)-12. It will be remembered that this ear gave a ratio of nine purples to seven non-purples. The seven non-purples would have the following formulæ: 1 PPcc, 2 Ppcc, 1 ppCC, 2 ppCc, 1ppcc. Crossing the particoloreds at random with pollen of individual plants of this lot should give on the average one ear with all purple seeds when pollinated by ppCC, two ears with 50% purple and 50% particolored when pollinated by ppCc, four ears pure particolored when pollinated by PPcc, Ppcc or ppcc. Nine ears were obtained of which one had all purple seeds, three had 50% purple and 50% particolored with a total of 308 purple seeds to 294 particolored seeds and five were all particolored. It should be mentioned, however, that the particolored seeds obtained by crosses with the whites of this (24 x 54) family in which the (ccPP) seeds were not particolored, gave seeds which averaged much lighter in appearance than the pure particolored. In other words particoloreds crossed with whites of other families show imperfect dominance of particolored. Some gene common to both parents of the (8 x 54) family, therefore, accounts for the production of the color.

These two families differed in no other endosperm character except presence and absence of starchiness. No correlation of any kind was observed between these two allelomorphic pairs.

TABLE 21*.

F_2 SEEDS FROM CROSS BETWEEN NO. 8 NON-PURPLE DENT STARCHY AND NO. 54 PURPLE NON-STARCHY.

Purple Seeds Planted.

Ear No.	CP	cP	Cp or cp	Total Purple	Total Non-Purple
(8 x 54)–1	297	75	146	297	221
(")–2	230	75	172	230	247
(")–3	...	..	...	302	239
(")–5	...	..	...	270	229
Total	...	..	...	1099	936

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* There were 1,514 starchy and 521 non-starchy seeds.

TABLE 21A*.

F_3 SEEDS OF EAR NO. (8 x 54)–1 OF TABLE 21.

Purple Starchy (PCS) Seeds Planted.

Ear No.	CP	cP	Cp or cp	Total Purple	Total Non-Pur.	Starchiness
(8 x 54)–1–1	233	70	. .	233	70	SS
(")–1–2	16	6	7	16	13	Ss
(")–1–3	238	69	. .	238	69	Ss
(")–1–4	321	86	. .	321	86	Ss
(")–1–6	. . .	. .	. .	312	. .	Ss
(")–1–8	239	106	93	239	199	Ss
(")–1–9	223	65	95	223	160	SS
(")–1–12	285	. .	66	285	66	SS
(")–1–14	160	33	111	160	144	Ss
(")–1–20	126	54	. .	126	54	SS

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* There were 1,362 starchy and 435 non-starchy seeds in the Ss ears.

TABLE 21B.

F_3 SEEDS OF EAR NO. (8 x 54)–1 OF TABLE 21.

Purple Non-Starchy (PCs) Seeds Planted.

Ear No.	CP	cP	Cp or cp	Total Purple	Total Non-Purple
(8 x 54)–1–1a	229	. .	79	229	79
(")–1–3a	236	44	116	236	160
(")–1–4a	295	. .	93	295	93
(")–1–6a	260	86	. .	260	86
(")–1–7a	86	20	38	86	58
(")–1–10a	239	89	. .	239	89
(")–1–11a	223	55	88	223	143

TABLE 21C*.

F_3 SEEDS OF EAR NO. (8 x 54)–1 OF TABLE 21.

Particolored Starchy (cPS) Seeds Planted.

Ear No.	cP	Cp or cp	Starchiness
(8 x 54)–1–2b	322	99	SS
(“)–1–5b	402	. .	SS
(“)–1–6b	115	70	Ss
(“)–1–7b	150	64	SS
(“)–1–10b	386	. .	SS
(“)–1–11b	254	92	Ss
(“)–1–13b	427	. .	Ss
(“)–1–14b	262	112	SS
(“)–1–15b	256	133	Ss

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* There were 1,026 starchy and 321 non-starchy seeds in the Ss ears.

TABLE 21D.

F_3 SEEDS OF EAR NO. (8 x 54)–1 OF TABLE 21.

Particolored Non-Starchy (cPs) Seeds Planted.

Ear No.	cP	Cp or cp
(8 x 54)–1–2c	149	110
(“)–1–3c	364	. . .
(“)–1–4c	168	89
(“)–1–5c	123	59
(“)–1–1w	230	115
(“)–1–12w	131	99

Family (15 x 54).

This family brings in a third allelomorphic pair namely presence and absence of yellow in the endosperm. No. 15 is Longfellow pure for the presence of starchiness and for a single yellow factor. This cross was made to find out whether there were further differences in the behavior of the purple factor when crossed with other non-purples, and it was thought that the yellow endosperm might prove a disturbing factor. This is not the case for the F_1 seeds were all purple with the exception that a few splashed purples, which behaved like the normal hybrid purples, also occurred in this family. Eight ears were obtained by growing the F_1 seeds, and starchiness and yellowness were found to segregate in a normal manner. There was a total of 1765 Y to 604 y and 1746 S to 623 s seeds.

There is only one fact of particular interest in this family. Table 22 shows the eight selfed F_1 ears grown from the purple-starchy hybrid seeds, containing the F_2 seeds. It will be noticed that in the table, six of the ears appear to show monohybrid segregation and two of them di-hybrid segregation. This is not really the case. All of the ears giving the 3 : 1 ratio were also di-hybrids. The figures in the column headed "Purple" contained purples, splashed purples and particoloreds. Some unknown cause produces many seeds in this cross that are heavily splashed with purple. These always behaved as heterozygous purples, although the heterozygous purples were not always splashed, but were generally full colored purples. The particoloreds are seeds containing the P factor but lacking the C factor as in cross (8 x 54). The difficulty here was to distinguish by sight *all* of the splashed purples (P C) from the particoloreds (Pc). They were all included in the table therefore as "Purples."

The ears (15 x 54)-2 and (15 x 54)-3 did not show particolored seeds, but that the same gametes were concerned is shown by the following data. Theoretically, if ear (15 x 54)-2 is a di-hybrid the purple seeds when selfed should give 1 ear with all purple seeds, 4 ears with 3 purple seeds to 1 non-purple seed and 4 ears with 9 purple seeds to 7 non-purple seeds. Twelve selfed ears were obtained in the next generation. One had all purple seeds. Seven had purple and non-purple seeds at the ratio of 9 : 7, there being a total of 1035 purple and 763 non-purple seeds. Four had purple and non-purple seeds at the ratio of 3 : 1, there being a total of 480 purple to 162 non-purple seeds. It should be remarked that in two of these ears a few very light particolored seeds were found, showing that the seemingly aberrant ear (15 x 54)-2 had a slight tendency to throw particoloreds like the other ears of the family. There is also some evidence that microscopical examination of the embryo stem would show particoloreds in the ordinary ratio.

The non-purples from this ear were also grown. Eighteen selfed ears were obtained. All of them were true to non-purple. Two of them had a few particolored seeds (6 in one case and 14 in another). These seeds might possibly have been produced by the contamination of a few grains of foreign pollen, but they might very well be white seeds of the formula Pc which were showing the racial tendency to a slight production of pigment (i. e. particoloreds).

Non-purples from the other aberrant ear No. (15 x 54)-3 were also grown and when selfed gave only non-purples. Three intercrosses and their reciprocals were made between different plants from this lot. It happened that no purple seeds were obtained as should be expected in a portion of the cases, as explained before. That the non-purples did differ in composition among themselves was shown however by crossing a pure particolored (PPcc) of the (8 x 54) family with pollen from one of our non-purples, ear No. (15 x 54)-3-10. The ear resulting from the cross had 179 purple seeds and 168 particolored seeds. This 1 : 1 ratio could only have been obtained from a non-purple heterozygous for C (Cc). As a non-purple with the formula Cc could only have been obtained from a di-hybrid cross, it is proved that all of the ears of this family were di-hybrids. The complete gametic structure of the hybrid seeds, speaking of endosperm characters only, is YySsCcPp.

TABLE 22.

F_2 SEEDS FROM CROSS BETWEEN NO. 15 NON-PURPLE YELLOW STARCHY AND NO. 54 PURPLE NON-STARCHY.

Purple Seeds Planted.

Ear No.	Purple*	Non-Purple	Ratio Approx.
(15 x 54)-1	138	45	9+3 : 3+1
(")-2	203	135	9 : 7
(")-3	109	83	9 : 7
(")-4	250	84	9+3 : 3+1
(")-6	201	61	9+3 : 3+1
(")-8	307	91	9+3 : 3+1
(")-11	254	93	9+3 : 3+1
(")-15	239	76	9+3 : 3+1

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* Every ear except ears 2 and 3 contained splashed purples which act as heterozygous purples in inheritance and particoloreds which act as if they had the gametic formula (cP), but the intergradations were so gradual that it was impossible to make an accurate classification. The matter is not worth mentioning here except for the reason that persons who had not had experience with the behavior of purple and non-purple crosses in other families would be utterly at loss for a classification and it would be necessary for them to grow each individual seed for another generation to determine its gametic formula.

Family (18 x 58)

No. 18, the female parent of this cross is a small non-purple sugar maize which usually has twelve rows. The purple parent is a small eight-rowed flint. The F_1 seeds were purple. Only one selfed ear was obtained from the F_1 plants through an unfortunate loss of pollen. The segregation of F_2 seeds is shown in Table 23. The hybrid seeds have the gametic formula Pp RrCc. The seeds with the formula PR and probably also with the formula P give particoloreds or very light purples as they did in family 8 x 54. They were very light however and the 138 seeds classed as whites or non-purples contained some particoloreds as is shown in the F_3 generation. Theoretically in the F_2 generation there should be 36 purples (27 PRC+9 PC), 9 reds (RC), 12 particoloreds (9 PR+3 P) and 7 whites (3 C+3

PLATE IX.

a. F3 color segregates from cross (18x58). 1. Pure extracted purple. 2, 3. Ears from heterozygous plants. 4. Pure extracted non-purple.

b. F3 color segregates from cross (18x58). 1. Pure extracted red. 2, 3. Ears from heterozygous plants. 4. Pure extracted non-red. Proper color values are not shown.

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R+1 prc). There is an excess of whites because the particoloreds could not be classified easily, so it might be said that there should be 36 purples: 9 reds: 19 particoloreds and whites. In the one ear obtained there is still an excess of the last class, but the behavior of the seeds in the F_3 generation proves the gametic constitution of the parents. Tables 23a and 23b give the results from planting purple F_2 seeds. The last four ears shown in Table 23a were planted from splashed purples but they gave the same result as the full purples. We may conclude therefore that splashed purples behave the same as self-colored purples in inheritance. Theoretically the entire lot of purples should have the following gametic constitutions and proportions:

Class 1. 1 P P R R C C = Pure purple.
" 2. 2 P p R R C C = 3 purple: 1 red.
" 3. 2 P P R r C C = Pure purple.
" 4. 2 P P R R C c = 3 purple: 1 white.
" 5. 4 P p R r C C = 12 purple: 3 red: 1 white.
" 6. 4 P p R R C c = 9 purple: 3 red: 4 white. 3 being particolored.
" 7. 4 P P R r C c = 12 purples: 4 white. 3 being particolored.
" 8. 8 P p R r C c = 36 purples: 9 reds: 19 whites. 12 being particolored.
" 9. 1 P P C C = Pure purple.
" 10. 2 P p C C }
" 11. 2 P P C c } = 3 purples: 1 white.
" 12. 4 P p C c = 9 purples: 7 whites and particoloreds.

These ears when selfed should give the proportions shown at the right of the above column. An examination of Tables 23a and 23b show that out of the 23 selfed ears obtained the expected ratios were followed rather well. There were two pure purple ears, Classes 1, 3 and 9; 2 ears of Class 2; 3 ears of Class 12; 4 ears of Classes 4, 7, 10 and 11 which collectively give 3 purples; 1 white; 3 ears of Class 8; 9 ears of Classes 5 and 6. The particolored seeds are very light in color and although they are classified as nearly as possible in the tables this classification should be considered only an approximation and not a reality.

Particoloreds and whites are considered together in determining the gametic constitution of the ears. The particolored seeds proved to be true particoloreds of the same nature as those of family (8 x 54). The selfed ears resulting from such seeds of ear No. (18 x 58)-1 of Table 23, gave no purples. Pure particoloreds ears and heterozygous particolored ears were obtained but no exact visual classification of the latter could be made and it was not considered worth while to determine their precise constitution by breeding.

The red segregates occurring in ear No. (18 x 58)-1 were also tested in the F_3 generation. Fifteen selfed ears were obtained and are shown in Table 23c. Among them were five pure red ears, six which threw reds and whites in the ratio of 9 : 7 and four which threw reds and whites in the ratio 3 : 1. The number of pure red ears obtained was slightly greater than should generally be expected but such a deviation should occur about once out of five times when dealing with lots of only fifteen ears. The selfed white segregates of ear No. (18 x 58)-1 of Table 23 yielded about one particolored ear either homozygous or heterozygous out of every four. This shows the error in trying to classify particolored and white seeds. There is no doubt however that when pure white segregates are planted they always breed true.

TABLE 23.

F_2 SEEDS OF CROSS BETWEEN NO. 18 NON-PURPLE NON-STARCHY AND NO. 58 PURPLE STARCHY.

Ear No.	Purple PCR + PC	Red RC	Particolored PR + P	Non-Purple + some P
(18 x 58)–1	191	56	42	138

TABLE 23A.

F_3 SEEDS OF EAR (18 x 58)–1 OF TABLE 23.

Purple Starchy Seeds Planted.

Ear No.	Purple	Red	* Parti-colored	Non-Purple
(18 x 58)–1–1	167	49	. .	84
(“)–1–2	18	4	. .	. .
(“)–1–3	41	13	. .	. .
(“)–1–6	211	72	. .	92
(“)–1–8	221	. .	66	96
(“)–1–8a	138	65	. .	83
(“)–1–11	80	. .	. .	. .
(“)–1–12	66	17	12	17
(“)–1–1s	240	. .	. .	78
(“)–1–2s	141	48	72	65
(“)–1–3s	121	38	. .	113
(“)–1–4s	93	21	15	60

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s Planted splashed purples.

* Particolored classification is only approximate.

TABLE 23B.

F_3 SEEDS OF EAR (18 x 58)–1 OF TABLE 23.

Purple Non-Starchy Seeds Planted.

Ear No.	Purple	Red	* Parti-colored	Non-Purple
(18 x 58)–1–2	49	25	. .	26
(“)–1–3	68	20	5	66
(“)–1–4	183	61	. .	73
(“)–1–5	240	. .	61	. .
(“)–1–6	22	7	. .	8
(“)–1–7	207	. .	. .	147
(“)–1–8	184	. .	24	140
(“)–1–9	360	. .	. .	. .
(“)–1–10	186	. .	68	. .
(“)–1–11	99	22	41	. .
(“)–1–12	84	. .	. .	34

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* Particolored classification is only approximate.

TABLE 23C.

F_3 SEEDS OF EAR (18 x 58)–1 OF TABLE 23.

Red Starchy Seeds Planted.

Ear No.	Purple	Red	Non-Purple	Ratio Approx.
(18 x 58)–1–1	...	222	162	9 : 7
(")–1–2	...	350	...	Pure
(")–1–3	...	222	80	3 : 1
(")–1–5	...	212	171	9 : 7
(")–1–6	...	195	115	9 : 7
(")–1–7	...	148	74	3 : 1
(")–1–12	...	187	102	9 : 7
(")–1–2s	...	300	...	Pure
(")–1–3s	...	350	...	Pure
(")–1–4s	...	276	...	Pure
(")–1–6s	...	209	63	3 : 1
(")–1–7s	...	44	35	9 : 7
(")–1–9s	...	237	141	9 : 7
(")–1–10s	...	361	...	Pure
(")–1–11s	...	206	61	3 : 1

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s Red sugar (s) seeds planted.

Family (7 x 54)

This cross introduces a combination of yellow endosperm and a dent character, the Leaming parent having long dented yellow seeds usually with formulæ $Y_1Y_1Y_2Y_2$. The current effect of the cross gave purple seeds some of which were splashed as in the previous case where yellow flint was the non-purple parent. There was nothing of special interest in the F_2 generation as the ears segregated purple and non-purple seeds in di-hybrid ratios without the appearance of particolored (cP) seeds. The characters in which these parent varieties differed segregated absolutely independently of each other.

Family (17 x 54)

The yellow flint which is the non-purple parent in this family is similar to No. 15. The ear is shorter, however, and has present a red pericarp color described under pericarp color as R_4. The F_1 seeds were purple. They were sometimes splashed purples but more rarely than in the other crosses. The F_2 seeds gave a simple mono-hybrid ratio but they were not followed into further generations. The plant of No. 17 used as the parent was homozygous therefore for either C. or P.

No. 54. Black Mexican sugar and No. 60 Tom Thumb pop above. Below F_1 ears with F_2 seeds. At left ear from the family without factor inhibiting the formation of color in aleurone cells. Other ears contain this inhibiting factor (heterozygous in mother plants).

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Family (19 x 54)

No. 19 the female parent of this cross is a large sugar corn comparable in size with the large dent varieties. The F_1 seeds were deep purple and the F_2 seeds segregated in ratios exceedingly close to the theoretic number for mono-hybrids.

Family (60 x 54)

No. 60 is a dwarf pop maize with a yellow endosperm, known as Tom Thumb. The individuals used as parents in the various crosses were pure Tom Thumbs but it is not certain that they were the product of a single selfed ear. They were grown from an ear which was self-pollinated, but because the silks appear in this variety while the young ear is entirely hidden in the axil of the leaf, it is less certain that foreign pollen was excluded from the bagged ears than it is in the case of our other crosses. The bags were slipped down into the leaf axil as firmly as possible but there was still some chance for cross pollination. This chance existed only among plants of the same variety, however, for no other pollen was mature at the same time. As several of these crosses were made upon different plants of variety No. 60 it is not strange therefore that one or two of the crosses acted as if parents with different gametic formulæ had been used. It does not follow that this was the case, for the Tom Thumb or the Black Mexican or both might have been heterozygous in some non-visible character.

The result of the immediate cross was different from our other crosses in which the purple aleurone cells were concerned; some of the seeds were dark purple, some were varying shades of light purple and some were white (i. e. non-purple). The behavior of the purple and non-purple hybrid seeds in the next generation showed conclusively that we were dealing neither with a reversal of dominance nor with a character in which the female gametes segregated normally and the male gametes abnormally as suggested by Correns, but with an entirely new dominant factor in which the Tom Thumb variety was probably heterozygous. This factor we take to be an actual inhibiting factor similar in action to the dominant white found in poultry. It is also analogous to the latter in that it does not always completely inhibit the development of color, in which case light purples similar in appearance to the particoloreds of earlier crosses develop. They are not like the particoloreds of family (8 x 54), however, for in the cross under consideration seeds with the gametic formula cP do not develop color. The light purples behave as if the inhibiting factor could vary zygotically so that in some cases light purples are developed while in others the color is completely inhibited, and also as if various amounts of color are developed in the presence of the inhibiting factor due to different combinations of other gametes. For example, it seems probable that more color may be developed in the presence of the inhibiting factor when the zygote is homozygous for the purple factor than when it is heterozygous. Further it seems less likely that any purple color develops when the inhibiting factor is homozygous than when it is heterozygous. This makes the segregating seeds of F_1 or F_2 ears very difficult to classify visually. The only accurate determination of the gametic structure of a seed is through its own further breeding.

Fifteen ears of Tom Thumb were crossed with the purple sugar corn, but only five crosses were selected from which to grow the F_2 generation. One of these, No. 60-5 x 54, had dark purple and non-purple seeds, while the other four crosses had only non-purple or very light purple seeds. It was a little unfortunate perhaps that this selection was made. The white seeds in cross 60-5 x 54 proved to be selfed Tom Thumbs, and the behavior of the purple hybrids showed that no inhibiting factor had been present in ear 60-5. The behavior of the crosses made on ears 60-2, 60-3, 60-8 and 60-11 showed that they had been homozygous in the inhibiting factor. No doubt a number of the other crosses would have shown that the maternal plants were heterozygous in the inhibiting factor.

For these reasons the data from cross 60-5 x 54, which may be called the purple family (without the inhibiting factor), have been listed in Table 24, while the other four crosses containing the inhibiting factor are shown in Table 25.

The resulting F_2 seeds obtained by selfing the purple F_1 seeds of cross 60-5 x 54 shown in Table 24 were purples, reds and non-purples. No light purples (particoloreds) appeared in this family. Splashed purples occurred as in other families, but as in other families all splashed purples were heterozygotes and not all heterozygotes were splashed purples, showing the phenomenon to be due — as before — to incomplete dominance *caused by other factors.* The only peculiar thing about this cross was the appearance and inheritance of the red color. The extracted reds bred true apparently and were hypostatic to purple as in other families, but they were purplish red (dark magenta) in appearance and not clear reds such as appear in other crosses. It is conceivable that this red is not the same red that appeared in the other crosses. It may be caused by something similar to what Wheldale (: 10) has suggested may occur in stocks; viz. that the blue oxygenase may act in conjunction with the red peroxydase or vice versa. The only difficulty in alining the results obtained with the ordinary behavior of the known factors, is the fact that almost none of the ears of the F_3 generation show the same ratios as the F_2 generation.

The ratio obtained in this generation, 1843 purples: 188 reds: 545 non-purples immediately suggests 12 : 1 : 3, which could be obtained from F_1 seeds with a formula Pp Xx CC RR where X is an inhibiting factor from the Tom Thumb which affects R but not P. Our failure to obtain ears in F_3 with segregates of 9 purple; 3 red; 1 non-purple caused this hypothesis to be discarded. The same ratio could be obtained by supposing that there is a partial gametic coupling between P and R similar to that obtained by Bateson and Punnet (: 08) between purple color and long pollen in the sweet pea. These authors suppose gametes to be produced after the general formula 7 AB: 1 Ab: 1 aB: 7 ab, from which result zygotes $3n^2$-(2n-1) AB: 2n-1 Ab: 2n-1 aB: n^2-(2n-1)ab. Such an interpretation, while it may represent Bateson's and Punnett's facts, throws no light on the mechanics of heredity for there is no reasonable way known at present for such a segregation to come about. In our own case * no such excess of purples was obtained in the F_3 generation. It seems better therefore to consider the results of the F_2 generation in the light of the breeding records of the F_3 generation. If this is done, the following interpretation fits the facts best. Tom Thumb, the female parent has the gametic formula pcR, and Black Mexican the male parent has

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* Bateson and Punnett have never reported their F_3 generation of sweet peas, although they state that it gave *conflicting results.*

the formula PCR. The F_1 generation is therefore PpCcRR. If this is the true formula there is the following theoretical expectation in the F_2 and F_3 generations:

	F_2	gives in F_3
9 Purple	1 P P C C R R	Pure Purple.
	2 P p C C R R	3 Purple : 1 Red.
	2 P P C c R R	3 Purple : 1 Non-purple.
	4 P p C C R R	9 Purple : 3 Red : 4 Non-purple.
3 Red	1 C C R R	Pure Red.
	2 C c R R	3 Red : 1 Non-purple.
4 Non-purple	1 P P R R	Pure Non-purple.
	2 P p R R	Pure Non-purple.
	1 p p c c R R	Pure Non-purple.

An examination of the F_3 segregates given in Tables 24a-e show how nearly the experimental results accord with the theory. First notice that out of 55 ears obtained by selfing purples of the F_2 generation, 20 segregated purples and non-purples without reds. This is more than our own theory calls for (theoretically 12 out of 55), so that here is clear evidence that we do not have to deal with partial gametic coupling of the kind described by Bateson and Punnett. But, since the deficiency of reds in the F_2 generation is too great to be due to chance and since there is a certain excess of purples in the F_3 generation, we must say frankly that we are dealing with something that we cannot yet explain.

The entire data from the 55 purple F_2 seeds from which selfed ears were obtained may be classified as follows: 8 ears pure purple; 20 ears segregating purples and non-purples in the ratio of 3 : 1; 9 ears segregating purples and reds in the ratio of 3 : 1; 1 ear each segregating purples and reds in ratios of 5 : 1, 6 : 1 and 12 : 1; 11 ears segregating purples, reds and non-purples in the ratio of 9 : 3 : 4; 3 ears segregating purples, reds and non-purples in the ratio of 48 : 3 : 13 or thereabouts.

From the red F_2 seeds 13 selfed ears were obtained. Out of these, 3 were pure red and 10 segregated reds and non-reds in about the ratio of 3 : 1. It should be remarked, however, that in three cases the heterozygous reds gave a greater excess of reds than usually should be expected with chance mating.

From the non-purple F_2 seeds 16 selfed ears were obtained: 15 were pure non-purple while one gave 12 purples to 49 non-purples. As this is a poor ear, the 12 seeds may be due to foreign pollen, or a chance pollen grain possessing the inhibiting factor may have produced the F_2 white seed from which the ear resulted.

With the plants resulting from non-purple F_2 seeds random intercrosses were also made. Of these 13 gave ears with all non-purple seeds and one gave an ear with 49 purples and reds and 140 non-purples.

These results generally follow our theory pretty closely, but there are abnormalities difficult to explain. We seem to be dealing with only two heterozygous factors — since 8 pure purples are obtained from 55 ears — yet tri-hybrids and tetrahybrids *are possible* which give such results. By our theory no whites should give purples when crossed at random. One such ear occurred. Was it an error? It is difficult to say. But if we were dealing with heterozygous red (Rr) we should expect more than one ear out of 14 to give purples on random crossing. Furthermore, it can be seen by inspection that there are many reasons why we cannot be dealing with simply a heterozygous red factor. It is not denied however that several other unknown factors *with* a heterozygous red factor might interpret the facts. It does not seem possible to explain the results by any reasonable system of gametic coupling or by selective mating. P and C certainly are present in an heterozygous condition. R is probably homozygous although it was not found in the Black Mexican in other crosses. But this is not peculiar since the Black Mexican used in the cross can only be said to be pure for purple. On the other hand, the red does not appear to the eye to be exactly the same red which appeared in the other crosses. It is more purplish in color, as if it were a modified purple. Nevertheless it always bred true after extraction.

Let us now turn to what may be called the white side of this family. As was stated before ears 60-2, 60-3, 60-8 and 60-11 gave no dark purples when crossed with No. 54. (Some seeds were afterwards found to be very slightly purple.) One may conclude therefore that they (the maternal plants) were either homozygous for a factor that inhibits the development of the purple color; or, that there is a reversal of dominance, which is improbable. There were other ears that gave both purple and non-purple seeds in crosses. These were either heterozygous for an inhibiting factor or exhibited dominance of both purple and non-purple on the same ear which is still more improbable. None of these ears were followed into the F_2 generation. but progeny of all four of the ears of the first type were grown.

The results of the F_2 generation from these ears are shown in Table 25. There is no reason why some of these families might not differ from others in invisible factors, for different plants of No. 60 were crossed with pollen from different plants of No. 54. They are placed in one table here but certain differences in their behavior in F_3 leads us to consider them separately. There is a total of 662 purples, 94 reds and 2838 light colored purples and non-purples. The reason for classing the light purples and non-purples together will be seen later.

The results of the F_3 generation as well as our experience with other crosses are such as exclude the possibility of a reversal of dominance. The purples did not breed true nor did the behavior of any of the classes indicate anything other than a normal Mendelian segregation involving several characters. Furthermore, a belief in reversal of dominance in our opinion strikes at the foundation stone of Mendelism. Not that dominance is an important part of Mendelism. It is not. Yet no analysis can be made of breeding records without following *every* individual for several generations if dominance is reversible. Of the thousands of extracted recessives that have bred true, many would have proved to be heterozygous dominants if dominance is reversible.

Taking the same F_1 gametic formula that served for the purple side of the family and adding an inhibiting factor I which comes from No. 60, gives the best interpretation of the data. This makes the F_1 formula PpCcIiRR. In F_2 the following classes would be expected:

27 P I C R	Color non-purple.
9 P I R	“ non-purple.
9 P C R	“ purple.
9 I C R	“ non-purple.
3 P R	“ non-purple.
3 I R	“ non-purple.
3 C R	“ red.
1 p c i r	“ non-purple.

The ratio is 9 purple : 3 red : 52 non-purple. The experimental results given in Table 25 show that here also there is a deficiency of reds. Many light purples also appeared, but these were classed as non-purples. This was done because in F_3 the light purples behaved as if they possessed the factor I in a heterozygous condition, the variation in color being due to the different combinations in which the factors P and C appeared.

With this theory the expectation in F_3 is 52 non-purples and light purples giving:

28 Producing all Non-purple seeds.
2 " 1 Purple : 3 Non-purple.
4 " 3 Purple : 13 Non-purple.
4 " 3 Purple : 1 Red : 12 Non-purple.
8 " 9 Purple : 3 Red : 52 Non-purple.
2 " 1 Red : 3 Non-red.
4 " 3 Red : 13 Non-red.

9 Purples giving:
1 Producing all Purple seeds.
2 " 3 Purple : 1 Red.
2 " 3 Purple : 1 Non-purple.
4 " 9 Purple : 3 Red : 4 Non-purple.

3 Reds giving:
1 Producing all Red seeds.
2 " 3 Reds : 1 Non-red.

Let us now examine Tables 25a-e which give the results from selfing the seeds of certain of the F_2 ears. Table 25a shows the progeny of ear (60-3x54)-1. This ear gave the smallest proportion of purple seeds in F_2, and such purples as were produced in F_2 were lighter in color than normal full purples. In F_3 the purples are again light in color. They are classed in with the non-purples in the last column, those showing some color being given first. The first two ears are progeny of the darkest purples; one has purple and non-purple seeds in the ratio of 3 : 1 and one is pure purple. The next two ears planted from lighter purples show a difference between themselves. One gives 3 light purples : 1 non-purple, the other gives 1 purple : 2 non-purple. The latter probably came from an ear heterozygous for the inhibiting factor and the former from a real purple. Of those ears resulting from white seeds, one has 33 red seeds dark enough to be classed as real reds and a number of very light reds classed with the non-purples — a total of probably near 25% reds, while another gives light purples (and possibly light reds) and non-purples. The remaining ears are non-purples. Five plants from non-purple F_2 seeds were also crossed, and gave all non-purple seeds. In reality, however, only two random crosses can be said to have been made, since the pollen of No. (60-3 x 54)-1-2 ES was used three times, while once the same plant was used as the mother. The progeny of ear (60-3 x 54)-1, therefore, behave like those of other ears of this family except that all of the progeny of purples are light in color. They give pure purples and purples segregating into 3 purples : 1 non-purple, but none are dark like normal purple ears. Some geneticists would probably interpret this as prepotency of the non-purple or rather lack of prepotency of the purple. But when one talks of prepotency he really confesses ignorance of the gametic constitution of his cultures. Is it not much more likely that the true reason for the production of these light purples lies in a fact more in keeping with what is known of hereditary phenomena? May not one say that here is a dominant purple character coming from the individual of unknown character of variety No. 54 which was used as the male parent? If the purple gene from the male parent was such as to give always a lighter purple in zygotic combinations where purple is visible then no dark purples would occur in the segregates resulting from crosees. Such results were obtained from four selfed plants. Two ears resulted from planting purples which were only slightly lighter than normal dark purples, such as the parents of ears (60-3 x 54)-1-1 and (60-3 x 54)-1-2, and two ears resulted from planting seeds quite light in color. (Table 25a).

Similar results were obtained from cross (60-8 x 54), of which the F_3 generation from ear (60-8 x 54)-8 are shown in Table 25e. Here eleven ears resulted from selfing seeds with the modified color if two red seeds are included. None of these ears had seeds dark in color, but the ratios are no doubt the same as those given in Tables 25 b-d. The general reduction of the amount of purple color, however, makes the error of classification too great for safe conclusions. There is even *some* doubt about the classification of the seeds from the F_2 generation of these two crosses (Table 25), but the results of the F_3 generation are such as to give us considerable faith in them.

Tables 25 b-d give a considerable number of F_3 progeny from F_2 seeds of three other ears. There seems to be no reason why they should not be considered together. From the purple F_2 seeds planted, twelve selfed ears were obtained. Three ears were pure purples of the normal shade. One ear gave a ratio of 3 purples : 1 red and two ears a ratio of 3 purples : 1 non-purple. The other six ears gave purple, red and non-purple segregates. Four of these ears were clearly of the ratio 9 : 3 : 4, but in the remaining two there was a considerable deficiency of red seeds. From the F_2 red seeds planted, only one selfed ear was obtained. This ear gave red and non-red segregates in the ratio of 3 : 1.

A large number of selfed ears were obtained from the F_2 light purple and non-purple seeds. Ears of each of the classes expected by the proposed theory were obtained, as will be seen by an examination of the Tables 25 b-d; but as the visual classification is arbitrary owing to the light color of most of the seeds, it could not be depended upon without further breeding. The light colored seeds are given first in the last column of the tables, followed by the seeds which were apparently non-purple. If one is a little charitable about the exactness of the classification the following conclusions can be drawn.

Both seeds which were apparently non-purple and seeds which were light purple in color in the F_2 generation gave light purple seeds among the F_3 segregates. This fact proves both the impossibility of exact classification and the gametic identity of seeds slightly different in their appearance.

Two plants from light red seeds (Table 25b) were selfed. One resultant ear showed a ratio of 1 dark red : 3 light red and non-red; the other ear showed only light red and non-red seeds which were classed together. Thirty-six plants from light purple and from non-purple F_2 seeds were selfed. Of these, fifteen ears resulted from planting seeds classified as non-purple in F_2. Only four of them threw dark purple segregates in F_2. On the other hand only two of the ears resulting from selfed plants which were progeny of seeds classified as light purples, threw no dark purple segregates. It seems to us that this shows a fair classification of seeds heterozygous for the inhibiting factor. A few seeds, however, were wrongly classified in the F_2 generation and proved their proper status in the F_3 generation.

Out of the total of 36 selfed ears from light purple and non-purple F_2 seeds, 23 threw dark purple segregates and 13 produced only light purple and non-purple seeds. Of those ears which threw purple segregates, none of them had ratios of purple to light purple plus non-purple greater than might reasonably be expected by chance mating. The different ratios expected in F_3 were followed rather well, although it is recognized that these ratios could not be determined accurately with such small numbers.

Conclusions.

There can be but little doubt that the factors I, C, P and R are concerned in this cross. Whether there is another factor which modifies the purple color or not, is a question that cannot yet be settled, because we have no data concerning the individual plant of No. 54 that formed the male parent; yet there seems to be no other way to account for the light purples in Table 25a and Table 25e. The ultimate analysis of the behavior of the R factor in this cross must also be left in abeyance. These unsettled questions however have no bearing upon two important conclusions which the evidence forces upon us. The first is that one should be exceedingly careful before he decides that the transmission of certain characters is an exception to the general law of Mendel. When a collection of white or non-purple aleurone strains are promiscuously crossed with a purple aleurone maize, the results seem almost impossible to bring into conformity with simple Mendelian results, yet this confusion is brought about simply by the gametic differences of the non-purple races. If such confusion can result in the case of a simple color inheritance, much more care must be taken to analyze the transmission of more complex characters before subsidiary hypotheses are submitted.

The second important fact is in regard to prepotency. It has been shown that certain families of purple and non-purple hybrids produce very light purples when P exists alone without C, while other families produce no color. No modification of the Mendelian ratio occurs, yet some transmissible difference in the two families gives this different result. Here is a probable explanation of prepotency. If these white families were mixed together, a mixture more easily imagined in the case of bisexual individuals, there would appear to be a difference in prepotency of the purple character. It therefore seems probable that prepotency is due only to a difference in gametic character which modifies somatic appearances and not to an actual modification of Mendelian chance ratios as others have suggested.

The behavior of the other families is so simple that we think there can now be no question but that the purple aleurone color behaves as a normal Mendelian character in inheritance.

TABLE 24.

F₂ SEEDS OF CROSS BETWEEN NO. 60–5 NON-PURPLE POP AND NO. 54 PURPLE SWEET.

Purple Seeds Planted.

Ear No.	Purple	Red	Non-Purple
(60–5 x 54)–2	271	28	57
(")–3	236	21	71
(")–4	92	11	33
(")–5	203	36	69
(")–6	272	33	71
(")–8	144	14	58
(")–10	198	21	55
(")–11	190	4	55
(")–12	237	20	76
Total	1843	188	545

TABLE 24A.

F₃ SEEDS OF EAR NO. (60–5 X 54)–2 OF TABLE 24.

Ear No.	Planted from	Purple	Red	Non-Purple
(60–5 x 54)–2–1	Purple S	277	23	..
(")–2–2	" S	16	..	5
(")–2–3	" S	176	45	69
(")–2–4	" S	233	48	92
(")–2–1	" s	209	49	72
(")–2–4	" s	396	..	..
(")–2–1	Red S	...	219	91
(")–2–5	" S	...	All	..
(")–2–1	L. Purple S	194	27	71
(")–2–2	" S	167	56	80
(")–2–1	Non-Pur. S	...	..	Pure
(")–2–2	" S	...	..	"
(")–2–3	" S	...	..	"
(")–2–4	" S	...	..	"
(")–2–2 BS x 2–1	Red x Red S	...	380	..
(")–2–3 AO x 2–2	Pur. x Pur. S	280	..	98
(")–2–3 CS x 2–1	L. Pur. x L. Pur. S	152	37	54
(")–2–5 ES x 2–4	Non-Pur. x Non-Pur. S	...	..	Pure
(")–2–2 EO x 2–1	Non-Pur. x Non-Pur. s	...	..	"

TABLE 24B.

F_3 SEEDS OF EAR NO. (60–5 x 54)–6 OF TABLE 24.

Ear No.	Planted from	Purple	Red	Non-Purple
(60–5 x 54)–6–3	Purple S	87	24	38
(“)–6–4	“ S	204	. .	60
(“)–6–6	“ S	262	. .	80
(“)–6–7	“ S	318	58	. .
(“)–6–4	“ s	265	83	. .
(“)–6–1	Red S	. . .	135	29
(“)–6–2	“ S	. . .	287	76
(“)–6–3	“ S	. . .	164	48
(“)–6–1	“ s	. . .	240	71
(“)–6–1	Non-Pur. S	. . .	. .	Pure
(“)–6–2 AS x 4	Pur. x Pur. S	384	. .	. .
(“)–6–5 AS x 7	“ S	420	. .	. .
(“)–6–2 AO x 3	“ s	200	. .	. .
(“)–6–2 ES x 1	Non-Pur. x Non-Pur. S	. . .	. .	Pure
(“)–6–4 ES x 5	“	. . .	. .	“

TABLE 24C.

F_3 SEEDS OF EAR NO. (60–5 x 54)–8 OF TABLE 24.

Ear No.	Planted from	Purple	Red	Non-Purple
(60–5 x 54)–8–1	Purple S	125	. .	. .
(“)–8–2	“ S	176	55	. .
(“)–8–3	“ S	212	60	. .
(“)–8–4	“ S	170	. .	. .
(“)–8–6	“ S	183	. .	60
(“)–8–1	“ s	180	28	65
(“)–8–2	“ s	182	35	. .
(“)–8–5	“ s	153	. .	35
(“)–8–6	“ s	217	. .	71
(“)–8–7	“ s	150	. .	. .
(“)–8–1	Red S	. . .	176	34
(“)–8–2	“ S	. . .	182	. .
(“)–8–2	“ s	. . .	156	57
(“)–8–1	Non-Pur. S	. . .	. .	180
(“)–8–4	“ S	12	. .	40
(“)–8–5	“ S	. . .	. .	250
(“)–8–1	“ s	. . .	. .	250
(“)–8–2	“ s	. . .	. .	220
(“)–8–3	“ s	. . .	. .	240
(60–5 x 54)–8–2 ES x4	Non-Pur. x Non-Pur. x	. . .	. .	400
(“)–8–7 ES x6	Non-Pur. x Non-Pur. x	49	. .	140
(“)–8–4 EO x 5	Non-Pur. x Non-Pur. x	. . .	. .	220

TABLE 24D.

F₃ SEEDS OF EAR NO. (60–5 x 54)–11 OF TABLE 24.

Ear No.	Planted from	Purple	Red	Non-Purple
(60–5 x 54)–11–3	Purple S	115	42	47
(“)–11–4	“ S	175	..	49
(“)–11–5	“ S	175	65	..
(“)–11–6	“ S	180	..	62
(“)–11–8	“ S	209	69	..
(“)–11–10	“ S	210	..	67
(“)–11–11	“ S	101	..	38
(“)–11–12	“ S	112	..	39
(“)–11–2	“ s	46	..	21
(“)–11–3	“ s	144	..	46
(“)–11–4	“ s	178	..	64
(“)–11–7	“ s	180	..	..
(*)–11–1	L. Purple S	116	25	49
(“)–11–3	“ S	204	41	75
(“)–11–5	“ S	124	..	52
(“)–11–1	“ s	218	29	75
(“)–11–2	L. Red S	...	163	38
(“)–11–1	Non-Pur. S	...	..	Pure
(“)–11–2	“ S	...	..	“
(“)–11–5	“ S	...	..	“
(“)–11–7	“ S	...	..	“
(“)–11–1	“ s	...	..	“
(“)–11–2	“ s	...	..	“
(60 x 54–5)–11–7 AS x 11-6	Pur. x Pur. S	142 Pur. and Red		56
(“)–11–3 ES x 11-5	Non-Pur. x Non-Pur. S	...	..	Pure
(“)–11–6 ES x 11-4	Non-Pur. x Non-Pur. S	...	..	“
(“)–11–2 EO x 11-3	“	...	..	“

TABLE 24E.

F_3 SEEDS OF EAR NO. (60–5 x 54)–12 OF TABLE 24.

Ear No.	Planted from	Purple	Red	Non-Purple
(60–5 x 54)–12–3	Purple S	206	78	80
(“)–12–4	“ S	201	44	67
(“)–12–4a	“ S	350	. .	. .
(“)–12–5	“ S	169	59	. .
(“)–12–6	“ S	245	65	. .
(“)–12–7	“ S	204	67	. .
(“)–12–8	“ S	191	49	88
(“)–12–9	“ S	187	. .	66
(“)–12–10	“ S	239	. .	66
(“)–12–13	“ S	248	. .	77
(“)–12–14	“ S	217	. .	77
(“)–12–1	“ s	240	. .	72
(“)–12–3	“ s	350	. .	. .
(“)–12–4	“ s	184	43	56
(“)–12–5	“ s	300	. .	. .
(“)–12–7	“ s	147	53	. .
(“)–12–1	Red S	. . .	229	76
(“)–12–3	“ S	. . .	280	. .
(“)–12–4	“ S	. . .	172	56
(“)–12–1	Non-Pur. S	. . .	. .	Pure
(“)–12–8	“ S	. . .	. .	“
(60–5 x 54)–12–2 AS x 3	Purple S	161	56	53
(“)–12–11 As x 6	“ S	175	. .	63
(“)–12–2 AO x 1	“ S	229	. .	74
(“)–12–3 ES x 5	Non-Pur. S	. . .	. .	Pure
(“)–12–4 ES x 5	“ S	. . .	. .	“
(“)–12–5 ES x 7	“ S	. . .	. .	“
(“)–12–7 ES x 5	“ S	. . .	. .	“
(“)–12–9 ES x 8	“ S	. . .	. .	“

TABLE 25.

F_2 SEEDS OF CROSS BETWEEN NO. 60 NON-PURPLE POP AND NO. 54 PURPLE SWEET.

Very Light Colored and White Seeds Planted.

Ear No.	Purple	Red	L. Pur. + Non-Pur.
(60–8 x 54)–1	83	5	66 + 135 = 201
(")–7	19	7	44 + 150 = 194
(")–8	35	4	41 + 215 = 256
(60–11 x 54)–2	22	4	22 + 40 = 62
(60–2 x 54)–1	68	15	96 + 159 = 255
(")–7	86	7	99 + 150 = 249
(")–10	89	14	69 + 148 = 217
(60–3 x 54)–1	26	..	76 + 282 = 358
(")–3	46	7	87 + 140 = 227
(")–5	54	12	102 + 159 = 261
(")–6	65	6	113 + 144 = 257
(")–7	69	13	117 + 184 = 301
Total	662	94	2838

TABLE 25A.

F_3 SEEDS OF EAR NO. (60–3 x 54)–1 OF TABLE 25.

Ear No.	Planted from	Pur.	Red	L. Pur. + Non-Pur.
(60–3 x 54)–1–1	Purple S	..	..	235 + 76 = 311
(")–1–2	" S	..	..	245 = 245
(")–1–1	L. Purple S	..	..	217 + 69 = 286
(")–1–2	" S	..	..	39 + 72 = 111
(")–1–5	Non-Pur. S	..	..	0 + 384 = 384
(")–1–6	" S	..	..	105 + 204 = 309
(")–1–7	" S	..	33L	380*
(")–1–9	" S	..	..	0 + 390 = 390
(")–1–10	" S	..	..	0 + 280 = 280
(")–1–1	" s	..	..	0 + 448 = 448
(")–1–2	" s	..	..	0 + 200 = 200
(")–1–3	" s	..	..	0 + 152 = 152
(")–1–4	" s	..	..	0 + 280 = 280
(60–3 x 54)–1–1 ES x 1–2	Non-Pur. x Non-Pur.	..	..	0 + 250 = 250
(")–1–2 ES x 1–1	"	..	..	0 + 258 = 258
(")–1–3 ES x 1–2	"	..	..	0 + 110 = 110
(")–1–4 ES x 1–2	"	..	..	0 + 308 = 308
(")–1–8 ES x 1–6	"	..	..	0 + 352 = 352

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* Light reds and non-reds.

TABLE 25B.

F$_3$ SEEDS OF EAR NO. (60–3 x 54)–5 OF TABLE 25.

Ear No.	Planted from	Pur.	Red	L. Colored + Non-Colored
(60–3 x 54)–5–1	Purple S	165	47	0+66 =66
(")–5–2	" S	250	. .	. . . .
(")–5–3	" S	203	62	. . . .
(")–5–4	" S	300	. .	. . . .
(")–5–6	" s	273	. .	0+ 95= 95
(")–5–1	" s	176	13	0+ 69= 69
(")–5–1	Red S	. . .	190	0+ 71= 71
(")–5–1	L. Purple S	58	. .	136+110=246
(")–5–2	" S	90	49	115+ 96=211
(")–5–2a	" S	104	. .	78+152=230
(")–5–3	" S	59	. .	91+145=236
(")–5–5	" S	80	. .	58+ 97=155
(")–5–5a	" S	79	. .	65+ 54=119
(")–5–6	" S	. . .	. .	88+ 26=114
(")–5–4	L. Red S	. . .	62	78+ 95=173
(")–5–2	" s	. . .	. .	75+ 51=126
(")–5–2	Non-Pur. S	. . .	. .	0+352=352
(")–5–4	" S	. . .	. .	0+ 30= 30
(")–5–7	" S	67	. .	138+193=331
(")–5–8	" S	. . .	. .	0+380=380
(")–5–9	" S	. . .	. .	0+345=345
(")–5–2	" s	. . .	. .	0+390=390
(60–3 x 54)–5–1 ES x 5–7	Non-Pur. x Non-Pur.	. . .	. .	125+313=438
(")–5–2 ES x 5–4	"	47	. .	14+126=140
(")–5–5 ES x 5–4	"	. . .	. .	0+320=320
(")–5–10 ES x 5–9	"	. . .	. .	90+154=244
(")–5–11 ES x 5–9	"	. . .	. .	43+ 87=130
(")–5–17 ES x 5–7	"	. . .	. .	109+144=253
(")–5–1 ES x 5–5	"	. . .	. .	0+360=360
(")–5–3 ES x 5–4	"	. . .	. .	0+104=104
(")–5–4 ES x 5–3	"	. . .	. .	0+254=254
(")–5–7 ES x 5–5	"	. . .	. .	0+400=400

TABLE 25C.

F_3 SEEDS OF EAR NO. (60–3 x 54)–6 OF TABLE 25.

Ear No.	Planted from	Pur.	Red	L. Colored + Non-Colored
(60–3 x 54)–6–2	Purple S	180	. .	0 + 71 = 71
(“)–6–2a	“ S	175	9	0 + 64 = 64
(“)–6–5	“ S	138	20	0 + 40 = 40
(“)–6–1	L. Purple S	31	5	36 + 38 = 74
(“)–6–1a	“ S	49	. .	68 + 49 = 117
(“)–6–3	“ S	27	. .	53 + 48 = 101
(“)–6–4	“ S	66	13	63 + 61 = 124
(“)–6–6	“ S	36	. .	144 + 130 = 274
(“)–6–1	“ s	90	. .	108 + 142 = 250
(“)–6–1	Non-Pur. S	. .	. .	0 + 250 = 250
(“)–6–3	“ S	25	. .	70 + 106 = 176
(“)–6–5	“ s	21	3	45 + 62 = 107
(“)–6–6	“ s	. .	. .	0 + 152 = 152
(“)–6–7	“ s	69	. .	53 + 105 = 158
(60–3 x 54)–6–3 AS x 6–2	Pur. x Pur.	51	. .	88 + 80 = 168
(“)–6–1 AO x 6–2	“	132	57	. . . .
(“)–6–2 ES x 6–3	Non-Pur. x Non-Pur.	14	. .	15 + 31 = 46
(“)–6–3 ES x 6–5	“	. .	. .	0 + 380 = 380
(“)–6–5 ES x 6–6	“	. .	4	21 + 117 = 138
(“)–6–6 ES x 6–5	“	. .	17	78 + 122 = 200
(“)–6–7 ES x 6–3	“	. .	. .	37 + 170 = 207
(“)–6–8 ES x 6–6	“	. .	4	65 + 61 = 126
(“)–6–2 EO x 6–1	“	. .	. .	84 + 222 = 306
(“)–6–4 EO x 6–3	“	. .	. .	33 + 26 = 59
(“)–6–3 EO x 6–4	“	80	25	24 + 171 = 195

PLATE XI.

Cross 60-5x54. F_3 generation not including red segregates. The combinations possible when parents differ in presence and absence of purple aleurone cells and presence and absence of starchiness. First three resulted from planting non-purple, the remainder from purple seeds. Ears 1, 4, 5 resulted from planting non-starchy, the remainder from starchy seeds.

INHERITANCE OF ALEURONE COLOR.

TABLE 25D.

F_3 SEEDS OF EAR NO. (60–8 x 54)–1 OF TABLE 25.

Ear No.	Planted from	Pur.	Red	L. Colored + Non-Colored
(60–8 x 54)–1–6	Purple S	135	36	0+ 56= 56
(")–1–7	" S	67	16	0+ 20= 20
(")–1–3	" s	280	. .	. . . .
(")–1–1	L. Purple S	20	. .	66+ 76=142
(")–1–1a	" S	51	17	63+164=227
(")–1–2	" S	35	1	31+ 84=115
(")–1–3	" S	40	20	67+124=291
(")–1–5	" S	20	16	27+ 21= 48
(")–1–1	" s	74	. .	101+115=216
(")–1–1	Very L. Pur. S	. .	. .	92+121=213
(")–1–2	" S	67	5	83+109=192
(")–1–4	Non-Pur. S	. .	. .	0+250=250
(")–1–5	" s	. .	. .	0+250=250
(")–1–6	" s	. .	. .	0+230=230
(")–1–7	" s	. .	. .	0+240=240
(60–8 x 54)–1–6 ES x 1–7	Non-Pur. x Non-Pur. S	. .	. .	0+100=100
(")–1–7 ES x 1–6	"	. .	. .	0+250=250
(")–1–8 ES x 1–9	"	. .	. .	43+ 66=109
(")–1–9 ES x 1–8	"	. .	. .	151+158=309
(")–1–10 ES x 1–9	"	. .	68	0+161=161
(")–1–1 EO x 1–2	Non-Pur. x Non-Pur. s	. .	. .	0+240=240
(")–1–3 EO x 1–7	"	. .	. .	½*+½ =1

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* Approximated.

TABLE 25E.

F_3 SEEDS OF EAR NO. (60–8 x 54)–8 OF TABLE 25.

Ear No.	Planted from	Pur.	Red	L. Colored + Non-Colored
(60–8 x 54)–8–2	Purple S	. .	†54L	236 + 100 = 336
(")–8–5	" S	. .	. .	212 + 0 = 212
(")–8–1	Red S	. .	200L	. . . .
(")–8–2	" S	. .	5L	26 + 8 = 34
(")–8–1	L. Purple S	. .	. .	110 + 104 = 214
(")–8–2	" S	. .	. .	224 + 123 = 347
(")–8–5	" S	. .	. .	50 + 148 = 198
(")–8–7	" S	. .	62L	0 + 248 = 248
(")–8–8	" S	70L	. .	84 + 164 = 248
(")–8–1	" s	. .	. .	90 + 202 = 292
(")–8–1 lower ear	" s	. .	. .	97 + 243 = 340
(")–8–3	Non-Pur. S	. .	. .	80 + 294 = 374
(")–8–9	" S	. .	. .	80 + 216 = 296
(")–8–2	" s	. .	37L	0 + 263 = 263
(60–8 x 54)–8–1 ES x 8–7	Non-Pur. x Non-Pur. S	. .	. .	0 + 300 = 300
(")–8–2 ES x 8–3	" S	. .	. .	39 + 260 = 299
(")–8–4 ES x 8–9	" S	. .	. .	49 + 214 = 263
(")–8–5 ES x 8–17	" S	. .	. .	0 + 200 = 200
(")–8–6 ES x 8–9	" S	. .	. .	0 + 300 = 300
(")–8–7 ES x 8–17	" S	. .	. .	24 + 270 = 294
(")–8–1 EO x 8–2	" s	. .	. .	88 + 174 = 262
(")–8–5 EO x 8–3	" s	. .	. .	48* + 153 = 201

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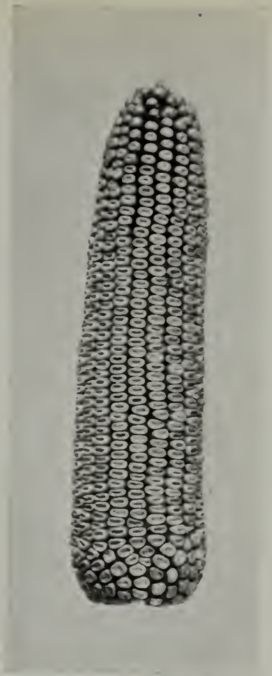
* Several seeds rather dark purple.

† Those marked L are light in color but not nearly as light as those given in the last column.

PLATE XII.

a. No. 21. Podded maize.



b. No. 7, non-podded maize.

c. Cross 21x7. F_1 above; pod character fully dominant. F2 below; complete segregation in monohybrid ratio.

Inheritance of "Podded" Character.

PART III.

XENIA.

The appearance of the endosperm in the F_1 generation in the crosses discussed in Part II really include almost all of our observations of true Xenia, but the subject is sufficiently important to warrant a more systematic arrangement of the facts.

The word Xenia was proposed by Focke to denote the effect, if any, produced by the action of pollen upon the maternal tissue of the seed plant. The classical example of such effect was the endosperm of maize. After the discovery by Guignard ('99) and Nawaschin ('99) that the endosperm is in reality a part of the filial generation formed by the development of the endosperm nucleus after fusion with the second male nucleus of the pollen cell, De Vries ('99), Correns ('99) and Webber (: 00) saw in this the explanation of the phenomenon in maize. These facts took away the only authentic illustration of Xenia in its original use — the effect of foreign pollen on maternal tissue. In this older sense the word is therefore of no value, and it may be used solely to describe the visible effect of the second male nucleus on the endosperm. Unfortunately, botanists have not been so prompt in discarding belief in the original meaning of Xenia as the zoologists in discarding telegony. In the experience of Correns, of Lock and of ourselves the effect of the second male nucleus has never extended to maternal tissue. One of the present authors has made several experiments in which pollination without fertilization (between infertile species) has had an effect on maternal tissue, (parthenocarpie), but this effect was simply that of a chemical stimulus or irritant producing cell division in the carpels.

The visible effects of double fertilization have been found in the following cases, in all of which the parents have been selfed strains that precluded errors in the observations. Non-starchy seeded plants crossed with starchy seeded plants always show starchiness. Starchiness is completely dominant, therefore the reciprocal cross, bringing in the "opposing" character, never shows Xenia.

Yellow endosperm is also completely dominant in most cases. Non-yellow crossed with yellow endosperm therefore shows Xenia while the reciprocal shows no Xenia. Three exceptions to this rule were found, however. In the large races of dent maize where the zone of soft starch at the summit of the seed is extensive, the heterozygous yellow is somewhat lighter in color than the homozygous yellow, and Xenia appears when the cross is made either way. It shows as a cap of lighter color than the homozygous yellow. When floury yellow races are crossed with floury white races this lighter color of the heterozygote extends throughout the seed. In this case difference in color is always great enough to be noticed by a careful observer in either cross, but where the cap only is floury the color intergrades to that of the homozygous yellow. When dealing with races with corneous endosperms, such as the flint and pop varieties, there is so little difference in color that the homozygous yellow is generally indistinguishable from the heterozygous yellow; therefore Xenia occurs only when the white is the female parent. Even here, however, we have found two different cases where a few heterozygous yellows were distinguishable from homozygous yellows when the latter were used as the female.

Both the red and the purple colors in the aleurone cells behave in the same way as regards Xenia. When the two parents differ only in these characters, they are completely dominant and Xenia occurs only when they are possessed by the male parent. Even in the race in which a slight purple color appeared when the color factor was absent (P c instead of P C) the same slight color appeared when it was used as the male upon a race in which P and C were both absent. Furthermore when this race was crossed either way with a white race bearing the color factor (P c x p C or p C x P c), the full purple developed and appeared as Xenia. The red color undoubtedly behaves in the same way although we have made no original crosses dealing with these conditions. Again, two pure white races (P c and p C) which show not the slightest color may bring together the two factors P and C necessary for full development of the purple color, and Xenia results when either is the female parent.

The next and last case in which we have observed Xenia is that in which the white parent possesses a character that inhibits the development of red or purple aleurone cells. Correns and Lock probably used races containing this character but they did not distinguish it from a recessive white or simple absence of the purple character. They therefore concluded that when a white race was crossed with a purple, Xenia sometimes results and sometimes does not result, and that no change occurs when the purple is the female parent. The true state of affairs is just the opposite of this. When a white containing the inhibitor is the male parent, a white seed results, and while the same result is obtained in the reciprocal cross it is of course unnoticed when the white is the female parent. Sometimes the purple is not fully inhibited and then a light purple results no matter which parent is the mother.

If one considers these observations as a whole, the following law regarding Xenia may be formulated:

When two races differ in a single visible endosperm character in which dominance is complete, Xenia occurs only when the dominant parent is the male; when they differ in a single visible endosperm character in which dominance is incomplete or in two characters both of which are necessary for the development of the visible difference, Xenia occurs when either is the male.

Correns observed that in every case where Xenia may be expected to occur, the seeds showing Xenia were always hybrids. This fact was assumed to prove that the second male nucleus always bears the same characters as the one that fuses with the egg cell to form the embryo. For this reason Mendelian segregation of the gametes must have occurred previous to the division of the pollen nucleus. Our observations are entirely in accord with those of Correns. The latter author and also Webber observed several cases where Xenia occurred in only one-half of the endosperm. These rare phenomena which are probably similar in nature to the gynandromorphs occurring in insects, they both interpreted as the independent development of the endosperm nucleus and the second male nucleus. We have observed many instances of this phenomenon and have grown a number of them to see if the tendency was inherited but without positive results. Correns' and Webber's explanation of the cause of these seeds is probably correct, yet the suspicion cannot be avoided that if the two nuclei can develop independently then the female nucleus ought sometimes to develop to the total exclusion of the male. If this were true a seed showing no Xenia where it is to be expected, should sometimes prove to be a hybrid. This has never occurred in our work, a fact in disagreement with the work of Webber. It may be possible then that the cause of these seeds is Mendelian segregation in somatic tissue, such as often occurs in bud sports. This could be proved if there occurred among the F_1 seeds of a cross in which the parents differed in two characters, an individual in which the characters were segregated differently: for example, if a white sweet maize were pollinated with a yellow starchy race, and a seed developed having one half yellow sweet and the other half white starchy. The matter is simply mentioned because it is important to biological theory, and it was thought that some experimentalist might happen upon such an individual.

It is thought that Webber's idea that seeds with splashed purple aleurone cells are due to mosaic development of cell descendants of the endosperm nucleus and of the second male nucleus, is incorrect. If this idea were true, in cases where the endosperm is heterozygous yellow, this character also should be mosaic. Such cases have never been reported. It therefore seems better to consider the splashed purples as cases of incomplete dominance caused by other factors as was explained in greater detail earlier in the paper.

PLATE XIII.

a. Podded maize. The four husks successively removed showing naked seed at right. The double rowed condition characteristic of all maize varieties is seen most clearly.

b. *c.*

Male spikes (tassels) showing development of seeds. *b*, a dominant F2 plant; *c*, a recessive F2 plant. Segregation is persistent in this cross, 21x7.

INHERITANCE OF "PODDED" CHARACTER.

a. At left, the color which develops in sunlight—R4; in center variegated or mosaic seeds—R2; at right, common red pericarp—R1.

b. Segregation of pericarp color R4 in F2 of cross 5x11. Amount of color developed is variable depending on light conditions during maturation.

PERICARP COLORS.

PART IV.

PLANT CHARACTERS.

In this part of the paper the inheritance of normal plant characters is considered. These characters in general have no effect upon the endosperm — the new generation — and therefore do not show as Xenia in the daughter seeds of the ear that has been crossed.

Podded and Podless Maize.

The inheritance of the podded character is interesting because it is a shining example of a case where a gross morphological character behaves as a simple Mendelian mono-hybrid. No. 21 a podded maize was crossed with a common Leaming dent like No. 7, but not of the same stock. The F_1 generation was as perfectly podded as the podded parent. There was of course some variation in the length of the husks of the seeds, a variation apparently physiological in character depending upon the vigor of the mother plant, but this variation was no greater in the F_1 generation than it was in the pure podded maize. The F_2 generation yielded 64 podded and 21 non-podded individuals. The latter were without any trace of husk and were not distinguishable from ordinary non-podded corn which had never been crossed with a podded variety. (See Plate XII.)

The F_1 generation was also crossed back with the recessive — the non-podded variety — and in the next generation yielded 41 podded ears and 50 non-podded ears. In other words Hh x h gave 50% Hh and 50% hh as was to be expected. The character was again strictly discontinuous. The extracted recessives proved absolutely true.

Pericarp Color.

There are various red sap colors appearing in the pericarp, the cob, the husks, the silks, the glumes and the anthers of maize. We have not been able to make a chemical study of them and so cannot say if they are due to the same compound, but the comparatively small amount of data regarding their inheritance that we have obtained is particularly interesting on account of the number of different organs in which color occurs. It has long been thought that such colors that manifest themselves in different parts of a plant, are single units as regards heredity, but are produced in visible quantities only when developmental conditions are favorable or when certain transmissible limiting factors or extension factors which effect their development, are present or absent. Our especial problem was to find out whether these red colors occurred and were transmitted separately or whether they were linked together in genetic or in chemical relationships. This work is therefore simply a report of progress.

The first red pericarp, which we will call R_1 was found in No. 27, a rice pop maize. It was the ordinary dark red color of the varieties commonly known as red corns. It did not have a red cob or red silks, although the glumes of the male flowers were sometimes reddish. Crossed with number 28, a rice pop with white pericarp, white cob and silks, it gave 75 red and 22 white ears in F_2. The color was inherited absolutely discontinuously, the reds being all dark and the whites showing no trace of color.

The only other cross with apparently this same dark pericarp color, was a peculiar ear found in a field of dent maize of unknown parentage. This ear, as shown in Plate XV, fig. a, had seeds with red pericarp on one side and seeds which were sometimes white and sometimes striped with red on the other side. The ear appeared in a field of white maize in which only white maize was planted. It must have been produced therefore by a hybrid seed R_1 r_1. Furthermore since it was the only ear in the field showing red pericarp, it is likely that it was nearly all pollinated by white. One would expect its seeds therefore to be half R_1r_1 and half r_1r_1, and that they would give in the next generation 50% red ears, 50% white ears. In order to test any possible transmission of the variation which appeared in this ear however, both the red seeds and the seeds from the side which had white and striped kernels were planted. From the dark red seeds were obtained 22 dark red ears and 22 white ears; from the white and striped seeds were obtained 15 ears showing a few red striped seeds and 15 ears with only white seeds. No difference was observed between the progeny of white and of striped seeds. Both kinds of seeds from this side of the ear gave striped ears and white ears. A selfed red ear of this generation gave a simple mono-hybrid ratio in the next generation — 75 red ears and 26 white ears. The explanation of this phenomenon evidently is the same as that of the bud variations that sometimes occur in perennials. They occur in annuals but are usually unnoticed. The plant due to produce a red ear varied somatically so that one-half of the ears was red and one-half striped. This variation was transmitted by seeds, but at the same time the hybrid character of its seeds was unchanged as shown by their segregation into reds and whites in the next generation and the normal segregation of the hybrid dark reds in a further generation. This strain had red cobs, and there was perfect coupling between the two characters in the next generation.

Two other red pericarp colors seemingly independent of red in other parts of the plant have been found, which may be called R_2 and R_3. R_2 is a dark red that occurs as irregular red stripes radiating from the point where the silk was attached; R_3 is a dirty red color more abundant at the base of the seed and almost wanting at the summit. The dye occurs in small amounts. The latter red, which occurs in Palmer's red-nosed yellow appears to be completely coupled * with red silks. It is almost certain that this red forms an allelomorphic pair with its absence that is entirely independent of R_1, R_2 and R_4, but our numbers are too small to make a full report on the matter. The mosaic red (R_2) is also one that has not been subjected to sufficient genetic study. Thus far (2 generations) it has not bred true but has thrown a percentage of non-reds.

Two other red pericarps have occurred, however, which are interesting because they are the same in appearance but are not allelomorphic to each other. The first is a rose red (R_4) characteristic of No. 5. It develops only in presence of light, hence the ears with thick husks show the color but faintly. When the husks are stripped away and the ear matures in full sunlight, however, the color appears over the entire ear as a bright

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* Coupling is proved by the fact that red silks occur *without* red pericarp in other combinations.

rose red. In numbers 2, 8 and 18 there appeared another red which we at first thought was the same as the above. It occurs in less amounts and on thick-husked ears can only be detected by careful examination. Since these two reds behave as separate allelomorphic pairs they are called R_4 and R_5.

The transmission of these two reds was shown by crossing No. 5 (R_4) with No. 18 (R_5). In F_1 all of the ears were red. In F_2 there were 131 red ears and 7 white ears. No. 5 (R_4) was also crossed with No. 2 (R_5) and gave similar results although the number of plants was small. In F_2 there were 52 red ears and 2 white ears.

It may be asked whether the red in No. 5 (R_4) acts as a simple mono-hybrid in crosses with strains having no red in the pericarp. We have only one cross of this kind in which data for pericarp color were taken. No. 11-2 (r_4) was crossed with No. 5 (R_4) and yielded 251 red ears and 91 white ears in F_2.

None of these varieties had the red color in other organs.

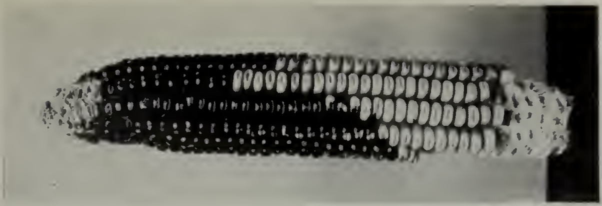
Cob Color.

Several crosses were made in which one parent had a red cob and one a white cob. None of the parents had dark red pericarps (R_1) but in one case R_4 was present (the light red pericarp developing in presence of light). In a cross between No. 5 and No. 6, F_2 yielded 277 ears, of which 212 had red cobs and 65 white cobs. It was strictly a mono-hybrid cross, and the character red-cob seemed not to be coupled with the pericarp color. This red we may call R_c.

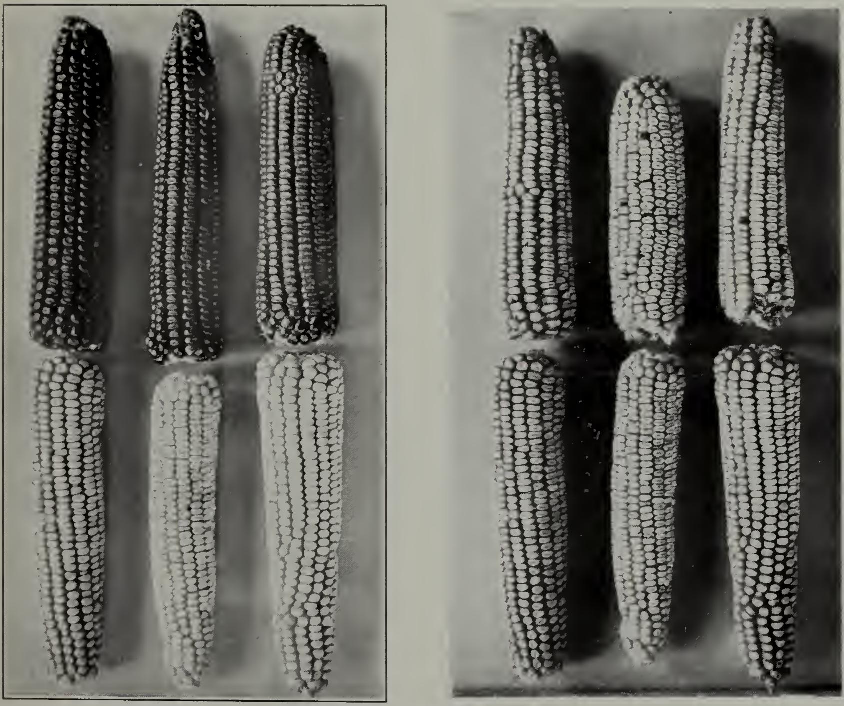
The parents in this case were tested for purity although there are strains of No. 6 in our possession that do not have red cobs. The results of the other crosses were similar and space will not be taken to report them in full. It must be noted however, that although no di-hybrid reds were found, it is not beyond probability that such might be found in an extensive series of crosses.

Silk Color.

Varieties are also obtained which have red silks although the red color is not manifested in other parts of the plant. In fact, No. 19, which has the darkest red silks of any variety in our pos-



a. Somatic or bud variation from dark red seeds to slightly variegated seeds in ear whose seeds were supposed to be half R_1, r_1, and half r_1, r_1.



b. Progeny of red seeds of *a.* Half dark red, half white.

c. Progeny of slightly variegated seeds of *a.* Half slightly variegated, half white.

d. Similar bud variation in which R2 is concerned.

Pericarp Colors and Somatic Segregation.

session, has white cob and pericarp. It is not quite clear, however, how this character is transmitted. The facts are obscured by the action of the bag over the ear to be hand-pollinated, which prevents the full development of the red color by shutting out the light. For this reason one cannot be certain whether the F_1 plants which are selfed are full reds or only red-haired silks.

An illustration of what is obtained in a cross between red silk and non-red silk varieties is as follows. No. 12-2 which is pure for non-red silks was crossed with No. 9-2 which is pure for red silks. In F_1 there were 110 plants with red silks and 27 with greenish-white silks with red hairs. In F_2 the progeny of 3 F_1 plants were grown. The first ear gave 123 plants with red silks and 40 with white silks. The progeny of the other two ears were of three classes; reds, greenish-whites with red hairs and greenish-whites in the numbers 198 : 29 : 94. We will not attempt to analyze this ratio. It is simply mentioned to show that the silk color does mendelize without the production of color in other parts of the plant.

Glume Color.

No plant has yet been obtained which has red glumes and yet shows no red color in other parts of the plant. One has been found however that is pure for red glumes and shows no red in other parts with the exception of the silks.

General Consideration of Red Sap Color.

It is difficult to put aside the thought that all of these red colors are localizations of the same general pigment. If this were true, there should be a series of varieties in which increasing extension of color is found, until red appears in all the organs in which it ever occurs. This is not true. Varieties exist, for example, with red pericarp and red cob, with red pericarp and white cob and with white pericarp and red cob. If these formed a series with increasing extension of red one might find the color localized in the cob and not found in the pericarp, but the theory could not account for the existence of varieties with red pericarp and white cob. It seems as if these facts would drive us to one of two conclusions. We are dealing either with different color compounds each of which manifests itself in only one organ, or with identical genes held in the germ cells in such different combinations that they may be manifested differently. The latter interpretation is more probable, and the natural assumption is that identical genes held by different chromosomes in some way accounts for the different manifestations. Yet there is an obstacle to this assumption which though not necessarily insurmountable, is at least important. One cannot quite understand why a red color should be manifested in different organs simply because its gene is held by different chromosomes.

Since the first draft of this paper was written Emerson * has reported important data from many crosses where certain of these red colors of maize are sometimes absolutely coupled in their inheritance and sometimes show spurious allellomorphism. For example, if a plant with a red cob and a red pericarp is crossed with one in which these colors are absent, there is segregation in F_2, but the colors remain together. On the other hand, if a cross is made between a plant having a red cob and a white pericarp and one having a white cob and a red pericarp, the colors show spurious allelomorphism. The spurious allelomorphism is shown by the F_2 generation, in which is produced 1 red pericarp-white cob : 2 red pericarp-red cob : 1 white pericarp-red cob. His idea is that in the case first mentioned the colors are both carried in the same chromosome while in the second case they are carried in different but homologous chromosomes. As Emerson himself has stated, this theory assumes the inevitable pairing of the two chromosomes carrying the colors, which is probable but unproved. Our own data show no facts diametrically opposed to this hypothesis but the criticism regarding genes held by different chromosomes that was made above would also apply here.

Physical Transformations of Starchiness.

Although presence and absence of starchiness behaves as a Mendelian allelomorphic pair in heredity, the physical condition of the starch is a different matter. Starchiness acts as a filial or endosperm character and shows as Xenia in individual seeds.

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* At meeting of Amer. Soc. Nat., Ithaca, N. Y., Dec. 29, 1910.

The physical condition of the starch behaves as a plant character affecting the entire ear. One may have ears which show a tendency towards the dented character in some seeds and a tendency towards the flint character in other seeds. Such ears are probably always heterozygous dent-flint combinations, and simply show zygotic variations. The different kinds of seeds give the same results in the next generation and show no tendency toward a real segregation of dent and flint .characters in the individual seeds.

The difference in the appearance of the starch in the different races of maize has been described earlier in this paper. The immediate cause of these differences is the amount and location of the soft starch formed in proportion to that of corneous or translucent starch. In the pop corns there is total absence of soft starch or at most only a small amount immediately surrounding the top and back of the embryo. As this amount of soft starch increases, the starch cells of the seeds lose their ability to hold the steam formed by the moisture they contain when heated, and can no longer evert their entire contents as cooked starch. They may pop slightly but they can no longer be considered commercial pop corns. They have passed into the flint corn class. This class includes varieties with varying amounts of soft starch up to those in which it covers the cap. The latter are dent corns, for the dent is simply formed by the greater percentage of contraction which the soft starch undergoes in drying. The amount of dentness is in direct proportion to the thickness of the soft starchy layer at the cap. A few varieties are known in which the soft starch has replaced *almost all* the corneous starch. They are known as semi-starchy corns. They are not so well known however, as the floury corns in which the corneous starch is absent.

As all of these varieties are known in *Z. mays curagua*, *Z. mays hirta* and *Z. mays tunicata*, it is obvious that·the proportions these two kinds of starch (in appearance at least) plays a great part in the commercial classification of maize. Also, since so many varieties are known in which every possible ratio of corneous starch to soft starch occurs, it is evident that the transmissible characters which cause these differences are relatively numerous and their interactions complex. For these reasons, it is perhaps too much to expect that the inheritance of this complex of characters will be cleared up until all possible combinations of these varieties have been made and studied. Our data serve only to establish certain general facts.

The first bit of evidence in the matter comes from a consideration of the behavior of the only class of maize varieties that apparently are beyond the scope of the subject in hand — the sugar varieties. When the latter are crossed with starchy varieties it is perfectly clear that starchiness is a separate character independent of the physical form in which it exists. Sugar varieties are found that are simply dents and flints which lack starchiness. We have also produced by crossing, sugar varieties that are characteristic pop corns lacking starch. No sugar varieties are known which would be soft starch varieties (Z. mays amylacea) if they contained the S factor, but it can hardly be doubted that such could be produced. The experimental evidence is as follows. When Black Mexican, Early Crosby and Golden Bantam are crossed with dent varieties, the Xenia starchy seeds, or F_1 generation are all flint-like in character. These when grown produce F_1 ears which have an appearance intermediate between dents and flints and give in F_2 ears which are characteristically flint in character. In the case of the cross between Black Mexican sugar No. 54 and Illinois High Protein dent No. 8, these flint segregates of F_2 were carried to the F_3 generation and bred true. Since pure dent varieties were the male parents of these crosses, the occurrence of flints in F_2 can only be accounted for by supposing that the sugar varieties that were used as the female parents of the crosses were latent flints. In the same way Stowell's Evergreen sugar and Late Egyptian sugar were proved to be latent dents by crossing them with starchy flint varieties. The Xenia seeds were dented and pure dents appeared in the F_2 generation. One peculiar thing occurred in the cross between Black Mexican sugar, No. 54 and Illinois High Protein dent, No. 8. In F_1 all of the ears were intermediate between dent and flint with a tendency toward dentness, except one. This ear was a pure flint in appearance. Only one of the intermediate ears was grown in the F_2 generation and it produced 91 dents and intermediates and 6 flints. The pure dents could not be separated from the intermediates but flints occurred in the ratio of one out of sixteen. The ear which was apparently flint in F_1 proved

PLATE XVI.

At left, No. 15, Longfellow flint. At right, No. 8 Illinois high protein dent. In center, F1 ears of cross 15x8, showing intermediate character of physical condition in which the starch is stored.

DENT-FLINT CROSSES.

to be an intermediate in F_2. Thirty-four ears were obtained, of which three were clearly dented, a number were intermediate, while from ten to twenty would ordinarily be classed as flints. Thirteen of the latter were grown in the F_3 generation and produced from 50 to 175 ears apiece. Nine out of the thirteen gave only flint ears in a total of 947 individuals. The other four ears produced a total of 264 ears of which between 10 and 20 were flints (i. e. ten were certainly flints and ten others were questionable). Therefore, since 9 out of 13 of the 20 ears classified as "probable flints" in F_2 proved to be true flints in F_3, we have 14 ears pure flint to 20 dents and intermediates in F_2. We do not know enough about this cross to say just what occurred here, but it is probable that one factor for dentness was missing in the pollen which produced the hybrid seed from which this lot F_2 ears came. In the other case a di-hybrid ratio appears.

Several other crosses were made between true dent and true flint races, that is, races in which the parents both were starchy. No. 15 Longfellow flint was crossed with No. 8 Illinois High Protein dent. The F_1 generation was intermediate in character. Through an unfortunate oversight data regarding the segregation in F_2 were taken on the progeny of only one ear of the three F_1 ears planted. This ear gave 33 dents and intermediates to 3 flints. About 200 ears were obtained from the other two F_1 ears planted and from our general field notes we can say that not less than 15 dents and intermediates to each flint ear were obtained. One flint ear gave a crop of 94 ears in F_3, all of which were flint. One dent ear grown in F_3 also proved to be pure. A better idea of these results is given by the photographs on Plates XVI and XVII, however, than can be given by written description.

Two crosses were made between No. 11, Sturgis' flint and No. 8, Illinois High Protein dent. Both were intermediate in F_1. In F_2, progeny of one F_1 ear of the first cross gave 44 dents and intermediates to 3 flints. In F_3, one ear from an intermediate of F_2 gave 23 dents and intermediates and 2 flints. Five F_1 ears of the other cross were grown in F_2 resulting in 175 dent and intermediate ears, and 17 flint ears. The ratio here is about 10 : 1, but if any error was made in the classification it certainly occurred by placing intermediates in the flint class.

Another cross of this kind was that of No. 5-5, Watson's flint with No. 2, Illinois Low Protein. The ears were intermediate in F_1. In F_2 there was segregation, for ears exactly like No. 2 were obtained. Out of the 101 ears obtained, however, no ears were produced that could be classed definitely as flints. One or two flint-like ears occurred which will be tested for purity this coming season. It is quite likely that we have here a tri-hybrid or possibly a tetra-hybrid.

The female parent of this cross, No. 5, was also crossed with No. 6, Leaming dent. F_1 generation was intermediate as before. Five F_1 ears were grown with the following results:

Dents and Inter.	Flints
98	16
71	17
51	5
42	7
Total, 262	45

These ears gave different ratios. Probably more ears were classed as flints than would prove to be such in the F_3 generation, yet they were classified similarly in each case and F_3 tests would probably only reduce the proportion of flints from each ear. Paradoxical as it may seem, however, different ratios are to be expected in F_2 if the general hypothesis concerning the applicability of Mendelian principles to cases where variation is apparently continuous, is true (East : 10). This is explained in the following paragraphs.

In the crosses described above three facts stand out definitely. The characters which give the flint or the dent appearance to maize are transmitted as plant characters to the entire ear and not as endosperm characters to the individual seed. They conform to the essential feature of Mendelism by showing segregation; and they are due to the action of more than one transmissible character. The question remains, can any or all of these characters be named?

Our experience suggests that the proportion of corneous starch to soft starch depends partially upon size and shape of the pericarp and upon the number of rows per ear. All of the

PLATE XVII.

a. F2 dent segregate above (frequency about 1 in 10). Random sample of its F3 progeny below.

b. F2 flint segregate above (frequency about 1 in 16). Random sample of its F3 progeny below.

Dent-Flint Crosses.

races (pop corns) in which soft starch is absent have small seeds, and the full corneous starch character cannot be transferred to large seeds by recombination through hybridization. On the other hand, by crossing a pop maize with a dent maize dent seeds may be obtained which are much smaller than many races with flint seeds. Further, dent races are known which have much larger seeds than some races in which the corneous starch is entirely absent (the flour corns). There is also some relation between the size of the plant and the amount of soft starch in their seeds. The floury or semi-floury corns are in general larger than the corneous starchy corns. Here again, however, there is an overlapping, for we have produced dent races by crossing with dwarf pop races, which are much smaller in size than the large pop and flint races.

Relationship between the physical character of the starch and shape of pericarp is much more intimate than it is between the former and size characters. In the rice pops the pericarp is drawn to a point at the place where the silk is attached. This makes the rice pop races have rather long slender seeds, but it is probably due to a separate character or characters. Leaving this complication out of consideration one may say that the pop corns have small seeds which are almost as broad as they are long. As the seeds become larger, if the ratio of length to breadth remains about unity or less, flint races are formed. If, instead, the ratio of length to breadth increases, dent races are formed. On the other hand, medium large to large seeded races may have almost any ratio of length to breadth and be either flint, dent or floury varieties.

Of course the shape of the pericarp depends somewhat on the number of rows, as the greater this number the more the seeds are crowded together and thus lengthened. Small-seeded pop and flint races exist with as high as 20 rows, but when the seeds are medium in size flint races are usually 8-rowed and 12-rowed, and never — in our experience — over 16 rowed. Dent races, on the other hand, seldom occur with less than 12 rows, but when large seeded they do exist with as few as eight rows. Floury races we have never seen with less than 10 rows, but they reach as high as 24 rows.

These relationships may simply be correlations and not direct causes of the proportion of corneous starch to soft starch that exists in various strains of corn. But even if they were directly concerned, they could not account for the large number of differences in varieties, for none of the correlations are sufficiently high. Many other characters, the exact nature of which is unknown, must be concerned in the matter. The simplest interpretation of the matter seems to be the interaction of independent allalomorphic pairs of the nature reported by Nillson-Ehle (: 10) and East (: 10) in earlier papers. If this interpretation be granted, one should expect that greatest difference in character pairs would exist in the case of pop and starchy races. Flint and dent races with about the same size seeds and small differences in number of rows should differ by fewer pairs of characters.

We have seen that in two of such crosses the evidence points to the existence of two allelomorphic pairs giving pure flints and pure dents in the F_2 generation once in every sixteen individuals. In another cross (5-5 x 2) at least three character pairs are concerned. It happened that in two of these cases the male parents were Illinois High Protein and Illinois Low Protein dent races, which gives us some idea as to why there was a di-hybrid ratio in one case and a higher ratio in the other case. These two strains were both isolated by selection from a commercial variety known as Burr's White. This variety, as are most commercial varieties, is a mixture of complex hybrids. By continued selection of ears high in protein and of ears low in protein with close interbreeding of the progeny these two strains were isolated. The high proteid race is characterized by a high percentage of corneous starch, bringing it into closer relationship to the flint corns. The low proteid race is characterized by a high percentage of soft starch, bringing it into closer relationship with the flour corns. It was the high proteid strain, that is, the one nearer the flint varieties, that gave the di-hybrid ratio when crossed with a flint race; while the low proteid strain, — the one nearer the flour corns, — gave the higher ratio.

This result is what one should expect, but can the 6 : 1 ratio obtained in the cross between No. 5 and No. 6 be explained so easily? We believe it presents no difficulties if the complex gametic constitution of No. 6 is properly appreciated. The individual which furnished the No. 6 pollen came from a selfed daughter ear of the original No. 6. Its sister ears varied in number of rows from 12 to 20 with the mode at 16. The individual furnishing the pollen in cross 5 x 6 was in all probability therefore a complex hybrid itself, and the cross instead of being simple was really a collection of crosses. There is no doubt that many intermediate ears were classed as flint in the table given above. If they could all be grown for another generation it is quite likely that a series of mono-di-tri and higher hybrids would be found. It may be asked why, if this is the case, were not the other crosses complex? The answer is that they undoubtedly were more complex than they seemed. For example, if a large number of F_1 ears were grown it is likely that *some* would give ratios other than those found. It was simply chance that gave us fairly good di-hybrid ratios from a *few* F_1 ears in two instances. The most important reason why the cross with No. 6 was likely to be more variable than the others, however, lies in the fact that *all* of the other strains had been inbred for much longer periods.

Size Characters.

The remainder of Part IV will be devoted to a discussion of the inheritance of size characters, — variations that have been considered to be and to casual observation are, continuous. Our studies have been concerned with the number of rows per ear, height of plant, length of ear and size of seed.

It is perfectly obvious to one familiar with the maize plant that it is almost impossible to work out in detail the inheritance of the complex factors that interact to cause the transmissible differences in the size of its organs. That size characters are complex in themselves is shown by the numerous varieties grown commercially. They each vary from their own means, but different variety means in height are found all the way from two and one-half to fourteen feet with but little actual difference between the most similar strains. Further to complicate matters, all size characters respond to environmental stimuli, and these non-inherited fluctuations obscure the analysis of pedigree cultures in a still greater degree. *For these reasons we do not attempt to analyze our results further* *than to say that they do show segregation in every case.* And segregation is held to be the important and essential feature of Mendelism. Therefore we believe that size characters mendelize.*

Let us now consider the hypothesis by which segregation in characters apparently continuous in their variation, could come about. Nillson-Ehle (: 09) has shown that black glumes in oats when crossed with their absence behave sometimes as monohybrids and sometimes as di-hybrids, and that presence and absence of red pericarp color in wheat sometimes behaves as a tri-hybrid. He further showed, although not quite so conclusively, that presence and absence of ligule in oats behaves as a tetra-hybrid. In this and in a former paper (East : 10) it has been shown that yellow endosperm, red pericarp and dented seeds (as opposed to flinty seeds) behave as di-hybrids, with so many data that the facts can hardly be questioned. We have also shown although less conclusively that other red pericarped varieties and other varieties which differ in their ratios of soft to corneous starch behave as higher hybrids.

TABLE 26.

INHERITANCE OF ROWS IN CROSS (5 x 6).

No.	Gen.	Rows of Parents	Row Classes								
			8	10	12	14	16	18	20	22	24
No. 5 flint (2 yrs.)	P	8	289	2	2						
No. 6 dent	P	18	. .	. .	6	31	51	18	4		
No. 5 x 6	F_1	8	13	36	53	10					
(5 x 6)–1	F_2	12	12	48	35	9	1				
(5 x 6)–2	F_2	10	7	22	15	2					
(5 x 6)–22	F_2	10	8	45	31	1					
(5 x 6)–23	F_2	12	4	25	60	18	4	2	1		

It should be clearly understood what this means to Mendelian theory. *Several genes for the same character may exist in the germ cells of one organism, the number being limited possibly by the number of chromosomes.* The limited number of cases thus

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* It is probable that the number of internodes per plant is one of the factors directly concerned in the inheritance of height of plant.

far found presumably is due to the fact that few size characters have been investigated, for nowhere would these phenomena be so likely to occur as in quantitative characters.

It is fortunate for us that it has been possible to prove the presence of several independent allelomorphic pairs due to produce the same somatic character, for characters like color where dominance is relatively perfect. Beginning with this as a basis, one can extend the theoretical possibilities of such facts to other cases and thus be better prepared for the paradoxical complexities that occur in actual pedigree cultures. When in a cross there is simple presence dominant to absence of one gene for a certain character, the ratio in F_2 is 3 dominant to 1 recessive; when two independent allelomorphic pairs producing the same character are concerned, the ratio in F_2 is 15 dominants to 1 recessive. In general then if *n* allelomorphic pairs are concerned, in F_2 there will be a ratio of 4^n-1 dominants to 1 recessive. It is not likely however that dominance is ever perfect in these complex hybrids. For example, in the case of the two yellow colors in the maize endosperm, the intensity of the yellow decreases in the following order $Y_1Y_1Y_2Y_2$, $Y_1y_1Y_2Y_2$ or $Y_1Y_1Y_2y_2$, Y_1Y_1 or Y_2Y_2, Y_1y_1 or Y_2y_2 and $y_1y_1y_2y_2$. In size characters dominance is probably very incomplete or absent. A heterozygous combination presumably produces half the effect of a homozygous combination. Then as dominance becomes less and less evident the Mendelian classes vary more and more from the formula $(3+1)^n$ and approach the normal curve of error $(\frac{1}{2}+\frac{1}{2})^n$. When there is no dominance

TABLE 27.

INHERITANCE OF ROWS IN CROSS (5 x 2).

No.	Gen.	Rows of Parents	Row Classes								
			8	10	12	14	16	18	20	22	24
No. 5 flint (2 yrs.)	P	8	289	2	2						
No. 2 dent	P	16	..	..	2	14	56	42	20	1	1
No. 5 x 2	F_1	8	1	9	20	4					
(5 x 2)–6	F_2	10	4	18	61	14	3	1			

and open fertilization, a state is reached in which the curve of variation simulates the fluctuation curve, with the difference that the gradations are heritable. The heritable variations are always more or less obscured, however, by the ever present fluctuation.

The experimental results may now be considered—remembering only that fluctuations are present and that in maize many genotypes are often present in one parent. In Table 26 are shown the results from a cross between a race practically pure to the eight rowed type, No. 5, and a dent No. 6, which varies from twelve to twenty rows with the mode at sixteen rows. The F_1 generation is intermediate and furnished four inbred ears that were grown in the F_2 generation. Now three of these four F_2 families show no greater range of variation than F_1, yet it is a noticeable fact that they vary in different ways. Ear (5 x 6)-1 shows a modal condition at ten rows. It may be considered that the crossed seed from which the F_1 ear that produced this crop came, contained the genes for lower numbers of rows from the varying parent, No. 6. Ear (5 x 6)-23, on the other hand, evidently contains genes from No. 6 that were due to produce higher numbers of rows.

TABLE 28.

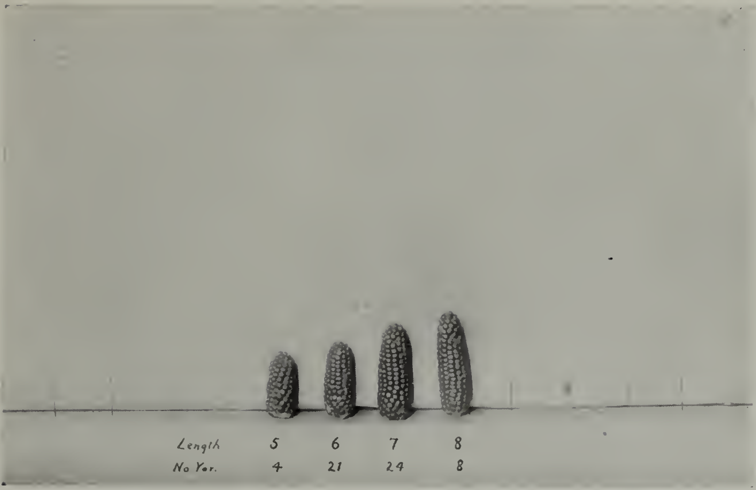
INHERITANCE OF ROWS IN CROSS (11 x 5).

No.	Gen.	Rows of Parents	Row Classes								
			8	10	12	14	16	18	20	22	24
No. 11 flint	P	12	1	4	387	7	1				
No. 5 flint	P	8	289	2	2						
No. 11 x 5	F_1	12	2	11	26	2					
(11 x 5)–8	F_2	12	10	38	107	23	8				
(11 x 5)–18	F_2	10	19	33	100	5					

Table 27 shows a slightly higher variability in F_2 than in F_1.

Table 28 is interesting because it shows the results of a cross between two varieties that have been selected for many years until they are relatively true to the 12-rowed and 8-rowed

PLATE XVIII.



a. No. 60, Tom Thumb maize, showing variation in length of ear. Class centers are even centimeters (1/6).

b. No. 54. Black Mexican sugar maize, showing variation in length of ear (1/6).

INHERITANCE OF LENGTH OF EAR.

PLATE XIX.

a. Variation in length of ear of F1, generation of cross between No. 60 and No. 54 (1/6).

b. Variation in length of ear of F2 generation of cross between No. 60 and No. 54. Family (60-5x54) (1/6).

INHERITANCE OF LENGTH OF EARS.

PLATE XX.

a. Variation in length of ear of F2 generation of cross between No. 60 and No. 54. Family (60-3x54) ($\frac{1}{6}$).

b. Variation in length of ear of F2 generation of cross between No. 60 and No. 54. Family (60-8x54) ($\frac{1}{6}$).

INHERITANCE OF LENGTH OF EARS.

a. Average size of seeds of No. 60 (upper left) and No. 54 (lower left) and the F1 generation of the cross between them. Extremes of the F2 generation at right.

b. Average ears of No. 60 (left) and No. 58 (right) with average of F1 generation in center. Extremes of F2 generation shown.

SIZE INHERITANCE.

TABLE 29.

INHERITANCE OF ROWS IN CROSS (11 x 18).

No.	Gen.	Rows of Parents	Row Classes								
			8	10	12	14	16	18	20	22	24
No. 11 flint	P	12	1	4	387	7	1				
No. 18 sugar (2 yrs.)	P	12	13	32	51	4					
No. 11 x 18	F_1	12	2	10	24	1	1				
(11 x 18)–4	F_2	12	1	9	78	10					
(11 x 18)–10	F_2	10	8	13	62	13					

conditions, respectively. F_2 shows a distinctly higher variability than F_1. It is expected that 8-rowed F_2 plants may breed relatively true.

Table 29 is given simply to show that a cross between two 12-rowed varieties does not show an extension of the row classes. Such a condition should sometimes be possible if our general hypothesis is true, yet it might not occur in more than one cross in hundreds.

Table 30 shows the results from a cross between another variety true to the eight rowed condition and a variety which varies from ten to eighteen rows with the modal condition at twelve. Unfortunately only a few plants matured in the F_1 generation and no conclusions can be drawn regarding its variability. The F_2 generation apparently shows a marked segregation. The

TABLE 30.

INHERITANCE OF ROWS IN CROSS (15 x 8).

No.	Gen.	Rows of Parents	Row Classes								
			8	10	12	14	16	18	20	22	24
No. 15 flint	P	8	100	1							
No. 8 dent	P	14	. .	3	54	36	12	2			
No. 15 x 18	F_1	8	. .	2	5						
(15 x 8)–2	F_2	10	14	15	28	9	1				
(15 x 8)–3	F_2	12	4	13	25	6	3				
(15 x 8)–2–10	F_3	14	. .	1	8	14	6	1	1		
(15 x 8)–2–1	F_3	8	32	35	23	4					
(15x 8)–2–5	F_3	12	4	41	116	15	3	1			

results in the F_3 generation are the most interesting, however, for the progeny of an eight rowed F_2 show a distinct tendency toward an 8-rowed condition, while progeny of F_2 ears having twelve and fourteen rows respectively, though highly variable, show a transmission of their parental qualities.

Our largest pedigree series for number of rows is shown in Table 31. The male parent is the same as was used in the previous cross. The female parent is an eight-rowed type but is not so pure for this condition as the eight-rowed varieties previously used. The general crop in F_1 was discarded before the data was taken upon the number of rows. This oversight is partially rectified by the records from 21 hand-pollinated ears, but the true variability is presumably somewhat greater. Two F_1 ears were grown in the F_2 generation, one having the moda condition at ten rows and the other at twelve rows. Nine ears from the F_2 progeny from (8 x 54)-1 produced F_3 crops. This table should be examined in order to appreciate the significance of the results of this generation. There is a marked tendency in different ears to segregate into twelve-row and eight-row types. Two of the ears have modal conditions at ten rows,

TABLE 31.

INHERITANCE OF ROWS IN CROSS (8 X 54).

No.	Gen.	Rows of Parents	Row Classes								
			8	10	12	14	16	18	20	22	24
No. 8 dent	P	12	. .	3	54	36	12	2			
No. 54 sugar	P	8	89	25	7						
No. 8 x 54	F_1	12	1	6	14						
(8 x 54)–1	F_2	12	9	22	16	1					
(")–5	F_2	12	1	3	16	1					
(")–1–1	F_3	10	. .	15	87	4					
(")–1–2	F_3	8	20	38	50						
(")–1–2a	F_3	10	61	48	54						
(")–1–3	F_3	10	32	75	15						
(")–1–3a	F_3	8	5	20	27	1					
(")–1–5	F_3	12	. .	33	158	26	3				
(")–1–6	F_3	12	4	36	109	8	2				
(")–1–10	F_3	8	Very irregular, mostly 8-rowed								
(")–1–13	F_3	10	96	43	8						

but their variability is so great that the presumption is that this represents simply the continuance of the heterozygous condition. In our opinion there is no question about segregation of number of rows but we are perfectly aware that the believer in selection would be justified in the criticism that that is the cause of the results obtained.

Table 32 shows the frequency distribution of the heights of two varieties Nos. 5 and 6, and the F_1 and F_2 generations of the resulting cross. A good idea of the possible segregation in the F_2 generation of such crosses as this, is obtained by the comparative size of the coefficient of variation of the F_1 and the F_2 generations. In every case it is at least 50% higher in the F_2 generation than in the F_1 generation. The F_1 generation is not intermediate between the two parents but is nearly as high as the taller parent. This fact is not to be regarded as in any way connected with dominance. It is due to the increased vigor which comes from crossing in maize as shown in a previous paper (East : 09). The total results of the F_2 generations show segregation from the lowest class range of the shorter parent to the highest class range of the taller parent. It must not be thought however that these segregates are regarded as pure types. Their behavior in further generations is still problematical. Continued selection of shorter or taller segregates presumably will give an approach toward the selected condition. The criticism that any such results would be due to selection and not segregation is not valid in this case, however, for segregates of extreme types that never appear in either of the parents alone have occurred here in the F_2 generation.

Table 33 shows similar segregation in heights of plants in another cross, No. (54 x 60). The frequency distribution of the heights in No. 54 was obtained from plants grown during the season and on the same soil upon which the F_2 generation was grown. The exact distribution of heights of No. 60 and of the F_1 ears was not taken because at that time another object was in view. The range of distribution as shown by the black lines, is correct. From notes recorded at the time we know that the F_1 generation was quite uniform, the measurements being distributed around classes 67 to 73. Here again the effect of crossing is observed in the relatively tall plants of this generation. The lowest plants in the F_2 generation reach the upper range of No. 60 while the highest plants are practically the height of the highest plants of No. 54. The reason that no plants were obtained in the lower range of No. 60 is due no doubt to their continued heterozygous condition in some of their characters and therefore an increased vigor.

Table 34 shows that the lengths of ears in the above cross segregate in a similar manner. The F_1 generation is not forced toward the long-eared parent as it is in the heights of the plants. In other words ear length does not show the increased vigor due to heterozygosis that is seen in the heights of plants. There can be scarcely a doubt that the greatly increased variability in F_2 is the direct result of segregation.

The segregations of weights of seeds in the above cross is shown in Table 35. The Black Mexican parent No. 54 shows somewhat distorted variation in this character as there are four classes of large sized seeds containing only six ears in all. No F_2 segregates occurred of this size. The reason is that the ears of No. 54 which produced this crop were commercial seed of which only three individuals were used in crossing. The F_1 generation in both Tables 34 and 35 were recorded from only one cross although three crosses were made. To be strictly fair, therefore, the F_2 generation of cross No. (60-5 x 54) is the only one that can be directly compared with the F_1 generation given. We have records, however, of a sufficient number of ears of the other two crosses to know that they differ but slightly if any from the one recorded in the tables. But even if we should be conservative and leave out of consideration the F_2 generations of crosses (60-8 x 54) and (60-3 x 54), there is still no question but that segregation has occurred.

TABLE 32*.

FREQUENCY DISTRIBUTION OF HEIGHTS OF PLANTS IN CROSS (5 x 6).

No.	Class Centers in Inches for Heights of Plants																					A.	S. D.	C. V.
	51	54	57	60	63	66	69	72	75	78	81	84	87	90	93	96	99	102	105	108	111			
5	3	2	4	5	11	17	17	18	10	6	4	..	..	..	..	..	..	..	..	..	..	68.22± .406	6.49± .324	9.51± .421
6	..	..	..	..	..	..	..	..	..	..	..	..	1	2	2	13	14	16	14	8	3	101.18± .400	5.07± .283	5.01± .279
(5 x 6) F_1	..	..	..	..	..	..	..	..	..	..	5	4	3	9	9	6	5	4	4	7	..	94.53± .740	8.21± .519	8.68± .553
(5 x 6)–1 F_2	2	1	3	6	3	5	10	10	22	16	10	7	11	13	6	10	8	4	4	2	..	81.00± .685	12.76± .486	15.75± .684
(5 x 6)–2 F_2	..	..	..	2	..	2	4	2	6	5	12	8	12	6	5	8	3	10	8	2	2	88.33± .802	11.78± .567	13.34± .675
(5 x 6)–8 F_2	2	..	..	..	4	4	12	7	9	13	12	15	10	8	8	1	1	1	..	..	..	79.46± .621	9.55± .438	12.02± .559
(5 x 6)–14 F_2	..	1	2	4	2	7	8	17	21	11	27	24	16	9	11	10	5	4	2	..	..	81.24± .513	10.28± .364	12.65± .450

* Grown side by side in same season.

TABLE 33*.

FREQUENCY DISTRIBUTION OF HEIGHTS OF PLANTS IN CROSS (54 x 60).

No.	Class Centers in Inches for Heights of Plants																								Total
	22	25	28	31	34	37	40	43	46	49	52	55	58	61	64	67	70	73	76	79	82	85	88	91	
No. 54	..	..	..	..	..	..	..	..	..	..	..	..	2	6	10	17	24	26	21	14	14	7	3	1	145
No. 60	<—	——	——	——	——	——	—>[1]	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..	100
(60-5 x 54) F_1	..	..	..	..	..	..	..	..	..	..	..	..	..	<—	——	——	——	——	——	——	——	—>[2]	..	..	55
(60-8 x 54) F_1	..	..	..	..	..	..	..	..	..	..	<—	——	——	——	——	——	——	——	——	——	——	—>[3]	..	..	26
(60-3 x 54) F_1	..	..	..	..	..	..	..	..	..	..	..	<—	——	——	——	——	——	——	——	——	——	——	—>[4]	..	30
[5](60-5 x 54) F_2	..	..	..	..	..	2	2	1	8	18	26	44	95	117	126	92	59	31	22	3	3	..	..	..	649
[6](60-8 x 54) F_2	..	..	..	..	..	1	..	1	5	6	13	13	35	29	36	27	21	20	9	6	4	1	1	..	228
[7](60-3 x 54) F_2	..	..	..	..	..	5	15	13	32	31	68	60	72	34	47	14	13	5	..	..	..	..	..	..	409

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* Distributions giving figures grown side by side in same season.

[1,2,3,4] Extremes were measured and number of plants counted. All strongly convergent around the central classes.

[5] Grown from five different ears.

[6] Grown from two different ears.

[7] Grown from three different ears.

TABLE 34*.

FREQUENCY DISTRIBUTION OF LENGTHS OF EARS IN CROSS (60 x 54).

No.	Class Centers in Cm. for Lengths of Ears.																	A.	S. D.	C. V
	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21			
No. 60	4	21	24	8	..	..	..	..	..	..	..	..	..	..	..	..	..	6.6± .073	.81± .051	12.27± .783
No. 54	..	..	..	..	..	..	..	..	3	11	12	15	26	15	10	7	2	16.8± .121	1.87± .088	11.13± .531
(60-5 x 54) F_1	..	..	..	..	1	12	12	14	17	9	4	..	..	..	..	..	..	12.1± .121	1.51± .088	12.48± .722
[1](60-5 x 54) F_2	..	..	4	5	22	56	80	145	129	91	63	27	17	6	1	..	..	12.7± .058	1.99± .037	15.67± .296
[2](60-8 x 54) F_2	..	..	1	10	19	26	47	73	68	68	39	25	15	9	1	..	..	12.9± .076	2.25± .053	17.44± .413
[3](60-3 x 54) F_2	..	..	2	5	17	33	33	33	27	21	13	10	11	12	1	2	1	12.6± .128	2.81± .087	22.30± .744

* No. 60 and F_1 gen. grown in 1909, F_2 gen. in 1910.

[1] Grown from five ears.

[2] Grown from two ears.

[3] Grown from three ears.

TABLE 35.

FREQUENCY DISTRIBUTION OF WEIGHTS OF SEEDS OF CROSS (60 x 54).

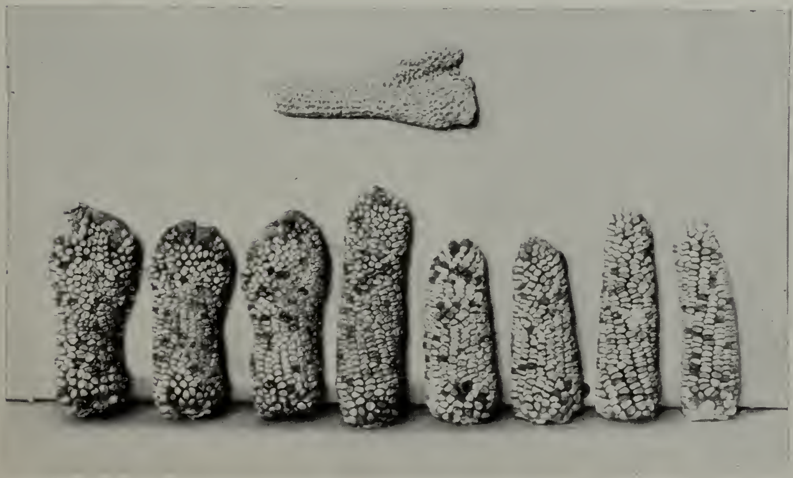
No.	Class Centers in Grams for Weights of 25 Seeds.																				A.	S. D.	C. V
	2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5	6.0	6.5	7.0	7.5	8.0	8.5	9.0	9.5	10.0	10.5	11.0	11.5			
60	7	22	28	4	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..	2.7± .034	.39± .024	14.44± .899
54	..	..	..	..	..	..	..	..	2	2	5	7	14	10	5	4	1	2	1	2	8.3± .107	1.21± .074	14.54± .963
(60-5 x 54) F_1	..	..	..	5	12	13	17	4	3	..	..	..	..	..	..	..	..	..	..	..	4.6± .059	.64± .041	13.91± .912
[1](60-5 x 54) F_2	..	1	7	17	49	98	113	83	69	43	27	13	8	1	..	1	..	..	..	..	5.3± .032	1.09± .022	20.56± .442
[2](60-8 x 54) F_2	..	..	..	1	3	15	16	17	23	23	18	12	8	9	1	1	..	..	..	..	6.2± .067	1.23± .047	19.84± .799
[3](60-3 x 54) F_2	..	..	..	4	10	24	41	53	40	52	21	19	9	8	2	3	..	..	..	..	6.0± .047	1.17± .033	19.50± .563

[1] Grown from five ears.

[2] Grown from two ears.

[3] Grown from three ears.

a. Fasciated ears. F2 generation of cross with normal showing dominants and heterozygotes.



b. F2 cob of heterozygote above; sample of F3 progeny below. From left to right first six show the abnormality in different degrees. Last two ears are normal.

INHERITANCE OF EAR FASCIATIONS.

PART V.

PLANT ABNORMALITIES.

A few abnormalities have appeared in the maize varieties under observation during the progress of these investigations. They have been studied with two objects in view. The first object was to see whether the manner of transmission of heritable monstrous characters gives any clue to the reason why monstrosities have seldom obtained a foothold in nature when in competition with normal types. The second object was commercial. If teratological specimens appear in commercial varieties of maize, it is desirable to know the easiest method to destroy them.

Dwarf Forms.

The first dwarf form appeared in the 1908 culture of No. 6 Leaming dent. This strain had been selfed for the two previous years without producing dwarfs. In the third generation, however, in a culture of 100 plants 5 dwarfs appeared. The plants were normal in appearance, having all parts correlated as in the full sized plants, as is shown in Plate XXV. They were from two to three feet in height and contrasted strangely with the other plants of the variety which were from nine to eleven feet in height. The female flowers seemed to be normal. At least cobs were formed and silks appeared. The pollen however was completely sterile. The dwarf plants were pollinated first with their own pollen and when no seeds formed were pollinated with pollen from normal-sized plants. A few seeds formed on two ears, which were planted the next season. From one ear which had been borne on a plant eighteen inches high only two plants resulted, one being a dwarf and the other of normal height. From the other ear which came from a plant three feet six inches high, seventeen individuals resulted, one of which was a dwarf. The dwarfs, as in the former year, had a normal correlation of parts. The leaves were opposite and the ear appeared in the axil of the sixth leaf from the top as in the normal plants. The pollen appeared to contain some normal grains this year and both of the plants were selfed. No seed set, however, and when pollinated with pollen from normal plants it was found that the silks had passed the receptive stage. This delay lost the strain. Seeds from the old ear of No. 6 had again been planted and had given two dwarfs out of sixty plants, but these had been lost in the same manner.

No. 69-5 a flint with a mosaic red pericarp also gave similar dwarfs with a ratio of 48 normal to 14 dwarf plants. The ear from which they came was a selfed ear from a commercial strain obtained the year before. The commercial strain had given no dwarfs but as only about 100 plants had been grown it is uncertain whether or not they had ever appeared before.

A different kind of dwarf plant appeared in a commercial strain of Stowell's Evergreen sugar corn in 1908. It was very short (18 inches) and had short leaves of the normal breadth. The joints were very close together and the whole appearance of the plant suggested a normal plant that had been pushed together like a telescope. An attempt to self this plant failed, but four days afterward it was pollinated with pollen from a normal strain of Stowell's Evergreen. A fairly good ear resulted which was planted in 1909. One dwarf like the maternal parent appeared out of thirty-seven plants. It was completely sterile, but a selfed normal plant from the same lot gave two dwarfs out of seventy-six plants in 1910. (See Plate XXIV.)

It is a matter of conjecture what occurred in these cases. In the first instance, at least, controlled cultures that had produced no dwarf plants, suddenly threw dwarfs. It was a much more definite occurrence than De Vries' Oenothera mutations for these were mutating when De Vries found them. If the normal type were completely dominant, one must conclude that one seed had been selfed in the case where the dwarf was pollinated with pollen from normal Stowell's Evergreen. In the other two cases the cross-pollination was made with pollen from plants of the same strain, and as only a small number of individuals were produced in the next generation, production of dwarfs was probably continued through the pollen gametes.

The variation was transmitted by plants normal in character, and whether one believes it to be a case of Mendelian dominance of normals or not, there was nevertheless definite segregation.

The fact that segregates appeared in ratios of less than one abnormal to three normal, may have been due to any one of several causes. Abnormal zygotes may have been formed and not have been able to develop, for the germinating power of the seeds formed on the dwarf plants was very low. On the other hand, it may be that this result was due to the same fact that probably gives rise to higher ratios in crosses that have been studied thoroughly; namely, more than one chromosome possesses the necessary material for normal height. There is also the possibility that many abnormalities, particularly those which show great latitude in their development, are due not to regular Mendelian segregation, but to some abnormal chromosome reduction. If some reductions took place normally and some abnormally through some disturbance of the plant's normal physiology, abnormal and normal plants might be produced without definite and constant ratios.

Regularity of Rows of Seeds on Cob.

The great majority of maize ears have rows of seeds running in straight regular rows from butt to tip. Sometimes two rows or even four rows may be dropped in going from the butt to the tip but even then a sufficient amount of regularity exists to call them straight-rowed ears. A varying percentage of ears in each variety, however, have the rows quite irregular, — the seeds often being squeezed together in such a hit and miss manner that the number of rows can only be counted by making cross sections of the cob. Experience with maize cultures shows that there are two distinct kinds of irregularity, one a physiological fluctuation which is not inherited, and one a definitely inherited character or possibly a set of characters. The non-inherited fluctuations are always present while the inherited irregularity may be present or absent. The latter kind has been isolated in several varieties, the most conspicuous being the Country Gentleman sugar corn.

Since the inherited irregularity can only be distinguished from the fluctuation by breeding and then with difficulty owing to the obscuring effect of the latter, it is difficult to come to any conclusion regarding the method of its transmission when dealing with mixed strains. It could undoubtedly be determined by careful work with a cross of which Country Gentleman formed one of the parents. We have not made such a cross, but observations of large commercial cultures of Country Gentleman lead us to believe that irregularity is a Mendelian dominant, although it may not act as a simple mono-hybrid.

Ears with irregular rows appearing in our cultures have been planted several times, but have proved to have been due to physiological fluctuation in all but one instance. An ear of strain 29-2 produced some ears with irregular rows, one of which happened to have been inbred. This ear gave 33 normal progeny and 12 with irregular rows in the next generation. One of these irregular ears gave 33 normal and 15 irregular ears in a further generation, while one of the regular rowed ears gave 125 normal and 5 irregular ears. One of these 5 irregular ears was selfed and will be tested next year. This is about the percentage of irregular ears that the variety gives in the commercial field, however, so the idea suggests itself, that these five ears were fluctuations. If we regard this as the true interpretation of the regular ears giving irregular ears, and reduce the number of irregularities in the progeny of the irregular ears in the same proportion, a ratio of 66 normal to 23 irregular ears is obtained. This looks like a case of mono-hybridism with reversed dominance. It is suggested, however, if this is a case of twice planting a heterozygous mono-hybrid; that it is an example of fluctuating dominance in which some apparently normal ears are really heterozygotes. One cannot even say that only homozygotes show dominance, for it was an irregular ear in each case that threw normals. There is no *a priori* reason why this hypothesis should not be true, but it seems probable that a more complex set of conditions exists. The one fact that stands out clearly is that if the percentage of irregular ears increases much over four percent in a commercial progeny row culture, the whole culture must be discarded to eliminate the undesirable "blood."

Bifurcated Ears.

Occasionally there is found among the eight rowed flint corns, ears which have only four rows. Their cobs are grooved so that they appear to be almost splitting. One of these individuals appeared in a culture of No. 17 (Palmer's Red-nosed yellow) that had been selfed for three generations. It was grown with the special object of finding out whether the four rowed condition is a final recessive condition as to number of rows. This proved not to be the case. The condition is a secondary effect of a heritable abnormality which causes the cob to show various conditions of splitting into two rowed sections at the base. The variations in this feature are shown in Plate XXIII, fig. a. From this ear, 34 ears abnormal in varying degrees and 12 normal ears were obtained. This ratio suggests the progeny of an ear heterozygous for presence and absence of the abnormality. It will be tested further.

A bifurcation of a different kind appeared in the progeny of No. 7 Leaming dent that had been selfed for four years. In its extreme form the tip of the growing ear becomes monstrously fasciate; but it may vary toward the normal to such a degree that the abnormality is shown only as a slight flattening of the ear when observed in cross-section. The ear in which this abnormality appeared was only slightly flattened; its progeny, however, showed 11 with divided tip and about 20 flattened ears out of 44. (See Plate XXII.)

The normal-eared grand parent of No. 7 had been crossed with No. 19, and from an extracted starchy ear of the F_2 generation there resulted the same abnormality. This ear, No. (19 x 7)-5-7 had a divided fasciate tip. It produced 29 ears with divided tip, 33 ears abnormally flattened and 23 normal ears, — a ratio of 62: 23. The illustration of this sort of fasciation shown in Plate XXII, fig. b, gives an idea of how gradually the abnormal ears intergrade with the normal ears. Yet this is a dominant character alternatively inherited. It is difficult to tell the pure normal ears by inspection but they appear to breed true when isolated.

Ears with Lateral Branches.

An illustration of an ear with lateral branches which is probably nearer the ancestral type of maize appeared in the original culture of No. 17. It is figured in Plate XXIII, fig. b. The ear was not hand pollinated and of course no conclusion can be drawn from the ratio in which the abnormality appeared in the next generation. As a matter of fact 4 ears out of 25 progeny were so affected. One of these happened to have been selfed, but it produced only a few seeds. Ten plants resulted from this poor individual, two of which were abnormal.

The only valid conclusion from these data is that the character does segregate. Normals and abnormals are produced; which fact suggests — as stated earlier in the paper — that the loss of the lateral branching character of maize occurred as a retrogressive mutation.

Plants with Striped Leaves.

Zea mays japonica is a race which produces leaves with longitudinal stripes with and without chlorophyll formation. In other words, the leaves are green with white stripes. Several races of this kind exist where the striping is apparently homozygous and the race breeds true. What experience we have had with striped races has been with another type of striping. The phenomenon has appeared several times in our cultures, and is clearly the same thing that Baur (: 09) obtained in pelargoniums. The full green type is dominant, the striped type is heterozygous, while the homozygous recessives are sometimes formed but cannot live because they lack assimilating organs. Crosses between the striped plants and normal green plants always gave all green progeny. Planting, in two cases, from plants that were striped when very young, 274 normal and 27 striped plants were obtained. This result might seem to indicate a more complex condition than Baur obtained. It is not necessarily so, however, for the plants were first examined for striping when about 18 inches high. This may have been too late to give them the proper classification, since it was found that many of the 27 striped plants became greener as they aged. Several plants without chlorophyll died when only a few inches high. These were probably homozygous recessives.

Hermaphrodite Flowers.

Perhaps it should be mentioned in passing that the immature sex organs, so called, of maize seem endowed with the power of becoming either stamens or carpels. One often finds a normal ear ending in stamens, and nearly every plant produces lateral branches which have carpels and stamens mixed together indiscriminately.

A number of cases have also been observed where a few of the ovules of an ear were surrounded by three stamens as in a perfect flower. The only instance we have seen where all of the ovules had three stamens within the glumes of the flower that is usually aborted, was that of the dwarfs with wide leaves mentioned under the heading "Dwarf forms." It might be supposed that this was an atavistic type representing some of the characters, at least, of the ancestral maize. We should prefer to believe, however, that this development of stamens is merely an accompaniment of the dwarfing due to an endeavor to retain physiologic balance. That is, this type is really a healthy luxuriant form producing very large ears for such a small plant. There may have been developmental energy present which when unable through inner limitations to produce a tall plant, manifested itself in producing stamens.

Considered together, these various abnormalities present several interesting features. It would be rash to make any dogmatic statements in regard to their inheritance, yet it is fair to say that if dominance shows progressive — and recessiveness retrogressive — variations, both types are present. Some of them are evidently simple in character — as far as inheritance goes — while others are complex. It may be that the same apparent type of variation will be found to be simple in some races and complex in others. By this it is meant that both 3 : 1 and higher ratios will be found affecting characters which to the eye are the same.

It does not seem probable that abnormal and degenerate types are always or even commonly extracted recessives in which absence of characters is concerned, as Davenport (: 08) has suggested. This statement has little basis from the data presented here, but the senior author has worked out certain dominant abnormal types in the genus Nicotiana which adds to our experience. The presumption is that they are more often dominant like most of the abnormalities found in man.

Perhaps the fact of prime importance from these data is the variable dominance of characters and their obscuration by physiological fluctuation. As stated once before, this shows the extreme importance of pedigree cultures to the commercial breeder, for some of these complex abnormalities cannot be distinguished from normal plants by gross inspection. It is possible that histological study might show points of difference but these methods are not at the command of the commercial grower.

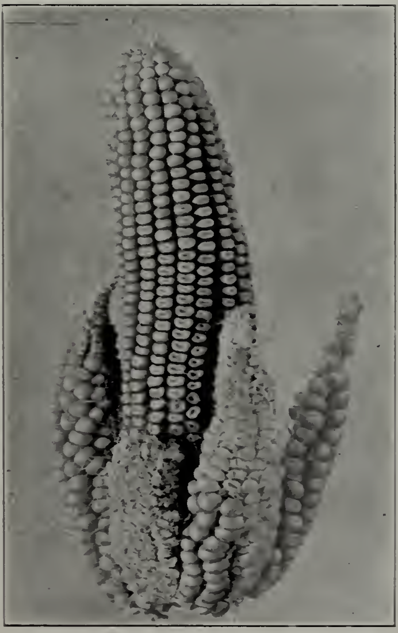
It will be noticed that several monstrous variations occurred in strains that had been selfed for several generations. The effects of inbreeding in maize will form the subject matter of another paper, but it might be well to suggest here a possible cause for their production. Inbreeding in maize gives the same effect as lack of nutrients, while cross-breeding gives the opposite effect. There is retardation or acceleration of cell division, respectively. Now such monstrosities as ears with divided tips, occur more frequently either in cross-bred plants that are over supplied with fertility, or in inbred plants. Perhaps the first case represents fluctuation only, and is uninherited; as to this point we have no data. But disregarding this possibility, might not abnormal distribution of chromatin produce these variations in both cases. The first kind could be caused by abnormally accelerated division and the second kind by abnormally retarded division.

General Conclusions.

The various points of genetic theory discussed in this paper are not sufficiently connected to make possible a short and at the same time intelligible recapitulation. We simply desire to mention our conclusions regarding the central problem of all genetic investigations, that of laws of heredity.

When Mendel's Law of Heredity was rediscovered in 1900, it was the general belief that it covered only a few isolated cases. Many apparent exceptions were cited. One by one, however, these exceptions have been found to yield to interpretation by simple extensions of the Mendelian notation when fully understood. In our experience as reported here, no exceptions to Mendelian interpretation have been found. Such exceptions may exist, yet it seems as unwise to say that Mendel's Law is not general as to conclude at once that it can be made to cover every possible case. One may say that Mendel's Law has

a. Heterozygous bifurcated cob above; dominant and heterozygous progeny below showing imperfect dominance.



b. Multiple ear. An imperfectly dominant character. Aboriginal maize probably possessed a similar character.

ABNORMALITIES.

A plant of the dwarf mutation appearing in Stowell's evergreen sugar maize compared with a normal ear of the latter.

DWARF FORMS.

A dwarf type which appeared in Leaming dent maize compared with a normal ear of that variety.

Dwarf Forms.

covered so many cases that its generality is rendered highly probable, although insufficient genetic investigation has been accomplished to place it on equal terms with any of the great laws of physics and chemistry. Yet some of the great laws of chemistry were accepted when surrounded by seeming exceptions. Some of these exceptions have been cleared up by such recent advances as the Ionic Theory and the Phase Rule; some still remain.

Is it not probable that other like generalities will be found in biology, which, although they may entirely change our general conception of the fundamental action of Mendel's Laws, will nevertheless leave the facts upon which it was based as useful and practicable as have been left the facts of chemical recombination in definite and multiple proportions in the light of the Electron Theory?

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[Reprinted from THE AMERICAN NATURALIST, Vol. XLVI., June, 1912.]

INHERITANCE OF COLOR IN THE ALEURONE CELLS OF MAIZE

IN those plants of which there is a considerable knowledge of the heredity of flower sap color, namely, *Antirrhinum, Lathyrus, Matthiola* and *Primula,* it has been found that an hypostatic color factor is often necessary for the production of an epistatic color. For example, a basic factor generally designated as C being present, a flower becomes red by the addition of a factor R, and becomes magenta or purple by the addition of still another factor P. The zygotic formula of a pure red flower is $RRCC$ and of a pure purple flower is $PPRRCC$; but a flower with the zygotic formula $PPCC$ is colorless.

On the other hand, certain seed coat and other colors of wheat, of beans and of other plants do not need the presence of the hypostatic factor for the formation of the epistatic color. For example, Nillson-Ehle crossed a black glumed oat $BBGG$ with a white glumed oat $bbgg$. In the F_2 he obtained 12 black : 3 gray : 1 white. The actual ratio was $9BG : 3Bg : 3bG : 1bg$, but as the black was produced whether the gray factor was present or not, the visible ratio was as given above.

The natural conclusion is that in the first category the epistatic factor is more specific in its action than it is in the second category. If one accepts the interpretation that color is formed by the action of an enzyme on a colorless chromogen, he must conclude that the epistatic enzyme of the first kind can only produce its action, if, by the presence of the hypostatic enzyme, the chromogen has already been carried through a necessary preliminary reaction. An epistatic enzyme of the second kind, however, is sufficient unto itself and is absolutely independent of the action of the hypostatic enzyme. One may even assume that the chemical bases upon which the two enzymes of the second category act are independent of one another.

Perhaps a concrete illustration will show the difference of action in these two cases better than description. In the black glumed oat $BBGG$, one can imagine the black color or the gray color wiped out mechanically. The other color remains. In the purple flower *PPRRCC*, if the red factor is wiped out no color is left.

In an earlier paper East and Hayes[1] found four independent gametic factors in maize, each of which affects the production of color in the aleurone cells of maize. These four factors are a basic color factor *C*, a reddening factor *R*, a purpling factor *P*, and an inhibiting factor *I* which prevents the development either of the red or of the purple color. Of the various points of interest in the interpetation of the inheritance of these factors, two have been investigated further. The first is the cause of modified colors. This will be discussed at length in another paper. The second is the action of the reddening factor *R* and the purpling factor *P*. It was then thought that the presence of the factor *P* together with *C* was all that was necessary for the production of the purple color. It can now be shown that the purple color develops only when the three factors *P*, *R* and *C* are present. The production of color in the aleurone cells of maize is therefore analogous to that in the flowers of the genera described above, which was designated as category one. This interpretation of the facts makes little difference in the general behavior of these colors in inheritance and is only interesting in so far as it unifies the interpretation of the aleurone colors in maize with the sap colors of certain flowers.

The following scheme will show the differences in behavior in the two schemes.

1. A purple crossed with a non-purple gives 3 purple : 1 non-purple in F_2. Here there is no difference in the two schemes. The proper interpretation gives this result from crosses

$$PPRRCC \times PPRRcc \text{ or}$$
$$PPRRCC \times PPrrCC.$$

2. A purple crossed with a non-purple gives 9 purple : 7 non-purple in F_2. The old interpretation was that this occurs when the F_1 has the formula *PpCc*. The present interpretation is that it occurs when the formula of the F_1 is *PPRrCc*.

3. A purple crossed with a non-purple gives the formula *PpRrCc* in F_1. If the *R* factor is unnecessary for the production of purple, the ratio in F_2 will be (*a*) 36 purple : 9 red : 19 white. If *R* is necessary for the production of purple the ratio in F_2 will be (*b*) 27 purple : 9 red : 28 white. A sample

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[1] "Inheritance in Maize," *Conn. Agr. Exp. Sta. Bull.*, 167: 1–141, 1911

family of F_2 segregates gave the following ratio which may be compared with the closest possible expectancy under each theory.

Actual result	191 purple : 56 red : 180 white.
Theory (*a*)	240 purple : 60 red : 127 white.
Theory (*b*)	180 purple : 60 red : 187 white.

This suggests theory *B*, but is not conclusive. Conclusive evidence comes from the F_3 generation. On theory *A*, every 36 purple F_2 seeds should give on the average the following results in F_3:

4 ears all purple.
10 ears segregating 3 purple : 1 white.
4 ears segregating 9 purple : 7 white.
2 ears segregating 3 purple : 1 red.
4 ears segregating 12 purple : 3 red : 1 white.
4 ears segregating 9 purple : 3 red : 4 white.
8 ears segregating 36 purple : 9 red : 19 white.

On theory *B*, every 27 purple F_2 seeds should give on the average these results in F_3:

1 ear all purple.
4 ears segregating 3 purple : 1 white.
2 ears segregating 3 purple : 1 red.
4 ears segregating 9 purple : 7 white.
8 ears segregating 9 purple : 3 red : 4 white.
8 ears segregating 27 purple : 9 red : 28 white.

The crucial test is the appearance of families showing the ratio 12 purple : 3 red : 1 white. No such family has ever appeared. On the other hand they did divide nicely into families with ratios of 9:3:4 and 27:9:28. Of the first type the total progeny of nine families was 935 purple : 318 red : 436 white. The closest theoretical ratio on the basis of 9:3:4 would be 950 purple : 317 red : 422 white. Of the second type the total progeny of four families was 423 purple : 127 red : 396 white. The closest possible ratio on the basis of 27:9:28 would be 400 purple : 133 red : 414 white.

All other tests made corroborated the interpretation that the *P* factor can produce purple only when *R* and *C* are present.

E. M. EAST

LABORATORY OF GENETICS,
BUSSEY INSTITUTION OF HARVARD UNIVERSITY

U. S. DEPARTMENT OF AGRICULTURE.
BUREAU OF PLANT INDUSTRY—BULLETIN NO. 243.
B. T. GALLOWAY, *Chief of Bureau.*

HETEROZYGOSIS IN EVOLUTION AND IN PLANT BREEDING.

BY

E. M. EAST,
*Assistant Professsor of Experimental Plant Morphology, Harvard University,
and Collaborator of the Bureau of Plant Industry,*

ASSISTED BY

H. K. HAYES,
Plant Breeder of the Connecticut Agricultural Experiment Station.

[In Cooperation with the Connecticut Agricultural Experiment Station and Harvard University.]

ISSUED JUNE 5, 1912.

WASHINGTON:
GOVERNMENT PRINTING OFFICE.
1912.

U. S. DEPARTMENT OF AGRICULTURE.

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BUREAU OF PLANT INDUSTRY.

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W. W. Garner, *Physiologist in Charge.*

E. H. Mathewson and G. W. Harris, *Crop Technologists.*

H. A. Allard, C. W. Bacon, E. G. Beinhart, D. E. Brown, C. L. Foubert, W. M. Lunn, E. G. Moss, and Otto Olson, *Assistants.*

J. S. Cuningham and B. F. Scherffius, *Experts.*

J. E. Blohm, *Special Agent*

B. G. Anderson, R P. Cocke, E. M. East, W. W. Green, E. K. Hibshman, and True Houser, *Collaborators.*

LETTER OF TRANSMITTAL.

U. S. Department of Agriculture,
Bureau of Plant Industry,
Office of the Chief,
Washington, D. C., January 20, 1912.

Sir: I have the honor to transmit herewith and to recommend for publication as Bulletin No. 243 of the series of this Bureau a manuscript entitled "Heterozygosis in Evolution and in Plant Breeding," by Dr. E. M. East, Assistant Professor of Experimental Plant Morphology, Harvard University, and Collaborator of this Bureau, and Mr. H. K. Hayes, Plant Breeder of the Connecticut Agricultural Experiment Station. This paper reports results from experiments that have at different times received aid from this Bureau, the Connecticut Agricultural Experiment Station, and the Bussey Institution of Harvard University and should be considered the product of their joint collaboration.

Respectfully,

B. T. Galloway,
Chief of Bureau.

Hon. James Wilson,
Secretary of Agriculture.

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sion. It is hoped that an adequate number of facts are cited to support the thesis, and it is sufficient on this occasion to say that not a single fact has been discovered that is irreconcilable with it.

THE PROBLEM.

The experimental data upon which the defense of our thesis is based have been obtained entirely from plants, but observations of animal hybrids and published records lead us to believe that the facts are the same among animals. We believe, therefore, that our conclusions apply alike to the animal and the vegetable kingdoms, for we believe the propositions upon which the arguments are based are applicable to all organisms reproducing sexually. These propositions are:

(1) Mendel's law—that is, the segregation of character factors in the germ cells of hybrids and their chance recombination in sexual fusions—is a general law.

(2) Stimulus to development is greater when certain, or possibly all, characters are in the heterozygous condition than when they are in a homozygous condition.

(3) This stimulus to development is cumulative up to a limiting point and varies directly with the number of heterozygous factors in the organism, although it is recognized that some of the factors may have a more powerful action than others.

We later in this bulletin take up briefly some of the specific reasons for extending these theories to the animal kingdom, but at present we shall confine ourselves to developing the botanical proof.

EARLY INVESTIGATIONS.

The number of cases in which hybridizers have noticed an increase in vigor in crosses between subvarieties, between varieties, and between species is so great that an extended citation of the facts is superfluous. Without exception the horticultural writers of the nineteenth century noted the phenomenon and many of them described it at great length. We have taken some trouble to find out its generality, and have found records of its occurrence in the gymnosperms (Darwin,[1] 1876; Focke, 1881) and pteridophytes (Focke, 1881) as well as throughout the angiosperms. In fact, out of 85 families of angiosperms in which artificial hybrids have been made, instances of hybrid vigor exceeding that of the parent species have been noted in 59.

Kölreuter (1763), the earliest botanist to study artificial plant hybrids—as Darwin notes—gives many exact measurements of his hybrids and speaks with astonishment of their "statura portentosa"

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[1] Citations to literature throughout this bulletin refer to the "Bibliography" on pages 49–51.

and "ambitus vastissimus ac altitudo valde conspicua." Later, after having been struck with certain natural adaptations for cross-fertilization, he made a passing remark which plainly showed that he thought nature had intended plants to be cross-fertilized and that benefit resulted therefrom. The hybridists that followed Kölreuter were all interested in the phenomenon, but up to the time of Darwin only Knight and Gärtner attempted to generalize from their observations. Perhaps this was because each one noted the fact that some species hybrids were small and weak. Knight (1799), however, made the somewhat generalized statement that nature had something more in view than self-fertilization and intended that sexual intercourse should take place between neighboring plants of the same species. On the whole, however, Gärtner has given the best expression of the views of the botanical experimenters down to 1849, and for this reason we have translated in full his section on "Wachstum, Luxuriation und Sprossungsvermögen der Bastarde" (Gärtner, 1849, p. 526). He writes as follows:

> One of the most conspicuous and common characteristics of plant hybrids is the luxuriance of all their parts, a luxuriance that is shown in the rankness of their growth and a prodigal development of root shoots, branches, leaves, and blossoms that could not be induced in the parent stocks by the most careful cultivation. The hybrids usually reach the full development of their parts only when planted in the open, as Kölreuter (1763) has already remarked; when grown in pots and thus limited in food supply their tendency is toward fruit development and seed production.
>
> Concerning the great vigor of hybrids all observers are agreed; on this point may be cited Kölreuter (1763), Sageret (1826), Sabine Berthollet (1827), W. Herbert (1837), Mauz (1825), and Lecoq (1845). The vigor of a plant can even serve to indicate its hybrid nature in a doubtful case, as Kölreuter has done with *Mirabilis jalapo-dichotoma*.
>
> Besides possessing general vegetative vigor, hybrids are often noticeable for the extraordinary length of their stems. In various hybrids of the genus Verbascum, for example *lychnitis-thapsus*, the stem shoots up 12 to 15 feet high, with a panicle 7 to 9 feet, the six highest side branches 2 to 3 feet, and the stem 1¼ inches in diameter at the base; in *Althaea cannabino-officinalis* the stem is 10 to 12 feet; in *Malva mauritano-sylvestris* 9 to 11 feet; in *Digitalis purpureo-ochroleuca* 8 to 10 feet, with panicles 4 to 5 feet; and in *Petunia nyctaginifloro-phoenicea* and *Lobelia cardinali-syphilitica* 3 to 4 feet each. Prof. Wiegmann also corroborates these observations.
>
> Hybrids in the genera Mirabilis and Datura are especially conspicuous for their enormous size, as Kölreuter has already stated. The different hybrids of *Datura—Stramonio-tatula*, *quercifolia-ferox*, *laevi-tatula*, and *laevi-ferox*—grew so large as to be almost treelike, with branches and leaves that nearly weighed down the stems, even before the time for developing their numerous blossoms. Likewise such species hybrids as *Nicotiana suavolenti-macrophylla*, *Nicotiana rustica-marylandica*, and *Tropaeolum majus-minus* reach a noteworthy height and circumference.
>
> The root system and the power of germination of hybrids are highly correlated with their great vegetative vigor. Many hybrids, therefore, which are not so luxuriant in growth as those just described, for example, Dianthus, Lavatera, Lycium, Lychnis, Lobelia, Geum, and Pentstemon hybrids, put forth stalks easily and therefore are readily propagated by layers, stolons, or cuttings. The observations of Kölreuter

(1763), Sageret (1826), and Wiegmann (1828) agree with ours in this respect. This extraordinary side branching and tillering, as well as the growth of the main stem, in most hybrids continues until late in the fall and in many until frost, as we have observed in *Lobelia syphilitico-cardinalis*, *Petunia nyctaginifloro-phoenicea*, *Nicotiana suaveolenti-macrophylla*, *Pentstemon gentianoideo-angustifolius*, *Digitalis purpureo-ochroleuca*, *Malva mauritiano-sylvestris*, *Althaea cannabino-officinalis*, etc. Sageret (1826) makes the same statement about *Nicotiana tabaco-undulata.* There are other hybrids, however, that are without this ability to form sprouts, such as *Matthiola annuo-glabra* and those between several Nicotiana species.

Luxuriation expresses itself at times as proliferation; for instance, in *Lychnis diurno-flos cuculi* the receptaculum is changed to a bud that puts forth branches and leaves. If, moreover, the vigor of the hybrids especially affects the stem and the branches, particularly their length, nevertheless the leaves take part in it by becoming larger. Hybrids in the genera Datura, Nicotiana, Tropaeolum, Verbascum, and Pentstemon are examples.

Kölreuter (1763) expresses the opinion that the strength and luxuriance of hybrids continued long after blooming rests upon the fact that the plants are not exhausted and worn out by the production of seed. Similarly, Edw. Blyth (1837) sees in the impotence or sterility of animal hybrids the explanation of their great muscular development, while the considerable size which these hybrids reach in comparison with their parents may be interpreted in the same manner, since capons are able to make a like growth.

But if we take into consideration that: (1) Such a sex condition may exist in diœcious plants without resulting in the luxuriance shown by hybrids, then the reason given above may be no adequate explanation of that phenomenon. (2) The luxuriance of the hybrid plants is already present and visible before the development of the flowers, although one may not doubt that the derangement of the sexual activities and of the development of those organs is not without consequences to the inner life of these plants and that there may obtain essential difference between the weakening or the entire suppression of one or the other of the sexual activities of the hybrids and of the normal separation of the sexes. (3) Not all partially fertile and sterile hybrids are gifted with an increased vegetative power, since we have observed several absolutely sterile hybrids with weakened and limited vegetative vigor; for example, *Nicotiana grandifloro-glutinosa*, *N. glutinosa-quadrivalvis*, *N. rustico-suavolens*, *N. suaveolenti-quadrivalvis*, *Dianthus barbato-deltoides*, *D. caucasico-arenarius*, *Verbascum blattaria-lychnitis*, etc.; at the same time many other hybrids keep the growth relationships of the parent plants unchanged. (4) Among all the hybrids that we have observed, those which show the greatest luxuriance in all their parts are precisely those which show the greatest fertility, for example, *Datura stramonio-tatula*, *Datura quercifolio-ferox*, *Tropaeolum majus-minus*, *Lavatera pseudolbio-thuringiaca*, *Lycium barbaro-afrum*, and *Mirabilis jalapo-dichotoma.* (5) Planting partially fertile hybrids, such as *Nicotiana rustico-paniculata* and *Dianthus barbato-chinensis*, etc., in pots makes the production of fruit and seed easier through limiting the vegetative growth, but a sterile plant is never made fertile by this method. Luxuriance is therefore a peculiar quality of several hybrids, although it is not possessed by all in the same degree.

Although the early hybridizers paid more attention to crosses between distinct species than they did to crosses between races that differed by only a few relatively unimportant characters, there is no question but that they noted a very great number of cases where crosses of the latter character gave plants that were remarkable for their vigor. In fact, we have found no record of intervarietal crosses where delicate or weak hybrids resulted. On the other hand, species crosses sometimes result in hybrids constitutionally feeble. It is obvious, therefore, that a reasonable interpretation of the facts must include an explanation of each category. This matter must be left until later, however, for the work of the early investigators is cited only to show the prevalence of the phenomena under discussion.

Gärtner's researches were followed by but little systematic study of cross and self fertilization in plants until the time of Darwin, and even Darwin's earlier work was confined to the natural means of plant pollination. This early work, mainly a study of pollination in orchids, was summed up in 1862 by the saying "Nature abhors perpetual self-fertilization," a dictum that has become known as the Knight-Darwin law. This important conclusion gave a great impetus to the study of the means of flower pollination throughout the angiosperms. A huge literature of several thousand titles was built up, from which at times important compilations, such as those of Müller (1873) and Knuth (1898), have been made. Every possible variation in flowering habit was argued into an adaptation for cross-fertilization with an ingenuity and zeal similar to that shown by zoologists in their work upon protective coloration and mimicry, and often with as enthusiastic prodigality of extravagant logic. The earnestness of these observers extended our knowledge of the mechanics of pollination in the angiosperms beyond that of any one phase of general botany, yet in the last half of the nineteenth century Darwin was the only scientist who made a systematic experimental inquiry into the physiological effect of cross-pollination compared with self-pollination. The net result of the work of the other observers was simply to show the widespread occurrence of means by which cross-pollination might take place. This fact may be taken to indicate that cross-fertilization is an advantage to a species, but it certainly does not prove that cross-fertilization is indispensable. The many plants naturally self-fertilized preclude it.

Darwin's later experimental work on this subject was so important, both from the standpoint of completeness and brilliancy of analysis, that it must be considered by itself. For this reason we will disregard chronology and conclude this part of our historical summary with the observations of the greatest hybridizer contemporary with Darwin, W. O. Focke. In Focke's fine work "Die Pflanzen-Mischlinge" he gives a chapter on the properties of hybrids, from which the following extract is taken:

> Crosses between different races and different varieties are distinguished from individuals of the pure type, as a rule, by their vegetative vigor. Hybrids between markedly different species are frequently quite delicate, especially when young, so that the seedlings are difficult to raise. Hybrids between species or between races that

are more nearly related are, as a rule, uncommonly tall and robust, as is shown by their size, rapidity of growth, earliness of flowering, abundance of blossoms, long duration of life, ease of asexual propagation, increased size of individual organs, and similar characters.

To undertake a closer examination of the above propositions, it will be necessary to cite a few examples. The following hybrids are abnormally weak: *Nymphaea alba* when crossed with foreign species, Hibiscus, *Rhododendron rhodora* with other species, *R. sinense* with Eurhododendron, Convolvulus, the polyhybrids of Salix, Crinum, and Narcissus. Moreover, it has often been noticed that other hybrid seedlings are somewhat delicate and are brought to maturity with difficulty. Really dwarf growths have been but seldom observed in hybrids; compare, however, certain hybrids of Nicotiana. (Page 285 above, and especially *N. quadrivalvis* × *tabacum macrophylla*, p. 292.) Giant growths are more frequent; note for example Lycium, Datura, Isoloma, and Mirabilis. In size the hybrids generally surpass both the parental species, or at the least they surpass the average height of the two; compare many hybrids of Nicotiana, Verbascum, and Digitalis. Development often goes on with great rapidity, as Klotzsch has emphasized in his hybrids of Ulmus, Alnus, Quercus, and Pinus. Further, the blossoms of hybrids often appear earlier than do those of the parent species, for example, *Papaver dubium* × *somniferum*, many Dianthus hybrids, *Rhododendron arboreum* × *catawbiense*, Lycium, *Nicotiana rustica* × *paniculata*, Digitalis, Wichura's six-fold Salix hybrids, Gladiolus, *Hippeastrum vittatum* × *reginae*, etc., and especially many hybrids of Verbascum. On the contrary, it must be admitted, there are several hybrids that blossom only after a long growth period or not at all, examples of which may be found in the genera Cereus and Rhododendron. Of earlier ripening of the seed independent of earlier blossoming only one example has come down to me, namely Nuphar. Very frequently, one might say very generally, an extraordinary numerical production of flowers has been observed, for example, Capsella, Helianthemum, Tropaeolum, Passiflora, Begonia, Rhododendron, *Nicotiana* (*rustica* × *paniculata*, *glutinosa* × *tabacum*, and others), Verbascum, Digitalis, many of the Gesneraceæ, Mirabilis, and Cypripedium. The size of the blossoms is often increased in hybrids. By crossing two species with flowers of different size, those of the hybrids very nearly reach (not seldom entirely reach) the size of the larger variety. Examples of hybrids with unusually large blossoms are *Dianthus arenarius* × *superbus*, *Rubus caesius* × *bellardii*, and hybrids of *Rosa gallica*, *Begonia boliviensis*, and *Isoloma tydaeum*.

A great capacity for vegetative propagation is very general in hybrids; among the good examples of such a phenomenon may be mentioned Nymphaea, hybrids of *Rubus caesius*, *Nicotiana suaveolens* × *latissima*, *Linaria striata* × *vulgaris*, and Potamogeton. Great longevity may be mentioned as a characteristic of a few hybrids of Nicotiana and Digitalis, ability to withstand cold is especially noticeable in *Nicotiana suaveolens* × *tabacum latissima*, while *Salix viminalis* × *purpurea* is more sensitive to frost than either of the parent species.

These facts point in part to a certain weakness of constitution which is a peculiarity of the hybrid as a result of its abnormal origin and in part to an extraordinary vegetative vigor. An explanation of the last phenomenon, which has been observed much more frequently than the weakness, has only recently been found. The noteworthy experiments of Knight, Lecoq, and others have been familiar for some time, but only through the painstaking experiments of Charles Darwin has the benefit of a cross between individuals and races of one and the same species been clearly demonstrated. The intensification of vegetative vigor in species hybrids is obviously a corresponding experience which requires no especial explanation on the basis of peculiar conditions in hybrids. It was formerly believed that the decreased sexual fertility of hybrids was compensated by a greater vegetative luxuriance, a conception the untenability of which, as Gärtner showed, is refuted in the simplest manner by the experience that many of the most fertile crosses (Datura, Mirabilis) are at the same time characterized by the most excessive stature.

THE WORK OF DARWIN.

Through Darwin's work we get a very different insight into the meaning of cross and self fertilization. At the beginning of his work the knowledge on the subject gained from the experiments and observations of the older hybridists might be summed up in one sentence: Crosses between varieties or between species often give hybrids with a greater vegetative vigor than is possessed by either parent. To be sure there was also a belief that ill effects result from inbreeding, but this belief was generally confined to the animal kingdom. At the end of Darwin's brilliant experiments, or, rather, brilliant analyses of simple but great experiments, not a single point of the many ramifications into which the problem may be divided but had been fully covered. Unfortunately Mendel's experiments were unknown, and the master key of the situation was not available to him. Had it been we can not doubt that he would have made good use of it.

Darwin's interest in the subject arose of course from its connection with the problem of evolution. If the offspring from a cross-fertilization has an advantage over the offspring of a self-fertilization in the struggle for existence, one can hardly doubt the power of natural selection in fixing the structures of flowers. And this being granted, it is obvious that in many flowers mechanical devices to procure cross-fertilization would have been developed. Having found this to be the case in several plants, he bent all his energies to interpreting all flower structures in the same manner. As stated before, the fascination of the work thus initiated has brought us a huge literature on the subject, some of the arguments of which are fantastic to say the least. Darwin himself never allowed his conclusions to get ahead of his facts, a trait that his followers did not always copy. He firmly believed that self-fertilization was so injurious that plants dependent upon it must ultimately perish, but he frankly admitted the obstacles which self-fertilized families like Leguminosæ placed in the way of his conclusions. If he had known of the vigorous plants that reproduce apogamously no doubt he would have regarded the obstacles more seriously than he did. Nevertheless one must admit that at that time, considering the importance of placing evolution on an impregnable foundation, Darwin did not overstate his conclusions. He proved conclusively the advantage of cross-fertilization and the numerous means by which it is obtained.

If he did not distinguish between the advantage a process may hold forth and the necessity of that process, it was because he was not in possession of all the facts. One does not criticize Darwin, therefore, if in a careful examination of his data in the light of modern knowledge many facts are found that may reasonably have somewhat different interpretations than those originally given.

The first point we will consider is the benefit arising from cross-fertilization. It must be granted from the data already presented that an increase in vigor generally results when different species or markedly different varieties are crossed. It is also perfectly obvious that many plants are naturally designed for cross-fertilization. It can hardly be argued, however, that specific crosses could have had a widespread value in the course of evolution. It must be shown, therefore, that in plants not widely different in character cross-fertilization shows an advantage over self-fertilization. In Table A (''Cross and Self Fertilisation,'' p. 240) Darwin's results on this subject are given. To be fair, 15 of these experiments should be discarded, because the number of plants measured in the comparison between those crossed and those selfed is less than five. There are 37 experiments left. Of these, the crossed plants were higher in 24 cases, provided an error of 5 per cent is allowed. In 13 cases, then, cross-fertilization showed no definite advantage.

In Table B, where the weights of entire plants are considered, cross-fertilization showed to advantage in 5 experiments out of 8. From these data it seems logical to argue that cross-fertilization between nearly related plants is often a benefit, yet since types that are self-pollinated in nature—legumes, wheat, tobacco, etc.—are among the most vigorous of living plants, it can not be said to be indispensable. Furthermore, about 25 of our most vigorous species of angiosperms have given up sexual reproduction either partially or entirely and have become apogamous.

Did the simple act of crossing produce these beneficial results? If so, why was the advantage due to cross-fertilization not general and without exception? Darwin himself answered these questions. He says (loc. cit., p. 269):

> A cross between plants that have been self-fertilized during several successive generations and kept all the time under nearly uniform conditions does not benefit the offspring in the least, or only in a very slight degree. Mimulus and the descendants of Ipomoea named Hero offer instances of this rule. Again, plants self-fertilized during several generations profit only to a small extent by a cross with intercrossed plants of the same stock (as in the case of Dianthus) in comparison with the effects of a cross by a fresh stock. Plants of the same stock intercrossed during several generations (as with Petunia) were inferior in a marked manner in fertility to those derived from the corresponding self-fertilized plants crossed by a fresh stock. Lastly, certain plants which are regularly intercrossed by insects in a state of nature and which were artificially crossed in each succeeding generation in the course of my experiments, so

> that they can never or most rarely have suffered any evil from self-fertilization (as with Eschscholtzia and Ipomoea), nevertheless profited greatly by a cross with a fresh stock. These several cases taken together show us in the clearest manner that it is not the mere crossing of any two individuals which is beneficial to the offspring. The benefit thus derived depends on the plants which are united differing in some manner, and there can hardly be a doubt that it is in the constitution or nature of the sexual elements. Anyhow, it is certain that the differences are not of an external nature, for two plants which resemble each other as closely as individuals of the same species ever do profit in the plainest manner when intercrossed if their progenitors have been exposed during several generations to different conditions.

In other experiments that Darwin performed it was shown conclusively that crosses between individual flowers borne on the same plant conferred no benefit whatever on the progeny. It is evident, therefore, since plants may differ in nonvisible transmissible characters, that differences in transmissible factors alone account for the benefit produced by crossing and are indispensable to its occurrence. This is clearly shown by the fact that even types naturally self-fertilized, such as the garden pea (*Pisum sativum*), showed a remarkable increase in vigor when entirely different strains were crossed. We may well believe, then, that if Darwin's plants used in his Table A had all been heterozygous at the start they would all have showed a considerable difference in favor of the progeny of those continually cross-fertilized. Furthermore, leaving out of consideration our own beliefs, a study of his own experiments (Ipomoea) shows that if his comparisons had been kept up for a considerable number of generations the cross-fertilized stocks would have become so nearly like the self-fertilized stocks in constitution that the advantage due to cross-fertilization would have been small. But to this point we shall again recur.

Let us now consider whether the known effects of inbreeding and crossbreeding are manifestations of the same phenomenon. In "Animals and Plants Under Domestication" he says (vol. 2, p. 89):

> The gain in constitutional vigor derived from an occasional cross between individuals of the same variety but belonging to different families, or between distinct varieties, has not been so largely or so frequently discussed as have the evil effects of too close interbreeding. But the former point is the more important of the two, inasmuch as the evidence is more decisive. The evil results from close interbreeding are difficult to detect, for they accumulate slowly and differ much in degree with different species, whilst the good effects which almost invariably follow a cross are from the first manifest. It should, however, be clearly understood that the advantage of close interbreeding, as far as the retention of character is concerned, is indisputable and often outweighs the evil of a slight loss of constitutional vigor.

It is obvious that Darwin believed in a definite accumulation of evil effects from self-fertilization, but his experiments do not justify this view. He is perfectly correct in saying that the good effects of crossing are immediately evident. This is clear when it is remembered that if two plants differ in several transmissible allelomorphs the first hybrid generation is heterozygous in all these characters, while future generations as a whole are heterozygous in only part of these characters. Furthermore, one may cross two plants differing but slightly and obtain only a small increase in size; he may then recross with a third plant of widely different nature and obtain a great increase. When one inbreeds, however, he relies on chance combinations to eliminate heterozygosis. This occurs through the action of the laws governing probabilities. Many heterozygous combinations are eliminated at once. This lowers the number of such combinations, and, while the percentage of elimination is the same, the effect of the inbreeding decreases. Complete homozygosis is approached as a variable approaching a limit. It may be illustrated by the old story of the dog decreasing the distance from the hare by half at each jump. The effects of inbreeding, therefore, appear to accumulate, while the effects of crossbreeding are immediately manifest. But is the apparent accumulation of evil effects real? And are the effects evil? Our interpretation is that the effects of inbreeding are not to accumulate ill effects, but to isolate homozygous strains. One does away with a stimulus due to heterozygosis, and one sometimes isolates strains with poor transmissible qualities. But one also isolates good strains; strains that remain good in spite of continued self-fertilization. In other words, the apparent evil effects of self-fertilization decrease directly with the number of generations it is practiced, due to the increase in homozygosis. On the theory entertained by us it should come to an end with complete homozygosis; practically, complete homozygosis is difficult to obtain.

Did such a decrease in deterioration actually occur in Darwin's experiments as they were increased in duration? They did. Darwin himself noted the point. He says ("Cross and Self Fertilisation," p. 55):

> As the plants which were self-fertilized in each succeeding generation necessarily became much more closely interbred in the later than in the earlier generations, it might have been expected that the difference in height between them and the crossed plants would have gone on increasing; but so far was this from being the case that the difference between the two sets of plants in the seventh, eighth, and ninth generations taken together is less than the first and second (and third) taken together.

This statement was made concerning his experiments with *Ipomoea purpurea,* which were continued for 10 generations. The ratio of heights of crossed to heights of selfed plants varied from 100 to 68 in the third generation to 100 to 86 in the fourth generation, but in the ninth generation the ratio was 100 to 79, which is higher than that of the first generation. The tenth generation was indeed low, but it may with all fairness be rejected, for Darwin states that the plants were diseased.

We know, further, that Darwin was not dealing with the same strain at the end of his experiments that he was at the beginning. This change was due, as we now know, to the elimination of Mendelian segregates. The plants in the beginning varied greatly in the color of their flowers. Indeed, they varied during the whole time of experimentation; but the color of the later generations was much more uniform than that of the earlier generations. The selfed generations were so uniform, in fact, that his gardener said "they did not need to be labeled."

In this experiment as well as in those with other species, such as *Mimulus luteus* and *Nicotiana tabacum*, remarkably vigorous self-fertilized types appeared. It may be that new transmissible variations arose, but it is unnecessary to assume it. One may account for every result obtained by Darwin by granting the isolation of homozygous Mendelian segregates, accompanied by loss of the vigor due to heterozygosis through self-fertilization.

RECENT INVESTIGATIONS.

Since the time of Darwin, several writers, whose results will be discussed later, have investigated the effect of inbreeding on animals. Botanists, however, have in general been interested only in the superficial results of inbreeding and crossbreeding and have made no attempts until recently to bring together and to correlate our knowledge regarding them.

In 1905, Shull and the senior writer each started independent investigations concerning the effects of inbreeding in maize, which may be regarded as an ideal cross-fertilized species. To supplement these experiments we have made a large series of crosses with species of the genus Nicotiana which are generally self-fertilized, as well as minor observations on other plants. We will not discuss our previous papers (East, 1907, 1908, 1909, 1910; Hayes and East, 1911) as the present paper gives a résumé of those experiments. Concerning Shull's work (1908, 1909, 1910, 1911), we wish to quote his own conclusions for they are stated very concisely. Furthermore, Shull's data and our own, independently obtained, are corroborative in every detail and therefore have greater weight than either alone. Even the additional conclusions drawn from the data presented in this paper are largely an application of the earlier analysis to the broader problems that are legitimately concerned.

Shull's conclusions up to the year 1910 are summarized by him as follows (Shull, 1910):

> (1) The progeny of every self-fertilized corn plant is of inferior size, vigor, and productiveness as compared with the progeny of a normally crossbred plant derived from

the same source. This is true when the chosen parent is above the average condition as well as when below it.

(2) The decrease in size and vigor which accompanies self-fertilization is the greatest in the first generation and becomes less and less in each succeeding generation until a condition is reached in which there is (presumably) no more loss of vigor.

(3) Self-fertilized families from a common origin differ from one another in definite hereditary morphological characters.

(4) Regression of fluctuating characters has been observed to take place away from the common mean or average of the several families instead of toward it.

(5) A cross between sibs (sister and brother) within a self-fertilized family shows little or no improvement over self-fertilization in the same family.

(6) A cross between plants belonging to two self-fertilized families results in a progeny of as great vigor, size, and productiveness as are possessed by families which had never been self-fertilized.

(7) The reciprocal crosses between two distinct self-fertilized families are equal and possess[1] the characters of the original corn with which the experiments were started.

(8) The F_1 generation from a combination of plants belonging to certain self-fertilized families produces a yield superior to that of the original crossbred stock.

(9) The yield and quality of the crop produced are functions of the particular combination of self-fertilized parental types and these qualities remain the same whenever the cross is repeated.

(10) The F_1 hybrids are no more variable than the pure strains which enter into them.

(11) The F_2 shows much greater variability than the F_1.

(12) The yield per acre of the F_2 is less than that of the F_1.

We should also like to quote Shull (1911) upon one important point upon which we have but few data:

> Necessary corollaries of the view that the degree of vigor is dependent on the degree of hybridity or, in other words, that it is dependent roughly upon the number of heterozygous elements present and not upon any injurious effect of inbreeding per se are (*a*) that when two plants in the same self-fertilized family, or within the same genotype, however distantly the chosen individuals may be related, are bred together there shall be no increase of vigor over that shown by self-fertilized plants in the same genotype, since no new hereditary element is introduced by such a cross; (*b*) that first-generation hybrids produced by crossing individuals belonging to two self-fertilized lines or pure genotypes will show the highest degree of vigor possible in progenies representing combinations of those two genotypes, because in the first generation every individual will be heterozygous with respect to all of the characters which differentiate the two genotypes to which the chosen parents belong, while in subsequent generations recombinations of these characters will increase the average number of heterozygous genes present in each individual; (*c*) that crosses between sibs (sister and brother) among the first-generation hybrids between two genotypes will yield progenies having the same characteristics, the same vigor, and the same degree of heterogeneity as will be shown by the progenies of self-fertilized plants belonging to the same first-generation family.
>
> All of these propositions have now been tested in a limited way. In 1910, nine different self-fertilized families were compared with nine crosses between sibs within the same self-fertilized family; ten crosses between sibs in F_1 families were compared

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[1] They are usually as vigorous or more vigorous than the original strains, but may or may not have the original characters. Some characters may have been entirely eliminated.—E. M. E.

with ten self-fertilizations in the same F_1 families; seven families were raised as first-generation hybrids between individuals belonging to different self-fertilized families; and ten families were grown in which self-fertilization had been entirely precluded during the past five years. The average height of plants in decimeters, the average number of rows per ear, and the average yield in bushels per acre in these 55 families are given in the following table:

	Selfed × self.	Selfed × sibs.	F_1.	F_2.	F_1 × self.	F_1 × sibs.	Cross-breds.
Average height	19.28	20.00	25.00	23.42	23.55	23.30	22.95
Average rows	12.28	13.26	14.41	13.67	13.62	13.73	15.13
Average yield	29.04	30.17	68.07	44.62	41.77	47.77	61.52

An examination of this table indicates to me that on the whole my self-fertilized families are not yet quite pure bred; for the sib crosses give on the average a slightly greater height, number of rows per ear, and yield per acre than the corresponding self-fertilized families as shown by a comparison of the first two columns of the table. The same fact is apparent from a comparison of the "F_1 × self" and "F_1 × sibs" columns, except that in this case the heights and number of rows per ear are essentially equal while the yield per acre is significantly higher in the sib crosses than in the self-fertilized families.

These statements should be sufficient to indicate Shull's work and point of view. Other writers have proposed methods designed to utilize commercially the increase in vigor shown by first-generation hybrids, and at least two other theoretical interpretations of this increase have been submitted (Jost, 1907; and Keeble and Pellew, 1910). These papers will be considered later. We will now take up the data obtained in our own experiments.

EXPERIMENTS ON A NORMALLY CROSS-FERTILIZED SPECIES, ZEA MAYS.

EFFECTS OF INBREEDING.

In these experiments over 30 varieties of maize, including all the varieties widely differentiated from each other, have been artificially self-fertilized for from one to seven generations. In every case a loss of vegetative vigor has followed. At least, following the earlier usage, one may say the result is a loss of vigor if it is kept clearly in mind that pathological degeneration is not what is meant. The universal decline in vigor consists simply in a somewhat less rapid cell division or slower growth and a smaller total amount of cell division resulting in smaller plants and plant organs.

Besides this phenomenon, to which there has been no exception, the progeny always become more or less differentiated in normal morphological characters, although this is less marked in some varieties than in others. For example, from the yellow dent variety known as Leaming various strains differing in the following characters have been isolated during the several generations that they have been inbred:

Red pericarp and colorless pericarp
Red cob and colorless cob.
Red silks and colorless silks.
Red glumes and colorless glumes.
Profusely branched tassels and scantily branched tassels.
Long ears and short ears.
Ears with various numbers of rows.
Ears with large seeds and ears with small seeds.
Ears with straight rows and ears with crooked rows
Ears high on the stalk and ears low on the stalk.
Stalks with many tillers and stalks with few tillers.

Other minor differences have been observed, but these will serve to show just what is meant by "normal differences." There were also differences in yield of seed—described later in this bulletin—some of which may not seem to be normal in character at first thought, but which we hope to show are not less normal than those given above.

Besides these variations, aberrant individuals appeared in a few strains with characters which might well be called abnormal; that is, they are monstrous characters. But this does not mean that they might not have originated in the same manner as normal characters, for they are transmitted as such. Two of these characters, fasciated ears and bifurcated cobs, show a simple Mendelian segregation with incomplete dominance; two others, striped leaves and dwarf plants, are probably recessives. It is possible, however, that one form of striped leaf is the heterozygote between pure white and normal green. It may be that the first two of these abnormalities are not simply isolated as Mendelian segregates. They have also appeared in commercial varieties grown on very fertile soil, a fact that suggests their origin through interference with normal processes of cell division, acceleration in one case and retardation in the other.

The variability of the strains in the above characters decreased as inbreeding was continued, until after four generations they were practically constant for all grosser characters. This does not mean that physiological fluctuation was not as great as in the original strain. It was not reduced in the least degree. Nor can it be said that no new heritable variations arose. Certain variations did appear which may have been new to the strain—witness the fasciated ears—but of this one could not be certain. Furthermore, it is not meant that after four or five generations of inbreeding a type is homozygous in all of its characters. Such a gametic condition is theoretical and could never be recognized in a pedigree culture. But near homozygotes or near homozygous genotypes are obtained without selection simply by inbreeding. The reason for this is simple.

Mendel in his original paper showed that if equal fertility of all plants in all generations is assumed and, furthermore, if every plant is always self-fertilized then in the n^{th} generation the ratio of any particular allelomorphic pair (A, a) would be $2^n-1\ AA : 2\ Aa : 2^n-1\ aa$. If we consider only homozygotes and heterozygotes, the ratio is $2^n-1:1$. Of course the matter is not quite so simple when several allelomorphs are concerned, but in the end the result is similar. Heterozygotes are eliminated and homozygotes remain. The probable number of homozygotes and any particular class of heterozygotes in any generation r is found by expanding the binomial $[1+(2^r-1)]^n$ where n represents the number of character pairs involved. The exponent of the first term gives the number of heterozygous and the exponent of the second term the number of homozygous characters. As an example, suppose we desire to know the probable character of the fifth segregating generation (F_6) when inbred, if three character pairs are concerned. Expanded we get

$$1^3+3[1^2(31)]+3[1(31)^2]+(31)^3.$$

Reducing, we have a probable fifth-generation population consisting of 1 heterozygous for three pairs; 93 heterozygous for two pairs; 2,883 heterozygous for one pair; 29,791 homozygous in all three character combinations.

From this illustration we think it is fairly easy to see that no matter in how many characters a plant is heterozygous, continued inbreeding will sooner or later eliminate them. Close selection, of course, tends toward the same end, but not with the rapidity or certainty of self-fertilization.

Inbreeding a naturally crossbred plant, then, has these results:

(1) There is partial loss of power of development, causing a reduction in the rapidity and amount of cell division. This phenomenon is universal and therefore can not be related to inheritance. Further, it continues only to a certain point and is in no sense an actual degeneration.

(2) There is an isolation of subvarieties differing in morphological characters accompanying the loss of vigor.

(3) There is often regression away from instead of toward the mean of the general population.

(4) As these subvarieties become more constant in their characters the loss of vigor ceases to be noticeable.

(5) Normal strains with such hereditary characters that they may be called degenerate strains are sometimes, though rarely, isolated.

(6) It is possible that pure strains may be isolated that are so lacking in vigor that the mechanism of cell division does not properly perform its function, and abnormalities are thereby produced.

The maize families shown in Table I illustrate some of these facts, if the yield of shelled corn per acre is taken as a basis of comparison of vigor. These families are not selected to fit a theory, but include representatives of four of the great subdivisions of the species out of those grown in sufficient quantity to give considerable confidence in the determination of yield. Many other types have been inbred for from one to four years, but neither land nor time was available to grow them in large quantities. Their behavior, however, was the same. Inbreeding always reduced the yield of seed and the height and delayed the time of flowering. In general, the decrease in vigor lessened with the inbreeding. Further, both good and bad strains were isolated.

TABLE I.—*Effect of inbreeding on the yield of maize.*

Variety.	Year grown.	Number of years inbred.	Yield in bushels per acre.
Watson's flint No. 5	1908	....	75.7
No. 5-8	1909	1	47.5
No. 5-8-3	1910	2	36.1
Starchy No. 10 [1]	1908	....	70.5
No. 10-3	1909	1	56.0
No. 10-3-7	1910	2	67.0
No. 10-3-7-3	1911	3	39.1
No. 10-4	1909	1	43.0
No. 10-4-8	1910	2	48.7
No. 10-4-8-3	1911	3	29.3
Stowell's sweet No. 19	1909	....	93.2
No. 19-4	1910	1	58.7
No. 19-4-7 [2]	1911	2	51.2
No. 19-8	1910	1	53.6
No. 19-8-2 [2]	1911	2	42.1
Leaming dent No. 1	1905	....	88.0
No. 1-6	1906	1	59.1
No. 1-6-1	1908	2	95.2
No. 1-6-1-3	1909	3	57.9
No. 1-6-1-3-4	1910	4	80.0
No. 1-6-1-3-4-4	1911	5	27.7
Leaming dent No. 1	1905	....	88.0
No. 1-7	1906	1	60.9
No. 1-7-1 [1]	1907	2	59.3
Leaming dent—Contd.			
No. 1-7-1-1	1908	3	46.0
No. 1-7-1-1-1	1910	4	63.2
No. 1-7-1-1-1-4	1911	5	25.4
No. 1-7-1-2	1909	3	59.7
No. 1-7-1-2-2	1910	4	68.1
No. 1-7-1-2-2-9	1911	5	41.3
Leaming dent No. 1	1905	....	88.0
No. 1-9	1906	1	42.3
No. 1-9-1	1908	2	51.7
No. 1-9-1-2	1909	3	35.4
No. 1-9-1-2-4	1910	4	47.7
No. 1-9-1-2-4-6	1911	5	26.0
Leaming dent No. 1	1905	....	88.0
No. 1-12	1906	1	38.1
No. 1-12-1	1907	2	32.8
No. 1-12-1-1 [1]	1908	3	46.2
No. 1-12-1-1-2	1909	4	23.3
No. 1-12-1-1-2-4	1910	5	16.5
No. 1-12-1-1-2-4-11	1911	6	2.0
No. 1-12-1-1-4	1909	4	28.7
No. 1-12-1-1-4-14	1910	5	9.5
No. 1-12-1-1-4-14-3	1911	6	2.0

[1] Two selections from the progeny of this ear grown.
[2] Probably a normal yield. Grown on a more fertile soil than the rest in 1911.

The different families were all planted on the same plat under uniform conditions each season, but, unfortunately, circumstances made it necessary to grow them upon different fields each season. It is therefore necessary to take into consideration the differences in soil fertility and meteorological conditions each year to see the truth of our conclusions, namely, that continued inbreeding caused only isolation of strains of varying potency. The greatest differences in the environmental conditions were in the years 1908, 1909, and 1911.

In 1908 the land used was highly fertile and the general environmental conditions much above the normal. Four stalks per hill were grown this season, but as only three were grown in other years the actual yields have been reduced one-fourth. Even at this disadvantage the yields in 1908 are probably somewhat high. For opposite reasons, poor soil and badly distributed rainfall, the yields of 1909 are somewhat too low and the yields of 1911 are very much too low. This will be appreciated if the low yields for 1911 are examined in Table III.

Since the data on the Leaming dent variety are the most interesting they are repeated in a somewhat different form in Table II. There they are shown in a regular line of descent.

TABLE II.—*Effect of inbreeding on a variety of Leaming dent maize.*

(Yield, in bushels, of shelled corn per acre.)

Parent variety.	Generations inbred and years in which grown.					
	1	2	3	4	5	6
88.0 (1905)....	59.1 (1906)	95.2 (1908)	57.9 (1909)	80.0 (1910)	27.7 (1911)	....
	60.9 (1906)	59.3 (1907)	46.0 (1908)	63.2 (1910)	25.4 (1911)	....
			59.7 (1909)	68.1 (1910)	41.3 (1911)	....
	42.3 (1906)	51.7 (1908)	35.4 (1909)	47.7 (1910)	26.0 (1911)	....
	38.1 (1906)	32.8 (1907)	46.2 (1908)	23.3 (1909)	16.5 (1910)	2.0 (1911)
				28.7 (1909)	9.5 (1910)	2.0 (1911)

The Leaming, a well-known commercial dent variety, yielded 88 bushels per acre the year before it was first inbred. The season was normal, and this yield may be considered fairly typical of what the variety will do on a moderately good soil. Four ears were inbred and were grown in 1906. This was again an average year. The four strains showed marked decreases in yield and notable differences in their characters. The year 1907 was again an average year, and the second inbred generations are normal. Two strains were not grown as second inbred generations until 1908, however, and they are therefore too high. In 1909 the yields are too low; in 1910 normal, and in 1911 much too low. With these facts in mind, an examination of the tables shows how the strains became more and more differentiated. The first strain, No. 6, is a remarkably good variety of corn even after five generations of inbreeding. It yielded 80 bushels per acre in 1910. The yield was low in 1911, but since all yields were low that year it can hardly be doubted that this strain will continue to produce good normal yields of grain. In the field, even in 1911, the plants were uniformly vigorous and healthy and were especially remarkable for their low variability. The poorest strain, No. 12, is partially sterile, never fills out at the tip of the ear and can hardly exist alone. In 1911 it yielded scarcely any corn but will no doubt continue its existence as a partly sterile variety. Plate I shows ears and tassels of an almost sterile strain isolated by inbreeding.

CROSSING INBRED TYPES.

When two of these inbred strains are again crossed, the F_1 generation shows an immediate return to normal vigor. The plants are earlier and taller, and there is a greater total amount of dry matter per plant. For example, in 1911 the average height of all the strains of inbred Leaming dent was 84 inches while the average height of the 16 hybrid combinations was 111 inches and the height of the shortest hybrid combination was 1 foot greater than that of the tallest inbred strain.

Table III gives the yields of shelled corn per acre of several inbred types, together with the yields of many first-generation crosses. Many interesting points may be learned from this table, provided it is remembered that maize is greatly influenced by environmental conditions and therefore only populations grown in the same season should be compared with each other. For this reason the comparisons between first-generation hybrids and the unselected commercial types from which the inbred strains came are not to be given too great weight. On the other hand, there is such an enormous difference between many of the first-generation hybrids and the normal commercial varieties that the conclusion that the former are often better is perfectly just.

TABLE III.—*Comparative yields of inbred types of maize and their first-generation crosses.*

Variety.	Year grown.	Number of years inbred.	Yield in bushels per acre.	Comparison between F_1 and unselected commercial strains.
White dent No. 8	1908	....	121.0	121.0
Leaming dent No. 1-7	1908	3	62.0	88.0
No. (8×1-7), F_1	1908	....	142.0	142.0
Flint No. 5	1909	1	47.5	75.7
Flint No. 11	1909	1	44.2	48.0
No. (5×11), F_1	1909	....	76.3	76.3
Flint No. 5	1909	1	47.5	75.7
Leaming dent No. 1–6	1909	3	57.9	88.0
No. (5×1–6), F_1	1909	....	88.9	88.9
No. (5×1–6), F_1	1910	....	105.5	105.5
No. (5×1–6)–1, F_2	1910	1	54.1	54.1
No. (5×1–6)–2, F_2	1910	1	48.9	48.9
Starchy No. 10	1910	2	48.7	70.5
Leaming dent No. 1–6	1910	4	80.4	88.0
No. (10×1–6), F_1	1910	....	139.0	139.0

Tassels and Ears of an Almost Sterile Strain of Corn Isolated by Inbreeding.

(Photographed by Emerson.)

WATSON'S FLINT AND LONGFELLOW FLINT CORN INBRED TWO YEARS WITH F_1 HYBRID.

(All ears hand-pollinated.)

TABLE III.—*Comparative yields of inbred types of maize and their first-generation crosses*—Continued.

Variety.	Year grown.	Number of years inbred.	Yield in bushels per acre.	Comparison between F_1 and unselected commercial strains.
Leaming dent No. 1–7	1910	4	65.5	88.0
Sweet No. 19	1910	1	53.6	93.2
No. (1–7×19), F_1	1910	....	142.7	142.7
Leaming dent No. 1–9	1909	4	23.3	88.0
Leaming dent No. 1–12	1909	3	35.4	88.0
No. (1–12×1–9), F_1	1909	....	110.2	110.2
No. (1–12×1–9), F_1	1910	....	117.5	117.5
No. (1–12×1–9)–1, F_2	1910	1	102.2	102.2
No. (1–12×1–9)–4, F_2	1910	1	91.5	91.5
No. (1–12×1–9)–12, F_2	1910	1	91.5	91.5
Leaming dent 1–6	1911	5	27.7	....
Leaming dent 1–7–1	1911	5	25.4	....
Leaming dent 1–7–2	1911	5	41.3	....
Leaming dent 1–9–2	1911	5	26.0	....
Leaming dent 1–12–2	1911	6	2.0	....
Leaming dent 1–12–4	1911	6	2.0	....
No. (1–6×1–7–1), F_1	1911	....	75.6	....
No. (1–6×1–7–2), F_1	1911	....	58.3	....
No. (1–6×1–9–2), F_1	1911	....	31.6	....
No. (1–6×1–12–2), F_1	1911	....	10.2	....
No. (1–7–1×1–6), F_1	1911	....	58.8	....
No. (1–7–1×1–7–2), F_1	1911	....	41.3	....
No. (1–7–1×1–9–2), F_1	1911	....	51.5	....
No. (1–7–1×1–12–2), F_1	1911	....	16.9	....
No. (1–7–1×1–12–4), F_1	1911	....	60.2	....
No. (1–7–2×1–6), F_1	1911	....	57.7	....
No. (1–7–2×1–12–2), F_1	1911	....	63.5	....
No. (1–9–2×1–6), F_1	1911	....	37.3	....
No. (1–9–2×1–7–1), F_1	1911	....	46.2	....
No. (1–9–2×1–12–2), F_1	1911	....	3.6	....
No. (1–12–2×1–7–2), F_1	1911	....	76.9	....
No. (1–12–2×1–12–4), F_1	1911	....	24.5	....

Attention is called first to the fact that in combinations (5×1–6) and (1–12×1–9) both the first and second hybrid generations are grown in the same year. The first hybrid generation gives an enormous increase over the inbred types. The second hybrid generation is also much greater than the inbred strains, but recombination with the production of homozygotes has taken place, and this generation gives much lower yields than when the greatest possible heterozygosity existed as in the first hybrid generation.

Attention should next be directed to the results of 1911, when nearly all the possible combinations of the inbred Leaming strains were made. The yields of the inbred types given are those with one more year of inbreeding than the real parents of the first-generation hybrids. But considering the amount of previous inbreeding to which they had been subjected this probably makes but little difference. As stated before, the yields in 1911 were very much reduced by the unfavorable season, and this too must be given due weight in studying the yields. As a whole the combinations into which No. 1–7 was introduced were the best while those into which the poor type No. 12 was introduced are the poorest. The combination (1–7–1×1–12–4) was, however, a very good cross.

Possibly a question may arise as to whether the fine yields of the combination (1–12 × 1–9) in 1909 and 1910 and the poor yields of combination (1–9–2 × 1–12–2) in 1911 are not due to a difference in the behavior of a reciprocal cross. This is probably not the correct reason, for in general there is no difference in reciprocals. No. 1–12 was further inbred when the combinations grown in 1911 were made and this is probably the cause of their poor showing. In the earlier combination, No. 1–12 undoubtedly had a somewhat different gametic constitution than when the later crosses were made. Some essential factor may have been eliminated, therefore, during the further inbreeding. On the other hand, the whole explanation may lie in the poor season of 1911.

The marked increase in productiveness of the F_1 hybrid over the parent inbred types of maize is well shown in Plates II and III, while Plate IV illustrates the falling off in productiveness of the F_2 generation as compared with the F_1 generation from inbred types. Plate V serves to show the striking increase in vigor of the F_1 generation from a cross of pure lines.

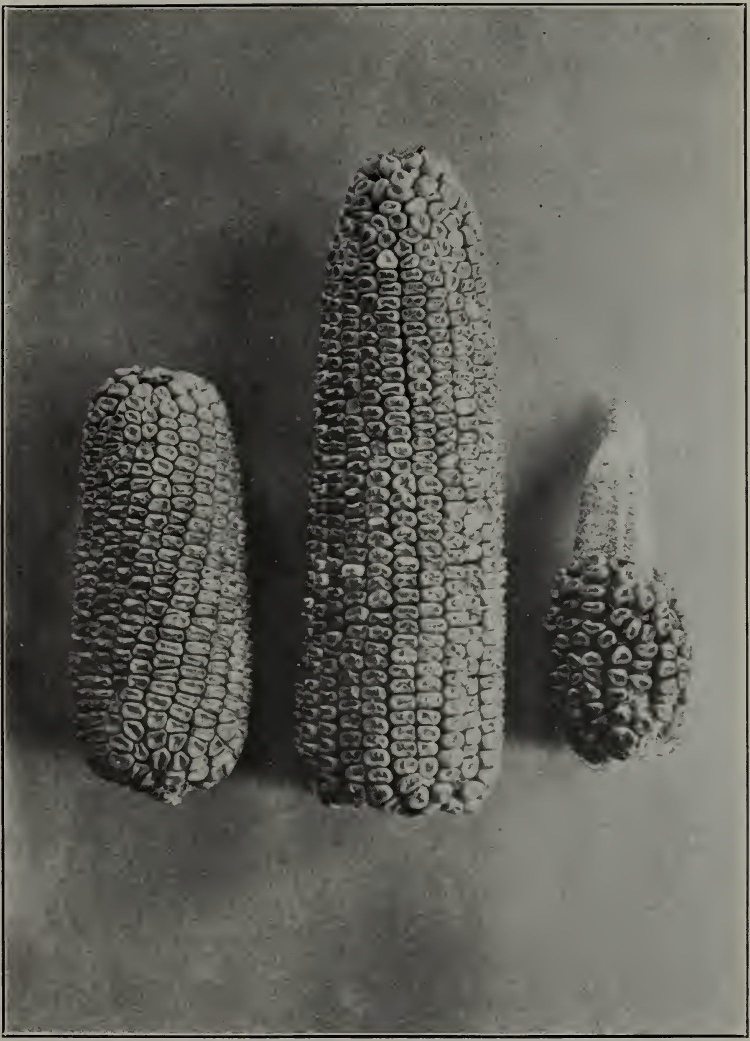
The logical conclusion from the facts brought out above is apparently that good inbred strains are better than poor ones in combination, but that good and poor strains crossed together may give very strong plants.

EXPERIMENTS ON SPECIES GENERALLY SELF-FERTILIZED.

As experimental material that contrasts well with maize, the genus Nicotiana was selected. This genus contains a large number of species and varieties, most of which have flowers adapted to self-fertilization. No doubt cross-fertilization sometimes occurs in most of them, but it is not the rule.

Seeds of several species and many varieties were obtained from various parts of the world through the kindness of a number of friends. The same species did not always arrive with the same name, and we have not been fortunate enough to have the aid of a Nicotiana specialist in their identification. We have, however, studied them in pure-line cultures during the past four years and have compared them with specimens in the Gray Herbarium of Harvard University. This gives us some confidence that the names used are in accord with the species as accepted and described by Comes in his "Monographie du Genre Nicotiana," Naples, 1899.

Many crosses have been made between different varieties within the two species *Nicotiana tabacum*, L., and *N. rustica*, L. Some of the varieties of *N. tabacum* have been practically identical as far as external appearance is concerned, although received under different names. When this has been the case, the results have been varied.



LEAMING DENT STRAINS OF CORN, NO. 9 (AT LEFT) AND NO. 12 (AT RIGHT), AFTER FOUR YEARS' INBREEDING, COMPARED WITH F_1 HYBRID (IN CENTER).

(All ears hand-pollinated.)

INBRED STRAINS OF LEAMING DENT CORN COMPARED WITH F_1 AND F_2 GENERATIONS.

(Grown in 1910.)

Strains 6 and 7 of Leaming Pure Lines of Corn (at Left) and F_1 Generation of Crosses 6 × 7-1 and 6 × 7-2 (at Right).

(Grown in 1911.)

FIG. 1.—NICOTIANA TABACUM VARIETY.

($\frac{1}{22}$ natural size.)

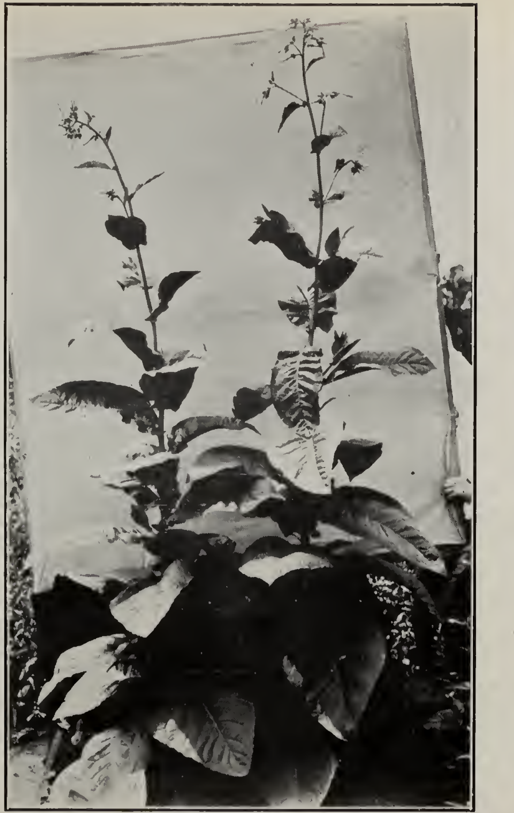


FIG. 2.—NICOTIANA TABACUM VARIETY × N. SILVESTRIS, F1 GENERATION.

($\frac{1}{30}$ natural size.)

FIG. 3.—NICOTIANA SILVESTRIS.

($\frac{1}{15}$ natural size.)

For example, two exceedingly similar varieties may give hybrids with no greater luxuriance of growth than the pure parent strains; other varieties as similar in appearance may give hybrids with as much as 25 per cent greater vigor than the average of the two parents. In this case the criterion of greater vigor is height of plant. If one accepts the old view that nonrelationship between the individuals used as parents is the reason for the increased vigor of the hybrids, there would be no logical reason why all such crosses should not show the same condition. If, on the other hand, the correct explanation is to be sought in the similarity or dissimilarity of the gametic constitution of the parents, it is quite evident that different crosses among varieties similar in external characters may behave in a different manner. Plants having a close genetic relationship with each other—that is, descendants of a previous cross—may be quite different in gametic constitution and therefore show an increased vigor in the F_1 hybrid; but genetically unrelated plants of practically the same gametic constitution may be obtained from different parts of the world under different names and not be expected to show an increased vigor in the hybrid.

An example of the amount of increase in height in crosses between *Nicotiana rustica brazilia* Comes and *N. rustica scabra* Comes, both obtained from Italy, is shown in Table IV.

TABLE IV.—*Height of crosses between Nicotiana rustica scabra (352) and N. rustica brazilia (349).*

Variety or cross.	Class centers in inches.																		
	24	27	30	33	36	39	42	45	48	51	54	57	60	63	66	69	72	75	78
349	4	10	22	14	7	....	....	....	....	....	....	....	....	....	....	....	....	....	....
352	....	....	....	....	....	....	2	1	5	11	16	17	6	....	....	....	....	....	....
352 × 349 F_1	....	....	....	....	....	....	....	....	....	1	3	0	5	5	5	6	1	1	....
349 × 352 F_1	....	....	...	....	....	....	....	....	....	....	....	3	5	2	4	6	5	1	2

The reciprocal crosses both showed a marked tendency to advance the mode until in each case it is higher than the highest plant of the taller parent. Different strains of *N. tabacum* var. "Sumatra," of *N. tabacum* var. "Havana," and of *N. rustica* var. brazilia, identical in external appearance, obtained both from the same locality and from opposite parts of the world, have also shown increased height when crossed. On the other hand, strains of *N. tabacum* varieties "Sumatra" and "Havana," from seed of plants grown in Connecticut, when crossed with like varieties from seed of plants grown in Italy have shown no increase in vigor. Accounts of other similar crosses could be given, but it seems unnecessary to multiply examples. We will therefore pass to a consideration of the specific crosses shown in Table V.

TABLE V.—*Condition of hybrids in crosses between species of Nicotiana.*

Cross.	Germination.	Fertility.	Condition of hybrid.
	Per cent.		
N. alata Lk. and Otto. var. grandiflora Comes:			
× N. forgetiana Hort. (Sand.).	100	Fertile	25 per cent in height; very vigorous and profuse in flowers.
× N. langsdorffii Weinm.	100	do.	105 per cent in height; vigorous and profuse in flowers.
× N. longiflora Cav.	100	Sterile	100 per cent in height; 100 per cent in vigor.
× N. paniculata L.	2	Slightly fertile.	80 per cent in height; 80 per cent in general vigor.
× N. tabacum L.	3	Sterile(?)	Very weak; seedlings died.
N. bigelovii Wats.:			
× N. alata grandiflora Comes.	0		
× N. longiflora Cav.	0		
× N. quadrivalvis Pursh.	100	Fertile	125 per cent in height; 100 per cent in general vigor.
× N. silvestris Speg. and Comes	0		
× N. tabacum L.	100	Sterile	120 per cent in height; 120 per cent in vigor; profuse in flowers.
N. forgetiana Hort. (Sand.):			
× N. alata grandiflora Comes.	100	Fertile	125 per cent in height; 130 per cent in general vigor; profuse in flowers.
× N. langsdorffii Weinm.	100	do.	160 per cent in height; 125 per cent in general vigor; profuse in flowers.
× N. tabacum L.	0		
N. glauca Grah. × N. tabacum L.	60	Sterile	80 per cent in height; less vigorous.
N. glutinosa L. × N. tabacum L.	(?)		Gärtner obtained plants higher and more vigorous than parents.
N. langsdorffii Weinm.:			
× N. alata Lk. Otto, var. grandiflora Comes.	100	Fertile	105 per cent in height; 100 per cent in vigor.
× N. bigelovii Wats.	100	Sterile	110 per cent in height; very vigorous.
× N. forgetiana Hort. (Sand.).	100	Fertile	110 per cent in height; 100 per cent in vigor; profuse in flowers.
× N. paniculata L.	0		
N. longiflora Cav. × N. alata Lk. and Otto. var. grandiflora Comes.	100	Sterile	100 per cent in height and general vigor.
N. paniculata L.:			
× alata Lk. and Otto, var. grandiflora Comes.	25	Slightly fertile.	95 per cent in height; rather weak.
× N. bigelovii Wats.	2	Sterile	100 per cent in height; 95 per cent in vigor.
× N. langsdorffii Weinm.	5	do.	15 per cent in height; very weak and stunted.
× N. longiflora Cav.	0		
× N. rustica L.	100	Partially fertile.	125 per cent in height; very vigorous and profuse in flowers.
× N. tabacum L.	5	Sterile(?)	Plants very weak and small.
N. plumbaginifolia Viv. × N. longiflora Cav.	100	Fertile	125 per cent in height; 110 per cent in general vigor.
N. quadrivalvis Pursh. × N. bigelovii Wats.	100	do.	110 per cent in height; 100 per cent in general vigor; profuse in flowers.
N. rustica L.:			
× N. alata Lk. and Otto, var. grandiflora Comes.	5		So weak that plants lived only about two weeks.
× N. langsdorffii Weinm.	80	Sterile(?)	110 per cent in height; 110 per cent in vigor; very profuse in flowers.
× N. paniculata L.	100	Partially fertile.	125 per cent in height; very vigorous; profuse in flowers.
× N. tabacum L.	5	Sterile	180 per cent in height; extremely vigorous; profuse in flowers.
N. silvestris Speg. and Comes:			
× N. longiflora Cav	0		
× N. tabacum L	100	Sterile	140 per cent in height; 120 per cent in vigor; profuse in flowers.
N. tabacum L.:			
× N. alata Lk. and Otto, var. grandiflora Comes.	2	do.	10 per cent of average of parents in height and in general vigor.
× N. bigelovii Wats.	100	Almost sterile.	120 per cent of average of parents in height and in general vigor.
× N. glauca Grah.	60	Sterile	85 per cent of average of parents in height and 80 per cent in general vigor.
× N. glutinosa L.	5		25 per cent of average of parents in height; Gärtner obtained plants more vigorous than parents.
× N. langsdorffii Weinm.	10	do	60 per cent of average of parents in height; 75 per cent in general vigor.
× N. longiflora Cav.	0		
× N. paniculata L.	1		Very small and weak; died before flowering.
× N. plumbaginifolia Viv.	0		
× N. silvestris Speg. and Comes	100	Sterile	135 per cent of average of parents in height; 120 per cent in vigor; profuse in flowers.

PLATE VII.

Fig. 1.—Nicotiana Rustica Texana.

($\frac{1}{8}$ natural size.)

Fig. 2.—Nicotiana Rustica Texana × N. Tabacum Variety, F_1 Generation.

($\frac{1}{26}$ natural size.)

Fig. 3.—Nicotiana Tabacum Variety.

($\frac{1}{18}$ natural size.)

FIG. 1.—NICOTIANA ALATA GRANDIFLORA.

(Twice scale of figure 2.)

FIG. 2.—NICOTIANA TABACUM VARIETY.

FIG. 3.—NICOTIANA ALATA GRANDIFLORA × N. TABACUM. WEAK AND STERILE HYBRID.

(Six times scale of figure 2.)

The voluminous data that have been collected on these hybrids have been condensed and approximated so that they include only facts germane to the matter in hand. Those crosses designated as not having germinated are crosses in which seed was obtained, but from which no plant was obtained from a planting of at least one hundred seeds. In some of these crosses the seed was poorly formed (without embryo) and one may say conclusively that they would never produce plants. Other crosses gave fully mature, perfect seed which did not germinate. Possibly the proper conditions for their germination were not obtained. At least it would be rash to conclude that all of the crosses of which the seed did not germinate would never produce plants under any conditions. But it is proper to say that some crosses are possible in which the hybrid plant reaches no further than the seed stage. A few hybrids, viz, *Nicotiana tabacum* × *N. paniculata*, *N. rustica* × *N. alata grandiflora*, etc., germinated and produced a few weak plants that died before flowering. There were still others that produced mature plants, but plants shorter than either parent and weak in character. By far the majority of the hybrids, however, were taller than the average of the parents and many were taller than either parent. The luxuriance of their growth was also such that they may be said to be more vigorous than either parent. Plate VI shows the result of a cross between *Nicotiana tabacum*, var., and *Nicotiana silvestris*.

One gets the idea from a survey of the crosses in this genus that there are (*a*) plants so different that they will not cross; (*b*) crosses that produce seed that contain no proper embryo; (*c*) crosses that produce seed with embryo, but which go no further than the resting stage of the seed; (*d*) crosses less vigorous than either parent; (*e*) crosses more vigorous than the average of the parents; and (*f*) crosses more vigorous than either parent. It seems probable, then, that actual fusion may take place between gametes either so different in character that the zygote can not develop or in which the male cell does not bring in the proper substance to stimulate development. On the other hand, when development does take place in a normal manner the great majority of cases show a stimulus greater in the hybrids than in the pure species. Compare Plate VII.

It might be supposed that the luxuriant development of many of these hybrids is due to their sterility, that is, due to the fact that no energy is used in seed formation. Such an idea was held by some of the earlier hybridizers, but was disproved by Gärtner. Nor is it justified by our own experience. Fertile crosses between plants differing in character either equal or exceed the parental vigor; sterile crosses may show a great increase in vigor or they may show a great diminution in vigor. Plate VIII represents a sterile hybrid showing decided diminution in vigor. But there need be no confusion in the interpretation of these facts. It is known that some plants are so unlike that there is mechanical or chemical obstruction to fertilization. In one case the stigmatic fluid may be poisonous to certain foreign pollen; in another case the pollen tubes can not penetrate the micropyle; sometimes nuclei do not enter the micropyle; frequently the two nuclei will not fuse. Such conditions absolutely prevent a cross. On the other hand, where crossing is possible, all of the physiological processes normal to the plant may not be carried out. The difficulty often lies in the maturation of the sex cells, the reduction of the chromatin, and the preparation for a new sexual act. In the proposed parent plants this has already taken place naturally. The male and female gametes are ready for fusion, and if nothing interferes this fusion takes place. But this does not mean that normal development can take place. Cell division may be so difficult that no embryo is formed, there being simply a pericarp formed by the reaction of maternal tissue to stimulation. Again, development of the embryo may take place, primarily because the difficulty of development is decreased through the nutrition furnished by the mother plant. But it may stop at this point. Thus it is obvious that where the parent plants are so different that normal somatic cell division can not take place, weak plants result even though they are heterozygous for many characters. If, however, cell division is normal we may believe that the vigor of the hybrid increases directly with the amount or the kind of heterozygosis present, without regard to whether the plant is sterile or fertile. Sterility, therefore, is often simply an inability to mature the sex elements properly, possibly because of mechanical obstruction to normal reduction of chromosomes differing widely in their character, and sometimes it is correlated with abnormal ontogeny.

We make the statement that hybrid vigor increases with the amount or with the kind of heterozygosis advisedly. The increased vigor may vary roughly with the number of heterozygous characters present, up to that limiting case where the action of other forces prevents or obscures their influence, or it may depend largely upon the quality of the characters that are heterozygous. This matter has not been determined; in reality it makes no difference with the thesis under discussion. It is an interesting problem, but can hardly be tested experimentally by crossing owing to the number of unknown characters that may be present in either a heterozygous or homozygous condition. The proof submitted here rests entirely upon the effects obtained by continued inbreeding as explained by the mathematical expectancy of homozygotes and heterozygotes under continued inbreeding.

One further point ought to be noted here. It has been shown that weak types are sometimes isolated from maize by inbreeding, their delicate constitution being due, it is assumed, to homozygosis of heritable characters that produce weakness and not to the mere fact of inbreeding. Does one obtain weak types in self-fertilized species? Undoubtedly such strains arise, but it is difficult to obtain examples because the weakness of individual plants is usually a physiological fluctuation due to external conditions and is not transmitted. This has been found to be true by growing seedlings from weak plants that have been self-fertilized. They usually give normal plants. Weak strains have been isolated, however, from *Nicotiana tabacum*, from *N. paniculata*, and from *N. attenuata* that continued to transmit their poor constitution. We may conclude, therefore, that weak strains arise in self-fertilized species, but are eliminated by natural selection.

THE CHARACTERS AFFECTED BY HETEROZYGOSIS

The term vigor has hitherto been used with the general meaning which the biologist readily understands. We will now endeavor to show in what plant characters this vigor finds expression. It is not an easy task because of the possibility of confusing the phenomenon of Mendelian dominance with the physiological effect due to heterozygosis. The confusion is due to a superficial resemblance only. Dominance is the expressed potency of a character in a cross and affects the character as a whole. A morphological character like the pods of individual maize seeds, or the product of some physiological reaction like the red color of the seed pericarp in maize may be perfectly dominant, that is, it may be developed completely when obtained from only one parent. Size characters on the other hand usually lack dominance or at best show incomplete dominance. The vigor of the first hybrid generation theoretically has nothing to do with these facts. This is easily demonstrated if one remembers that the increased vigor manifested as height in the F_1 generation can not be obtained as a pure homozygous Mendelian segregate, which would be possible if due to dominance. Furthermore, the universality with which vigor of heterozygosis is expressed as height shows the distinction between the two phenomena. If the greater height were the expression of the meeting of two factors $(T_1t_2 \times t_1T_2)$ both of which were necessary to produce the character, one could not account for the frequency of the occurrence. Nevertheless, in practice the confusion exists, and while we have considerable confidence in the conclusions drawn from our experiments, we have no intention of expressing them dogmatically.

It has been stated that the vigor due to heterozygosis is primarily an increase and an acceleration of cell division; in other words, an increased power of assimilation. This is first of all expressed by the increased size of the root system, a fact noticed by Kölreuter and Gärtner as quoted on page 9. This is the first noticeable difference, for the size of the cotyledons of the hybrid is largely influenced by the size of the maternal pericarp, yet there is a slight increase in the cotyledon size, as we have found in experiments with species of the genus Impatiens and with the tomato, *Lycopersicum esculentum*. Hybrid seedlings next show the increased vigor by their rapidity of growth tending toward an earlier maturity. This feature is the acceleration of cell division referred to above. Ultimately, however, there is not only acceleration but increased cell division, resulting in taller plants. Data supporting this fact have already been shown in papers on maize (East, 1911, 1911a). The increased size is entirely internodal. Neither in crosses between maize varieties nor between varieties of *Nicotiana tabacum* is there any tendency to increase the number of nodes. This stem growth is comparatively much greater than is increased leaf surface in the plants investigated (*N. tabacum*), although the latter can be definitely traced.

The size of the flower is not affected, at least not certainly. The fruit also does not seem to be affected where there is a small natural amount of cell division, as in the capsule of tobacco. In fleshy fruits like the tomato or eggplant there is a marked increase. This is probably true also of the large pomes and pepos, but this is only a surmise by analogy.

The increased vigor of the whole plant makes it possible for more flowers and fruit to be produced, as we have determined in tobacco and tomato. A more or less indeterminate inflorescence is always prolonged, which probably accounts for the increased size that is found in the ears of maize hybrids.

There are many less important plant characters upon which no data have been gathered, but the action of heterozygosis is known well enough to justify the former statement that it affects the amount and rapidity of assimilation as expressed by cell division.

THEORETICAL INTERPRETATION OF RESULTS.

At this point it may be well to stop, collect our facts, and discuss their theoretical interpretation, notwithstanding a certain repetition it will involve. We believe it to be established that—

(1) The decrease in vigor due to inbreeding naturally cross-fertilized species and the increase in vigor due to crossing naturally self-fertilized species are manifestations of one phenomenon. This phenomenon is heterozygosis. Crossing produces heterozygosis in all characters by which the parent plants differ. Inbreeding tends to produce homozygosis automatically.

(2) The phenomenon exists and is in fact widespread in the vegetable kingdom.

(3) Inbreeding is not injurious in itself, but weak types kept in existence in a cross-fertilized species through heterozygosis may be isolated by its means. Weak types appear in self-fertilized species, but are eliminated because they must stand or fall by their own merits.

The logical interpretation of all of these facts rests, we believe, on the acceptance of Johannsen's (1903, 1909) "genotype conception of heredity." This conception in turn is an extension of Weismannism[1] without Weismann's mechanistic speculations, supported by Mendelism. Johannsen (1911) gives the essential points of this conception in these paragraphs:

> The personal qualities of any individual organism do not at all cause the qualities of its offspring, but the qualities of both ancestor and descendant are in quite the same manner determined by the nature of the "sexual substances"—i. e., the gametes—from which they have developed. Personal qualities are then the reactions of the gametes joining to form a zygote; but the nature of the gametes is not determined by the personal qualities of the parents or ancestors in question. This is the modern view of heredity.
>
> The main result of all true analytical experiments in questions concerning genetics is the upsetting of the transmission conception of heredity, and the two different ways of genetic research, pure-line breeding as well as hybridization after Mendel's model, have in that respect led to the same point of view, the "*genotype conception*" as we may call the conception of heredity just now sketched.

A simple illustration of what is meant by the above statement is as follows: Suppose a maize with red pericarp (RR) be crossed with one with a colorless pericarp (rr). In the hybrid the gametes R and r are formed in equal quantities. By chance mating $1RR:2Rr:1rr$ are obtained. Now the homozygous dominant RR is exactly like the heterozygote Rr in appearance, but the one breeds true to red pericarp and the other again throws about 25 per cent white progeny. In other words, the gametic composition of the zygotes determines whether the resulting plants shall have ears with red or with colorless pericarps, but the fact that a plant has an ear with a red pericarp does not show what kind of gametes it will form.

The genotype conception of heredity, as stated before, rests on the noninheritance of somatic modifications and the general truth of Mendelism. The first part of the proposition now has almost universal support. All data point to a germ-cell-to-germ-cell hereditary transmission. In certain animals it has been demonstrated that there is an early segregation or setting apart of the material designed

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[1] One need become a Weismannian only so far as to agree with the observed facts which have shown that the transmission of acquired characters must be so relatively infrequent as to make the possibility negligible in experimental genetics and plant breeding.

to become the germ cells. This fact naturally has been proved in but few animals, but from it one must infer that in all metazoa there is a relative independence of soma and germ plasm undreamed of a few decades ago. In the higher plants no visible difference between germ plasm and soma plasm has been proved, yet the recent experiments of Baur and of Winkler on periclinal chimeras or false-graft hybrids have shown that one of the subepidermal layers is probably alone responsible for the sexual cells. In recent years few biologists have believed that surrounding conditions did not occasionally modify gametic structures. On the other hand, fewer and fewer investigators have maintained that any sort of somatic adaptation would impress the germ plasm with the ability to transmit the same modification.

The experimental work on the genotype conception of heredity has been largely a demonstration of the last statement. It has shown that in general fluctuations caused by ordinary environmental changes are not inherited. The idea involved is comparatively old. Vilmorin's promulgation of his "isolation principle" in plant breeding in the middle of the nineteenth century might be called its starting point. Vilmorin used the average character of a plant's progeny as the index of that particular plant's breeding capacity. This is the genotype conception, pure and simple. Since that time all plant breeding by selection which has been at all profitable has been done in this way, although the theoretical interpretation of the results obtained was unknown. This was given by Johannsen through his work upon barley and beans.

Since then corroborative results have been obtained by Jennings (1908, 1910) on Paramaecium, Hanel (1907) upon Hydra, Pearl (1909, 1911) upon fowls, Barber (1907) upon yeasts, Woltereck (1909) upon Daphnia, Jensen (1907) upon bacteria, East (1910a) upon potatoes, Love (1910) upon peas, and Shull (1911) and East (1911) upon maize. And no one to my knowledge has made a successful attack upon the position taken. It is true that attacks have been made by Pearson (1910) and Harris (1911), but their main argument is that the genotype theory is wrong, because it antagonizes the utterly erroneous biometric idea that heredity is measured only by the correlation between parents and progeny in somatic characters.

To be sure a caveat has been filed by Castle ("Heredity", New York, 1911) to the effect that unit characters so called can sometimes be modified by selection. This is no real criticism of the genotype conception of heredity, however, for Castle firmly believes in the generality of Mendelism and the general noninheritance of somatic modifications. It must simply be understood that, like most chemical compounds, characters are generally stable under ordinary conditions, but also like chemical compounds they may sometimes be modified. This modification then becomes a new character or is the old character in a slightly different form, depending on the point of view.

The second part of the proposition rests upon the law of segregation and recombination of gametic factors, which is the essence of Mendelism. Every day the generality of this law becomes more probable. Leaving out of consideration experiments on apogamous and parthenogenetic species almost every paper published since 1900 dealing with crosses between varieties fertile inter se in which qualitative differences have been studied has shown that factors representing these characters segregate in the germ cells of the hybrid and recombine in the next generation. The few exceptions have been papers dealing with characters evidently quantitative, treated from a biometrical standpoint and not proving or disproving anything.

Recently there have also been investigations (Emerson, 1910; East, 1910, 1911; East and Hayes, 1911; Lang, 1911, Tammes, 1911) showing that size or quantitative characters also segregate. Of course all selection experiments on cross-fertilized species using Vilmorin's isolation principle and the investigations just cited in support of Johannsen have really proved segregation and recombination of size characters, else strains differing in such characters could not be isolated from complex hybrids. The senior writer (1910), however, has shown how such segregation can be given a strict Mendelian interpretation by postulating absence of dominance and multiplicity of determinants affecting the same general characters. The experimental basis upon which it rests is the investigations of Nillson-Ehle (1909) upon oats and wheat and his own upon maize.

It is possible that there are many apparent exceptions to the law of segregation; it is even possible that practically there are real exceptions, but these exceptions are likely to be in the nature of changed conditions which modify the action of Mendel's law through new sets of conditions. Our meaning is shown by parallels in the domain of physics and chemistry, where certain laws act perfectly only under ideal conditions which are very often not fulfilled in nature. For example, De Vries (1907) states that Burbank's and Janczewski's bramble hybrids have bred true. Without any data upon which to base a critical judgment one does not know what to say, but taking the statement at full value, any number of conditions may cause this hybrid constancy without invalidating the law of segregation. There may be apogamy, all zygotes may not develop, selective fertilization may occur, or the action of the law may be opposed or suspended by other conditions of which we know nothing.

Personally we consider the genotype conception not as a theory but as a fact. Considering it as a fact, how does it aid the interpretation of the results obtained by inbreeding and by crossing inbred types of maize? Maize as a cross-fertilized species of great variability is in a constant state of hybridization. It is a collection of complex hybrids. Its usual mode of pollination through the agency of the wind keeps up this state of hybridization. Inbreeding, however, tends to produce homozygous types. As already shown, if one assumes equal fertility for all plants and that each plant lives and produces offspring in the n^{th} generation there is a ratio 2^n-1 pure dominants, 2 heterozygotes and 2^n-1 pure recessives for each allelomorphic pair.

This theoretical state of affairs may not occur for other reasons (as unpaired chromosomes) and the large number of allelomorphic pairs in a complex hybrid may prolong the time required for isolation of strains that are completely homozygous, but final isolation of strains completely homozygous is the goal toward which inbreeding tends. These completely homozygous strains are Johannsen's homozygous genotypes. Perhaps no one has ever isolated a real homozygous genotype, but strains homozygous for many characters are constantly being separated. This, indeed, is the sole function of selection.

Weismann assigned two purposes to the gametic fusion termed sexual reproduction; one being to mingle the hereditary characters carried by the two germ cells, the other to stimulate development of the zygote. This general statement was so obviously a fact that biologists were unanimous in its acceptance and two distinct lines of investigation have developed from it. Research concerning transmission phenomena has been almost divorced from the study of the physiology of development in its intimate connection with sexual reproduction. This separation, in view of the subject of this bulletin, seems unnecessary and unwise, for it may permit only a partial and distorted view of the results of reproduction. At any rate the data given here are of interest from both view points, since they deal with a purely physiological result brought about by a strictly morphological transmission phenomenon.

The hypotheses in regard to the way by which the act of fertilization initiates development are numerous, but since they are entirely speculative it is not necessary to discuss them here. The only conclusion that seems justified is that they are not immediately psychological or vitalistic in nature. Loeb's remarkable researches prove this. But whatever may be the explanation of the means by which the process is carried out, the statement can be made unreservedly that the heterozygous condition carries with it the function of increasing this stimulus to development. It may be mechanical, chemical, or electrical. One can say that greater developmental energy is evolved when the mate to an allelomorphic pair is lacking than when both are present in the zygote. In other words, developmental stimulus is less when like genes are received from both parents. But it is clearly recognized that this is a statement and not an explanation. The explanation is awaited.

The developmental stimulus is to a certain degree cumulative. In other words, the expression "the greater the degree of heterozygous condition the greater is the vigor of the resulting plant" roughly expresses the facts. This does not mean that the possession of certain allelomorphic pairs in a heterozygous condition is not more necessary than others of normal development. Castle and Little (1910), for example, have shown the probability that zygotes which are potentially homozygous yellow mice are formed but do not develop. Baur (1909) has shown that homozygous recessives of pelargoniums that lack the necessary mechanism for chlorophyll formation are formed but can live only a few days. Of course in the latter case there is actual absence of a physiological mechanism that is absolutely essential to development. Whether the condition is similar in the yellow mice is unknown. It is quite possible that lack of normal or presence of abnormal factors will account for many cases of improper development, but these facts must not be confused with the phenomenon under consideration. What we are concerned with here is that developmental stimulus due to heterozygosity increases roughly with the number of heterozygous allelomorphic pairs, even though some of these pairs may produce a much greater stimulus than others.

Inbreeding, then, tends to isolate homozygous strains which lack the physiological vigor due to heterozygosity. Decrease in vigor due to inbreeding lessens with decrease in heterozygosity and vanishes with the isolation of a completely homozygous strain. Moreover, these homozygous strains can be quite different from each other in natural inherent vigor. From a single strain of Leaming dent maize one isolated type is a good profitable corn after four generations of inbreeding, having yielded at the rate of 80 bushels per acre in 1910; another type is partially sterile and can hardly develop to maturity after five generations of inbreeding, and yielded in 1910 only 9.5 bushels per acre. Thus we see the true explanation of the apparent degeneration that so many observers have attributed to inbreeding per se.

When species that are naturally close fertilized produce variations that are weak and degenerate, they perish in the natural struggle for existence or are not allowed to propagate by man. Since only the experimental breeder sees the origin of degenerate strains of close-fertilized species (as we have done in the genus Nicotiana), biologists have left them out of their consideration and have concluded that some exception to the natural laws of physiology has been made in their favor so that they could stand the inbreeding for which they are naturally fitted. Nothing could be further from the facts. Species which through their flower structure must be self-fertilized produce as many degenerate strains as any species. They are produced, but they do not survive; they are lost and forgotten. Species which through their flower structure are naturally cross-fertilized, on the other hand, produce strains poor in natural vigor, degenerate strains, and they are kept from sight. They survive the scythe of natural selection; they are selected for propagation by man because they are crossed with other strains and are vigorous through heterozygosity. Inbreeding tears aside their mask. They must then stand or fall on their own merits. Those strains with a high amount of inherent natural vigor, due to gametic constitution, lose the added vigor due to a heterozygous condition, but are still good strains, ready to stand up forever under constant inbreeding. The poor strains that have had the help of hybridization with good strains, combined with the added vigor due to heterozygosity, are stripped of all pretense, shown in all their weakness, and inbreeding is given as the cause for their degeneracy. At least inbreeding has until recently been given as the cause, but it is hoped that the newer interpretation will now be accepted as logically interpreting all the facts.

Although the increased power of growth of hybrids and the decreased vigor attending inbreeding have not been recognized as the same phenomenon until the work of Shull and the senior writer, nevertheless there has been a so-called interpretation of the increased vigor of hybrids current among plant physiologists. It is the theory of rejuvenescence or renewal of youth in the protoplasm. Continued self-fertilization is thought to be comparable to vegetative reproduction and continued vegetative reproduction is supposed to bring about a senile condition in the protoplasm. This theory was borrowed from zoology, having long since been proposed by Bütschli to account for conjugation in protozoa. It can not be considered a theory that helps in interpreting the vigor of hybrids, for it tells us nothing. Moreover, it may be based upon wrong premises. It is not at all certain that conjugation is an absolutely necessary phenomenon. Woodruff (1911) has demonstrated that protozoa can be kept in healthy condition without conjugation for at least 2,300 generations. Jennings has been unable to make certain genotypes of Paramaecium conjugate. Nuclear fusions sometimes occur in some of the ascomycetes and basidiomycetes, but in general these fungi reproduce asexually and possibly have produced hundreds of species in this manner. In the higher plants there are many species in which either no seed is produced or sexual propagation is seldom resorted to, and yet they seem to be in no danger of degeneration. Among them may be mentioned the banana, hop, strawberry, sugar cane, and many of the grasses. There are also certain apogamous genera, such as Taraxacum and Hieracium, that are exceedingly vigorous. From these facts it is reasonably conclusive that sexual reproduction may be a benefit, but is not a necessity.

Keeble and Pellew (1910) have recently suggested that "the greater height and vigor which the F_1 generation of hybrids commonly exhibit may be due to the meeting in the zygote of dominant growth factors of more than one allelomorphic pair, one (or more) provided by the gametes of one parent, the other (or others) by the gametes of the other parent." We do not believe this theory is correct. The "tallness" and "dwarfness" in peas which Keeble was investigating is a phenomenon apparently quite different from the ordinary transmissible size differences among plant varieties. Dwarf varieties exist among many cultivated plants, and in many known cases dwarfness is recessive to tallness. It acts as a monohybrid or possibly a dihybrid in inheritance, and tallness is fully dominant. Varietal size differences generally show no dominance, however, and are caused by several factors. Transmissible size differences are undoubtedly caused by certain gametic combinations (East, 1911), but this has nothing to do with the increase of vigor which we are discussing. The latter is too universal a phenomenon among crosses to have any such explanation. Furthermore, such interpretation would not fitly explain the fact that all maize varieties lose vigor when inbred.

EXTENSION OF THE CONCLUSIONS TO THE ANIMAL KINGDOM.

Can the conclusions in regard to heterozygosis be extended to animals? The answer is affirmative as far as an interpretation of the known facts is concerned. No experimental attack from the standpoint taken in this paper has been made, but the older work furnishes many data that readily fit this view. As a matter of fact, however, it is questionable whether it is necessary to make formal proof in the matter. Sexual reproduction has undoubtedly arisen several times in the vegetable kingdom and at least once independently in the animal kingdom. Why or how it arose, one need not inquire; having arisen, the purposes served are essentially the same if the similarity of the methods is an argument. The duplex nature of organisms, the halving of the chromatin and the production of simplex cells at the maturation of the sex cells, the fusion of two simplex cells as the starting point of a new organism, the general result of this fusion in the matter of development, and the transmission of heritable characters, are so similar in their main points that it would be quite wonderful if the process both in plants and animals did not now fulfill like requirements.

Since our conclusions are based upon the generality of Mendelism, which has been rendered highly probable by the multiplicity of zoological researches, it seems only necessary to show that heterozygosis in animals does cause (or accompany) an increase in vigor. It is easier to do this than to attack the still widespread belief that inbreeding is injurious per se. We have seen fertile crosses between different varieties of cattle, of swine, of sheep, and of domestic birds that were more vigorous than either parent. There are several swine raisers in the Middle West who make a practice of selling only first-generation crosses on account of their size. A number of very vigorous sterile hybrids of both domestic and wild animals might also be cited, but with these crosses a complication is encountered. In plants we found that the presence or absence of normal sexual organs made little if any difference in the amount of vigor induced by heterozygosis. In animals the case is undoubtedly different. From their very mode of development—animals being closed forms and plants open forms—internal secretions play a great rôle. And it is a matter of common knowledge that castration, in vertebrates at least, causes an extraordinary development of the body. In the human race this development is especially noticeable in the femur bones, so that Havelock Ellis states that the eunuchs of Cairo can be readily picked out of a crowd by their great stature. It is obvious, therefore, that there are two causes of vigorous somatic development, elimination of sexual organs and heterozygosis. In sterile hybrids, therefore, one can not say how much of the induced stimulation is due to each cause, but in fertile crosses there is no question about its source.

It is much more difficult to argue against the supposed injurious effects of inbreeding. Abhorrence of incest, which probably had a religious origin among our ancestors, is so difficult to eradicate from our minds that judgment is made before the facts are heard. This belief is not universal in the human race if Westermarck, the greatest authority on the history of marriage, is to be trusted, but the retort is made that the races that approve incestuous unions are low in intelligence. The answer does not prove anything, however, as low races with both beliefs are found, and, furthermore, as disapproval of incestuous relations is both religious and esthetic, it would only be expected in races of some intelligence. Nor is the answer germane, for it is not shown that incestuous tribes are less well developed physically than related tribes with different customs, which is the real matter under examination.

But let us confine the discussion to the lower animals. If this is done there are two things to consider, the closeness of matings and their result. The statement is often made that self-fertilization in plants is a much closer sexual relationship than can obtain in bisexual animals. With a germ-to-germ transmission conception of heredity it is doubtful if this is true. After a wide cross, a self-fertilized plant of the F_1 generation produces markedly different progeny, due to recombinations of gametic factors. After continuous self-fertilization for many generations, the gametic factors tend to become homozygous and their matings are close in relationship. Thus it is perfectly clear that it is not kinship of the two organisms furnishing the sex cells that determines the closeness of the mating, but the similarity of the constitution of the cells themselves. There is no a priori reason, therefore, why bisexual animals may not be bred as thoroughly in-and-in as plants.

On this account the statement must be made very emphatic that investigations such as studies of cousin marriages in the human race amount to nothing. A cousin marriage may be a wide cross, it may be very narrow.

There is a possibility that has not been mentioned, however, that may prove to be an essential difference between the reproduction of bisexual animals and hermaphroditic plants. There is no question but that sex in the higher animals is essentially Mendelian in its behavior. There is no necessity of tying its interpretation to the chromosomes or to the accessory chromosome in particular. Castle's (1909) simple explanation that the female is gametically[1] the male plus a theoretical X factor has interpretated so many facts that its correctness—possibly somewhat modified—is highly probable. Under this interpretation one sex is always heterozygous. No similar explanation has been advanced to account for hermaphroditism. Possibly the same thing accounts for the differentiation into microgamete and macrogamete in plants, although not accompanied there by somatic changes. Since we are ignorant of the facts in plants, we can not say that sex furnishes a real reason for believing bisexual animal matings

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[1] Note the words "gametically the male." This is not at all the same thing as saying *the male plus* something else. The X may produce many important changes during ontogeny.

There are two classes of facts; in one the male is homozygous, having no X factors, while the female has one. In the other the male is heterozygous, having one X factor, while the female is homozygous, with two X factors The human race probably belongs to the second type.

less incestuous than plants. The facts are simply given for what they are worth.

We are now ready to take up the actual effect of inbreeding in animals. In the statements of Darwin's correspondents we find throughout the tendency to mix esthetic feelings and facts. But here and there an independent observer maintained that breeding good stocks in-and-in had no evil effect. Undoubtedly there is sometimes a slight loss in vigor (we should say vegetative vigor as we have done in plants, because constitutional vigor is not lost), but there is no degeneration. On the other hand, there is segregation toward homozygous strains, and these strains differ in constitutional vigor. The greatest breeds of horses, cattle, swine, and sheep have been developed by in-and-in breeding. Breeders have worked for homozygous strains, for they desired strains that bred true. Inbreeding has been accused of producing everything undesirable in many of these strains, but the accusations are extremely illogical. Consider one or two examples. The race horse has undoubtedly been inbred more than the draft horse. Did inbreeding produce the nervousness and delicate constitution of the former? Certainly not. It is absolutely essential that the race horse be nervous. It has been thus selected for generations. Again, the delicate constitution of the Boston terrier or even the toy terrier is pointed out as the effect of inbreeding. We doubt very much if there has been any more inbreeding in the case of the Boston terrier than with the St. Bernard, but the selective ideals have been quite different.

The necessity for heterozygosis may be very different in various species of animals. In some the stimulus to zygotic development may be insufficient when like germ cells conjugate; in others, it may produce normal development. Weismann has made much of the fact that hermaphroditic animals are always cross-fertilized at times. It may be necessary in these species or it may be coincidence. Possibly hermaphroditic species will be found that are always self-fertilized yet retain their vigor even as in plants. At any rate Weismann's arguments seem to have little force, considering the widespread prevalence of parthenogenesis in the animal kingdom. It seems reasonable to conclude that in animals as in plants cross-fertilization may be advantageous but is not a necessity.

The actual experiments of Crampe (1883), Ritzema Bos (1894), and Von Guaita (1898) on mammals, of Fabre-Domengue (1898) on birds, and of Castle et al (1906) on the fly *Drosophila ampelophila* Low may all be interpreted in this way. Fertility was decreased in some strains. Those strains needed the stimulus due to a certain amount of heterozygosis for their proper development. Other strains were perfectly fertile in spite of inbreeding. Sometimes combinations of hereditary characters resulted in relatively weak strains; other combinations of characters gave strong strains. In no case was there absolute and universal degeneration due directly to inbreeding.

As a final example of the simple way in which these experiments on animals fit the heterozygosis theory, we will take a case that puzzled Weismann (1904). Nathusius allowed the progeny of a Yorkshire sow to inbreed for three generations. Weismann says: "The result was unfavorable, for the young were weakly in constitution and were not prolific. One of the last female animals, for instance, when paired with its own uncle, *known to be fertile with sows of a different breed,* produced a litter of 6 and a second litter of 5 weakly piglings; but when Nathusius paired the same sow with a boar of a small black breed, which boar had begotten 7 to 9 young when paired with sows of his own breed (the black breed evidently near homozygous through close breeding), the sow of the large Yorkshire breed produced in the first litter 21 and in the second 18 piglings."

VALUE OF HETEROZYGOSIS IN EVOLUTION.

Before undertaking to discuss the part that heterozygosis may have played in evolution, emphasis must be laid upon one point of criticism directed against almost all speculative evolutionary philosophy. Unconsciously, perhaps, many of the conditions that are widespread among living forms have been spoken of as having been selected to continue their existence in nature because they are indispensable to the organism. This is certainly untrue. One has only to recall other epochs of geology to appreciate the fact. The huge reptiles of the Cretaceous period were long in developing their peculiar specializations, yet they were swept away. In a present-day post-mortem we can assign many reasons why they were eliminated from the organic world, but if their characters were so unfit for their environment, how did they come to be developed? It is said the environment changed and left them too specialized for adaptive response. This is plausible enough, but, nevertheless, possibly untrue.

Must we not be just as skeptical about the question of sexual differentiation? It has arisen several times; it has persisted. Having arisen, it undoubtedly has a function. Perhaps it was necessary; perhaps it was a fundamental blunder, as was once humorously stated. Speculation is, of course, futile. We merely wish to point out that in discussing a function intimately connected with sexual reproduction it is absolutely unnecessary to suppose that sex fulfills a protoplasmic necessity or demand.[1] We do not say that the belief is untrue, but that it is not known to be true and therefore should not be treated as a fact.

In other words, electric drills and hammers are very useful in building a bridge, but good bridges have been built without them. Sexual reproduction serves a purpose, but several of the most vigorous genera of our higher plants have given it up. It is evidently unnecessary to them. We must cast a vote, therefore, against the belief in the rejuvenescence theory of sexual reproduction. Furthermore, we believe that any hypothesis in which an endeavor is made to twist the phenomena attending sexual reproduction into requisites indispensable to the evolution of all species possessing it is without basis. All one can do is to suggest how it may have been beneficial at times to some species.

Transmissible variations are produced in great numbers by apogamous genera such as Taraxacum and Hieracium, so that sexual reproduction is not the cause of variation. Johannsen's (1906) and many other pedigree-culture studies have shown that it presumably never increases variation. But it does permit recombination of the gametic factors of the parents, and this has no doubt been of great service in evolution. Galton and Quetelet (Weismann, 1904) have argued that the intercrossing thus allowed is a means of keeping the species constant, but even with the old idea of blended inheritance this seems to us to be an exaggeration. Greatest constancy in the actual descendants, if new heritable variations are disregarded, would come from asexual reproduction. If the species group is considered as a whole and comparatively free from competition, a great amount of intercrossing—as in a naturally cross-fertilized strain—would help toward a general fixation of type, even though it did not contribute toward the production of homozygous factors; but if a rigid competition is allowed, new and better combinations of characters would replace the old. Perhaps this matter may be made clearer by an illustration drawn from our maize studies. Height is a complex due to many contributing factors. Some of them are probably correlated in inheritance, but a sufficient number are transmitted independently to give the

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[1] Vitalistic interpretations of the origin of characters, though largely confessions of ignorance of ultimate causes, deserve consideration for calling attention to that fact; yet one must admit that if everything is accounted for by some "perfecting principle" this creative force has made many trials and errors. Of course things do not just happen. The chemist sees certain series of compounds give similar reactions under like conditions, while other series give other reactions under those conditions. More complex chemicals under the general term protoplasm probably act in the same manner and produce variations through their reactions. Some of these variations are widespread—that is, they are general reactions; others are less general—that is, they are specific reactions. Personally this analogy helps in the conception of certain orthogenetic phenomena, but the conception leads back to the same blank wall of ignorance. The vitalist and the believer in mechanico-chemical theories reach the same point, but the latter is pleased if he is able to reduce a series of facts to the shorthand of a formula; the former is worried because knowledge stops at the most interesting place.

example validity. There is no dominance, and when two individuals differing in stature are crossed there is a blend in the first hybrid generation. There is a real segregation, however, resulting in an increased variability in the F_2 generation. In the F_1 generation there is a normal frequency deviation due to noninherited fluctuations. In the F_2 generation there is a similar curve, but with greater variability, due to fluctuating variability plus the variability due to the recombination of gametic factors. This condition of affairs tends toward the maintenance of a general mean in height, but this mean is false. It is false because the modal class which Galton and Quetelet took to be the type toward which the species is tending actually contains more heterozygous individuals and individuals heterozygous for more factors than any other. An individual selected from this class is less likely to breed true than one selected from the extremes. Cross-fertilization, therefore, may tend toward the production of a mean that gives falsely an appearance of fixity of type.

This preliminary discussion has necessarily been rather long in order to have a basis for considering the part that heterozygosis may have played in evolution. We shall confine ourselves to the higher plants, although we think a portion of the statements made are equally true when applied to animals. It can hardly be doubted that heterozygosis did aid in the development of the mechanisms whereby flowers are cross-fertilized. Variations must have appeared that favored cross-fertilization. These plants producing a cross-fertilized progeny would have had more vigor than the self-fertilized relatives. The crossing mechanism could then have become homozygous and fixed, while the advantage due to cross-fertilization continued. But was this new mechanism an advantage? It must have been often an advantage to the species as a whole. In competition with other species, the general vigor of those which were cross-fertilized would aid in their survival. But the mechanism may not have been useful in evolving real vigor in the species, because of the survival of weak strains in combination. In self-fertilized species, new characters that weakened the individual would have been immediately eliminated. Only strains that stood by themselves, that survived on their own merits, would have been retained. On the other hand, weak genotypes in cross-fertilized species were retained through the vigor that they exhibited when crossed with other genotypes. The result is, therefore, that self-fertilized strains that have survived competition are inherently stronger than cross-fertilized strains. On this account weak genotypes may often be isolated from a cross-fertilized species that as a whole is strong and hardy.

VALUE OF HETEROZYGOSIS IN PLANT BREEDING.

First-generation hybrids of many economic plants give a yield sufficiently greater than pure strains to pay for their production and leave a profit. This is true only of crops where crossing is easy and where profit is made from accelerated and increased cell division or number of fruits. In general, it is not true where the selling price is greatly increased by the possession of some special quality. As Collins has remarked, value may at times accrue also from the fact that a plant breeder who has found a great increase in yield from growing the first hybrid generation of a particular cross may keep the parents a secret and maintain a justly remunerative business by selling hybridized seed or seedlings. A few suggestions as to the crops to which this method may be applied are given below.

MAIZE.

Maize is our most important field crop, and an increase of one bushel per acre to the average yield would add many millions of dollars annually to the nation's resources. The methods now in general use for its improvement all follow Vilmorin's isolation principle. Progeny-row tests are grown from individual ears. This means that good strains are isolated, but it also means that the longer selection is carried on the nearer is a homozygous condition approached. Thus the increased stimulus due to heterozygosis is lost. Since from both Shull's tests and our own, strains made almost homozygous by artificial inbreeding have yielded as high as 250 per cent increase over the average of the parents, this stimulus is not to be lightly disregarded. Of course these tests were made with strains so nearly homozygous that they gave very low yields. But we have obtained yields of ear corn very much higher than are ever given on land of like fertility by commercial types. Shull (1909) has therefore suggested that near-homozygous strains be produced by self-fertilization, the best combination determined by experiment, and hybridized seed of this combination sold. This procedure is undoubtedly the best in theory, because the greatest degree of heterozygosis is thereby obtained. Perhaps it can be made practical, but we are afraid very few commercial men would undertake it.

As a method whose practicability outweighs its theoretical disadvantage, the senior writer (East, 1909) has suggested that combinations of commercial varieties be made, testing them until the most profitable combination is found. Since maize is monœcious, this method can be used on a large scale at a small cost. It is only necessary to take two varieties, *A* and *B*, plant them in alternate rows, and detassel all of the plants of one variety. The seed gathered from this detasseled variety is all crossed seed and will give, in general, a greater yield than the average of the two parents. Crossed seed can be produced in this manner at an additional cost over natural seed of not over 9 cents per bushel. If it averages two bushels per acre increase in yield, the producer can sell it at one dollar advance over natural seed and still allow the buyer a good profit. The method is given in greater detail in another paper (Hayes and East, 1911).

This plan we thought original, but Collins (1910) has shown that it is comparatively old. It has been suggested time and again without gaining a foothold in agricultural practice. Let us hope that the time is now ripe for a scientific method to be understood, appreciated, and used.

It is fortunate that we have at hand data from many agriculturists showing the value of using first-generation hybrids in maize. They are very convincing. We will not discuss them in detail, but refer the reader to Collins's paper (1910). We may say, however, that the following researches have shown that a commercial use of the method is possible: Beal at the Michigan Experiment Station in 1880, Ingersoll at the Indiana Experiment Station in 1881, Sanborn at the Maine Experiment Station in 1889, Morrow and Gardner at the Illinois Experiment Station in 1892, Shull of the Carnegie Institution Station for Experimental Evolution in 1909, East at the Connecticut Experiment Station in 1909, Collins and his assistants of the United States Department of Agriculture in 1910, Hayes and East at the Connecticut Experiment Station in 1911, and Hartley and his assistants of the United States Department of Agriculture in 1912.

TRUCK CROPS.

In some truck and garden crops, such as beans and peas, the difficulty of making artificial crosses absolutely precludes a commercial use of the stimulus due to heterozygosis. Other crops, such as pumpkins and squashes, are too plentiful and cheap to be worth the trouble. Besides, these crops are so often crossed naturally that they are always more or less heterozygous. On the other hand, there are garden crops that are in demand at all seasons of the year and are grown under glass during the winter with profit. Some of them are easily crossed and will pay for their crossing. As examples, tomatoes and eggplants may be cited. Both are easily crossed and are worth crossing. We grew a cross between Golden Queen and Sutton's Best of All tomatoes in 1909. It outyielded both parents. Further, we are informed that several unpublished experiments at the New York Experiment Station by Wellington have shown that crossed seed is worth its production.

Eggplants have another advantage that should be mentioned. Varieties exist whose fruits are so large that the buyer does not care for them, the seller makes no profit, and the plant produces a very limited number. Other varieties have very small fruit. Now fruit size is intermediate in the first hybrid generation, while the number produced is increased and the time of ripening advanced.

PLANTS REPRODUCED ASEXUALLY.

The one type of plants where heterozygosis has been utilized, though not purposely, is that class which is reproduced asexually by cuttings, grafts, etc. Potatoes and grapes are good examples. Commercial varieties are always hybrids, and the reason, we think, is because the hybrids yield so profusely. The cross is made and the best plant of the first generation is simply multiplied indefinitely by division. This method could be applied more generally to bush fruits, such as gooseberries, raspberries, blackberries, etc., and to the larger fruits, like apples, pears, and peaches.

FORESTRY.

There is one other class of economic plants where it seems possible to make a practical use of heterozygosis. We refer to trees used for lumber. Many plans for breeding forest trees have been suggested, yet we have never seen the use of first-generation hybrids suggested. This omission seems strange, for as early as 1855 (Darwin, "Animals and Plants," vol. 2, p. 107) M. Klotzsch crossed *Pinus sylvestris* and *nigricans, Quercus robur* and *pedunculata, Alnus glutinosa* and *incana, Ulmus campestris* and *effusa* and planted the crossed seeds and seeds of the pure parent species in the same place and at the same time. The result was that after eight years the hybrids averaged one-third taller than the parent trees. Further, the quick-growing hybrid walnuts produced by Luther Burbank undoubtedly owe that valuable quality to heterozygosis.

A large amount of experimental work will be necessary before definite recommendations can be made as to what species can be crossed, what result may be expected, and what extra cost must be allowed for the production of hybrid seed. It is perfectly evident that hybrid seed will be impossible in many cases, and even where hybrids can be produced comparatively few can be crossed at a small enough cost to make the scheme a commercial success. On the other hand, we have no doubt that with many good lumber trees crossing would be found easy and hybrid seed could be sold with a wide margin of profit both to producer and to forester.

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PROFESSOR E. M. EAST

NEW YORK
1912

[Reprinted without change of paging, from the AMERICAN NATURALIST, 1912.]

[Reprinted from THE AMERICAN NATURALIST, Vol. XLVI., Nov., 1912.]

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PROFESSOR E. M. EAST

BUSSEY INSTITUTION, HARVARD UNIVERSITY

As I understand Mendelism[1] it is a concept pure and simple. One crosses various animals or plants and records the results. With the duplication of experiments under comparatively constant environments these results recur with sufficient definiteness to justify the use of a notation in which theoretical genes located in the germ cells replace actual somatic characters found by experiment. This is done wholly to simplify the description of the experimental results. If one finds that the expression $DR \times DR = 1DD + 2DR + 1RR$ adequately represents the facts in numerous breeding experiments, he is then able to use the knowledge and the expression in predicting the results of other similar experiments. Mendelism is therefore just such a conceptual notation as is used in algebra or in chemistry. No one objects to expressing a circle as $x^2 + y^2 = r^2$. No one objects to

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[1] I do not speak here of the new biological facts discovered by Mendel or by his followers. Facts are always facts. Alternative inheritance and character recombinations were important facts, but I think no one will deny that the greatest value of Mendel's facts arose from the mathematical treatment he gave them. This mathematical notation remains conceptual just as does the chemical formula, but it must have as much basis of fact as there are pertinent facts extant.

saying that $BaCl_2 + H_2SO_4 = BaSO_4 + 2HCl$. No one should object to saying that $DR + RR = 1DR + 1RR$. We push things into the germ cells as we place the dollars in the magician's hat. Hocuspocus! They disappear! Presto! Out they come again! If we have marked our money we may find that that which appears from the magician's false-bottomed hat is not the same as that which we put in. But it looks the same and is good coin of the realm. We have a good right therefore to poke our characters into the germ cell and to pull them out again if by so doing we can develop—not a true conception of the mechanism of heredity—but a scheme that aids in describing an inheritance. We may do this even as we may use algebraical and chemical notations, if we remember that $x^2 + y^2$ does not give us a circle, that a chemical equation does not represent a true reaction or prove the atomic theory, that we have not pulled something new and astonishing out of the germ cell, that a unit factor represents an idea and not a reality, though it must have a broad basis of reality if it is to describe a series of genetic facts.

The facts of heredity that one describes in the higher organisms are the actual somatic characters, variable things indeed, but still things concrete. Their potentialities are transmitted to a new generation by the germ cells. We know nothing of this germ cell beyond a few superficial facts, but since a short description of the breeding facts demands a unit of description, the term unit factor has been coined. As I hope to show, a factor,[2] not being a biological reality but a descriptive term, must be fixed and unchangeable. If it were otherwise it would present no points of advantage in describing varying characters. The only obvious reason for poking it into the germ cell is to distinguish thus the actual parent (the cell) from the putative parent (the carrier).

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[2] I hope this statement is not confusing. The term factor represents in a way a biological reality of whose nature we are ignorant just as a structural molecular formula represents fundamentally a reality, yet both as they are used mathematically are concepts.

If we forget ourselves and begin to speak of unit factors as particles, only a confusion follows similar to that caused by Nägeli, Spencer and Weismann. Nothing is gained and even facts are obscured.

The Scope of Mendelism

How far may we carry this conceptual notation? My answer is: just as far as the notation interprets the facts of breeding and is helpful. Interest in the scope of Mendelism is now focused on two phases, complete and partial coupling and the interpretation of so-called size characters. Complete coupling in the transmission of characters apparently non-related has been shown in a large number of cases. Perhaps those best worked out in animals are the sex-coupled or sex-limited characters observed by Morgan in *Drosophila*. In plants, cases observed by Emerson and by Bateson and his coworkers are equally clear. Emerson has shown beyond a reasonable doubt that the characters he describes are independent of each other, and can not be represented by one factor. Bateson has recently corroborated the observation on other characters. Besides this phenomenon, Bateson has discovered partially coupled characters. All three of these writers, have subsidiary hypotheses to account for their facts. Bateson, when discussing perfect couplings, merely says that the characters come out in F_2 coupled in the way they went in in the grandparents, which naturally is only a restatement of the facts. Morgan and Emerson deal in pictures of carrying bodies. Both of their theories fit their own facts as they necessarily would. Emerson and, I may say, myself believe Morgan's theory incompatible with that of Emerson. Morgan believes his theory adequate for both cases. Without discussing the merits of these particular hypotheses I think it is agreed that some characters do go into the F_1 generation and come out from it together that are in other cases independent. The importance of the phenomenon is greater than the theory at present.

It has been questioned whether one has the right to continue to couple characters in large numbers to interpret facts, because by proper coupling one may interpret almost any fact, and place himself in a logically unassailable position. But this is no reason for not coupling factors as much as one pleases if it is helpful and if all of the facts fit. *A propos* of this statement I might say that I have recently remade the historical old cross first made by Kölreuter in 1760, *Nicotiana rustica* × *Nicotiana paniculata*. These species differ in many details—habit of growth, size, shape and hairiness of leaf, inflorescence, and size and shape of flower and fruit. Both of the parent species have been reproduced exactly from a partially fertile F_1 in a total number of less than 200 F_2 plants. One may formulate an hypothesis of selective elimination of gametes combined with selective fertilization that helps to describe the facts, but unless large numbers of factors are coupled together I believe it to be impossible to account for all the facts by the usual Mendelian notation.

Before leaving this subject it might be mentioned that Bateson's theory, originated to account for partial coupling, keeps the idea of factors segregating from their absence, but instead of A and a being formed in equal quantities as in "regular" Mendelian notation, they are to be formed in series represented by the scheme $n — 1AB : 1AB : 1aB : n — 1ab$. I do not believe one should hasten to accept this description, although Bateson's F_2 generation facts certainly fit and have been recently supported by Baur. My reason for making this statement is that as yet Bateson's F_3 facts do not fit the theory. Some of them would even make necessary two or more different kinds of factorial distribution in the same plant varieties. On this score the helpfulness of our notation[3]

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[3] Here is a good illustration of the Mendelian notation as a concept. Supposing the gametic distribution $n — 1AB : 1Ab : 1aB : n — 1ab$ were to fit all the facts in the case, then no one could object to its use. If it were to be demonstrated that segregation occurred at the reduction division, however, the scheme no longer fits the facts and must be abandoned.

is impaired and this is the only excuse for its existence. Furthermore, while it has not been proved that the phenomenon we call segregation occurs at the reduction division, the presumption is in favor of that view. The work of Webber, Correns, Lock, Emerson and myself on *Xenia* in maize indicates that segregation does not take place immediately after reduction, while the work of the Marchals on regeneration in mosses indicates that it does not take place before reduction.

Now to turn to the kinds of variation that may be described by the Mendelian notation. Owing to its youth, we can all remember how we wondered, as each new case came up, whether Mendelian phraseology would fit. Since qualitative characters were the ones that could be divided into definite categories they were the ones attacked. One by one they were analyzed. The phraseology did fit. Qualitative characters however form a very small proportion of the characters in animals and plants. The numerous characters are the quantitative, the size characters. If Mendel's law is to be worth anything as a generality, therefore, it must describe the inheritance of these characters.

To some of us Mendel's law from the first seemed destined to be a notation generally useful in describing inheritance in sexual reproduction. This conclusion was indicated by the simple fact that Mendel's law described many cases in both the animal and the vegetable kingdom. It was inconceivable that this should be the result of coincidence. It was therefore still more inconceivable that only a small portion of the facts in each kingdom should come under the scope of Mendelism.

A basis for the inclusion of quantitative characters was obtained when Nilsson-Ehle and the writer showed that certain qualitative characters gave ratios of 15:1, 63:1, etc., in the F_2 generation, and in other ways behaved so that they might be described only by assuming more than one independent gametic factor as the germ cell representative of the character, if the orthodox idea of segregation were retained. From these phenomena it was immediately seen that where dominance is absent and such multiple factors are assumed, size characters can be interpreted as coming under the Mendelian law. When dominance is complete the mathematical representation of an F_2 generation is $(3/4+1/4)^n$ where n represents the number of factorial differences involved; as the manifestation of dominance becomes less this formula approaches the type $(1/2+1/2)^{2n}$. The difference between the heredity of qualitative characters and quantitative characters is therefore only one of degree, for there is absence of dominance in cases of simple monohybrid qualitative characters and there is presence of multiple factors in cases of qualitative characters showing dominance. But it is manifestly absurd to expect size characters to appear in natural groups as do many qualitative characters. The marked effect of environment and our ignorance of the exact effect to attribute to each factor precludes it. One can determine whether size inheritance compares with the inheritance of qualitative characters only by the use of arbitrary biometrical methods. In theory, homozygotes with size differences when crossed should give an intermediate F_1 of low variability and an F_2 of high variability. Various F_3 populations should differ in their mean and in their variability. The difference in the variability of F_2 over F_1 should decrease as the heterozygosity of the parents increases. Sometimes parents of the same size should differ in the factors they contain and the F_2 generation should contain individuals smaller and individuals larger than either of the parents. Each of these requirements has been satisfied by experiment. East and East and Hayes have tested it for number of rows per cob, height of plant, length of ear and size of seed in maize, Shull for number of rows in maize, Emerson for fruit sizes in maize, beans and gourds, Tammes for various characters in flax species, Tschermak for time of blooming in beans, Hayes for number of leaves in tobacco, Belling for certain characters in beans, Phillips for body size in ducks, MacDowell for body size in rabbits. In these investigations every test possible for the theory has been satisfied. No criticism could be made except that certain of the characters used varied considerably in the mother varieties and therefore were presumably not homozygous for all character factors. This criticism is apparently answered by a recent investigation of the writer's, as yet unpublished, where two species, *Nicotiana forgetiana* and *Nicotiana alata grandiflora* were crossed. As seen by the table, the corolla length is very slightly variable in either species, nor is it affected to any extent by environment, yet each species was absolutely reproduced by recombination in the F_2 generation.

TABLE I

FREQUENCY DISTRIBUTIONS FOR LENGTH OF COROLLA IN A CROSS BETWEEN *Nicctiana forgetiana* (314) AND *N. alata grandiflora* (321).

Designation	Class Centers in Millimeters														
	20	25	30	35	40	45	50	55	60	65	70	75	80	85	90
314	9	133	28												
321										1	19	50	56	32	9
(314×321)F_1				3	30	58	20								
(314×321)F_2		5	27	79	136	125	132	102	105	64	30	15	6	2	

Coefficients of variation are: 314 = 8.86 ± .33 per cent.; 321 = 6.82 ± .25 per cent.; (314 × 321) F_1 = 8.28 ± .38 per cent.; (314 × 321) F_2 = 22.57 ± 39 per cent.

I do not believe that biologists have sufficient facts as yet to warrant any concrete meaning being given to their notation as regards germ-cell structure, but I do maintain that the Mendelian notation satisfies the facts of size inheritance as well as it satisfies the facts of qualitative inheritance. As a description, it goes the whole way. If qualitative inheritance is Mendelian, quantitative inheritance is Mendelian; if quantitative inheritance is not thus described, qualitative inheritance is described not a whit better.

All writers do not agree with this statement; nevertheless, speaking for myself only, I believe it to be beyond question. Castle (AMER. NAT., 46: p. 361) says:

> It is quite possible that we are stretching Mendelism too far in

> making it cover such cases. Dominance is clearly absent and the only fact suggesting segregation is the increased variability of the second as compared with the first hybrid generation. This fact however may be accounted for on other grounds than the existence of multiple units of unvarying power.
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> If size differences are due to quantitative variations in special materials within the cell, it is not necessary to suppose that these materials are localized in chunks of uniform and unvarying size, or that they occur in any particular number of chunks, yet the genotype hypothesis involves one or both of these assumptions. Both are unnecessary. Variability would result whether the growth-inducing substances were localized or not, provided only that they were not homogeneous in distribution throughout the cell. Crossing would increase variability beyond the first generation of offspring because it would increase the heterogeneity of the zygote in special substances (though not its total content of such substances) and this heterogeneity of structure would lead to greater quantitative variation in such materials among the gametes arising from the heterozygote. Thus greater variability would appear in the second hybrid generation.

I can not agree with this statement as I understand it, though this disagreement may be due to my own limitations. We do not stretch Mendelism and we do not make it cover such cases. The facts of breeding have been obtained and the Mendelian notation expresses them. That is all that it is necessary to claim. It is not precisely true, however, to say that increased variability in the second hybrid generation is the only fact to be expressed. It is of paramount importance that various F_2 individuals giving F_3 populations differing in mean and in variability, should be included in the Mendelian description. They are included.

Again, Castle states that the genotype conception assumes the localization of the hypothetical factors either in chunks of uniform and unvarying size, or that they are carried by a particular number of chunks. I am unaware of any such assumptions. It is true that some such picture has been suggested as a diagram helpful to the imagination in its conception of the scheme as a mechanical process, but this is purely and simply a diagram. The real matter under discussion is that the breeding facts are adequately described in a notation essentially Mendelian.

Of course Castle's scheme of expressing the facts by heterogeneity in the germ cell might serve. He produces increased variability in the second hybrid generation by greater differentiation among the gametes arising from the heterozygote. But one can also describe inheritance of qualitative characters in the same way, and one gains no system by it. It is a return to the type of expression used by Nägeli, Naudin and De Lage in pre-Mendelian days. It is simply a trans-nomination possessing no advantages.

Before leaving this phase of the subject, I must speak of Davis's recent fine paper (AMER. NAT., 46: p. 415) on his crosses between *Œnothera biennis* and *Œnothera grandiflora*. As I have had the advantage of seeing his cultures many times in the past two years, I am in a fair position to draw my own conclusions as to the meaning of his data. In regard to his F_2 generation from the hybrid plant marked 10.30 L *b* he says:

> 1. In the immensely greater diversity exhibited by the F_2 generation over that of the F_1 is clearly shown a differentiation of the germ plasm expressed by the appearance in the F_2 plants of definite tendencies in directions toward the two parents of the cross. This seems to the writer the essential principle of Mendelism and does not necessarily involve the acceptance of the doctrine of unit characters and their segregation in either modified or unmodified form.
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> 2. Certain characters of the parent species have appeared in the F_2 segregates in apparently pure condition. but the very large range of intermediate conditions indicates that factors governing the form and measurements of organs (if present at all) must in some cases be concerned with characters so numerous and so small that they can not be separated from the possible range of fluctuating variations. If this is true such characters seem beyond the possibility of isolation and analysis and the unit character hypothesis for these cases has little more than a theoretical interest.
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> 3. Both cultures certainly showed marked progressive advance in the range of flower size, the largest flowers having petals somewhat more than 1 cm. longer than those of the *grandiflora* parent. There was a similar advance in the size of the leaves and the extent of their crinkling. These progressive advances would seem to demand on the unit character hypothesis either the modification of the old or the creation of new factors.
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> 4. The absence of classes among the F_2 hybrids (except for the

dwarfs) further works against the unit character hypothesis as of practical value in the analysis of a hybrid generation of this character. It should be remembered, however, that there were in this cross no sharply contrasted distinctions of color, anthocyan (stem) coloration proving most unsatisfactory for the purpose of a genetical study.

These four paragraphs are practically a résumé of Davis's genetic facts: I take exception only to some of the implied conclusions. It is quite evident that Dr. Davis believes that many breeding facts are expressed in shorthand by the Mendelian notation. His statements, however, imply a feeling of loss of caste or something of the kind if he makes definite use of Mendelian phraseology. His F_2 generation was exactly what would be expected when several Mendelian units without dominance segregate and recombine. The advance in size of corolla was predicted by me in 1910 (Amer. Nat., 44: p. 81) as a direct consequence of size inheritance. It has since been demonstrated by Tschermak for time of blossoming of beans and clearly analyzed by Hayes for number of tobacco leaves. It demands neither modification of old nor the creation of new factors. It occurs whenever *AABB* (size factors) is crossed with *CCDD,* and each factor is allelomorphic to its own absence, to use the ordinary phraseology.

As to the difficulty of precise analysis into factors, I agree with Dr. Davis, but that there is no advantage in showing that this behavior is described in typical Mendelian terms I can not admit. One holds the same practical advantage here—though the case is complex—that one holds in all Mendelian inheritance. He knows that somatic appearance is not the criterion of breeding capacity, but that it is determined in some way by gametic constitution, although no germ cell architecture is presupposed. He knows that recombination of some kind of factors occurs and has some idea of the number of progeny to be grown to obtain the desired combination. In other words, the blend in F_1 does not indicate complete loss of extremes.

Mendel's Law and Galton's Law

The above statement leads into a discussion of Mendel's law of heredity as compared with Galton's law, for in itself it is almost a statement of the difference. As Bateson was the first to emphasize, organisms inherit from parental germ cells only, therefore a law of ancestral heredity is a fallacy and a misnomer. The simple illustration that of two individuals alike in appearance one is homozygous for a character and the other heterozygous for the same character, shows the superficial reasoning that leads to the correlation coefficient as a measure of heredity. Parental and filial populations may show correlation, but that is only a matter of averages and not a measure of the inheritance.

Professor Castle has recently disclosed the probable Mendelian basis for Galton's data on coat color of Bassett hounds by showing the inheritance of tricolor coat in guinea-pigs, yet he makes the surprising statement that "as regards height, however, and other size characters, Galton's law is quite as good a basis for predicting the result of particular matings as is Mendel's." The arch priest of biometry, Karl Pearson, does not claim that Galton's law can predict the result of individual matings. Similarly, Mendel's law predicts only by averages. It says that where DR meets DR, there will be on the average $1DD:2DR:1RR$ produced. Where the classes are larger the prediction in increasingly complicated. But the prediction is as good for size characters as for qualitative characters of the same complication. And there are such qualitative complications, as is manifest by Castle's formula of $AACCUUIIYYBBBrBrEE$ for a wild rabbit's coat color. The difference between Galton's law and Mendel's law is that the true criterion of the germ plasm of any individual is its breeding *power* and not the somatic appearance of its back ancestry. This is as true of size characters as of any other characters.

The Genotype Conception of Heredity

Expressed in Johannsen's words, the basis of the modern conception of heredity is: "Personal qualities are the *reactions of the gametes joining to form a zygote;* but the nature of the gametes is not determined by the personal qualities of the parents or ancestors in question." The quotation expresses well the idea that I have just tried to convey, and from it one sees plainly that it is the correlation that necessarily appears to a greater or less extent between the somatic qualities of two generations when they exist in large numbers that gave the basis for Galton's superficial law.

This quotation is Johannsen's slogan for the genotype conception of heredity. As there stated, it is merely a generalized expression of the essential features of the Mendelian notation. Johannsen, therefore, was the first to admit the broadness of its scope. In his exposition of his position, however, he adds two subsidiary propositions that we will now discuss; the first is the perennial question of the possibility of the inheritance of acquired characters, the second is a question which from its illusiveness is likely to take on a perennial habit—that of the relative constancy of unit characters.

In regard to the first question I must be content here with a mere general statement. Like Osborn I would emphasize the possibly delusively static condition of organisms when tested during the infinitesimal time usually devoted to experiment. The inheritance of acquirements in some subtle way unknown to us may have been of immense importance in evolution. On the other hand, some sort of an orthogenesis may account for all the facts without the inheritance of acquired characters. It scarcely seems possible that everything is mere chance, though one who has studied plant teratology is astounded at the almost infinite number of characters that have appeared that were absolutely dangerous to the individual in its contest for survival. Be that as it may, I simply wish to acknowledge unbelief in any so-called proof that the inheritance of acquired characters is *impossible*. At the same time one must admit that no unquestionable proof of such inheritance has ever been submitted. Experimental evidence is woefully negative. It seems only reasonable, therefore, considering the available corroborative evidence, to relegate the expression of new characters to variations that have affected the potentialities of the germ cells. We can simply divide variations into the classes inherited and non-inherited without any admission as to their cause. We can call the inherited variations mutations if we will, or we can give them any other name. We must simply remember that they are both large and small.

One can hardly agree with Osborn that *large* variations which are not in an orthogenetic line have had little value in evolution, or that teratological phenomena are of little consequence. The production of identical quadruplets in the armadillo can scarcely be a gradually perfected character. Zygomorphism in flowers is lost as a unit and although this does not prove its birth as a unit, still that is to be presumed. One could fill pages with such data, but this is hardly the place for it. We will therefore consider the relative constancy of what we know as a unit character.

THE CONSTANCY OF UNIT FACTORS

The first thing one does if he wishes to oppose the idea of a unit character is to ask for a definition. A perfect definition of a unit character is as difficult to formulate as for a flower, yet one can obtain an idea of a flower by proper application. If one describes a unit character as the somatic expression of a single gametic factor or heredity unit, he at once gets into trouble. As the factor and not the character is the descriptive unit, a unit factor may affect a character but that character may never be expressed except when several units cooperate in ontogeny. I should prefer to disregard the word character therefore in formulating the problem. The real problem is: Are the facts of heredity adequately described by unvarying hypothetical factors? It is my thesis that if they can not be so described, the Mendelian notation fails.

Johannsen was the first to show the relative constancy of characters by his beautiful experiments on beans. Since that time, experiments designed to show change, if present, have yielded negative results on bisexual animals such as poultry (Pearl), on plants such as peas (Love), beans (Johannsen's later work), maize (Shull, East, Emerson), on asexual animals such as hydra (Hanel), paramæcium (Jennings) and on asexual plants such as bacteria (Barber and others), and potatoes (East).

Three critics have appeared. Karl Pearson took up the gage of battle because Johannsen's work shows the utter untenability of the correlation coefficient as a measure of heredity. He has produced no evidence to uphold his view. Harris, following Pearson for a like reason, has concluded against Johannsen, but has not yet presented his data for public criticism. There remains the work of Castle, which he believes is supported by the work of Woltereck. The question to consider then is whether the work of these two investigators justifies the contention.

Castle states that by selection he has modified a unit character. No one questions that under certain conditions changes in characters are made manifest by selection. It has been done again and again. The question as I see it is the following: Are not the facts presented by Castle and the facts of the pure-line workers described most concisely and in a way most helpful to investigation, by the reactions of fixed and unchanging units? If they can not be thus described the use of units is an absurdity, for one can not measure or describe by changing standards.

Castle's principal work on selection is with a fluctuating black and white coat pattern—the so-called hooded rat. In writing of these experiments, Castle says (*l. c.*, p. 355):

I shall speak first of the case least open to objection from the genotype point of view, which requires:

1. That no cross breeding shall attend or shortly precede the selection experiment, lest modifying units may unconsciously have been introduced, and

2. That only a single unit-character shall be involved in the experiment.

These requirements are met by a variety of hooded rat which shows a particular black and white coat pattern. This pattern has been found to behave as a simple Mendelian unit-character alternative to the self-condition of all black or of wild gray rats, by the independent investigations of Doncaster, MacCurdy and myself. The pigmentation however in the most carefully selected race fluctuates in extent precisely as it does in Holstein or in Dutch Belted cattle. Selection has now been made by Dr. John C. Phillips and myself through 12 successive generations without a single out-cross. In one series selection has been made for an increase in the extent of the pigmented areas; in the other series the attempt has been made to decrease the pigmented areas. The result is that the average pigmentation in one series has steadily increased, in the other it has steadily decreased. The details of the experiment can not be here presented, but it may be pointed out (1) that with each selection the amount of regression has grown less, *i. e.,* the effects of selection have become more permanent; (2) that advance in the upper limit of variation has been attended by a like recession of the lower limit; the total range of variation has therefore not been materially affected, but a progressive change has been made in the mode about which variation takes place.

3. The plus and minus series have from time to time been crossed with the same wild race. Each behaves as a simple recessive unit giving a 3:1 ratio among the grandchildren. But the extracted plus and the extracted minus individuals are different; the former are the more extensively pigmented.

4. The series of animals studied is large enough to have significance. It includes more than 10,000 individuals.

The conclusion seems to me unavoidable that in this case selection has modified steadily and permanently a character unmistakably behaving as a simple Mendelian unit.

This conclusion, from the writer's standpoint, is not only avoidable, but unnecessary. No direct or implied denial of these facts is made, but a shift is made in the point of view. It seems to me a logical necessity that hypothetical units used as measurement or descriptive standards be fixed. The problem to be solved is the simplest means of thus expressing the facts. If the most definite characters—*i. e.*, certain pure-line homozygotes—are sufficiently constant in successive generations to be expressed by a fixed standard, well and good. The whole heredity shorthand is then simple. If such is not the case, the *character* must still be described by some fixed standard, but in that case recourse must be had to complex mathematical expressions and not to a single unit to describe the most constant somatic expressions. Furthermore, if these mathematical expressions served any practical purpose, it would be necessary to prove that all somatic variability of homozygotes under uniform conditions (if there is any) may be expressed by very few formulas.

Such an attitude does not seem to be in harmony with the progressive spirit of the times. I believe that we may describe our results simply and accurately by holding that unit factors produce identical ontogenetic expressions under identical or similar conditions. If under identical conditions the expression *is* different, then a new standard, a new unit, must be assumed; that is, factor *A* by any change becomes factor *B*. The results of the pure-line investigations are the warrant for this interpretation, for they are the investigations of successive generations of somatic expressions with the least attendant complication. From them one may assume that a succession of individuals homozygous in all characters and kept under identical conditions will be alike.[4] To be sure there are numerous changes in the expression of characters when external and internal conditions are not so uniform as the above, but I believe that these changes can all be described adequately and simply by ascribing them to modifying conditions both external and internal. When external we recognize their usual effect in what we called non-inherited fluctuations, when internal we recognize their cause in other gametic factors inherited independently of the primary factor but

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[4] Possibly even under these conditions rare variations that are exceptions to this rule might occur. In other words, mutations might occur having no external cause and therefore to be left for vitalistic interpretation, but this would not affect the general situation.

modifying its reaction during development. This is a physiological conception of heredity, as it recognizes the great coöperation between factors during development. It is a very simple conception of heredity, moreover, for it allows a multitude of individual transmissible differences with the assumption of a very few factors. Some illustrations will be given later that will show the idea underlying this theory. Let us now see whether Castle's work can be described properly by it.

Castle started with a peculiar character. It fluctuates continually and has never been bred to as small a variability as have many other characters. I have worked with a somewhat similar character in maize. It is a variegated pericarp color. In experimenting with it I have raised over a thousand progeny in one generation, a thing manifestly impossible with rats. Both solid colored ears and white ears have been obtained, and while at present it would be unwise to draw definite conclusions, it appears that both solid red ears and white ears of this kind give again variegated progeny. In other words, neither the red ear nor the white can behave like a normal red or white ear, but as if the pattern had fluctuated so widely that it can not appear on the ear (this explanation was suggested by Emerson). At any rate, we may conclude that the rat pattern fluctuates widely and is therefore markedly affected by some condition either internal or external.

Castle began therefore with a character in a fluctuating condition, possessed by a race which had not recently been crossed with a different race. This does not mean, however, that the various individuals forming his original stock did not differ in several factors that in their different combinations might have an effect upon the developing pattern. Suppose for the moment that this were actually the case. If he had been able to produce a fraternity by a single mating numbering several thousands, he would have produced individuals with all of these combinations of other genes. It is probable that he would then have obtained his progressive extremes in one generation, extremes that were never seen when but few individuals were produced. This sort of a thing is not hypothetical. It is mathematically demonstrable that with the same variability $(a + b)^n$ expanded gives an increase in the number of classes as the total number of individuals increases. It is, moreover, supported by the experimental evidence of De Vries on selecting for higher number of rows in maize. I, myself, by using greater numbers obtained an increase in protein in maize in one generation comparable to that obtained by the Illinois Agricultural Station in three generations.

Castle further argues that decrease in regression toward the original mean supports his view. On the other hand, this is exactly what should take place on assuming the truth of the fixed factor conception, as has been shown by Jennings.

Again, the selected races when crossed with wild races both act as simple recessives, but the extracted plus individuals are more pigmented. This is what I should expect. The extracted plus individuals would be more pigmented when existing in small numbers, because the modifying factors are several. If several thousand progeny were grown, however, recombinations would show a more varied result. And as a matter of fact, extracted recessives from the plus race are not precisely comparable in their fluctuation to the selected race with which the wild was crossed. They are more variable than the progeny of an inbred hooded individual of the same grade as the parent used in the cross with the wild. I do not think that one has a right to say, therefore, that there were no modifying genes present in various combinations in the extracted recessives.

When the selected lines were crossed together, moreover, the resulting progeny were somewhat intermediate and variable. The grandchildren were *more* variable. This is what should result from our assumptions. The animals are homozygous as far as having a pattern is concerned, but they differ in several genes that affect the development of the pattern.

Taking into consideration all the facts, no one can deny that they are well described by terminology which requires hypothetical descriptive segregating units as represented by the term factors. What then is the object of having the units vary at will? There is then no value to the unit, the unit itself being only an assumption. It is the expressed character that is seen to vary; and if one can describe these facts by the use of hypothetical units theoretically fixed but influenced by environment and by other units, simplicity of description is gained. If, however, one creates a hypothetical unit by which to describe phenomena and this unit varies, he really has no basis for description.

The facts obtained when working with pied types are complex. They are evidently not thoroughly understood as is evidenced by a different interpretation made by every worker who has investigated them. Doncaster and Mudge see two types of Irish rat. Why not three or four? Crampe obtained hooded rats from cross of self-colored and albino, the hooded coming only from heterozygotes having some white. No adequate explanation has been given. Cuénot concluded regarding pied mice with several degrees of piedness that each was recessive to the other of next higher grade. In fact the behavior of self colors and spotted colors among mammals as among plants is pretty well "confused," as in several species spotted types dominant to self color are known.

Castle's other experiments in selection—the formation of a four-toed race of guinea-pigs starting with one animal with a rudimentary fourth toe, and the perfection of a silvered race of guinea-pigs from an animal in which the character was feebly expressed—need not be considered here. Both were necessarily crossed with normals at the start, and gradual isolation of races having the proper gene complex for complete expression of the characters is to be expected. There have been numerous selection experiments of this type—such as those of De Vries, the Vilmorins, the Illinois Agricultural Experiment Station, etc.—that have yielded results.

But these results, with one possible exception, were open to the criticism that they probably had to do with mixed lines and could therefore be described by the notation we have used. The experiments on pure lines have given no such results. One should not be asked to accept the results of the unguarded experiments and disregard the results of the guarded investigations.

The one possible exception alluded to above refers to the experiments of Woltereck (*Deut. Zool. Gesell.*, 19: 110–173, 1909) on parthenogenetic strains of *Hyalodaphnia* and *Daphnia* where there can be no question of gametic recombination. This experiment is not beyond criticism as will be seen later, but if it were our position would not be affected. The results would still have to be described by some *fixed standard* but the description would be complicated. *Since it is not beyond criticism, there is yet no reason for such a complication.*

Woltereck's work was primarily to show whether or not acquired characters are inherited. It was a secondary object to find out whether small variations or distinct sports occurred in the species. Those who use the work as an argument for unit factor modification, therefore, should also accept his inheritance of characters acquired.

Woltereck tested the effect of selection on seven characters. Selection gave *no* results in five cases. The first supposedly successful case is for difference in head height. In different pure lines he found an enormous effect of environment. He therefore endeavored to plot curves for different kinds of environment, food, temperature, generation number, etc. By comparing these curves he makes an argument for the inheritance of small acquired variations. In the absence of control cultures, and from the fact that culture conditions very uniform to Dr. Woltereck may have been somewhat extreme to Mr. A. Daphnia, the argument has only the value of the other numerous scholastic defences of inherited acquirements. It is criticized by Tower in a recent publication. Woltereck did obtain one inherited head variation. It apparently arose suddenly. He calls it a mutation.

The only result that can be considered seriously from the standpoint taken in this paper is the result when selecting for a rudimentary eye. *Daphnia* has been distinguished from *Hyalodaphnia* by the presence of a rudimentary eye. The distinction does not seem to be valid, for Woltereck noticed rudimentary eyes several times in pure line cultures of *Hyalodaphnia* and they have also been seen by others in wild cultures. He regards the phenomenon as a reversion to a preexisting condition. *He found that the presence of the rudimentary eye is periodic. In the spring it appears, in the summer it again disappears.* Either kind can produce progeny of the other kind. From this fact it seems reasonable to believe that environment or generation number has much to do with the expression of the character, although Woltereck in one place inclines to the opinion that external factors affect it but little. He performed several experiments on the effect of light and temperature, however, and says that provisionally they gave no result free from objection—" . . . gegaben einstweilen kein einwandfreies Resultat." Almost any interpretation can be given this statement.

From a pure line in which this variable eye spot appeared, he isolated a mother and grandmother with the character well developed. Ninety per cent. of the progeny had the eye well developed. The rapidity of his results and the fact of *periodicity* in the expression of the character makes any cumulative effect of selection exceedingly questionable. One is not justified therefore in accepting it as proof without corroboration.

CONCLUSION

In conclusion, it may be asked if it is not reasonable to accept simply as a nomenclature the description of the whole facts of inheritance in sexual reproduction given by the Mendelian system? Is it wise to turn backward and to give up this handy and helpful notation right in the midst of a useful career? The experiments least open to objection (the pure-line experiments) have shown the wisdom of assuming a stable unit factor, this factor being representative of the stability manifested by a character complex when no interfering conditions intervene. Let us accept this simple interpretation provisionally, appreciating the fact that the stability of the characters that have been represented by fixed units may be only a static appearance due to limited experiments; but that this appearance justifies our neglecting any infinitesimal fluency of our factor standards in experiments of like duration, since taking them into account would necessitate a change of standard, a new fabric of hypotheses and a more complicated system. Let us take a physiological view of heredity. Factors are assumed to be stable. Characters are somewhat unstable owing to the effect that other factors have upon their expression. Factor *A*, for example, is potentially able to produce a typical expression in ontogeny under certain definite conditions of environment, but the presence or absence of *B* or *C* or *D* or *B*, *C*, and *D* are responsible for slight changes in the expression of *A*. This conception gives us a picture of heredity in real accordance with physiological facts, in contradistinction to the non-biological and fixed physical conception—the mosaic organism conception—that critics often say is held by some geneticists.

One may answer that this conception is all right for quantitative characters, but do the facts uphold it for qualitative characters? They do. I will give examples from my own experiments on the inheritance of the purple aleurone cells in maize. Here one obtains progeny by the thousands and sees phenomena that are obscured by lesser numbers.

Crosses of the purple variety with three different whites have given three different results. One shows that the purple may be represented by the schematic description *PPRRCC*. Crossed with *pp rr cc* it gives purples, reds and whites in the F_2 generation, as all three factors are necessary for the production of the purple color. How many other factors (present also in the whites) may be necessary one can not say. In another white, the *R* factor is present and purples and whites in the ratio of 9:7 result. In another white, both *P* and *R* are present. In another white, both *P* and *C* are present. Both give monohybrid ratios when crossed with the purple.

This is not the sum total of whites, however; several others have been found. One has an intensifying factor. We get darker purples together with the normal purples, but no one can doubt that the purple is still the same pigment modified in its expression. Another white has a dominant inhibiting factor. In the heterozygous condition it does not always inhibit the color entirely, but in the homozygous condition color never develops. The dominance of this factor is proved by the fact that extracted colored recessives are still heterozygous for presence of color.

In still other whites I have demonstrated the presence of at least three modifying genes $M_1M_2M_3$. They are independent of each other, yet each and all affect the purple color. One is dominant, as if it were a partial inhibitor, the others are recessive, as if they were the loss of intensifying factors. Purples of all different degrees can be isolated and breed true. The lightest is such that the color can be distinguished only with a lens. But they are all strictly alternative in their transmission and somewhere near the expected ratios of darks, lights, very lights, etc., appear. It is too much to ask that exact ratios be obtained for with this kind of modification all shades appear, yet conclusive evidence has been obtained by F_3 and F_4 generations.

The qualitative characters do act the same as quantitative characters, therefore, and one can not make a real distinction between them.

RESEARCH BULLETIN NO. 2.

THE UNIVERSITY OF NEBRASKA.

BULLETIN
OF THE
AGRICULTURAL EXPERIMENT STATION
OF
NEBRASKA.

THE INHERITANCE OF QUANTITATIVE CHARACTERS IN MAIZE.

BY COLLABORATION OF
R. A. EMERSON,
THE UNIVERSITY OF NEBRASKA,
AND
E. M. EAST,
BUSSEY INSTITUTION OF HARVARD UNIVERSITY.

ACCEPTED FOR PUBLICATION AUGUST 16, 1912.
DISTRIBUTED APRIL 1. 1913.

LINCOLN, NEBRASKA
U. S. A.

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CONTENTS.

THE INHERITANCE OF QUANTITATIVE CHARACTERS IN MAIZE.

BY COLLABORATION OF R. A. EMERSON AND E. M. EAST.*

INTRODUCTION.

When Mendel's law of heredity was rediscovered by Correns, de Vries, and von Tschermak, most biologists were justly skeptical of the possibility that it might be general in scope. The early disclosure that a wide range of breeding facts in both the animal and the vegetable kingdom were made orderly and consistent by using the Mendelian notation† brought about only a slight change in this attitude. A few optimistic minds believed that the similarity of the breeding phenomena in animals and in plants indicated that the Mendelian mechanism was intimately associated with that fundamental process common to both kingdoms,—sexual reproduction. Those who reasoned in this manner suspected a broad general operation of the law of segregation and recombination of unit factors. The remaining great majority held to their position of respectful distrust.

There were several reasons for this situation. A goodly portion of the responsibility for it rests upon de Vries, who, from data of doubtful significance, drew the broad conclusion that varietal characters and specific characters obey quite different laws of heredity. Unpaired or unisexual characters he believed to have sole claim to the rank of specific differences. These characters, he thought, are incapable of Mendelian segregation. Segregating characters he called paired or bisexual qualities, and these he believed worthy of only varietal rank. The fallacy in de Vries' reasoning is obvious, now that Bateson's presence and

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* Harvard University, Bussey Institution, Jamaica Plain, Mass.

† The prevailing Mendelian terminology is followed in this paper, but it must not be assumed that the writers regard Mendelian formulæ as other than a helpful descriptive shorthand convenient for describing breeding facts. Hypothetical germ cell factors are substituted for somatic characters because they are useful in exactly the same manner that hypothetical formulæ are useful in describing chemical reactions. To establish the contention that quantitative characters are essentially Mendelian in their inheritance, therefore, it is only necessary to show that the notation adequately describes the breeding facts.

RESEARCH BUL. 2, AGR. EXP. STATION OF NEBR. Published also as a contribution from the Laboratory of Genetics, Bussey Institution of Harvard University.

absence notation has come into general use. In truth one must accept a reversal of de Vries' views, for the presence and absence notation is based on the fact that the hereditary transmission of a factor can only be determined when the factor is present in one of the parental gametes and absent from the other.

A second reason for limiting the operation of Mendel's law was the earlier misconception of the importance of the phenomenon of dominance. This reason shortly disappeared when it was found that perfect dominance was the exception rather than the rule and that cases where dominance was entirely lacking were frequently found.

These were important concessions that contributed much toward recognition of the importance of the Mendelian conception of heredity, but the main stumbling-block to Mendelism lay in the comparative frequency of quantitative variations. The variations whose behavior in crosses yielded to a simple factorial interpretation were all qualitative in character. They could reasonably be regarded as being due to the presence or absence of something in the gamete. Such varietal differences existed by the score, yet their number was insignificant when compared with the multitude of variations expressed as differences in size of organs common to the individuals under consideration. Many of these differences, it is true, were fluctuations due to environment which were non-heritable,* but the remaining transmissible variations were legion. About three years ago, however, a hypothesis was proposed which would bring even this broad category of facts under the scope of the law of unit-factor segregation and recombination. The experimental data upon which this hypothesis was then based and the data that have been collected subsequently are comparatively few, yet the fact that the experiments of several independent investigators have been corroborative in every detail makes it plausible to believe that its main contention will soon be established beyond a reasonable doubt.

If this statement be granted for the moment, one might hazard the suggestion that the probable limits that circumscribe Mendelian phenomena really do coincide with the limits within which transmissible character potentialities are transferred by typical sexual reproduction. In other words, Mendelian phenomena are in some way bound up in and coincident with that preparation of the reproductive cells called maturation and their subsequent fusion to form a new organism. This, it is thought, is an admissible induction from the conspicuous diversity and profusion

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* It has been shown that if adaptive responses are ever heritable, the phenomenon is so rare that it may be neglected in experimental pedigree cultures.

of Mendelian phenomena, and from the fact that the only indisputable cases of non-mendelizing characters are (1) that discovered by Correns in *Mirabilis jalapa*, where a pathological condition affecting a leaf function is carried by the cytoplasm and inherited only thru the mother, and (2) that reported by Baur for *Pelargonium zonale*, in which vegetative segregates (sectorial chimeras) occur in F_1 plants and breed true in F_2.

The acceptance of this point of view depends largely upon whether a correct interpretation of the inheritance of quantitative variations has been reached. The authors, therefore, desire to submit a somewhat complete discussion of the inheritance of this category of variations, supported by data from experiments on maize that have been gathered independently. If there be criticism that the conclusions drawn are too broad since the facts have been gathered from a single plant species, the authors may answer that each has made corroborative investigations upon other species which they expect to publish in the near future.

The experiments conducted by one of the writers were begun at the Connecticut Agricultural Experiment Station at New Haven in 1906 and removed to the Bussey Institution of Harvard University at Forest Hills, Massachusetts, in 1909. The materials employed in this study consisted principally of crosses of Tom Thumb pop with Black Mexican sweet and of Watson flint with Leaming dent. The number of rows per ear were noted in several other crosses, the parents of which are listed later in this paper. For help in gathering and compiling the data from these crosses, we are indebted to H. K. Hayes, plant breeder of the Connecticut Agricultural Experiment Station, and to O. E. White and D. L. Davis, graduate students in genetics at Harvard University.

The experiments of the other writer were begun at the Nebraska Agricultural Experiment Station at Lincoln in 1908, where they have been continued to the present time. A part of the material studied in 1911 was grown at the Bussey Institution at Forest Hills, Massachusetts. The plants used in this study were crosses of Tom Thumb pop with Missouri dent and of the latter variety with California pop. For assistance in connection with this work, we acknowledge our indebtedness to R. F. Howard, now assistant horticulturist of the Wisconsin Agricultural Experiment Station, to F. C. Miles, graduate student in genetics at the University of Nebraska, to E. H. Herminghaus, student assistant, and particularly to E. R. Ewing, who has computed most of the statistical constants from the data obtained from the Nebraska experiments.

THE THEORY OF MULTIPLE FACTORS AFFECTING A SINGLE CHARACTER COMPLEX.

Bateson and later other writers have shown that development of qualitative characters may depend upon the interaction of two or more gametic factors that are transmitted independently. When one of these essential factors is absent from the zygote the visible character does not develop. Thus color in the sweet pea, in stocks, in snapdragons, in primulas, in beans, and in the hair of various animals does not develop unless a basic factor, generally known as the color factor *C*, is present in addition to the color determiner which fixes the shade of the color. When either the color factor or the specific determiner is absent, color does not develop.

In two papers published in 1908 and 1909, Nilsson-Ehle showed that certain colors in wheat and in oats are determined not by single gametic factors but by several, the presence of any one of which serves to determine the color. In other words, these gametic factors, altho affecting the same zygotic character, are not allelomorphic to each other, but to their own absence. Nilsson-Ehle in 1908 showed how such facts gave a foundation for a theory interpreting the inheritance of *quantitative* characters, altho no experimental data were submitted to support the hypothesis.

A little later, East, in ignorance of Nilsson-Ehle's 1908 paper,* submitted a similar case of color inheritance in maize, and developed a similar theory.

Fortunately this type of inheritance was discovered in characters showing dominance, otherwise its proof would have been very difficult. We will describe these cases first, and afterwards show their bearing on the so-called "blending" inheritance characteristic of quantitative characters.

Nilsson-Ehle (1909) crossed together many varieties of oats differing in glume color. The results from all but one of these crosses were quite simple. Presence of color dominated absence. Where the parents differed in one character pair, such as black *(B)* and absence of black *(b)*, the ratio in F_2 was 3:1. Where the parents differed in two allelomorphic pairs, black *(B)* and absence of black *(b)*, and gray *(G)* and absence of gray *(g)*, the ratio in F_2 was 12 black : 3 gray : 1 white, because the classes *BG* and *Bg* could not be distinguished. So far there were no

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* East learned of Nilsson-Ehle's second paper, which does not give the full development of the quantitative-inheritance theory, after the first draft of his own paper was written, but Nilsson-Ehle deserves full credit for the theory that is based upon his beautiful results on qualitative characters.

complications. In one of these crosses between a black glumed and a white glumed race, however, the F_2 generation was 630 black : 40 white, clearly a ratio of 15 black : 1 white. Here there appeared to be two factors for black, each dominant to its own absence, segregating and recombining independently. The zygotic formula of the black race may be expressed as $B_1B_1B_2B_2$ and that of the white race as $b_1b_1b_2b_2$. The F_1 generation brings both of these factors to the heterozygous condition, $B_1b_1B_2b_2$, and in F_2 gives $9B_1B_2 : 3B_1b_2 : 3b_1B_2 : 1b_1b_2$. But as the first three classes are alike in appearance, the ratio of F_2 is 15 black : 1 white.

Again, experimenting with wheat, Nilsson-Ehle found a dihybrid Mendelian ratio. A brown headed wheat crossed with a white headed variety gave in F_2 1410 brown headed plants to 94 white headed plants, a ratio exactly 15 : 1. There is hardly a doubt that the brown headed variety had the formula $B_1B_1B_2B_2$ and the white headed variety the formula $b_1b_1b_2b_2$. The proof of the interpretation comes from a more surprising experiment of the same investigator.

He made six crosses between a red-seeded wheat and a white-seeded variety. The F_1 plants were all red-seeded: *the 384 plants of the* F_2 *generation were also red-seeded.* This result was so peculiar that selfed seed of all of the 78 F_2 plants of one cross were grown and the progeny of each recorded in the F_3 generation. This result was obtained:

50 F_2 plants gave only red-seeded plants.
8 F_2 plants gave approximately 3 red : 1 white plants.
15 F_2 plants gave approximately 15 red : 1 white plants.
5 F_2 plants gave approximately 63 red : 1 white plants.

The interpretation of this result was that a trihybrid had been produced by crossing $R_1R_1R_2R_2R_3R_3$ with $r_1r_1r_2r_2r_3r_3$, each R factor being able to produce the red color in the seed, but segregating with its own absence independently of the other factors. The red F_1 generation would have the formula $R_1r_1R_2r_2R_3r_3$ and should by recombination produce 63 red : 1 white plants in F_2. To be sure, no white-seeded plants were obtained in the 384 individuals forming the total F_2 generation, but such a result might be expected frequently in chance matings of this type following the Law of Error, unless more F_2 progeny were grown. The real proof of the interpretation comes from the F_3 generation as noted before. The F_2 generation, theoretically, consists of 63 reds to 1 white. The 63 red plants, tho similar in appearance, should have the different zygotic formulae expected of an ordinary tri-hybrid, and therefore should give different results in F_3 as follows:

	F_2 gives in	F_3
Class 1.	1 $R_1R_1R_2R_2R_3R_3$	red only.
Class 2.	2 $R_1r_1R_2R_2R_3R_3$	red only.
Class 3.	2 $R_1R_1R_2r_2R_3R_3$	red only.
Class 4.	2 $R_1R_1R_2R_2R_3r_3$	red only.
Class 5.	4 $R_1r_1R_2r_2R_3R_3$	red only.
Class 6.	4 $R_1R_1R_2r_2R_3r_3$	red only.
Class 7.	4 $R_1r_1R_2R_2R_3r_3$	red only.
Class 8.	8 $R_1r_1R_2r_2R_3r_3$	63 red : 1 white.
Class 9.	1 $R_1R_1R_2R_2r_3r_3$	red only.
Class 10.	2 $R_1r_1R_2R_2r_3r_3$	red only.
Class 11.	2 $R_1R_1R_2r_2r_3r_3$	red only.
Class 12.	4 $R_1r_1R_2r_2r_3r_3$	15 red : 1 white.
Class 13.	1 $r_1r_1R_2R_2R_3R_3$	red only.
Class 14.	2 $r_1r_1R_2r_2R_3R_3$	red only.
Class 15.	2 $r_1r_1R_2R_2R_3r_3$	red only.
Class 16.	4 $r_1r_1R_2r_2R_3r_3$	15 red : 1 white.
Class 17.	1 $R_1R_1r_2r_2R_3R_3$	red only.
Class 18.	2 $R_1r_1r_2r_2R_3R_3$	red only.
Class 19.	2 $R_1R_1r_2r_2R_3r_3$	red only.
Class 20.	4 $R_1r_1r_2r_2R_3r_3$	15 red : 1 white.
Class 21.	1 $R_1R_1r_2r_2r_3r_3$	red only.
Class 22.	2 $R_1r_1r_2r_2r_3r_3$	3 red : 1 white.
Class 23.	1 $r_1r_1R_2R_2r_3r_3$	red only.
Class 24.	2 $r_1r_1R_2r_2r_3r_3$	3 red : 1 white.
Class 25.	1 $r_1r_1r_2r_2R_3R_3$	red only.
Class 26.	2 $r_1r_1r_2r_2R_3r_3$	3 red : 1 white.

Collecting the statistics, we find that the 63 red plants of the trihybrid minimum of 64 in F_2 should give in F_3, when selfed, 37 breeding true to red, 6 throwing reds and whites in the ratio of 3:1, 12 throwing reds and whites in the ratio of 15:1, and 8 throwing reds and whites in the ratio of 63:1. From these data one can calculate the most probable result in the experiment where 78 plants were followed to the F_3 generation. It is as follows:

Type	Per cent	Actual result	Most probable expectancy
Plants breeding true to red	58.8	50	46
Plants giving 3 red : 1 white.........	9.5	8	7
Plants giving 15 red : 1 white.........	19.0	15	15
Plants giving 63 red : 1 white.........	12.7	5	10

The actual results are so close to those expected on the theory that it would be a most wonderful coincidence if the theory were not correct.

The original basis for the theory developed by East (1910) was of this same character. Two factors for the production of yellow color in the endosperm of maize were found. The presence of either factor produced yellow seeds identical in appearance, but each was allelomorphic to its own absence in crosses. An ear with the zygotic formula Y_1Y_1 was exactly like an ear with the zygotic formula Y_2Y_2. It is true that there was a cumulative effect so that ears with the zygotic formula $Y_1Y_1Y_2Y_2$ were darker in color than those homozygous for a single factor, but those zygotes homozygous for either single factor were indistinguishable. A zygote from any cross which produced the constitution $Y_1y_1Y_2y_2$ gave approximately 15 yellows: 1 white in the next generation when selfed. A total of 5,222 segregates were recorded, of which 4,880 were yellow and 342 white. This ratio is very nearly 15:1, the closest possible ratio being 4,895:327. The actual results were therefore astonishingly close to theoretical expectation.

These segregates were followed to the F_3 generation, and gave decisive evidence that they varied in zygotic constitution in accordance with the scheme which has just been outlined. Theoretically the 15 yellows of F_2 should have the following zygotic formulae:

	F_2	gives in	F_3
9	1 $Y_1Y_1Y_2Y_2$		all yellow.
	2 $Y_1Y_1Y_2y_2$		all yellow.
	2 $Y_1y_1Y_2Y_2$		all yellow.
	4 $Y_1y_1Y_2y_2$		15 yellow : 1 white.
3	1 $Y_1Y_1y_2y_2$		all yellow.
	2 $Y_1y_1y_2y_2$		3 yellow : 1 white.
3	1 $y_1y_1Y_2Y_2$		all yellow.
	2 $y_1y_1Y_2y_2$		3 yellow : 1 white.

Each of these classes provided for by the theory were actually obtained. Those ears having the formula $Y_1Y_1y_2y_2$ were indistinguishable from those ears having the formula $y_1y_1Y_2Y_2$, but where Y_1Y_1 and Y_2Y_2 were both present in the same zygote the seeds were a much darker yellow than where either yellow alone was present.

It should be noted that Emerson (1911) has found two yellow colors in the endosperm of maize that seem to be unlike in appearance. One is a golden yellow and one a light yellow. It is possible that further observations will show that the orange yellow is due to the cumulative action of two similar or identical yellows as described above, but it is also possible that action of a hitherto unobserved *intensity* factor is responsible for the phenomenon.

East and Hayes (1911) reported one other case of this kind where dominance is comparatively complete. Two varieties of maize breeding true to that form of red pericarp which comes to complete development only in sunlight were crossed. The F_2 generation, instead of breeding true to this color as was expected, gave a fraternity composed of ears with and without the red pericarp in the ratio of 15:1. The extracted recessives bred true, but no further observations were made on the segregates having red ears. It can hardly be doubted, nevertheless, that here again were two factors producing apparently the same visible character, each segregating from its own absence and not allelomorphic to each other.

Let us now consider what peculiarities such a scheme of inheritance presents as a whole. If there is absolute dominance and each dominant unit factor affects the zygote as fully as all do when combined, it would take but a few character pairs to present the appearance of breeding true in F_2. This we saw to be the case in the wheats. The F_2 generation of 384 plants appeared to show no segregation. Ordinarily six pure recessives might be expected from this number of plants when dealing with three interacting factors in a heterozygous condition, but if four units $A_1A_2A_3A_4$ were crossed with their absence $a_1a_2a_3a_4$, only one pure recessive could reasonably be expected out of 256 F_2 zygotes and in the numbers generally reported from pedigree cultures they would often be lacking. This sort of occurrence may explain the classical (for some unknown reason, because uncorroborated) strawberry crosses of Millardet (1894) where the F_1 generation is supposed to have resembled the male parent and bred true. A striking character in the male parent due to four or five allelomorphic pairs showing perfect dominance would appear to give such a result. One should be careful, at least, about reporting heterozygotes as breeding true without sufficient evidence.

Another important class of facts resting on this theoretical basis may be illustrated by the case of the red pericarps in maize that has just been described. Two maize varieties, both breeding true for this peculiar pericarp red that develops only in a certain amount of light, were crossed. The F_1 plants, when selfed, unexpectedly gave one plant with colorless pericarp on the ears out of every sixteen. Now one of these varieties had been crossed previously with a white variety and had given the usual monohybrid ratio of 3 : 1 in F_2. This character was therefore represented by the presence of a single factor R and behaved as it should when it met its absence r. But since the cross between the two red varieties gave the dihybrid ratio in F_2, one must suppose that, tho alike in appearance, they were not allelomorphic to each other. One was $R_1R_1r_2r_2$ and the other $r_1r_1R_2R_2$. Each bred true to red within its variety, but when crossed, each factor met its absence and an F_1 was produced heterozygous in both. The resulting F_2 generation for this reason was exactly the same as if a red $R_1R_1R_2R_2$ had been crossed with a white $r_1r_1r_2r_2$.

Just imagine such a case when more pairs of factors are in action. If units $A_1A_2A_3a_4$ met units $a_1a_2a_3A_4$ in hybridization, there is again the apparent paradox of two individuals which have bred true for a particular character, giving pure recessives in F_2 when crossed. But this time the zygote without the character occurs only once in 256 progeny. When one considers the rarity with which dominants or recessives pure for all factors are obtained when three factors or more interact, he can hardly avoid the suspicion that here is a perfectly logical way of accounting for many cases of so-called atavism that are not explained by the interaction of two dominant factors. No definite cases can be pointed out, but it is not uncommon for a florist to have a new type appear with regularity tho with extreme rarity for many generations after a cross. No doubt proper analyses of controlled cultures would elicit simpler explanations of most of these cases, but some of them surely must be referred to the class of phenomena we have just discussed. It is quite within the range of possibility that some of de Vries' Oenothera mutants have originated in this manner.

The colored zygotes, in the glume color of oats and head color and seed color of wheat investigated by Nilsson-Ehle, have been treated as if they were exactly alike in appearance. This was not always the case. The color produced by additional factors was often somewhat cumulative, particularly so in the case of the colored wheat heads. Where the two dominant factors were present there was usually more color developed than where only one was present. The classes could not be distinguished visually, but the general tendency was cumulative. This fact is nicely illustrated by the F_2 segregates of the maize cross in which yellow endosperm behaved as a dihybrid. All of the classes having different gametic constitutions vary in the intensity of their yellow color. The yellow becomes lighter in shade in the following order: $Y_1Y_1Y_2Y_2$, $y_1y_1Y_2Y_2$ or $Y_1Y_1Y_2y_2$, Y_1y_1 or Y_2y_2, and $Y_1y_1Y_2y_2$.

These facts open up two new and important phases of Mendelian inheritance. The first is the possibility of having new characters formed under operation of the Mendelian law. The matter is purely speculative, nevertheless it is suggestive. We do not know why two factors should exist that apparently cause identical characters to be formed in the same plant organ, but different factor-carrying apparatus of some kind which makes possible independent segregation and recombination in the germ cells is indicated. If one accepts—for the sake of argument only—the idea that Mendelian factors are expressions of chromosome functions (Emerson 1911a), then the location of any factor *A* in chromosome number one would give a monohybrid ratio when paired with a homologous chromosome in which *A* was absent. the location of the same factor in both chromosomes 1 and 2 would give the dihybrid ratio when paired with chromosomes in both of which *A* was absent, and so on to the limit of the gametic number of chromosomes. Now if in any line of descent a factor *X* should become located in different chromosomes or in any other way be so affected as not to be allelomorphic to itself in *all* combinations, and further if additional expressions of the factor should result in a cumulative effect, one might have new quantitative characters or even apparently new qualitative characters formed. This hypothesis we consider very important as it gives the first reasonable explanation of the production of new characters otherwise than by mutations of unknown cause.

A second speculation is that when one considers the difficulty of distinguishing the zygotes having various formulae even when dominance is comparatively perfect, he might expect a population of F_2 individuals with almost continuous quantitative variation if dominance is imperfect or absent. This gives a clue to a Mendelian interpretation of the inheritance hitherto known as blended.

The simplest case of this kind yet explained is the inheritance of a peculiar physical condition of the starch in maize (East and Hayes 1911). The varieties of maize known as flinty have the endosperm starch in a hard translucent or corneous condition. Those varieties known as dent have that portion of the starch immediately surrounding the embryo and that at the cap of the seed in a soft condition so that its great shrinkage while drying gives the kernel a dented appearance.

An eight-rowed yellow flint was crossed with a white dent and gave in F_1 an intermediate condition as regards the physical appearance of the starch. The selfed seeds of this generation gave ears in F_2 that ran the whole gamut from flint to dent. A few were like pure flint ears and a few were like pure dent ears, but the great majority were variously intermediate. When followed to the F_3 generation, however, one ear out of sixteen of the F_2 ears bred true to the dented condition and one out of sixteen bred true to the flinty condition. One cannot describe any particular character factors here, or say such and such a factor was contained by either variety. There is no doubt, however, but that the presence (or the absence) of two factors of some kind or other in the pure condition caused the one dent ear out of sixteen to breed true, and that the absence (or the presence) of these two factors in the pure condition caused the one flint ear out of sixteen to breed true. Furthermore, there is good evidence that factorial difference (without dominance) of *more than two* allelomorphic pairs causes the difference in physical appearance of the starch in other maize varieties.

From these facts, it is clear that one may give a valid Mendelian description to the behavior of those quantitative characters that give a blend in the F_1 generation. Two adequately supported assumptions must be made; first, that dominance is absent and that two doses (*i. e.,* the homozygous condition) of a factor have twice the effect of one dose; second, that independent factors cumulative in their operation are paired with their absence in the hybrid.

Let us assume a case of the so-called "blended" inheritance where all fluctuations due to environment are eliminated. A plant 12 inches tall is supposed to be crossed with a plant 28 inches tall. The difference between them is 16 inches. If this difference is due to one allelomorphic pair in which dominance is absent, the F_1 generation is all intermediate—about 20 inches—and the F_2 generation falls into three classes in which two represent the grandparental forms and one represents the F_1 form. Twenty-five per cent are 12 inches tall, fifty per cent are 20 inches tall, and twenty-five per cent are 28 inches tall.

Again, let us suppose this 16-inch difference between the parents to be represented by two allelomorphic pairs instead of one. The F_1 generation is again 20 inches tall, but instead of there being three classes in F_2, there are five classes, viz., 12, 16, 20, 24, and 28 inches, and they appear in the ratio 1 : 4 : 6 : 4 : 1. The grandparental types each appear once out of sixteen times.

The way this ratio is obtained is by simple recombination, but as dominance is absent, each time a *single* "presence" factor is added, the height is increased four inches.

$$9\begin{cases} 1\ AABB = 28 \text{ inches.}\\ 2\ AaBB = 24 \text{ inches.}\\ 2\ AABb = 24 \text{ inches.}\\ 4\ AaBb = 20 \text{ inches.}\end{cases}$$

$$3\begin{cases} 1\ AAbb = 20 \text{ inches.}\\ 2\ Aabb = 16 \text{ inches.}\end{cases}$$

$$3\begin{cases} 1\ aaBB = 20 \text{ inches.}\\ 2\ aaBb = 16 \text{ inches.}\end{cases}$$

$$1\begin{cases} 1\ aabb = 12 \text{ inches.}\end{cases}$$

If three independent size factors instead of two were involved in this cross, the F_1 individuals would fall in the same class as before, but the F_2 classes would be seven in number and the grandparental sizes would each be recovered only once out of sixty-four times. For four factors there would be nine classes of F_2 individuals, and the grandparental types would each occur only once out of 256 times; while with only eight factors, the forms of the grandparents would each appear only once out of 65,536 times, and it would be quite remarkable if they were ever recovered from an ordinary cross.

The entire scheme of this type of inheritance can be expressed in mathematical form just like ordinary Mendelian inheritance with full dominance. Let us recall that the F_2 Mendelian expression for n allelomorphic pairs when dominance is complete is the expanded binomial.

$(3 + 1)^n$ or $(\frac{3}{4} + \frac{1}{4})^n$
$n = 1 \quad (3 + 1)^1 = 3 + 1$
$n = 2 \quad (3 + 1)^2 = 3^2 + 3 + 3 + 1 = 9 + 3 + 3 + 1$
$n = 3 \quad (3 + 1)^3 = 3^3 + 3(3)^2 + 3(3) + 1 =$
$27 + 9 + 9 + 9 + 3 + 3 + 3 + 1$

Likewise the expanded binomial $(\frac{1}{2} + \frac{1}{2})^{2n}$ or $(1 + 1)^{2n}$ gives the numerical relationships when dominance is absent and n represents the number of allelomorphic pairs. The expression $(\frac{1}{2} + \frac{1}{2})^{2n}$ instead of $(\frac{1}{2} + \frac{1}{2})^n$ is used because it is supposed that the presence of any allelomorphic pair in the heterozygous condition produces one-half the visible effect on the character that is produced when the genes are present in the homozygous condition. When n is very large the frequencies with which the different classes occur form a regular curve called the normal curve of error. This is the curve that is produced when the errors in any physical measurement are similarly plotted using as classes any constant deviation from the average, as a, 2a, 3a, etc. This same curve is also produced when one plots the fluctuations of any organic character produced by the infinite complexity of external conditions.

If no non-heritable fluctuations intervened to obscure the class to which any particular zygote belongs, therefore, one should expect the following classes in F_2 when parents of different sizes differing in n allelomorphic pairs are crossed. The extremes represent the grandparental types in each case, and the intermediate classes theoretically divide the difference between the parents into aliquot parts. It should be noted, however, that this is theory only; in reality the influence of one factor might be somewhat different from that of another factor.

$n = 1$					1	2	1					= 4
$n = 2$				1	4	6	4	1				= 16
$n = 3$			1	6	15	20	15	6	1			= 64
$n = 4$		1	8	28	56	70	56	28	8	1		= 256
$n = 5$	1	10	45	120	210	252	210	120	45	10	1	= 1024

Let us now note a few of the practical difficulties in interpreting breeding results that may follow this method of inheritance. In the theoretical example that we have used for the sake of clearness, it was assumed that there were no nonheritable fluctuations due to environment. Unfortunately this is not the case in nature. Fluctuations are everywhere present. They would obscure the classes to which individuals belong even if these class differences were very large. And since they are usually small, the change of individual form due to environmental causes makes it impossible to separate an F_2 population into the true classes to which each belongs theoretically. Nor is this the whole trouble. If the table showing the expected results with two pairs of size characters is examined, it is found that not all the individuals that belong to a particular size class have the same zygotic formula. For this reason one cannot pick out zygotes of a certain size and expect them to breed the same. Their potentialities are likely to be different. Furthermore, practical breeding results are undoubtedly complicated by cases of correlation. This correlation need not be gametic, tho such cases in all likelihood do occur; they may be merely physiological. For example, a maize plant might have the gametic possibilities of small plant size and large ear size, but it would be foolish to expect that a plant capable of only a limited amount of development could bear as large an ear as if it were, as a whole, capable of greater size development. Thus it must not be expected that theoretical possibilities are always expressed perfectly in nature, any more than it should be expected that theoretical physical calculations concerning known laws should agree perfectly with experimental data. The reproductive cells of plants and animals may indeed be described as mosaics of independently transmissible factors, but a plant or animal certainly can not be considered a mosaic of independent unit characters. These factors that appear to be independent in heredity act and react upon one another in complex ways during their development.

There is a means, however, by which it can be determined whether segregation does or does not occur in cases of seemingly blended inheritance. This is by comparing the variability* of the

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* In order to understand the data from these experiments it is necessary to be familiar with the meaning of several mathematical terms used

F_1, the F_2, and the F_3 generations. The fact that a blend occurs in F_1 does not mean that blended inheritance obtains. If it did, the F_2 and future generations should breed true to the type obtained in F_1. If, however, the variability of the F_2 generation is much greater than that of the F_1 generation, segregation and

in the treatment of statistics. Should any reader be unfamiliar with their use, it is hoped that the following short explanation will make them clear:

When variations which are continuous in character are investigated, it is necessary to treat them arbitrarily as discontinuous variations. For example, in one of our studies of heights of maize plants we have put them into three-inch classes. The names of the classes are called the *class centers*. This means that the class whose center is 58 inches includes all the individuals from 57 inches to 59 inches inclusive. Any series of things thus measured and thrown into classes is known as a *frequency distribution*, the number in each class being the frequency with which the class occurs. Then if each class value is multiplied by the frequency with which it occurs, and the sum of these products divided by the total number, the average or *Mean* is obtained. Expressed mathematically,

$$A = \frac{\text{value} \times \text{frequency}}{\text{number}}.$$

With these data we are then able to compute a single number that expresses the variability of the frequency distribution from this mean. This number is called the *Standard Deviation* and is always denoted by the Greek letter Sigma (σ). It is found by getting the deviation of each class from the mean,—all values to the left of the mean being negative and all to the right positive,—squaring them, multiplying the squared deviations by their frequency, dividing the sum of these products by the total number of individuals, and extracting the square root of the quotient. The formula is $\sigma = \sqrt{\frac{\Sigma D^2 \times f}{n}}$, where Σ is the sign of summation. The value thus obtained is in terms of the unit values used. For example, in the distribution of plant heights in the table just mentioned, the standard deviation is in inches. To reduce this concrete value to an abstract one so that inches may be compared with pounds, centimeters, and so on, one has only to divide the Standard Deviation by the mean and multiply by 100. This gives us a measure of variability expressed in per cent known as the *Coefficient of Variability*. It is the best method we have of expressing variability as a single arithmetical term.

When terms like the Mean and the Standard Deviation are used, it is also convenient for us to know how much confidence to place in them. If we have 100 plants in one frequency distribution and 500 plants in another, common sense tells us to rely more upon the second distribution than upon the first. But to say just how much confidence to place in any calculated value, we determine what is known as the *Probable Error*. This value is preceded by the plus-or-minus sign ±. It means that there is an even chance that the true value is found within the limiting values made by adding to or subtracting from the calculated value the Probable Error. For instance, we say that a certain coefficient of variability is $6.49 \pm .32$. This means that the chances are even that the true coefficient of variability is within or without the values $6.49 + .32 = 6.81$ and $6.49 - .32 = 6.17$.

The class of greatest frequency is termed the *Mode*. When the frequency distribution is not symmetrical with respect to the mode it is said to be *skewed*.

TABLE 1.—F_2 *heights, frequencies, and formulae, and* F_3 *ranges, where parents differ in four factors.*

Classes in inches...	10	12	14	16	18	20	22	24	26	F_2 Nos.	F_3 Range	F_2 Groups
F_2 Frequencies	1	8	28	56	70	56	28	8	1	256	16 inches	
Zygotic Formulae and Frequencies in F_2 and Range of Variation in F_3, the latter in parentheses	I 1 *aabbccdd* – 1 **(Breeds true)**	VI 2 *Aabbccdd* 2 *aaBbccdd* 2 *aabbCcdd* 2 *aabbccDd* – 8 **(10=14 inches)**	II 1 *AAbbccdd* 1 *aaBBccdd* 1 *aabbCCdd* 1 *aabbccDD* – 4 **(Breed true)** X 4 *aaBbCcdd* 4 *aaBbccDd* 4 *aabbCcDd* 4 *AaBbccdd* 4 *AabbCcdd* 4 *AabbccDd* — 24 **(10=18 inches)**	VII 2 *AABbccdd* 2 *AAbbCcdd* 2 *AAbbccDd* 2 *AaBBccdd* 2 *aaBBCcdd* 2 *aaBBccDd* 2 *AabbCCdd* 2 *aaBbCCdd* 2 *aabbCCDd* 2 *AabbccDD* 2 *aaBbccDD* 2 *aabbCcDD* — 24 **(14=18 inches)** XIII 8 *aaBbCcDd* 8 *AabbCcDd* 8 *AaBbccDd* 8 *AaBbCcdd* — 32 **(10=22 inches)**	III 1 *AABBccdd* 1 *AAbbCCdd* 1 *AAbbccDD* 1 *aaBBCCdd* 1 *aaBBccDD* 1 *aabbCCDD* – 6 **(Breed true)** XI 4 *AAbbCcDd* 4 *AABbccDd* 4 *AABbCcdd* 4 *aaBBCcDd* 4 *AaBBccDd* 4 *AaBBCcdd* 4 *aaBbCCDd* 4 *AabbCCDd* 4 *AaBbCCdd* 4 *aaBbCcDD* 4 *AabbCcDD* 4 *AaBbccDD* — 48 **(14=22 inches)** XV 16 *AaBbCcDd* — 16 **(10=26 inches)**	VIII 2 *aaBbCCDD* 2 *aaBBCcDD* 2 *aaBBCCDd* 2 *AabbCCDD* 2 *AAbbCcDD* 2 *AAbbCCDd* 2 *AaBBccDD* 2 *AABbccDD* 2 *AABBccDd* 2 *AaBBCCdd* 2 *AABbCCdd* 2 *AABBCcdd* — 24 **(18=22 inches)** XIV 8 *AABbCcDd* 8 *AaBBCcDd* 8 *AaBbCCDd* 8 *AaBbCcDD* — 32 **(14=26 inches)**	IV 1 *aaBBCCDD* 1 *AAbbCCDD* 1 *AABBccDD* 1 *AABBCCdd* – 4 **(Breed true)** XII 4 *AABbCcDD* 4 *AABbCCDd* 4 *AABBCcDd* 4 *AaBbCCDD* 4 *AaBBCcDD* 4 *AaBBCCDd* — 24 **(18=26 inches)**	IX 2 *AaBBCCDD* 2 *AABbCCDD* 2 *AABBCcDD* 2 *AABBCCDd* – 8 **(22=26 inches)**	V 1 *AABBCCDD* – 1 **(Breeds true)**	16 64 96 64 16	Breed true 4 inches 8 inches 12 inches 16 inches	I-V VI-IX X-XII XIII, XIV XV

recombination of gametic factors of some kind must have taken place, for in no other way could this greater variability be explained. Contributions toward such proof have been given by the essential agreement of the investigations reported by at least eight investigators.

Perhaps before giving a brief résumé of the results of these investigations, however, it may be well to illustrate by concrete hypothetical examples just what sort of complexities the data from actual crosses are likely to show if the theory of multiple factors is correct. The practical difficulties of proving this theory have been mentioned in general terms; let us see in what form these difficulties are likely to present themselves to us in our actual experiments.

Returning to a simple case like that used before, in which the difference in height between a 10-inch plant and a 26-inch plant is supposed to be due to four independent, interchangeable, allelomorphic pairs, let us examine the results that are to be expected in the F_1, F_2, and F_3 generations and in back crosses between an F_1 individual and either parent. In this example the presence of any *single* factor is supposed to add 2 inches to the height of a zygote and all effects of environment on development are assumed to be absent.

Plant A, 10 inches high, has the zygotic formula (x =10 in.) $+ aabbccdd = 10$ inches, the plant B has the zygotic formula $(x = 10 \text{ in.}) + AABBCCDD = 26$ inches. Since all F_1 plants have the zygotic formula $(x = 10 \text{ in.}) + AaBbCcDd$, they are all 16 inches high. If we apply the formula $(\frac{1}{2} + \frac{1}{2})^8$ for determining the frequency distribution of the F_2 generation, the following result is obtained:

Classes in inches	10	12	14	16	18	20	22	24	26
No. of individuals	1	8	28	56	70	56	28	8	1

Thus, while the plants of the F_1 generation are alike, the F_2 generation is variable. The class of greatest frequency is the same as that of the F_1 generation, but it is really made up of individuals unlike in zygotic constitution and which therefore breed very differently in F_3 as is shown in Table 1.

One can see clearly what different progenies these various zygotes will produce. Zygotes selected at random from the extremes will be more likely to breed comparatively true to their type than will those near the class of greatest frequency. Thus of the 74 zygotes 10 to 14 and 22 to 26 inches high, 10, or 13.5 per cent, will breed true; 16, or 21.6 per cent, will produce progenies with ranges of variation of only four inches; and 48, or 64.9 per cent, will produce progenies with ranges of eight inches. Of the 182 zygotes 16 to 20 inches high, only 6, or 3.3 per cent, will breed true; 48, or 26.4 per cent, give progenies with four-inch ranges; 48, or 26.4 per cent, give progenies with eight inch ranges; 64, or 35.2 per cent, give progenies with twelve-inch ranges; and 16, or 8.7 per cent, give progenies with sixteen-inch ranges. The old biometrical idea that the class of greatest frequency is the *type* of the population toward which all selected individuals tend to revert is therefore erroneous. It is true, however, that if a large number of size factors were heterozygous in the F_1 generation, the grandparental types would be recovered so infrequently that practically there would be a sort of blended inheritance. One could obtain races that bred comparatively true for all of the grades intermediate between the parents, but the nearer he was working for an absolute blend the easier would be his work when he was able to raise only a few individuals in each generation. He could, nevertheless, cross extreme sizes and expect to recover either grandparental size combined with any other good quality possessed by the other grandparent if he could deal with sufficiently large numbers.

Passing now to the question of back crosses, the interesting problem arises as to whether the F_1 crossed with either parent gives data which are easier to analyze than are those obtained from breeding the F_1's *inter se.* When parents representing a single allelomorphic difference in a qualitative character are crossed, the "$F_1 \times$ the recessive parent" gives heterozygous dominants and homozygous recessives in equal numbers. It is easier to determine a 1:1 ratio than a 3:1 ratio; therefore backcrossing is a popular method in genetics. But in the cases we are discussing, no aid to factoral analysis is obtained from backcrossing. Backcrossing F_1 with either parent gives a frequency curve exactly like that obtained from breeding the F_1 individuals *inter se.* When crossed with the small parent, the distribution of the progeny is from the size of the small parent on the one hand to the size of the F_1 on the other; when crossed with the large parent, the distribution is from the size of the F_1 on the one hand to the size of the large parent on the other. The frequencies of the classes between these extremes is found by expanding the binomial $(\frac{1}{2} + \frac{1}{2})^n$ instead of $(\frac{1}{2} + \frac{1}{2})^{2n}$ as is done in the case of interbreeding the F_1 individuals.

Unfortunately one can never meet in practice a case as simple as our hypothetical one. The first perplexing factor is environmental influence. External conditions influence the whole development of both plants and animals, but nowhere is their effect so great as on size characters. In studying the heredity of size characters, therefore, one is not only confused by the fact that individuals of a certain theoretical size have different genetic possibilities, but is literally overwhelmed with difficulties by the knowledge that these theoretical sizes are *pushed upward and downward* by external causes. One does not know when he selects an individual of a certain size *whether it even belongs in the class in which it appears.* Besides this trouble, the stimulus to development due to heterozygosis also causes confusion. It is well known that hybrids often exceed their parents in size. This phenomenon is not one of heredity but one of conditions due to heredity. There is a greater stimulus to development when a character is in the heterozygous condition than when it is in the homozygous condition. Moreover this stimulus is in some degree cumulative. It increases roughly with the number of heterozygous factors. For this reason it is a force that aids environment in *pushing* individuals that theoretically should be in one size class into a different class.

But these are not the only troubles. One is not often—if ever—able to cross two plants in which any particular size difference can be represented by a notation like $(X = 10 \text{ inches}) + aabbcc$ and $(X = 10 \text{ inches}) + AABBCC$. A great majority of plants are frequently crossed thru insect or other agencies, while a comparatively large number have flower structures of such a character that they are always cross-fertilized. Heterozygosis is also favored by bisexuality wherever it occurs. Many plant varieties are therefore hybrid complexes from which types of different kinds can be isolated by inbreeding. Other plants which are especially adapted to self-fertilization may be homozygous in nearly all their gametic factors, for self-fertilization tends to eliminate heterozygosis. But such cases are not common. Ordinarily one may presume that the two individuals he wishes to cross are heterozygous for a considerable number of factors. This reduces the excess of variability which the F_2 generation might be expected to show over that shown by the F_1 generation. Furthermore, it gives an F_1 generation in which the zygotic constitution of each individual is not the same as would be the case were both parents completely homozygous. A series of F_2 generations raised from different F_1 individuals, for this reason, may be quite different in their variability.

On the other hand, it may sometimes happen that the variability of the F_2 generation exceeds that of the F_1 generation in a degree unsuspected by the difference in size of the parents. East (1910) predicted that if the theory of multiple interchangeable segregating factors gave a true interpretation of the behavior of quantitative characters in crosses, cases would be found when parents identical in size would show marked size recombinations in F_2. The reasons for such a phenomenon are these. Two plants are the same size. The size in plant No. 1 is due to the influence of factors "*AABBCC*," the size in plant No. 2 is due to the influence of factors "*DDEEFF*." Three interchangeable factors in the homozygous condition have produced the same size effect in each case. But no one of these factors in plant No. 1 is allelomorphic to a factor in plant No. 2. The F_1 generation is therefore theoretically about the same size as either parent, for there are six factors present in each case. The fact that in one case these six factors are made up of three homozygous allelomorphic pairs, and in the other case by six heterozygous allelomorphic pairs, makes no difference in the size produced (leaving out of consideration the stimulus due to heterozygosis). In F_2, on the other hand, recombination gives a frequency distribution far above and far below this typical size. It is the distribution obtained by expanding the binomial $(\frac{1}{2}+\frac{1}{2})^{12}$. In fact, it is exactly what would have occurred if a plant "*aabbccddeeff*" had been crossed with a plant "*AABBCCDDEEFF*."

Recently Hayes (1912) has reported a fulfillment of this prediction. He crossed two varieties of *Nicotiana tabacum* and studied the inheritance of number of leaves. In both varieties the mean number of leaves was about 20 (see Table 2), and the model class about 20. The variability of each variety was small: for the "Cuban" variety it was 7.53 ± .293 in the first year studied and 5.29 ± .227 in the second year studied, while for the "Havana" variety it was 6.98 ± .272 the first year and 8.87 ± .345 the second year. The mean, the mode, and the variability of the F_1 generation were about like those of the parents. The coefficient of variability was 6.10 ± .237. The F_2 generation, however, showed a range of variability so much greater than the F_1 generation that it can hardly be questioned that segregation and recombination of several "leaf-number" factors has occurred. The coefficient of variability was 15.84 ± .549.

Cases of the same kind are reported later in this paper. (See Table 33 and Table 38).

TABLE 2.—*Frequency distribution of number of leaves per plant of cross between two varieties of Nicotiana tabacum.*

Number	Year grown	Leaves of parent	Leaf classes																				Total	Mean	Stand. Dev.	Coef. Var.
			14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33				
405 Cuban	1910	......	......	......	1	7	16	37	36	35	12	4	1	1	......	......	......	......	......	......	......	......	150	19.9 ± .082	1.50 ± .059	7.53 ± .293
405-1	1911	21	......	......	......	......	3	16	34	46	20	5	......	......	......	......	......	......	......	......	......	......	124	20.6 ± .066	1.09 ± .047	5.29 ± .227
402, Havana	1910	......	......	......	......	3	22	44	42	22	10	6	1	......	......	......	......	......	......	......	......	......	150	19.8 ± .076	1.38 ± .054	6.98 ± .272
402-1	1911	20	......	......	1	10	11	23	34	31	16	12	3	2	......	......	......	......	......	......	......	......	143	20.3 ± .101	1.80 ± .072	8.87 ± .354
(402x405) F_1	1910	......	......	1	1	3	8	39	60	30	7	......	......	1	......	......	......	......	......	......	......	......	150	19.8 ± .067	1.21 ± .047	6.10 ± .237
(402x405)-1 F_2	1911	20	3	4	8	8	20	18	30	24	25	17	16	5	4	3	1	1	1	1	2	1	192	20.9 ± .161	3.31 ± .114	15.84 ± .549

Note. 405-1 is a frequency distribution of the progeny of a single self-fertilized plant of 405, and 402-1 is a frequency distribution of a single self-fertilized plant of 402.

Let us now consider the experimental evidence that is thought to show segregation and recombination of factors of the kind that have just been discussed.

Nilsson-Ehle in 1908 stated that he had found greater variability in F_2 than in F_1 in numerous crosses where the parents differed in quantitative characters. Data were presented, however, only on qualitative characters.

East (1910), in his independent proposal of this method of interpreting the inheritance of quantitative characters by a Mendelian notation, showed how certain data on the inheritance of different numbers of rows of seeds on the ear of maize could be thus analyzed. His observations were not sufficiently numerous, however, to be convincing.

A little later Emerson's (1910) paper on inheritance of quantitative characters appeared. Here segregation and recombination of size factors was clearly shown in three species; viz, *Cucurbita pepo*, *Phaseolus vulgaris*, and *Zea mays*.

Cucurbita pepo. A cross between a long, narrow Crookneck squash and a round flat Scallop squash gave an F_1 generation intermediate in all respects and an F_2 generation showing a complete series of sizes and shapes running from that of the Crookneck to that of the Scallop. The Means and Coefficients of variation were as shown below. The Probable Errors were not calculated but the number of individuals was large enough for one to have considerable confidence in the results.

Race or hybrid	Means		Coefficients of variation	
	Length	Diameter	Length	Diameter
	Cm.	*Cm.*	*Per cent*	*Per cent*
Crookneck	39.6	11.4	17.0	12.0
Scallop	7.4	17.8	15.8	12.6
F_1 hybrid	17.5	17.5	19.0	12.6
F_2 hybrid	19.6	13.2	42.7	58.8

A cross between a long narrow Spoon gourd and a shorter, much thicker Horned gourd gave a similar increase of variability in F_2 over that shown by F_1.

Race or hybrid	Means		Coefficients of variation	
	Length	Diameter	Length	Diameter
	Cm.	*Cm.*	*Per cent*	*Per cent*
Horned	10.3	9.0	9.4	9.9
Spoon	14.0	4.2	15.6	16.0
F_1 hybrid	12.9	5.6	15.8	15.7
F_2 hybrid	15.7	5.5	37.5	21.2

Phaseolus vulgaris. A large number of crosses between beans differing in length, breadth, weight, and thickness were made. These, one and all, showed segregation and recombination in F_2. A few cases recorded behaved as shown in the following table. It should be noted, however, that the F_2 generation was grown in the winter in a greenhouse, while the F_1 generation was grown in the open field. Any effect that this difference of environment would have on the variability would probably be towards a smaller variability for the greenhouse-grown plants.

Race or hybrid	Means				Coefficients of variation			
	Weight	Length	Breadth	Thickness	Weight	Length	Breadth	Thickness
	Cg.	*Mm.*	*Mm.*	*Mm.*	*Per cent*	*Per cent*	*Per cent*	*Per cent*
Fillbasket .	32.2	13.8	7.6	4.4	7.93	3.22	2.20	3.78
Longfellow	28.3	12.9	5.6	4.6	6.09	3.27	3.57	4.54
F_1 hybrid..	28.4	13.0	6.5	4.7	7.63	3.53	1.43	3.37
F_2 hybrid..	36.8	14.1	7.0	5.0	17.43	8.83	10.33	9.45
Fillbasket .	32.2	13.8	7.6	4.4	7.93	3.22	2.20	3.78
Snowflake .	16.4	8.3	5.7	4.7	9.40	2.89	3.68	5.38
F_1 hybrid..	25.4	11.4	6.4	4.5	9.90	3.47	2.70	3.83
F_2 hybrid..	28.6	11.3	6.9	4.8	24.48	8.32	5.96	8.95

Emerson also reported segregation of size of seed and height of stalk in F_2 when maize varieties differing in those characters were crossed. These results will not be discussed for they are reported in full later in this paper. For the same reason, the papers of East (1911) and East and Hayes (1911) reporting segregation of number of rows, height of stalk, size of ear, and size of seed in maize will not be considered here.

Shull (1910) also reported observations on the number of rows on the maize ear, showing that while an F_1 population is no more variable than the pure strains from which it came, the F_2 population is much more variable.

In 1911 an excellent paper of Tammes (1911) appeared. Miss Tammes made many crosses between varieties and between species in the genus *Linum*, and studied the inheritance of such characters as length and breadth of seed and length and breadth of the petals. A cultivated flax known as Egyptian with seeds varying from 5.0 mm. to 6.9 mm. in length was crossed with *Linum angustifolium* whose seeds vary from 2.4 mm. to 2.6 mm. in length. The seeds of the F_1 generation were intermediate, varying from 3.6 mm. to 4.2 mm. The range of variability in the F_2 generation, on the other hand, was very large, ranging from 2.8 mm. minimum to 4.7 mm. maximum. A second cross between another cultivated flax with a minimum seed length of 3.5 mm. and a maximum seed length of 4.5 mm., with *Linum angustifolium* gave F_1 seeds varying in length from 3.0 mm. to 3.5 mm. and F_2 seeds varying in length from 2.3 mm. to 4.2 mm. In opposition to these species crosses giving a comparatively uniform F_1, a cross between the two varieties Common Flax and Egyptian flax and its reciprocal gave a variable progeny, tho here, too, an intermediate character was manifested. Further, the F_2 was but little more variable than the F_1. Altho this result might easily be explained by gametic impurity of the parents, it was so interesting that the progeny of extreme individuals was followed to the F_3 generation. There was no doubt, from the data obtained in this generation, that various F_2 plants did have different gametic formulae, for the progeny produced were different. A somewhat similar result was obtained when Egyptian flax was crossed with *Linum crepitans.*

Length and breadth of petals was studied in a cross between Egyptian flax and *Linum angustifolium* and its reciprocal. In both cases the variability of F_2 was greater than that of F_1.

In Nilsson-Ehle's extended monograph of 1909, it was stated that crosses of wheats and of oats were in progress in which such characters as number of seeds per head, hairiness, height, length of glume, length of spike, size of seed, hardiness, and resistance to rust were being studied. In 1911 that portion of his results relating to the internodal length of wheat spikes and resistance to rust was reported. Some of the various crosses had been followed to the F_3 generation. The data show clearly that varietal differences in these characters depend on several independent gametic factors, altho their exact behavior is not known. Possibly complete analysis of such a thing as resistance to a fungal parasite cannot be cleared up by field work, as Nilsson-Ehle believes, for there is evidence that genotypic differences in the parasites exist which complicate matters.

Hayes' (1912) paper on tobacco has already been mentioned in another connection. He presented data upon number of leaves per plant, height of plant, and size of leaf. In each case there was a greater variability in F_2 than in F_1, and this difference he shows clearly can be most logically described by assuming segregation and recombination of "size" factors.

Von Tschermak (1902) some years ago found that an early blossoming pea crossed with a late blossoming variety gave an intermediate F_1 and a variable F_2. Afterwards he found (1904) that only a portion of the "early" and "late" segregates bred true, and that this behavior could not be expressed by a simple Mendelian formula. In a recent paper (1912) he gives a full analysis of his later results on this character complex, and expresses them with the Mendelian notation by assuming multiple factors.

Belling (1912) has reported a study of a cross between two bean varieties that differed in many characters. In such quantitative characters as time of flowering, size of pod, and size of seed, there seems to be no question but that factors have segregated and recombined. In the F_2 generation there was invariably great variability, and in several cases extremes appeared that were beyond the grandparental types. The author does not commit himself further than to say that segregation has occurred, but the data that he presents are exactly what should be expected on the multiple factor interpretation.

Phillips (1912) has studied the inheritance of body weight in a cross between the large French Rouen duck and the small domestic mallard. The first named variety is more than double the weight of the latter. He concludes that: "The amplitude of variation of the F_2 animals is greater than that of the F_1 animals, but does not extend beyond the nearer limit of the respective grandparental races." As a matter of fact, with an F_2 population of only 33 individuals it is remarkable that the variability was so great.

In addition to these papers giving the results of actual investigations, Lang (1911) has published a long article showing how the results of Castle (1909) on ear length of rabbits might have a Mendelian interpretation by using Nilsson-Ehle's scheme. As Lang might have suspected, Castle had already acknowledged (in public lectures) the possibility of such an interpretation, an acknowledgment that was afterwards published (Castle 1911).

METHODS AND MATERIALS USED.

The methods employed in making these studies, including the growing of the plants, guarding the pollination, recording the data, etc., have been given in detail in previous papers (East and Hayes 1911 and Emerson 1911). A brief statement here will therefore suffice. In so far as possible the different generations of the various crosses and representatives of the parent varieties have been grown together in the same season. Where comparisons are made between lots grown in different years the fact is indicated in the tables or accompanying text. The various lots to be compared have always been grown on the most nearly uniform soil available. In most cases certain plantings have been duplicated in different parts of the field, but it has not been possible to carry out this plan as fully as might be desired. Careful planting of seed, placing label-stakes in the field, and recording the position of the different lots on planting plans have practically removed the possibility of mistakes in identification.

In the production of hand-pollinated seed, both staminate and pistillate inflorescences have been protected with heavy manilla bags. In the transfer of pollen from the tassel-bags to the silks great care has been exercised to prevent the entrance of undesired pollen. After pollination each ear has been rebagged and a label tag attached to it. The experimental error in regard to pollination is believed to be not greater on the average than about one seed per ear. (See East and Hayes 1911.)

The measurements of height of plant were made in the field. The main stalk of each plant was chosen for measurement and the distance from the surface of the ground to the top of the tassel determined. The number of nodes was determined by counting the leaves, except near the base of the stalk, where it was necessary to make a more careful examination. It is probable that not all the basal nodes were noted, but any slight error of that sort is not serious, for, since all counts for the several families of any one series were made by one person, the same error would likely be made in case of each lot. The ears were measured after having been harvested and allowed to dry. Time of flowering was noted at approximately the same time each day. At each examination any plant whose staminate flowers had begun to open was tagged with a label bearing the date and the individual number of the plant.

The frequency distributions and the computations of statistical constants have all been checked over once, and, where practicable, this has been done by a second person.

It is not claimed that no errors have been made in connection with the studies, but great care has been taken to avoid them and it is our belief that the data presented are essentially accurate.

The plants of California pop and of Tom Thumb pop used in making crosses with Missouri dent and also the plants from which the later representatives of these varieties were grown were from seed obtained from seed dealers. The Missouri dent plants used in crossing and those from which all later lots of this variety came were from a single open-fertilized ear obtained from an exhibitor at the National Corn Exposition held at Omaha. Neither of these varieties, therefore, had been self pollinated before the experiments were begun. The other varieties used in these experiments were in most cases selfed for at least two generations before being crossed, but in some cases there were used parent ears obtained from commercial growers who made a specialty of the types furnished. In general, the descriptive notes below include only such characters of the several varieties as were made use of in the experiments reported in this paper.

The strain of Tom Thumb used in the cross with Missouri dent is a very dwarf and very early pop corn. The plants tiller considerably and have short stalks with few nodes. The tillers are mostly shorter than the main stalk and frequently end in an ear instead of in a tassel. The silks protrude from the leaf sheaths in advance of the ear-shoots and some days before the pollen is shed. The ears are very short and usually have about twelve rows of small, smooth, flinty grains.

California pop is also a dwarf corn, but the stalks are considerably taller than those of Tom Thumb and have nearly twice as many nodes. The tillers are more numerous than in Tom Thumb, are mostly of about the same height as the main stalk, and like it have one or more ears and end in a tassel. The ears are even smaller than those of Tom Thumb and contain numerous, irregularly arranged grains. The latter are sharply rice-pointed and considerably smaller than the grains of Tom Thumb. Unlike Tom Thumb this variety is very late in flowering and in ripening.

Missouri dent is a large late corn. It blossoms at about the same time as California pop, but matures somewhat later. The stalks are tall and stout and have few tillers. The latter are often short and without tassels or ears and, when tall, almost always end in a tassel, but frequently lack ears. The ears are rather short for dent corn, but are large in diameter and contain numerous rows of kernels—most commonly sixteen to twenty. The grains are of a fairly smooth dent type and medium broad for corn of that type. They are, of course, very much larger than the grains of either one of the two pop corn varieties.

The strain of Tom Thumb (No. 60) used in crosses with Black Mexican was similar to the strain described above in height of stalks, number of tillers, earliness, etc., but had slightly smaller ears and seeds.

Black Mexican sweet (No. 54) has medium tall stalks and fairly numerous tillers. Its ears are long and almost always have eight rows of rather broad, wrinkled grains.

Watson flint (No. 5) has medium short stalks and very long ears with eight rows of grains as the prevailing type.

Leaming dent (No. 6) has very tall stalks and long ears with from 12 to 20 rows, 16 rows being the modal condition.

Of the varieties used in the other crosses, only one character—number of rows—is of direct concern to us here. Illinois low protein (No. 2) is a dent corn with 12 to 20 rows and a mode of 16 rows. Illinois high protein (No. 8) is a 12-rowed dent with a range from 10 to 18 rows. Early Crosby (No. 18) is a 12-rowed sweet corn ranging from 8 to 14 rows. Sturges' flint (No. 11) is strongly 12-rowed in type, only a few ears having other row numbers ranging from 8 to 16. Longfellow flint (No. 15) is an 8-rowed type, rarely ever having ears with any other number of rows.

INHERITANCE OF NUMBER OF ROWS PER EAR.

The maize ear may be regarded either as a fusion of four or more spikes, each joint of the rachis bearing two spikelets, or as meristic variations or repetitions of a rachis bearing two spikelets. The rows are paired, and no case is known where a member of a pair has been aborted. De Vries has regarded the number of rows on the maize ear as continuous variations which are inherited in a somewhat temporary manner. He came to this conclusion from a selection experiment in which he was able to increase the number of rows in the variety with which he was working, from an average of about 13 rows per ear to an average of about 20 rows per ear in seven years, only to find that the selected variety gradually reverted to the type of the original variety when selections ceased. We do not question these facts but the interpretation that de Vries gave to them. The work of East, Emerson, Hayes, and Shull has shown conclusively that the reversion in de Vries' selected strains came about simply thru the intercrossing of the progeny of plants that were somatically alike but genotypically different. The evidence presented here shows that the character complex of number of rows per ear is made up of a series of cumulative unit factors independent in their inheritance.

Altho there seems to be no good reason why this character complex should not be considered to be of the same nature as that of other size characters in which variation appears to be continuous, it possesses the advantage of being divisible into a discontinuous series of numbers. From such a numerical series one can obtain some idea of the prevalence and magnitude of non-inherited fluctuations. A large number of counts of the number of rows of both ears on stalks that bore two ears has shown a variation which is usually ±2 rows, altho it may be as high as ±4 rows. An ear which appears to belong to any particular class, therefore, may be pushed by external conditions into the wrong class, but the proper class will be one of five in which the apparent class is the central one. All ears, however do not have the same ability to respond to external stimuli. The 8-rowed condition, which is the last subtraction form in which maize appears, may be selected until the strain breeds very true. In a count of the population of an isolated maize field where Longfellow, an 8-rowed flint, had been grown for many years, 4 four-rowed, 993 eight-rowed, 2 ten-rowed, and 1 twelve-rowed ears were found. On the other hand, a Leaming dent, selected for many years and inbred by hand for six generations, still gave a range of variation from 12 to 20 rows. Part of this difference in behavior shown by races with a low number of rows and races with a high number of rows is undoubtedly due to correlation with other characters—both physiological and gametic—but at the same time it is more reasonable to suppose that an ear which can vary in any one of eight spikes will show a greater degree of fluctuation than one which can vary only in any one of four spikes. For this reason it is likely that strains with a high number of rows will never show the low variability seen in strains with a low number of rows.

Taking into consideration the probable amount of fluctuating variation, several interesting points are presented in the tables showing the frequency distributions of the number of rows per ear in the various crosses.

Tables 3, 4, and 5 show the results from three different crosses in which No. 5,* an 8-rowed white flint, was crossed with other varieties. In Table 3 the male parent was No. 6, the well known Leaming dent. In the particular strain from which this male parent was taken, the modal condition (that is, the class of greatest frequency) was sixteen. Four F_2 families were grown, of which only one was markedly more variable than the F_1 generation. The modal condition of F_1(12) was intermediate between the two parents. In three of the F_2 families, the modal condition was ten; in the remaining family it was twelve. In Table 4, the male parent, a white dent, was also typically twelve rowed. The F_1 and F_2 generations were comparable to those in Table 3. While the F_2 generation was much more variable than the F_1 generation, no great confidence can be placed in the results, owing to the meager numbers of the F_1 generation. Table 5 shows a peculiar result. Here No. 5 was the male parent, while the female parent was a 12-rowed flint which is very low in variability. Both the F_1 and F_2 generations were typically twelve rowed.

These data form the basis for several speculations. In two crosses the typical condition of the hybrid was intermediate. In this last cross the 12-rowed condition appears to be dominant. Is this due to the fact that the 12-rowed parent was the mother? Is it due to the fact that the maternal individual was gametically 16 rowed? Is it due to there being few differences between the parents by which complications thru correlation might arise, for both are flints? It seems probable that the last two suggestions are likely to be at the basis of the phenomenon, but there is no way of deciding from the evidence at hand.

Table 6 merely shows that this same 12-rowed variety when crossed with another variety which is typically twelve rowed shows no greater variability than one of the parents.

Table 7 gives the data from a cross between an 8-rowed flint and a very variable 12-rowed dent. The description of the F_1 generation was not taken at the time, as the cross was made primarily to study another question. The omission is partially

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* For description of these varieties see page 29.

covered by the data from nine ears that were selfed artificially. The interesting part of the record is decidedly the F_3 families, for they arose from three ears, each having a different number of rows. The 8-rowed mother gave progeny with a few more 10-rowed ears than 8-rowed ears, but one can see what a tendency there is toward the 8-rowed condition. It would probably be very easy to recover a typical 8-rowed race in the next generation. On the other hand, the progeny of the 14-rowed mother is a variable family with the modal condition at 14 rows. The number of ears exceeding this condition is sufficient to lead one to believe that a race like the No. 8 parent would be as easy to obtain as the one like the No. 15 parent. But this could not be done, we believe, by selecting ears having the flint condition. There is certainly some sort of a correlation between the dent condition and a high number of rows, for all of the ears with a high number of rows are dent in character.

This dent, No. 8, was also crossed (Table 8) with an 8-rowed sweet, No. 54 (Black Mexican). *Nine F_3 families were grown in this cross, and strains exactly like each parental condition were recovered.* The progeny of (8x54)1-13 are as typically 8-rowed as the Black Mexican variety, while the progeny of No. (8x54)1-5 and No. (8x54)1-6 are practically like No. 8, in number of rows, tho they both differ in other characters.

Tables 9, 10, and 11 are similar to Table 8. Different individuals belonging to the commercial varieties known as Tom Thumb and Black Mexican were crossed. This gives one some idea of the various gametic possibilities within what might be considered single races. No. 54 is a comparatively pure 8-rowed race, but No. 60, since it produces individuals having from 8 to 16 rows, may be expected to have various potentialities.

Table 9 lists the data from the cross known as 60-3x54. The F_1 generation is very variable, if confidence can be placed in a population of only 33 ears. Three F_2 families were grown. Family (60-3x54)-1 grew from a 16-rowed ear, and shows clearly that it had within it possibilities different from those of ears (60-3x54)-5 and (60-3x54)-6, the other two ears that produced F_2 families. In fact, tho ear (60-3x54)-1 produced no daughter ears with *less* than 12 rows, ear (60-3x54)-6 produced no daughter ears with *more* than 12 rows. No further proof is necessary to show that one of the parental plants was heterozygous for certain of the factors necessary for the production of rows greater than eight in number. The F_1 individuals, in other words, were different in gametic constitution.

Even the family (60-3x54)-1 contained individuals heterozygous for several factors, moreover, tho apparently it was somewhat low in variability. This is shown by the varied appearance of the progeny of the four ears from it that were followed into the F_3 generation. The progeny of ear (60-3x54)1-1EO, a 16-rowed ear, showed a modal condition of 16 rows, with ears having both 18 and 20 rows. None had less than 12 rows, but this undoubtedly was an accidental circumstance because the 12-rowed class occurred 15 times. In a perfect sample of the population, therefore, one would expect to find individuals with both 10 and 8 rows. At the same time the modal condition of this family was unquestionably sixteen rows, while that of the other three families was twelve, fourteen, and twelve. This result is very striking when compared with the F_3 families (60-3x54)-5-3 and (60-3x54)6-5, where the modal conditions are 10 and 8, respectively.

Five F_2 families of the cross 60-5x54 (Table 10) were grown. These were again different in character. Family (60-5x54)-6 was characteristically twelve rowed, while family (60-5x54)-11 was characteristically 8 and 10 rowed. Unfortunately only a single 8-rowed ear from family (60-5x54)2 was carried to the F_3 generation and its progeny tended to form an 8-rowed type. Most remarkable for its tendency to form a non-variable family, however, was family (60-5x54)-11. Five ears were grown and in each there was a notable tendency toward an 8-rowed strain. It may be doubted very seriously whether selection toward a 16-rowed type in all of these families would be successful. Success might come in one or two instances,—for gametic possibilities of a 16-rowed race may still exist in some of the families,—but it would not be likely in every family. On the other hand, the two fraternities raised from ears (60-5x54)12-1ES and (60-5x54)-12-2ES are very high in variability and show that they contain potentialities of yielding races averaging from 16 to 20 rows.

In cross 60-8x54, Table 11, a race characteristically 8 rowed and a race characteristically 12 rowed were obtained in the F_3 generation.

In Table 12 are presented data derived from a cross of Tom Thumb pop and Missouri dent. Few records are available except those of F_2 and F_3 families grown in 1911. No F_1 generation was grown in 1911 and the records of F_1 ears produced in 1910 were accidently destroyed. There are entered in the table, however, under the designation of F_1 generation, such records as were obtainable from the ears kept as samples. Tho both parent varieties were grown in 1911, no record was obtained of the ears of Missouri dent since this variety failed to mature in Massachusetts. Owing to poor germination only a few plants of Tom Thumb pop were grown in 1911 and these, because of the unfavorable weather in early summer, while they were developing, and perhaps also because of previous selfing, produced few ears. The entries in the table under the designation of Tom Thumb pop and of Missouri dent, like those of the F_1 generation, were obtained from such ears of these varieties as had been preserved as samples.

Such data as are available indicate a 12-rowed type for Tom Thumb pop. The range of variation indicated for Missouri dent suggests that the stock of that variety used may have been heterozygous for several factors for number of rows, with a tendency to produce 16 to 20 rows per ear. The three F_2 families, 510 grown in Nebraska in 1910 and 1127 and 1128 grown in Massachusetts in 1911, were somewhat different in the mean number of their rows and in their ranges of variation. Such behavior as this was to have been expected if either or both of the parents were heterozygous for any of the factors concerned in determining number of rows per ear. The range of variation in the F_2 fraternities extended somewhat beyond the extreme ranges of the parent varieties—if indeed the records available can be said to show the ranges of the parents.

The best evidence of real segregation in F_2 of the factors for number of rows is furnished by the F_3 families. As pointed out more fully later, in the discussion of inheritance of height of stalk, the differences between the several F_3 lots under consideration here cannot be ascribed to the possible heterozygous condition of the parent plants used in this cross, because all the F_3 families are descendants of one F_2 fraternity, No. 510, which in turn, of course, was the progeny of a single F_1 plant. What other F_2 progenies or other F_1 plants may have been like or even what other sorts of gametes the parent plants may have produced, in addition to the two gametes whose union resulted finally in these F_3 families, is absolutely immaterial. Since all these F_3 families were related in this way, their differences are ascribable only to segregation of size factors in F_2. Among the F_3 lots, there were included at least one 12-rowed type, 1142, like Tom Thumb pop, several lots of a 16-rowed type, and two or three of a 20-rowed type, particularly 1140 and 1141, with perhaps also 14-rowed and 18-rowed types, 1144, 1134, 1135, etc. That a type with more than 20 rows might be isolated by selection from a family like 1140 seems probable, but the possibility can be tested only by further breeding.

TABLE 3.—*Frequency distribution of number of rows per ear in cross (5x6).*

Designation	Gen.	Rows of parents	Classes for number of rows per ear								
			8	10	12	14	16	18	20	22	24
No. 5 flint (2 yrs.)	P	8	289	2	2						
No. 6 dent	P	18	......	..	6	31	51	18	4		
No. 5x6	F_1	8	13	36	53	10					
(5x6)-1	F_2	12	12	48	35	9	1				
(5x6)-2	F_2	10	7	22	15	2					
(5x6)-22	F_2	10	8	45	31	1					
(5x6)-23	F_2	12	4	25	60	18	4	2	1		

TABLE 4.—*Frequency distribution of number of rows per ear in cross (5x2).*

Designation	Gen.	Rows of parents	Classes for number of rows per ear								
			8	10	12	14	16	18	20	22	24
No. 5 flint (2 yrs.)	P	8	289	2	2						
No. 2 dent	P	16	...	..	2	14	56	42	20	1	1
No. 5x2	F_1	8	1	9	20	4					
(5x2)-6	F_2	10	4	18	61	14	3	1			

TABLE 5.—*Frequency distribution of number of rows per ear in cross (11x5).*

Designation	Gen.	Rows of parent	Classes for number of rows per ear								
			8	10	12	14	16	18	20	22	24
No. 11 flint	P	12	1	4	387	7	1				
No. 5 flint	P	8	280	2	2						
No. 11x5	F_1	12	2	11	26	2					
(11x5)-8	F_2	12	10	38	107	23	8				
(11x5)-18	F_2	10	19	33	100	5					

TABLE 6.—*Frequency distribution of number of rows per ear in cross (11x18).*

Designation	Gen.	Rows of parents	Classes for number of rows per ear								
			8	10	12	14	16	18	20	22	24
No. 11 flint	P	12	1	4	387	7	1				
No. 18 sugar (2 yrs.)	P	12	13	32	51	4					
No. 11x18	F_1	12	2	10	24	1	1				
(11x18)-4	F_2	12	1	9	78	10					
(11x18)-10	F_2	10	8	13	62	13					

TABLE 7.—*Frequency distribution of number of rows per ear in cross (15x8).*

Designation	Gen.	Rows of parents	Classes for number of rows per ear								
			8	10	12	14	16	18	20	22	24
No. 15 flint	P	8	100	1							
No. 8 dent	P	14	...	3	54	36	12	2			
No. 15x18	F_1	8	1	2	5	1					
(15x8)-2	F_2	10	14	15	28	9	1				
(15x8)-3	F_2	12	4	13	25	6	3				
(15x8)-2-10	F_3	14	...	1	8	14	6	1	1		
(15x8)-2-1	F_3	8	32	35	23	4					
(15x8)-2-5	F_3	12	4	41	116	15	3	1			

TABLE 8.—*Frequency distribution of number of rows per ear in cross (8x54).*

Designation	Gen.	Rows of parents	Classes for number of rows per ear								
			8	10	12	14	16	18	20	22	24
No. 8 dent	P	12	.	3	54	36	12	2			
No. 54 sugar	P	8	89	25	7						
No. 8x54	F_1	12	1	6	14						
(8x54)-1	F_2	12	9	22	16	1					
(8x54)-5	F_2	12	1	3	16	1					
(8x54)-1-1	F_3	10	..	15	87	4					
(8x54)-1-2	F_3	8	20	38	50						
(8x54)-1-2a	F_3	10	61	48	54						
(8x54)-1-3	F_3	10	32	75	15						
(8x54)-1-3a	F_3	8	5	20	27	1					
(8x54)-1-5	F_3	12	..	33	158	26	3				
(8x54)-1-6	F_3	12	4	36	109	8	2				
(8x54)-1-10	F_3	8	Very irregular, mostly 8-rowed								
(8x54)-1-13	F_3	10	96	43	8						

TABLE 9.—*Frequency distribution of number of rows per ear in cross (60-3x54).*

Designation	Gen.	Rows of parents	Classes for number of rows per ear						
			8	10	12	14	16	18	20
60	P	12	1	13	11	1	1		
54	P	8	25	2	1				
(60-3x54)	F_1	10	8	10	12	2	1		
(60-3x54)-1	F_2	16	..	..	4	8	5		
(60-3x54)-5	F_2	12	7	8	14	2	1		
(60-3x54)-6	F_2	12	5	11	8				
(60-3x54)-1-1AS	F_3	12	1	6	23	40	29		
(60-3x54)-1-1EO	F_3	16	..	..	15	16	25	6	1
(60-3x54)-1-5	F_3	16	2	10	38	21	10		
(60-3x54)-1-7	F_3	12	2	30	90	40	12		
(60-3x54)-5-1	F_3	12	10	22	44	10			
(60-3x54)-5-2	F_3	10	4	7	15	2	1		
(60-3x54)-5-3	F_3	8	42	73	25				
(60-3x55)-6-1	F_3	10	23	29	15	2			
(60-3x54)-6-5	F_3	10	55	33	6				

TABLE 10.—*Frequency distribution of number of rows per ear in cross (60-5x54).*

Designation	Gen.	Rows of parents	Classes for number of rows per ear						
			8	10	12	14	16	18	20
60	P	12	1	13	11	1	1		
54	P	8	25	2	1				
(60-5x54)	F_1	12	2	6	54	7	1		
(60-5x54)-2	F_2	12	2	7	6	1			
(60-5x54)-6	F_2	14	0	1	10	2			
(60-5x54)-8	F_2	12	3	13	4				
(60-5x54)-11	F_2	10	14	14					
(60-5x54)-12	F_2	12	1	4	14	10	2		
(60-5x54)-2-3CS	F_3	8	46	20	4				
(60-5x54)-11-2CS	F_3	10	43	44	14	1			
(60-5x54)-11-3AS	F_3	10	56	10					
(60-5x54)-11-5AS	F_3	8	65	26	1				
(60-5x54)-11-5CS	F_3	10	50	24	7				
(60-5x54)-11-10AS	F_3	8	71	33	10				
(60-5x54)-12-1ES	F_3	12	12	34	51	28	20	6	2
(60-5x54)-12-2ES	F_3	12	13	44	62	28	2		

TABLE 11.—*Frequency distribution of number of rows per ear in cross (60-8x54).*

Designation	Gen.	Rows of parents	Classes for number of rows per ear						
			8	10	12	14	16	18	20
60	P	12	1	13	11	1	1		
54	P	8	25	2	1				
(60-8x54)	F_1	10	3	5	2	1			
(60-8x54)-1	F_2	10	20	3					
(60-8x54)-8	F_2	14	2	2	14	10			
(60-8x54)-1-1ES	F_3	8	61	10	1	1			
(60-8x54)-8-8CS	F_3	14	20	43	53	7	1		

Table 12.—*Frequency distribution of number of rows per ear in cross between Tom Thumb pop and Missouri dent.*

Designation	Year	Gen.	Rows of parents	Classes for number of rows per ear											Mean
				8	10	12	14	16	18	20	22	24	26	28	
Missouri dent		P				1	1	7	9	6	1				17.7 (?)
Tom Thumb pop		P			2	15	3								11.4 (?)
F_1 generation		F_1						2	2	1					(?)
510	1910	F_2		1	2	16	36	54	31	21	9	1			16.30 ± .15
1127	1911	F_2			1	13	26	15	6	2					14.57 ± .18
1128	1911	F_2				3	8	15	19	15	1	2			17.46 ± .22
1142	1911	F_3	12	1	7	46	17	3							12.38 ± .11
1132	1911	F_3	16		2	15	7	5	3						13.50 ± .26
1144	1911	F_3	14		1	15	29	33	3						14.54 ± .13
1134	1911	F_3	18			13	19	10	1						13.95 ± .16
* 1150	1911	F_3	18			8	15	14	1	1					14.56 ± .20
1131	1911	F_3	20			6	16	21	4	3					15.28 ± .19
* 1147	1911	F_3	20			9	22	19	11	2	1				15.31 ± .19
1133	1911	F_3	12			5	13	22	8	2					15.56 ± .18
1143	1911	F_3	14			3	15	28	14	1					15.84 ± .15
1145	1911	F_3	18			4	15	32	8	6					15.91 ± .17
1146	1911	F_3	14			3	26	28	22	5					16.00 ± .14
1149	1911	F_3	16			4	16	26	5	6					15.75 ± .18
1138	1911	F_3	18			3	12	33	20	8	2				16.62 ± .16
1139	1911	F_3	16			3	10	23	26	13	7				17.39 ± .18
1135	1911	F_3	16			5	13	16	18	8	3	4			17.07 ± .25
1137	1911	F_3	18				4	11	9	16	12	1			18.90 ± .24
1141	1911	F_3	20				2	17	16	20	8	4	1		18.91 ± .21
1140	1911	F_3	20					1	10	31	20	9	3	1	21.04 ± .17

*The brace indicates that the families joined by it are duplicates grown in different parts of the field from seed of the same ear.

INHERITANCE OF LENGTH OF EARS.

Length of the ear of maize is perhaps as good a character complex for inheritance studies as is found in the species. It is not affected by heterozygosis to as great a degree as height, altho some effect may be traced. Like other size characters, length of ear is intimately connected in development and in heredity with other complexes, yet it is not so markedly affected by external conditions as the height of plant. This statement, however, must be considered as somewhat tentative. Length of ear may be greatly changed by external conditions, yet if soil

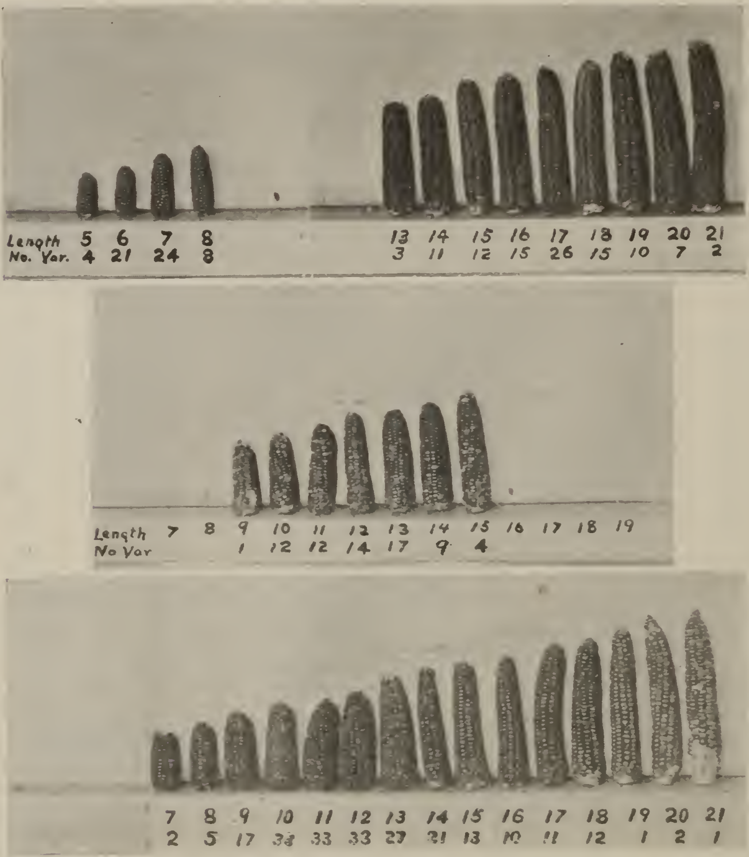

Fig. 1. Size of ears of Tom Thumb pop and Black Mexican sweet (top row), the F_1 generation of the cross between these varieties (middle row), and the F_2 generation of the same cross (bottom row),—1-9 natural size.

and moisture conditions are as uniform as is possible in field experiments and if these conditions are suitable for normal development, unavoidable changes in meteorological conditions do not change the ear development to any great extent. The greatest changes occur thru physiological correlation. Number of rows of seeds, and therefore total number of seeds, is inversely correlated with ear length. Size of seed and possibly the character of the seed may also have some effect, tho just what this effect is we are not prepared to say. Size of plant is directly correlated with length of ear.

How much of these correlations is gametic in the sense of true gametic couplings is unknown. It is apparently physiological in most cases, tho this physiological correlation is imposed by gametic constitution. In other words, a plant which is gametically large and a plant that is gametically small have different possibilities of ear development even when the same genes immediately affecting ear development are present. Expressed in genetic language, zygote (tall + ear length *AABBCC*) gives longer ears than zygote (short + ear length *AABBCC*).

Tables 13, 14, and 15 give the data on lengths of ear for the crosses (60-3x54), (60-5x54), and (60-8x54). These, it will be remembered, are three crosses made between individual plants of Tom Thumb pop (No. 60) and Black Mexican sweet (No. 54).

Unfortunately the data on the F_1 generations were taken only for the cross (60-5x54). It can be stated definitely that the range of variability in the other two families was no greater, but not that the percentage of variability was no greater. It is possible that a slight error is introduced, therefore, in comparing the F_2 generations of the crosses (60-3x54) and (60-8x54) with the F_1 data taken for cross (60-5x54). From what is known of the F_1 generations, however, it is thought that the error is very small.

In cross (60-3x54), three F_2 families were grown from selfed ears of the F_1 generation. If 12.48 ± .72 per cent represents the correct variability of the F_1 generation (Table 13), then the variability of each F_2 generation is much greater, namely, 16.70 ± .67 per cent, 15.87 ± .63 per cent, and 15.54 ± .90 per cent respectively. Perhaps it is wrong to compare the variability of the total population in F_2 with that of F_1, for the F_1 ears followed to the F_2 generation may have had different gametic formulae. The figures 22.30 ± .74 per cent are simply given for what they are worth. There is no way of disregarding the fact that one F_2 family, (60-3x54)-1, had a greater range of variability than F_1 by *five* classes. Individuals were obtained, therefore, that were exactly like typical individuals of both of the parental varieties.

Let us see how these individuals behaved in the F_3 generation.

Disregarding the progeny of ear (60-3x54)5-2CO because there were only 29 individuals, there is a range of variability from 12.66 ± .68 per cent to 19.61 ± .98 per cent. There is no question then but that the progenies of different F_2 ears show great deviations in variability of ear length, as should be expected if segregation of multiple factors occurs. The most interesting fact, however, is the differences in mean length of ear shown by the various frequency distributions of F_3. The mean length of the progeny of ear (60-3x54)6-5EO is only 9.7 ± .11 cm. They compare very favorably with Tom Thumb pop, the smaller parent, tho they are sweet segregates breeding true. Perhaps here again extremely small segregates are less common than they should be theoretically on account of the slight increase in size due to heterozygosis. Perhaps also the *combination of genes representing other character complexes* that would be most favorable to small size was not obtained.

On the other hand, the progeny of ear (60-3x54)1-7ES average 15.9 ± .15 cm. They have nearly the same length as the long-eared parent, Black Mexican. They average about twelve rows per ear, and are starchy segregates, however, so perhaps they are not as long as they would be with the same zygotic structure for ear size, if they were typically 8-rowed and wrinkled like the Black Mexican. At least there is good reason for suspecting a physiological correlation between long ears and few rows per ear.

Cross (60-5x54), Table 14, does not show such uniform increase in variability in F_2 over that shown by F_1 as does the cross just described. There is an increased range of variability and an increase in the calculated coefficient of variability in each of the five F_2 families grown, but in three cases out of five this increase is not significant. If one adds together the entries in all of the five families, he obtains a frequency distribution with a coefficient of variability of 15.67 ± .30 per cent. This coefficient is considerably larger than that of F_1, 12.48 ± .72 per cent, but the procedure is questionable.

The nine F_3 families grown show great range in variability. The smallest coefficient is 10.46 ± .60 per cent and the largest coefficient 21.68 ± .88 per cent. It is unfortunate that no extremely short ears and but one extremely long ear were selfed, but this should not be regarded as the fault of the experimenter. When the ear is bagged for selfing, one is unable to tell whether it will be large or small. Over 300 ears were selfed in the F_2 generation and of these about 275 matured, but among them were found but one normal ear of 7 cm. and but one ear of 18 cm. Mice destroyed the short ear, and recourse to ears 9 cm. and 10 cm. in length was necessary. The average of the progeny of

Fig. 2. Representative ears of an F_3 generation of a cross between Tom Thumb pop and Black Mexican sweet—2-9 natural size.

Fig. 3. Representative ears of an F_3 generation of a cross between Tom Thumb pop and Black Mexican sweet—2-9 natural size.

Fig. 4. Representative ears of an F_3 generation of a cross between Tom Thumb pop and Black Mexican sweet—2-9 natural size.

Fig. 5. Representative ears of an F_3 generation of a cross between Tom Thumb pop and Black Mexican sweet—2-9 natural size.

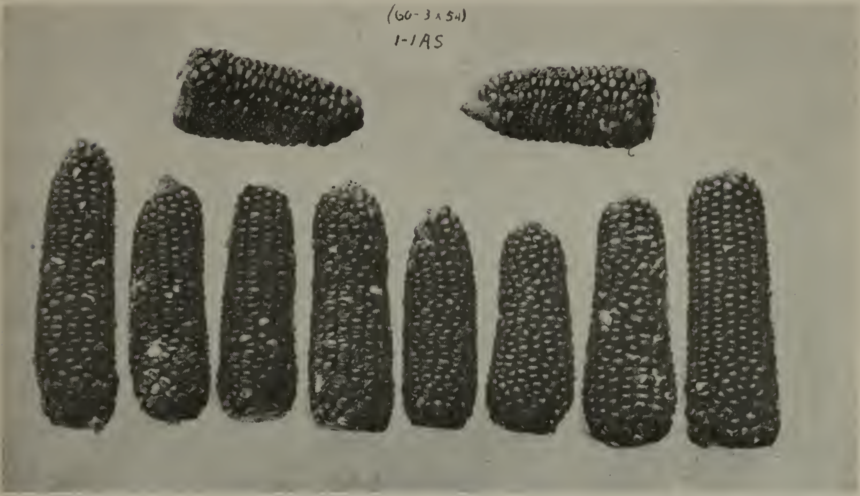

Fig. 6. Representative ears of an F_3 generation of a cross between Tom Thumb pop and Black Mexican sweet—2-9 natural size.

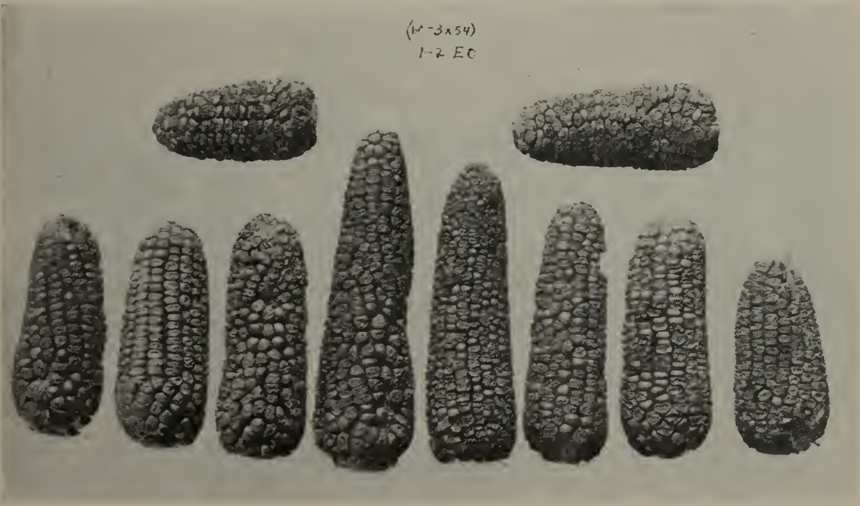

Fig. 7. Representative ears of an F_3 generation of a cross between Tom Thumb pop and Black Mexican sweet—2-9 natural size.

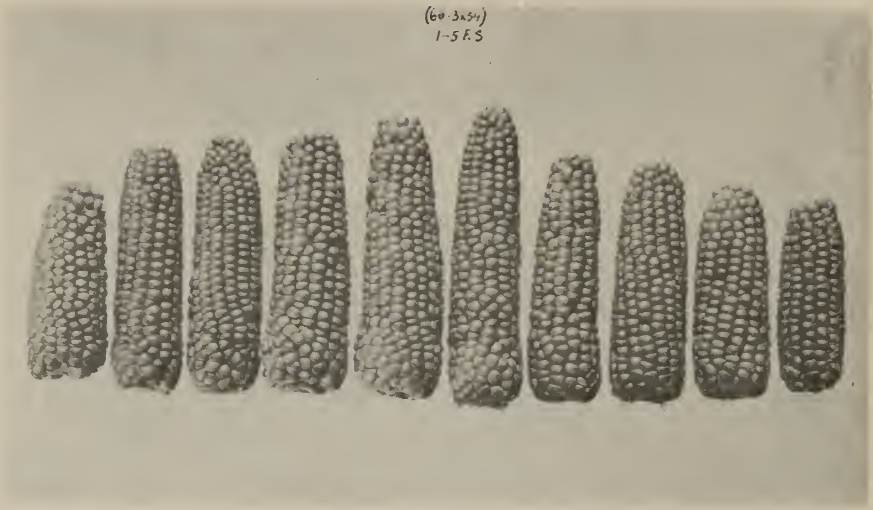

Fig. 8. Representative ears of an F_3 generation of a cross between Tom Thumb pop and Black Mexican sweet—2-9 natural size.

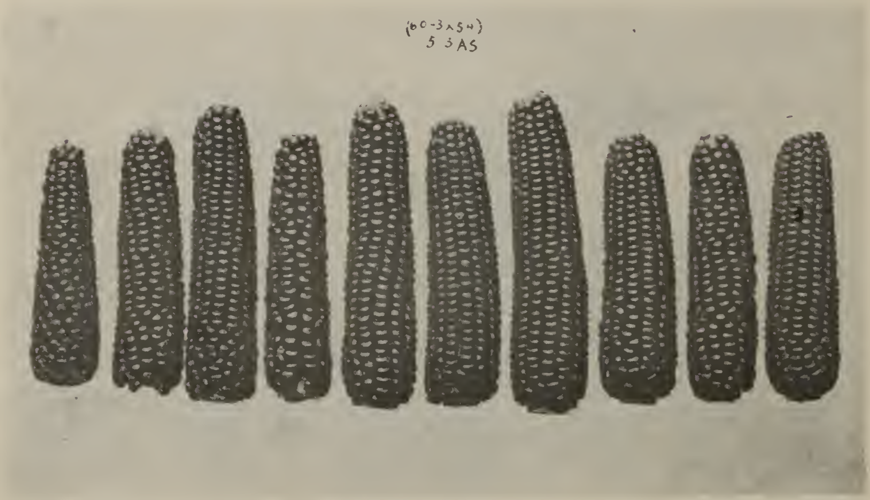

Fig. 9. Representative ears of an F_3 generation of a cross between Tom Thumb pop and Black Mexican sweet—2-9 natural size.

Fig. 10. Representative ears of an F_3 generation of a cross between Tom Thumb pop and Black Mexican sweet—2-9 natural size.

Fig. 11. Representative ears of an F_3 generation of a cross between Tom Thumb pop and Black Mexican sweet—2-9 natural size.

ear (60-5x54)12-1ES, which was itself 9 cm. long, was 10.0 ± .12 cm. The average of the progeny of ear (60-5x54)-12-2ES, an ear of 9 cm. long, was 9.2 ± .07. These families of short ears contrast noticeably with the family produced by ear (60-5x54)-11-2AS an ear 18 cm. long, for this family averaged 15.8 ± .13 cm. In fact, the extreme variates of the shortest family and the longest family scarcely overlap.

Cross (60-8x54) is notable for the extreme variability of F_2. Two families were grown, in which the coefficients of variability were 14.80 ± .69 per cent and 18.95 ± .89 per cent respectively. The total number of F_2 individuals gave a coefficient of variability of 17.44 ± .41 per cent. It happened that no long F_2 ears were selfed in this cross. Two medium length ears were grown primarily for other purposes, however, and their progeny measured. One family was very short, averaging 9.3 ± .07 cm. and the other family very variable, showing a coefficient of variability of 21.52 ± 1.23.

The data for ear length in the cross of Tom Thumb with Missouri dent are given in Table 16. As in the case of number of rows, the parents and F_1 are represented by measurements taken from ears kept for samples. The records of the F_1 generation grown in 1910 were accidently destroyed and no F_1 was grown in 1911. The Missouri dent, owing to its lateness of maturity, did not ripen in 1911 and Tom Thumb produced few ears that year. As a matter of fact the samples preserved represent the parents and F_1 fairly well. The F_1 ears as a whole were practically as long as those of Missouri dent. The F_2 families differed in ear length as they did in other characters to be noted later. The parent stocks evidently were heterozygous in some factors influencing ear length. None of the F_2 ears were as short as the average Tom Thumb pop ear but many were shorter than any ears of Missouri dent or of the F_1 generation. The F_3 families, all of which came from a single F_2 family and therefore, from a single F_1 ear, differed noticeably in ear length. The shortest average length was a little over 10 centimeters and the longest a little over 18 centimeters. The longest ears of some families were only slightly longer than the shortest ears of other families. It seems probable that types of ear length like the parent types as well as various intermediate types could readily be established from this material.

Table 17 includes all available data in regard to ear length from the cross of California pop with Missouri dent. F_1 families 502 and 505 were grown under fairly favorable weather conditions in 1910, while all the other families listed were grown under the conditions of extreme drouth prevailing at Lincoln, Nebraska, in 1911. The general appearance of the Missouri dent plants, No. 833, indicated that they suffered more seriously from the dry weather than did the California pop plants. Many of the Missouri dent plants produced no ears and a considerable number of the ears produced were little better than "nubbins." The F_1 plants, families 836 and 837, were apparently much less seriously affected by the unfavorable weather than any of the other lots, which doubtless, in part at least, accounts for the fact that their ears averaged quite as long as the ears of the Missouri dent parent. The mean ear lengths of the F_2 families were distinctly intermediate between the means of the parents. Altho the range of variation in the F_2 families was not extreme, the variability in F_2, as measured by the coefficient of variation, was in general considerably greater than that of the F_1 families.

TABLE 13.—*Frequency distribution of lengths of ears in cross (60-3x54).*

Designation	Gen.	Parent class	Class centers in cm. for lengths of ears																	Mean	Coef. var.
			5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21		
60	P	......	4	21	24	8	......	......	......	......	......	......	......	......	......	......	......	......	......	6.6 ± .07	12.27 ± .78
54	P	......	......	......	......	......	......	......	......	......	3	11	12	15	26	15	10	7	2	16.8 ± .12	11.13 ± .53
(60-5x54)	F_1	......	......	......	......	......	1	12	12	14	17	9	4	......	......	......	......	......	......	12.1 ± .12	12.48 ± .72
(60-3x54)-1	F_2	......	......	......	1	4	10	11	24	27	26	27	12	6	2	1	......	......	......	12.4 ± .11	16.70 ± .67
(60-3x54)-5	F_2	......	......	......	......	......	5	9	9	29	28	33	26	18	13	8	1	......	......	13.8 ± .11	15.87 ± .63
(60-3x54)-6	F_2	......	......	......	......	6	4	6	14	17	14	8	1	1	......	......	......	......	......	11.7 ± .26	15.54 ± .90
(60-3x54)-total	F_2	......	......	......	1	10	19	26	47	73	68	68	39	25	15	9	1	......	......	12.6 ± .13	22.30 ± .74
(60-3x54)-1-1AS	F_3	11	......	......	4	3	13	13	18	23	9	5	4	3	3	1	......	......	......	11.4 ± .15	19.61 ± .98
(60-3x54)-1-1EO	F_3	14	......	......	......	......	3	8	14	13	16	8	1	......	......	......	......	......	......	11.9 ± .12	12.06 ± .74
(60-3x54)-1-2AS	F_3	11	......	......	......	2	5	6	6	11	13	15	11	9	2	......	......	......	......	13.0 ± .17	16.99 ± .93
(60-3x54)-1-5ES	F_3	10	......	......	......	4	6	16	20	16	11	5	3	......	......	......	......	......	......	11.3 ± .12	14.68 ± .79
(60-3x54)-1-7ES	F_3	18	......	......	......	......	......	6	9	10	8	18	14	26	24	28	12	10	4	15.9 ± .15	17.87 ± .68
(60-3x54)-5-1BO	F_3	8	......	......	......	......	1	8	5	13	10	18	18	11	2	2	......	......	......	13.6 ± .14	14.77 ± .77
(60-3x54)-5-2CO	F_3	7	......	......	......	1	......	7	10	7	4	......	......	......	......	......	......	......	......	11.2 ± .14	9.94 ± .89
(60-3x54)-5-3AS	F_3	12	......	2	3	3	17	16	19	25	26	17	7	4	......	2	......	......	......	11.8 ± .13	14.27 ± .59
(60-3x54)-6-1ES	F_3	10	......	1	......	14	15	24	10	2	2	1	......	......	......	......	......	......	......	9.7 ± .11	14.37 ± .84
(60-3x54)-6-5EO	F_3	11	......	......	2	3	4	14	19	26	19	7	......	......	......	......	......	......	......	11.5 ± .11	13.66 ± .68

TABLE 14.—*Frequency distribution of lengths of ears in cross (60-5x54).*

Designation	Gen.	Parent class	Class centers in cm. for lengths of ears																	Mean	Coef. var.
			5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21		
60	P	......	4	21	24	8	......	......	......	......	......	......	......	......	......	......	......	......	......	6.6 ± .07	12.27 ± .78
54	P	......	......	......	......	......	......	......	......	......	3	11	12	15	26	15	10	7	2	16.8 ± .12	11.13 ± .53
(60-5x54)	F_1	......	......	......	......	......	1	12	12	14	17	9	4	......	......	......	......	......	......	12.1 ± .12	12.48 ± .72
(60-5x54)-2	F_2	......	......	......	......	......	1	5	8	27	32	31	28	13	5	2	......	......	......	13.6 ± .16	12.70 ± .50
(60-5x54)-6	F_2	......	......	......	1	2	6	9	22	43	24	14	11	......	3	3	1	......	......	12.4 ± .11	16.22 ± .67
(60-5x54)-8	F_2	......	......	......	1	2	3	8	9	17	19	14	10	7	7	1	......	......	......	13.0 ± .16	17.60 ± .87
(60-5x54)-11	F_2	......	......	......	......	......	3	5	7	15	21	19	10	6	2	......	......	......	......	13.1 ± .13	13.59 ± .70
(60-5x54)-12	F_2	......	......	......	2	1	9	29	34	43	36	13	4	1	......	......	......	......	......	11.7 ± .08	13.36 ± .49
(60-5x54)-total	F_2	......	......	......	4	5	22	56	80	145	129	91	63	27	17	6	1	......	......	12.7 ± .06	15.67 ± .30
(60-5x54)-2-3AS	F_3	11	......	......	......	......	1	5	8	9	25	20	2	......	......	......	......	......	......	12.7 ± .11	10.46 ± .60
(60-5x54)-11-2AS	F_3	18	......	......	......	......	......	......	......	9	7	13	18	25	20	6	1	......	......	15.8 ± .13	12.56 ± .57
(60-5x54)-11-2CS	F_3	10	......	1	......	8	22	25	15	22	7	2	......	......	......	......	......	......	......	10.5 ± .10	14.92 ± .72
(60-5x54)-11-3AS	F_3	12	......	......	1	1	3	6	10	6	17	12	7	3	......	......	......	......	......	12.5 ± .17	15.89 ± .96
(60-5x54)-11-5AS	F_3	12	......	......	......	1	1	3	9	17	28	14	13	5	1	......	......	......	......	13.1 ± .12	12.62 ± .64
(60-5x54)-11-5CS	F_3	10	......	1	1	1	9	18	16	13	16	5	1	......	......	......	......	......	......	11.2 ± .13	15.36 ± .83
(60-5x54)-11-10AS	F_3	10	......	......	......	......	4	1	6	11	24	18	17	16	11	4	1	1	......	14.2 ± .14	15.23 ± .70
(60-5x54)-12-1ES	F_3	9	......	5	13	20	28	28	30	10	7	6	3	2	1	......	......	......	......	10.0 ± .12	21.68 ± .88
(60-5x54)-12-2ES	F_3	10	......	3	10	33	47	40	18	3	......	......	......	......	......	......	......	......	......	9.2 ± .07	13.55 ± .53

TABLE 15.—*Frequency distribution of lengths of ears in cross (60-8x54).*

Designation	Gen.	Parent class	Class centers in cm. for lengths of ears																	Mean	Coef. var.
			5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21		
60	P	......	4	21	24	8	......	......	......	......	......	......	......	......	......	......	......	......	......	6.6 ± .07	12.27 ± .78
54	P	......	......	......	......	......	......	......	......	......	3	11	12	15	26	15	10	7	2	16.8 ± .12	11.13 ± .53
(60-5x54)	F_1	......	......	......	......	......	1	12	12	14	17	9	4	......	......	......	......	......	......	12.1 ± .12	12.48 ± .72
(60-8x54)-1	F_2	......	......	......	......	2	4	1	7	17	16	17	10	10	11	12	1	2	1	14.3 ± .17	18.95 ± .89
(60-8x54)-8	F_2	......	......	......	2	3	13	32	26	16	11	4	3	......	......	......	......	......	......	10.9 ± .10	14.80 ± .69
(60-8x54)-total	F_2	......	......	......	2	5	17	33	33	33	27	21	13	10	11	12	1	2	1	12.9 ± .08	17.44 ± .41
(60-8x54)-1-1ES	F_3	12	......	3	4	1	2	3	3	6	11	19	12	6	3	......	......	......	......	12.9 ± .22	21.52 ± 1.23
(60-8x54)-8-8CS	F_3	11	......	5	3	19	42	39	14	2	......	......	......	......	......	......	......	......	......	9.3 ± .07	13.16 ± .57

TABLE 16.—*Frequency distribution of length of ear in cross between Tom Thumb pop and Missouri dent.*

Designation	Year	Gen.	Parent class	Class centers in centimeters for length of ears																					Mean
				5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	
Missouri dent....	....	P	....	......	......	......	......	......	......	......	......	1	4	4	5	3	4	2	2	1	......	......	......	......	16.6 (?)
Tom Thumb pop	....	P	....	1	1	5	6	3	2	......	......	......	......	......	......	......	......	......	......	......	......	......	......	......	7.8 (?)
F1 generation......	....	F1	....	......	......	......	......	......	......	......	......	......	......	......	1	..	...	2	1	1	......	......	......	......	(?)
510..............	1910	F2	....	......	......	4	3	......	2	8	14	14	19	34	34	20	18	11	5	..	1	1	......	......	15.15 ± .14
1127..............	1911	F2	....	......	......	......	......	......	......	1	2	3	3	5	7	9	11	11	6	2	1	......	1	1	17.40 ± .23
1128..............	1911	F2	....	......	......	......	......	......	2	1	1	5	6	5	6	9	11	10	2	4	1	......	......	......	16.66 ± .23
1132..............	1911	F3	13	......	5	1	1	3	5	4	8	2	2	1	......	......	......	......	......	......	......	......	......	......	10.34 ± .30
* 1150..............	1911	F3	16	......	2	1	1	......	2	1	2	5	10	6	3	3	2	1	......	......	......	......	......	......	13.62 ± .33
* 1134..............	1911	F3	16	......	......	......	2	2	1	5	4	4	7	6	3	5	2	2	......	......	......	......	......	......	13.81 ± .29
1140..............	1911	F3	11	......	......	......	1	......	3	8	8	19	12	13	6	2	2	1	......	......	......	......	......	......	13.56 ± .16
1133..............	1911	F3	12	......	......	......	1	1	3	9	3	14	11	5	3	......	......	......	......	......	......	......	......	......	12.82 ± .17
1137..............	1911	F2	13	......	......	......	......	1	2	4	4	12	20	8	2	......	......	......	......	......	......	......	......	......	13.38 ± .14
1135..............	1911	F3	14	......	......	......	1	......	1	......	2	5	12	6	8	11	12	6	3	....	......	......	......	......	15.97 ± .20
1143..............	1911	F3	14	......	......	......	......	......	......	2	5	7	13	12	15	6	1	......	......	......	......	......	......	......	14.67 ± .14
1141..............	1911	F3	16	......	......	......	......	......	1	2	3	4	4	14	12	11	10	4	......	......	......	......	......	......	15.76 ± .17
1149..............	1911	F3	16	......	......	......	......	......	......	1	3	4	13	8	7	8	8	2	1	1	......	1	......	......	15.70 ± .21
* 1147..............	1911	F3	17	......	......	......	......	......	......	1	1	1	3	3	11	14	15	10	4	......	1	......	......	......	17.17 ± .16
* 1131..............	1911	F3	17	......	......	......	......	......	......	......	......	1	3	2	5	7	10	8	7	4	2	1	......	......	18.14 ± .21
1138..............	1911	F3	17	......	......	......	......	......	......	......	1	2	5	8	12	15	12	8	10	2	3	......	......	......	17.32 ± .17
1142..............	1911	F3	17	......	......	......	......	......	......	......	1	3	5	9	21	12	14	4	4	1	......	......	......	......	16.55 ± .14
1144..............	1911	F3	15	......	......	......	......	......	......	......	1	2	4	6	15	21	16	10	4	2	......	......	......	......	17.05 ± .13
1139..............	1911	F3	20	......	......	......		....	......	......	1	5	4	5	16	16	15	12	3	4	1	......	......	......	17.07 ± .16
1145..............	1911	F3	19	......	......	......	......	......	......	......	......	6	8	7	21	12	6	2	2	1	...	......	......	......	16.03 ± .15
1146..............	1911	F3	17	......	......	......	......	......	......	......	......	......	6	10	9	13	16	12	10	4	3	......	1	......	17.73 ± .16

* Duplicates.

TABLE 17.—*Frequency distribution of length of ears in cross between California pop and Missouri dent.*

Designation	Year	Gen.	Class centers in centimeters for length of ears																	Mean	Coef. var.
			4.25	5.25	6.25	7.25	8.25	9.25	10.25	11.25	12.25	13.25	14.25	15.25	16.25	17.25	18.25	19.25	20.25		
834	1911	P	3	10	21	5	....	....	....	....	....	....	....	....	....	....	....	....	....	5.97 ± .09	13.13 ± 1.02
835	1911	P	5	17	4	....	....	....	....	....	....	....	....	....	....	....	....	....	....	5.21 ± .08	11.25 ± 1.08
833	1911	P	....	....	....	....	....	2	1	2	1	1	6	1	3	2	2	1	1	14.64 ± .42	20.61 ± 2.14
836	1911	F_1	....	....	....	....	....	1	...	...	...	4	6	13	8	3	2	....	....	15.25 ± .18	10.56 ± .84
837	1911	F_1	....	....	....	....	....	....	1	1	2	1	8	5	3	6	1	1	....	15.15 ± .26	13.58 ± 1.23
829	1911	F_2	....	....	1	6	3	7	11	12	9	7	4	3	....	....	....	....	....	11.00 ± .19	20.00 ± 1.26
830	1911	F_2	....	....	1	5	4	4	8	7	10	4	3	....	....	....	....	....	....	10.68 ± .21	19.67 ± 1.44
831	1911	F_2	2	1	2	7	3	4	6	10	3	8	2	1	....	....	....	....	....	10.19 ± .26	26.55 ± 1.94
832	1911	F_2	....	1	...	2	8	6	8	6	9	8	6	1	....	....	....	....	....	11.05 ± .21	20.44 ± 1.38
839	1911	F_2	....	....	....	....	2	3	8	7	14	8	4	2	....	....	....	....	....	11.88 ± .16	14.03 ± 1.00
840	1911	F_2	....	....	1	2	3	10	13	10	7	8	5	1	....	....	....	....	....	11.03 ± .17	17.91 ± 1.14

INHERITANCE OF DIAMETER OF EARS.

Diameter of ear is directly correlated with number of rows and with seed size and probably also, negatively, with length of ear. In general, ears show less fluctuation in diameter than they do in length.

Table 18 exhibits the data obtained from the cross of California pop and Missouri dent. As grown in 1911, the former, Nos. 834 and 839, had a mean ear diameter of a little less than 25 millimeters and the latter, No. 833, a mean diameter of a little over 50 millimeters. The mean diameter of F_1 ears grown the same year, Nos. 836 and 837, was nearly 42 millimeters, or about 4 millimeters greater than the mid-parental value. It seems likely that this small increase over the mid-parental diameter is due to the increased vigor accompanying heterozygosis, for the mean diameters of the F_2 families averaged only a little over 37 millimeters, or almost exactly half way between the parents. The F_1 families grown under the more favorable conditions of 1910, Nos. 502 and 503, naturally had somewhat greater ear diameters than the F_1 families of 1911.

The ranges of variation of the F_2 families were not great, having extended only from a little above the mean of California pop to a little below the mean of Missouri dent. While the number of individuals of no one F_2 family was large, the total number of individuals of the six F_2 families was 321, or more than a sufficient number to give an even chance of obtaining all possible combinations of factors if the parents differed by only four factors. It is probable, however, that neither the seed size nor the number of rows of the parents differed by so few as two factors and, since diameter of ears is directly related to both of these other ear characters, it would not be strange if the difference in diameter of ears between Missouri dent and California pop were due to as many as seven or eight factors. In the latter case, something like 65,000 F_2 individuals would have to be grown to afford an even chance of obtaining a single plant each, gametically like the two parent types as regards factors influencing diameter of ears. In this case, some of the smallest and some of the largest F_2 ears of the 321 should prove heterozygous for diameter factors and should, therefore, produce F_3 plants more extreme than any plants of F_2.

This apparently is exactly what happened in case of the cross of Missouri dent with Tom Thumb pop, the data for which are presented in Table 19. Several of the F_3 families contained individuals with ears of smaller diameter than any F_2 ear. In fact the mean diameter of No. 1132 was practically equal to the diameter of the smallest ears of the F_2 families grown the same year, Nos. 1127 and 1128, and also equal to the particular F_2 ear from which it came. Whether types with still smaller ears can be isolated by selection from family 1132 remains to be seen.

Many of the F_2 families of this cross were distinctly different from others in ear diameter. There were six families, for instance, the smallest ears of which were larger than the largest ears of family 1132. The mean diameter varied from nearly 31 to nearly 46 millimeters.

TABLE 18.—*Frequency distribution of diameter of ears in cross between California pop and Missouri dent.*

Desig-nation	Year	Gen.	Class centers in millimeters for diameter of ears																		Mean	Coef. var.
			22.5	24.5	26.5	28.5	30.5	32.5	34.5	36.5	38.5	40.5	42.5	44.5	46.5	48.5	50.5	52.5	54.5	56.5		
833	1911	P	....	....	....	....	....	....	....	....	....	....	2	2	2	4	2	4	5	2	50.33 ± .60	8.46 ± .84
834	1911	P	8	20	8	2	1	....	....	....	....	....	....	....	....	....	....	....	....	....	24.86 ± .20	7.26 ± .55
835	1911	P	7	12	6	1	....	....	....	....	....	....	....	....	....	....	....	....	....	....	24.58 ± .21	6.57 ± .61
836	1911	F_1	....	....	....	....	....	1	2	2	2	4	15	7	3	1	....	....	....	....	41.91 ± .39	8.29 ± .65
837	1911	F_1	....	....	....	....	....	....	....	2	6	7	6	5	3	....	....	....	....	....	41.53 ± .36	6.88 ± .61
829	1911	F_2	....	....	....	1	....	5	5	15	8	15	5	6	3	....	....	....	....	....	38.82 ± .33	10.06 ± .61
830	1911	F_2	....	....	1	...	4	5	10	7	5	7	4	2	1	....	....	....	....	....	36.80 ± .43	11.77 ± .84
831	1911	F_2	....	....	....	3	9	7	8	12	7	2	1	....	....	....	....	....	....	....	34.58 ± .33	9.91 ± .68
832	1911	F_2	....	....	1	2	3	8	7	11	12	6	4	1	....	....	....	....	....	....	36.32 ± .36	10.77 ± .70
839	1911	F_2	....	....	....	....	....	5	1	10	8	9	8	4	3	....	....	....	....	....	39.42 ± .37	9.66 ± .67
840	1911	F_2	....	....	....	....	7	5	7	14	10	4	8	4	1	....	....	....	....	....	37.33 ± .37	11.29 ± .70

TABLE 19.—*Frequency distribution of diameter of ears in cross between Tom Thumb pop and Missouri dent.*

Designation	Year	Gen.	Parent class	Class centers in millimeters for diameter of ears																					Mean
				20.5	22.5	24.5	26.5	28.5	30.5	32.5	34.5	36.5	38.5	40.5	42.5	44.5	46.5	48.5	50.5	52.5	54.5	56.5	58.5	60.5	62.5	
Mo. dent		P															1	1	3	6	2		2	1		52.9 (?)
Tom Thumb		P		4	5	6	3																			23.4 (?)
F1 Generation.		F1											1	2	2											(?)
510	1910	F2				<													>†							37.34 ± .22
1127	1911	F2							1	4	4	2	8	9	7	11	8	4	4	1						42.09 ± .44
1128	1911	F2								3	3	5	7	10	9	5	13	3	3	2						42.37 ± .42
1132	1911	F3	30.5				5	5	7	11	4															30.75 ± .30
1133	1911	F3	32.5				1	3	7	17	12	6	2	2												33.38 ± .28
1142	1911	F3	32.5					4	7	16	20	21	4	2												34.31 ± .21
1139	1911	F3	34.5					1	2	10	22	28	11	6	2											35.94 ± .20
1146	1911	F3	34.5					1	2	3	17	21	19	17	3	1										37.29 ± .22
* { 1134	1911	F3	38.5					1	1	5	12	9	8	6	1											36.22 ± .31
1150	1911	F3	38.5						1	2	6	10	9	4	6	1										37.83 ± .35
1135	1911	F3	38.5						1	3	7	17	15	10	7	3	4									38.65 ± .30
1137	1911	F3	36.5						2	4	6	14	9	11	6	1										37.74 ± .30
1141	1911	F3	36 5							9	6	23	22	7	1											36.94 ± .20
1138	1911	F3	38.5								3	4	13	18	19	17	1	2	1							41.50 ± .24
1144	1911	F3	40.5								1	1	7	13	23	22	10	3	1							43.02 ± .21
1140	1911	F3	40.5									3	8	18	18	20	7	1								42.34 ± .21
1145	1911	F3	42.5									1	8	17	22	10	6	1								42.16 ± .21
* { 1147	1911	F3	44.5									1	4	14	21	11	9	3	1							43.03 ± .24
1131	1911	F3	44.5										5	9	12	15	7	1	1							43.18 ± .26
1143	1911	F3	42.5										4	9	13	13	11	4	7							44.40 ± .29
1149	1911	F3	44.5										2	3	7	14	18	5	5	3						45.76 ± .29

* Duplicates.

† Measurements were taken in tenth-inches. The frequency distribution from 1.0 to 2.0 inches was: 1, 4, 14, 24, 41, 47, 26, 19, 8, 0, 3.

INHERITANCE OF WEIGHT OF SEEDS.

The behavior of seed weight in the crosses (60-3x54), (60-5x54), and (60-8x54) may be considered together. The detailed data are shown in Tables 20, 21, and 22. In making the weights for this study, it was necessary to use a scheme by which the sugary or wrinkled seeds of the Black Mexican parent, No. 54, and of certain recessive segregates from the cross, could be weighed as starchy seeds. This end was accomplished by having starchy families growing near the wrinkled families. Crossed seeds which had become starchy thru xenia were usually obtained in sufficient numbers to make data upon 25 seeds possible. An insufficient number of hybrid starchy seeds, however, accounts for the meager data in certain families. It must be understood that this method of weighing is correct in theory, since the size of the seed is governed by the development of the pericarp and since this is unaffected by the pollen from a starchy race. The pericarp is merely filled out with starch by the dominance of starchiness in the F_1 endosperm.

In discussing these results, the weight of 25 seeds is taken as the unit quantity. When the mean weight of any particular family is stated, therefore, this is what is meant.

Tom Thumb No. 60 has an extremely small seed. The mean weight is 2.7 ± .03 grams. The mean weight of the Black Mexican is 8.3 ± .11 grams, but this is probably too high. These data were taken from plants raised from commercial seed, which was in a very mixed condition. The last three positive classes should probably be discarded for this reason. The ears upon which they were borne showed plainly that they had come from hybrid seed, —the male parent evidently being some large dent variety.

Small numbers in the frequency distributions of both parents and hybridity in the Black Mexican undoubtedly make the coefficients of variability—14.44 ± .90 per cent and 14.54 ± .96 per cent respectively—too high; but, even leaving this fact out of consideration, the ten F_2 families grown show increased variability over that of the F_1 generation in every case. The range of variability in each case overlaps that of the grandparents. The extreme F_2 individuals do not reach the extremes of either parent, but if one leaves out of account the three higher classes of the larger parent, for the reason given above, the extreme size of the Black Mexican is generally obtained. This may be a questionable procedure, but, as a matter of fact, the individuals used as the male parents of the crosses were only average in this character. The F_3 families often gave segregates with seeds as small as the smallest Tom Thumb seeds.

The F_3 families having the smallest mean seed weight were

(60-3x54)-6-1ES, (60-5x54)-2-3AS, and (60-5x54)-12-2ES, with means of 4.9 ± .08, 4.8 ± .07, and 4.6 ± .04 respectively. These means do not approach the mean of the Tom Thumb race very closely, yet the appearance of the families, is, as a whole, very like the Tom Thumb. Furthermore, the great difference in variability among the F_3 families, extending as it does from 13.90 ± .67 per cent to 24.96 ± 1.35 per cent, indicates a difference in heterozygosity from which we might well argue the ease of producing races like the original parental strains with a little more selection.

The family (60-5x54)-11-2AS, coming from a large segregate of F_2, gave a fraternity with a mean seed weight of 7.3 ± .08 grams. This is probably very close to the true mean seed weight of a well-selected Black Mexican race.

TABLE 20.—*Frequency distribution of weights of seeds in cross (60-3x54).*

Designation	Gen.	Parent class	Class centers in grams for weights of 25 seeds																				Mean	Coef. var.
			2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5	6.0	6.5	7.0	7.5	8.0	8.5	9.0	9.5	10.0	10.5	11.0	11.5		
60	P	......	7	22	28	4	......	......	......	......	......	......	......	......	......	......	......	......	......	......	......	......	2.7 ± .03	14.44 ± .90
54	P	......	......	......	......	......	......	......	......	......	2	2	5	7	14	10	5	4	1	2	1	2	8.3 ± .11	14.54 ± .96
(60-5x54)	F1	......	......	......	......	5	12	13	17	4	3	......	......	......	......	......	......	......	......	......	......	......	4.6 ± .06	13.91 ± .91
(60-3x54)-1	F2	......	......	......	......	3	7	13	25	28	20	18	3	3	......	2	......	......	......	......	......	......	5.5 ± .06	16.94 ± .75
(60-3x54)-5	F2	......	......	......	......	......	1	6	8	14	13	30	16	14	9	6	2	3	......	......	......	......	6.6 ± .07	17.67 ± .79
(60-3x54)-6	F2	......	......	......	......	1	2	5	8	11	7	4	2	2	......	......	......	......	......	......	......	......	5.5 ± .09	16.15 ± 1.22
(60-3x54)-total	F2	......	......	......	......	4	10	24	41	53	40	52	21	19	9	8	2	3	......	......	......	......	6.0 ± .05	19.50 ± .56
(60-3x54)-1-1AS.	F3	5.0	......	......	4	1	5	2	13	23	7	25	9	8	......	......	1	1	......	......	......	......	5.9 ± .08	20.40 ± 1.02
(60-3x54)-1-1EO.	F3	5.0*	......	......	1	......	7	4	17	13	11	8	1	......	1	......	......	......	......	......	......	......	5.9 ± .07	14.97 ± .92
(60-3x54)-1-2AS.	F3	6.0	1	......	1	......	4	......	16	11	9	17	4	12	2	1	......	1	......	......	......	......	6.1 ± .09	20.50 ± 1.15
(60-3x54)-1-5ES.	F3	4.5	......	......	1	......	1	3	9	10	14	25	8	5	5	......	......	......	......	......	......	......	6.2 ± .06	13.92 ± .75
(60-3x54)-1-7ES.	F3	5.0	......	......	......	......	3	2	21	26	36	35	10	28	4	4	......	......	......	......	......	......	6.3 ± .05	15.31 ± .57
(60-3x54)-5-1BO.	F3	6.0*	1	......	4	3	2	5	9	5	10	16	5	17	6	2	2	1	......	......	......	......	6.2 ± .11	24.96 ± 1.35
(60-3x54)-5-3AS.	F3	5.0	......	......	3	1	3	6	16	30	26	29	14	9	3	1	......	......	......	......	......	......	6.0 ± .06	16.81 ± .69
(60-3x54)-6-1ES.	F3	4.5	1	......	3	1	14	13	13	18	4	1	1	......	......	......	......	......	......	......	......	......	4.9 ± .08	20.99 ± 1.26
(60-3x54)-6-5EO.	F3	6.0*	......	......	......	3	8	4	15	21	15	13	7	3	3	1	......	......	......	......	......	......	5.7 ± .07	17.31 ± .88

*Ear with wrinkled seeds. Weight estimated in terms of starchy seeds.

TABLE 21.—*Frequency distribution of weights of seeds in cross (60-5x54).*

Designation	Gen.	Parent class	Class centers in grams for weights of 25 seeds																				Mean	Coef. var.
			2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5	6.0	6.5	7.0	7.5	8.0	8.5	9.0	9.5	10.0	10.5	11.0	11.5		
60	P	......	7	22	28	4	......	......	......	......	......	......	......	......	......	......	......	......	......	......	......	......	2.7 ± .03	14.44 ± .90
54	P	......	......	......	......	......	......	......	......	......	2	2	5	7	14	10	5	4	1	2	1	2	8.3 ± .11	14.54 ± .96
(60-5x54)	F_1	......	......	......	......	5	12	13	17	4	3	......	......	......	......	......	......	......	......	......	......	......	4.6 ± .06	13.91 ± 91
(60-5x54)-2	F_2	......	......	......	1	3	7	20	31	19	18	13	3	1	......	......	......	......	......	......	......	......	5.3 ± .05	16.10 ± .73
(60-5x54)-6	F_2	......	......	1	2	6	11	21	17	12	12	8	12	4	1	......	......	......	......	......	......	......	5.3 ± .08	22.54 ± 1.09
(60-5x54)-8	F_2	......	......	......	......	2	11	14	18	11	11	5	1	......	......	......	......	......	......	......	......	......	5.1 ± .06	15.98 ± .91
(60-5x54)-11	F_2	......	......	......	3	3	6	8	10	10	11	7	8	8	5	1	......	1	......	......	......	......	5.8 ± .11	24.48 ± 1.35
(60-5x54)-12	F_2	......	......	......	1	3	14	35	37	31	17	10	3	......	2	......	......	......	......	......	......	......	5.1 ± .04	14.40 ± .57
(60-5x54)-total	F_2	......	......	1	7	17	49	98	113	83	69	43	27	13	8	1	......	1	......	......	......	......	5.3 ± .03	20.56 ± .44
(60-5x54)-2-3AS	F_3	4.0	2	......	......	......	21	11	9	17	9	......	1	......	......	......	......	......	......	......	......	......	4.8 ± .07	18.67 ± 1.10
(60-5x54)-11-2AS	F_3	7.0	......	......	......	......	1	......	2	3	14	10	16	25	5	16	2	4	2	1	......	......	7.3 ± .08	15.94 ± .78
(60-5x54)-11-2CS	F_3	4.0	......	......	......	......	4	6	25	31	10	21	3	1	1	......	......	......	......	......	......	......	5.6 ± .05	13.90 ± .67
(60-5x54)-11-3AS	F_3	7.0	......	......	......	......	3	2	7	12	8	9	4	13	5	2	......	1	......	......	......	......	6.4 ± .09	17.43 ± 1.05
(60-5x54)-11-5AS	F_3	4.0	......	......	......	2	7	7	21	15	13	15	2	7	2	......	......	......	......	......	......	......	5.6 ± .07	18.29 ± .94
(60-5x54)-11-5CS	F_3	4.0	......	......	1	1	16	12	14	16	16	3	2	......	......	......	......	......	......	......	......	......	5.1 ± .06	16.50 ± .89
(60-5x54)-11-10AS	F_3	7.5	......	......	......	......	1	8	8	23	26	3	25	7	9	1	2	......	1	......	......	......	6.3 ± .07	17.94 ± .83
(60-5x54)-12-1ES	F_3	4.0	......	......	10	8	22	32	20	20	8	20	6	4	1	2	......	......	......	......	......	......	5.1 ± .07	24.06 ± .98
(60-5x54)-12-2ES	F_3	3.5	......	1	7	8	41	36	15	33	7	1	......	......	......	......	......	......	......	......	......	......	4.6 ± .04	17.25 ± .69

TABLE 22.—*Frequency distribution of weights of seeds in cross (60-8x54).*

Designation	Gen.	Parent class	Class centers in grams for weights of 25 seeds																				Mean	Coef. var.
			2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5	6.0	6.5	7.0	7.5	8.0	8.5	9.0	9.5	10.0	10.5	11.0	11.5		
60	P	......	7	22	28	4	......	......	......	......	......	......	......	......	......	......	......	......	......	......	......	......	2.7 ± .03	14.44 ± .90
54	P	......	......	......	......	......	......	......	......	......	2	2	5	7	14	10	5	4	1	2	1	2	8.3 ± .11	14.54 ± .96
(60-5x54)	F_1	......	......	......	......	5	12	13	17	4	3	......	......	......	......	......	......	......	......	......	......	......	4.6 ± .06	13.91 ± .91
(60-8x54)-1	F_2	......	......	......	......	......	1	4	7	2	15	13	14	11	6	5	......	......	......	......	......	......	6.6 ± .08	16.39 ± .91
(60-8x54)-8	F_2	......	......	......	......	1	2	11	9	15	8	10	4	1	2	4	1	......	......	......	......	......	5.8 ± .10	20.93 ± 1.26
(60-8x54)-total	F_2	......	......	......	......	1	3	15	16	17	23	23	18	12	8	9	1	......	......	......	......	......	6.2 ± .07	19.84 ± .80
(60-8x54)-1-1ES	F_3	6.0	......	......	......	......	1	......	4	8	14	10	13	16	7	......	......	......	......	......	......	......	6.6 ± .07	13.73 ± .78
(60-8x54)-8-8CS	F_3	5.0	1	......	1	1	8	8	24	19	26	17	14	1	3	......	......	......	......	......	......	......	5.7 ± .06	17.99 ± .80

INHERITANCE OF BREADTH OF SEEDS.

In case of the crosses of California pop with Missouri dent and of the latter with Tom Thumb pop, the breadth of twenty-five kernels from an ear of each plant was determined. Tables 23 and 24 contain these data. Of the varieties of corn known to us, California pop has by far the smallest seeds. As grown in 1910 and 1911 (Table 23), the average breadth of twenty-five kernels of this variety was only about 85 millimeters, while the breadth of twenty-five Missouri dent kernels averaged about 216 millimeters. The F_1 seeds were distinctly intermediate in size. In 1910 the difference between the largest twenty-five seeds of California pop and the smallest class of F_1 was 20 millimeters and there was the same difference between the largest class of F_1 and the smallest of Missouri dent. In 1911 the F_1 families had somewhat larger seeds than in 1910. The variation in the F_2 families was noticeably greater than in the F_1 lots. The range of variation, however, was not sufficient in all the F_2 families together completely to close the gap between even the inner extremes of the parents. The greater variation in F_2 was doubtless due to a segregation of size factors, and types like the parents would probably have appeared had a sufficient number of F_2 plants been grown. How many F_2 plants it would be necessary to grow in order to have an even chance for the appearance of forms like the parents cannot be told. More than enough were grown to realize this result if the parents differed by only four factors influencing breadth of kernels. Obviously the difference between them must be considerably more than four factors. If they differ by five factors, over three times as many plants as were grown would have been necessary; if by six factors, nearly thirteen times as many; if by seven factors, over fifty times as many, and if by eight factors, over two hundred times as many. It is out of the question to grow in ordinary pedigree cultures any such number of individuals as would be necessary to regain the original types of seed size in F_2 if the parents differ by as many as seven or eight factors. But, by selecting F_2 plants with the smallest seeds and continuing the selection in F_3 and F_4, if necessary, it should be possible to regain the parent types in a few generations without growing excessively large numbers in any generation.

The F_3 families of the cross of Missouri dent with Tom Thumb pop (Table 24) illustrate the possibility of accomplishing this isolation gradually without growing extremely large numbers. True the parents of this cross did not differ so greatly in seed size as did Missouri dent and California pop and all the intervening classes were occupied by F_1 individuals. Furthermore the F_2 families taken together contained a few individuals with seeds smaller than the mean seed size of Tom Thumb pop and also a few with seeds larger than the mean seed size of Missouri dent. But none of the more than 300 F_2 plants had seed as small as the smallest of Tom Thumb pop or as large as the largest of Missouri dent, and neither one of the three F_2 families alone overlapped appreciably even the inner extremes of both parent varieties. F_2 family 510 with 188 plants represented in Table 24 had a few plants with seeds nearly as small as the mean seed size of Tom Thumb pop but none with seeds larger than the very smallest seed-size class of Missouri dent. From this one F_2 fraternity, the seven F_3 lots included in the table were grown. No F_3 family came from the large-seeded F_2 plants, so that very large-seeded F_3 lots were not to be expected. F_3 families 1139 and 1140 were the descendants of comparatively small-seeded, tho not the smallest seeded, plants of 510. Their mean seed sizes were slightly less than the seed size of their F_2 parents and their smallest seeded members were quite the equal of the smallest seeded Tom Thumb pop plants. The rather pronounced variation shown by these F_3 families makes it seem possible that a stable type with seeds even smaller than those of Tom Thumb pop might be established by further selection.

TABLE 23.—*Frequency distribution of breadth of 25 seeds in cross between California pop and Missouri dent.*

Designation	Year	Gen.	Class centers in millimeters for breadth of 25 seeds																																		Mean	Coef. var.
			73	78	83	88	93	98	103	108	113	118	123	128	133	138	143	148	153	158	163	168	173	178	183	188	193	198	203	208	213	218	223	228	233	238		
499, 500...	1910	P		4	4	8	3	2																													86.81 ± .90	6.87 ± .72
353	1910	P																										1	2	2	2	3	1	1		1	214.54 ± 1.99	4.95 ± . 5
502	1910	F_1													1	5	9	2	6	7	1	4	1														151.06 ± 1.17	6.90 ± .55
505	1910	F_1											1		3	5	9	12	6	5	1	3	1														148.33 ± 1.07	6.83 ± .48
834	1911	P	1	6	14	7	8	3																													86.59 ± .68	7.26 ± .55
835	1911	P		8	10	5	2	1																													83.77 ± .70	6.31 ± .59
833	1911	P																										3	1	1	3	5	5	2	2		216.86 ± 1.49	4.76 ± .48
837	1911	F_1															1	3	3	3	6	8	3	1	1												162.83 ± 1.18	5.79 ± .51
836	1911	F_1															2	2	5	7	6	6	2	2													160.66 ± 1.06	5.56 ± .50
829	1911	F_2												2	2	7	5	14	6	4	8	4	3	4	1	2	1										155.30 ± 1.29	9.80 ± .59
830	1911	F_2								1		1	1	3	2	2	6	6	6	7	6	3	1														150.07 ± 1.61	10.75 ± .76
831	1911	F_2									1	1	2	4	1	6	4	9	6	6		4	4		1												148.31 ± 1.51	10.54 ± .73
832	1911	F_2										2	2			4	5	3	11	2	4	12	2	3	1	2	1										157.26 ± 1.56	10.78 ± .70
839	1911	F_2											2	3	2	1	1	6	4	6	9	3	5	4	1	1											157.17 ± 1.57	10.25 ± .71
840	1911	F_2										2	1	10	5	14	14	5	6	2		1															139.67 ± .86	7.09 ± .46

TABLE 24.—*Frequency distribution of breadth of 25 seeds in cross between Tom Thumb pop and Missouri dent.*

Designation	Year	Gen.	Parent class	Class centers in millimeters for breadth of 25 seeds																										No.	Mean	Coef. var.
				113	118	123	128	133	138	143	148	153	158	163	168	173	178	183	188	193	198	203	208	213	218	223	228	233	238			
353...	1910	P	....	......	......	......	......	......	......	......	......	......	......	......	......	......	......	......	......	......	1	2	2	2	3	1	1	......	1	13	214.54 ± 1.99	4.95 ± .65
508...	1910	P	....	......	1	2	3	3	4	4	1	......	......	......	......	......	......	......	......	......	......	......	......	......	......	......	......	......	......	18	134.39 ± 1.29	6.05 ± .68
509...	1910	F1	....	......	......	......	......	......	......	......	......	1	1	2	3	2	4	3	6	1	1	......	1	......	......	......	......	......	......	25	175.00 ± 1.79	7.56 ± .72
510...	1910	F2	....	......	......	......	......	......	5	7	12	10	31	23	24	15	26	12	12	6	5	......	......	......	......	......	......	......	......	188	167.55 ± .70	8.51 ± .50
1128...	1911	F2	178	......	......	......	1	1	......	6	3	10	5	11	3	7	7	1	3	1	......	......	......	......	......	......	......	......	......	59	162.23 ± 1.24	8.70 ± .54
1127...	1911	F2	198	......	......	......	......	......	......	......	......	1	4	5	7	12	11	7	5	5	1	4	......	1	......	1	......	......	......	64	178.78 ± 1.17	7.74 ± .46
1139...	1911	F3	143	1	6	1	6	10	13	15	9	8	6	4	1	......	......	......	......	......	......	......	......	......	......	......	......	......	......	80	141 31 ± .93	8.70 ± .46
1140...	1911	F3	148	1	1	......	5	13	12	13	12	10	5	2	1	......	......	......	......	......	......	......	......	......	......	......	......	......	......	75	142.73 ± .81	7.28 ± .39
1135...	1911	F3	158	......	......	1	3	2	4	7	7	7	10	12	6	2	4	......	......	1	......	......	......	......	......	......	......	......	......	66	154.97 ± 1.16	8.98 ± .53
1132...	1911	F3	158	......	......	......	......	......	2	2	7	4	2	7	1	1	1	......	......	......	......	......	......	......	......	......	......	......	......	27	155.03 ± 1.30	6.46 ± .59
1134...	1911	F3	168	......	......	......	......	......	......	2	2	2	6	5	9	6	7	3	......	1	......	......	......	......	......	......	......	......	......	43	167.20 ± 1.15	6.67 ± .49
1142...	1911	F3	173	......	......	......	......	......	......	1	......	4	4	5	4	9	7	6	1	2	......	......	......	......	......	......	......	......	......	43	170.91 ± 1.17	6.68 ± .49
1143...	1911	F3	178	......	......	......	......	......	......	......	1	1	5	7	5	9	10	13	2	......	......	......	......	......	......	......	......	......	......	53	172.90 ± .90	5.60 ± .37

INHERITANCE OF HEIGHT OF PLANTS.

Tables 25, 26, and 27 contain the data on the heights of the plants in the three crosses 60-3x54, 60-5x54, and 60-8x54,—Tom Thumb pop crossed with Black Mexican sweet. As stated before, these crosses differ only in the fact that separate individuals were the parents. In a cross-fertilized species like maize, however, this might give as varied results as crosses between different commercial varieties. The crosses having been made originally for the purpose of studying other characters, the height frequencies of No. 60 and of the F_1 generation were not recorded. It is only known that the extreme individuals of No. 60 were 22 and 40 inches and that the extreme individuals of the F_1 generation of cross 60-3x54 were 55 and 88 inches. But one is comparatively safe in supposing that both populations exhibited variation approaching that of a normal curve. Granting this assumption, the modal classes of No. 60 and No. 54 were 31 inches and 73 inches, respectively. There was a difference, then, of 42 inches between the classes of greatest frequency of the two parents, and a difference of 18 inches between the plus extreme of the shorter parent and the minus extreme of the taller parent. If the F_1 generation was exactly intermediate between the two parents, its modal class would be about 52 inches. In reality, however, it is about 70 inches. In other words, it has been pushed up about six classes above the intermediate position. This phenomenon is not to be regarded as an expression of dominance. It is due to the increased vigor (largely internodal) that appears when many gametic factors are in the heterozygous condition (East. 1909).

Since no coefficient of variability of the F_1 generation can be calculated, it is useless to calculate it for the F_2 and F_3 generations. There are a number of facts germane to our thesis to be discovered by a simple inspection of Table 25. Three F_2 families were grown and in every case the extremes of these distributions overlap those of the parental distributions. If segregation has occurred, F_3 families from F_2 parents of different height should show different average heights and different amounts of variability. Such results were obtained. The progeny of several short F_2 individuals were, in general, short, and the distributions were different in their variability. The mode of the daughters of (60-3x54)-5-3 was 37 inches, while the mode of the daughters of (60-3x54)-1-7 was 58 inches. The parental types were not recovered in the F_3 generation, but there is good reason—from the appearance of the F_3 distributions—to believe this could be done by further selection.

The F_2 distribution in cross 60-5x54 (Table 26) was much more variable than the F_1 distribution. Only one family from this cross was followed to the F_3 generation, however, so no comparison between progenies of large and of small plants can be made.

Passing to cross 60-8x54 (Table 27) one finds the same increased variability in F_2 over that shown by the F_1 generation. There two plants were followed to the F_3 generation. One was a comparatively short plant 43 inches high; the other was a tall plant 85 inches high. The range of the first fraternity was from 28 inches to 55 inches with the mode about 40 inches. It is clearly a larger type than the original Tom Thumb. Part of this height may be due to the vigor of heterozygosis, but certainly the maternal parent is not reproduced. The other distribution, that from the tall stalk, is remarkable for its extreme variability. Its members range from 37 inches to 91 inches with the mode at about 61 inches. The modes of the two families differ therefore by something like 21 inches. One can scarcely doubt the possibility of recovering a very tall strain from this highly variable family, by continued selection.

Table 28 shows the results from a cross between a small flint, No. 5, and a large dent, No. 6. The mean of the first variety is 68 inches and the mean of the second variety 101 inches. The coefficient of variability of the F_1 generation is about the same as that of the flint parent. The coefficients of variability of the four F_2 families are significantly greater. One has only to examine the range of variation to see this. Individuals were obtained with the complete range from the smallest individuals of the small variety to the largest individuals of the large variety. The coefficient of variability of F_1 is $8.68 \pm .55$, while those of the F_2 families are $15.75 \pm .68$, $13.34 \pm .68$, $12.02 \pm .58$, and $12.65 \pm .46$, respectively.

No F_3 families can be reported for this cross, because the location of the experiment was of necessity moved northward and the plants would not mature.

Table 29 contains the data secured from a cross of Tom Thumb pop and Missouri dent, very short and rather tall strains respectively. The cross was made in the greenhouse in the winter of 1908-9 and the F_1 generation grown the following summer as No. 170. The two parent plants grown in the greenhouse were from open pollinated ears and no selfed seed was obtained from either plant. There was grown, however, along with the F_1 plants in 1909, representatives of the two parents—146, Missouri dent, and 151, Tom Thumb pop—from the same open-pollinated ears from which the two greenhouse plants used in the cross were grown. The plants of all three of these families were measured, but the data were recorded in six-inch classes and therefore can not be presented in a table in which one-decimeter classes are used. The extreme fluctuations of the families are, however, shown in the table by means of arrows. F_1 is plainly intermediate between the parents but somewhat nearer the large than the small parent.

Fig. 12. Height of plants in 1910. *A*, Missouri dent; *C*, Tom Thumb pop; and *B*, F_1 generation of cross between these varieties.

Fig. 13. Height of plants of the F_2 generation of Tom Thumb × Missouri dent, as grown in 1910.

In 1910 the parent varieties and a small number of F_1 plants were again grown for comparison with the F_2 generation. No. 509, the F_1 generation, is from the same crossed ear that produced No. 170, No. 508 from the same ear as No. 151, and No. 363 from the same ear as No. 146. Again the F_1 generation was intermediate between the parents in height of stalks with a comparatively small range of variation. The F_2 generation, family 510, contained some plants quite as small as the smallest of Tom Thumb pop and some as large as the average of the Missouri dent, with all gradations between. Since no selfed seed was obtained from the two original plants used in the cross it cannot be known in how far they were heterozygous for size genes. That the seeds of the original ear of Missouri dent had different size potentialities is indicated by the difference between the four fraternities, 353, 354, 357, and 358, which were grown from selfed seed of different individual plants of No. 146.

In 1911 the parent stocks, two F_2 families, and several F_3 lots were grown side by side. The F_2's again showed greater variation than the parents as indicated both by the range of heights and by the coefficients of variation, tho no individuals were quite so small as those of the small parent and few so large as those of the large parent. None of the F_3 progenies exhibited the range of variation shown by the F_2 fraternities, tho as measured by the coefficient of variation some of them were practically as variable. Measured by the same standard, however, some of them were even less variable than the parent stocks, but whether any of them were homozygous in all or most of their size factors can only be told by growing another generation. No F_3 family was so small as Tom Thumb pop and

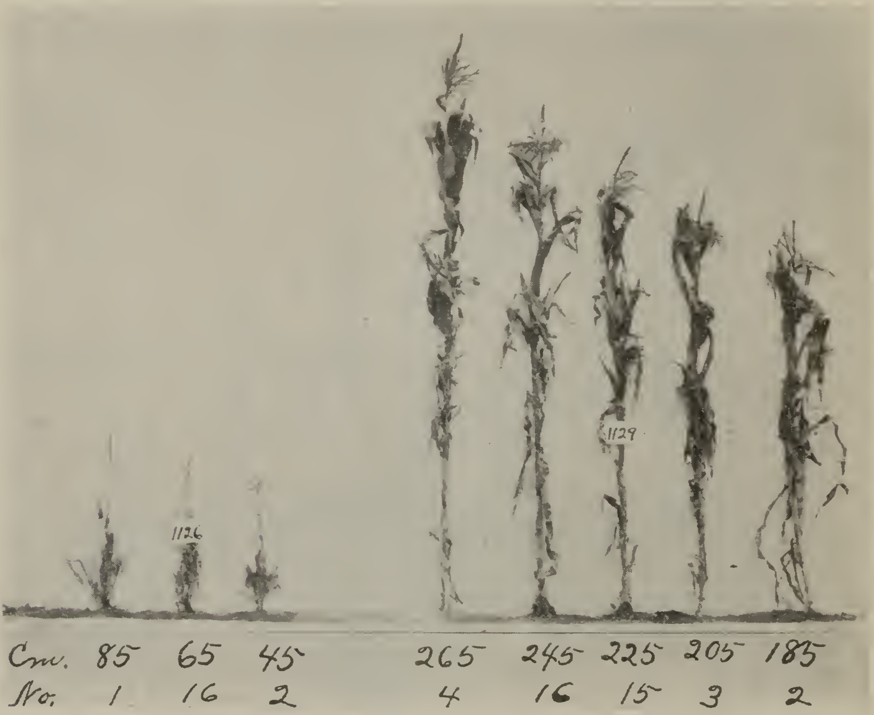

Fig. 14. Height of plants in 1911—Tom Thumb pop at the left and Missouri dent at the right.

none such was to have been expected, since none of them came from the smallest F_2 plants. One of the F_3 families, 1149, from the tallest F_2 plant of the previous year, however, exceeded the Missouri dent parent stock in height. This fact is probably not to be ascribed to any new combination of height factors not found in the tall parent variety but rather to the fact that the vigor of the Missouri dent families grown in 1911 had been reduced by previous selfing. Nos. 1129, 1130, and 1148, the latter two duplicates, were from selfed ears of No. 353, which was grown from a selfed ear of No. 146.

Fig. 15. Height of plants of the F_2 generation of Tom Thumb pop × Missouri dent, as grown in 1911.

The variation in height within any lot of the parent varieties and within the F_1 generation would necessarily be influenced by heterozygosity in the size factors of the parent stocks, the variation increasing with an increasing number of heterozygous size genes. The same condition might also result in noticeable differences between F_2 families grown from different F_1 ears as well as between different selfed strains of the parent varieties. *Nothing of the sort, however, can explain the great variation within any one F_2 progeny, nor the pronounced differences between the several F_3 fraternities arising from a single F_2 family.*

All the F_3 families from which data are at hand came from a single F_2 fraternity, No. 510, which, of course, came from a single F_1 plant, and this plant from the union of a single gamete from each of the parents. Whether all the other gametes produced by those two parent plants were like the two concerned in this cross or whether they were of several different sorts can obviously have no bearing on the behavior of the one F_1 plant, the single F_2 family, and the several F_3 families in question, because these other sorts of gametes, if there were such, had no connection with this line of progenies. In other words, the possible, or even probable, fact that the parent plants may not have been homozygous in all size factors can in no way lessen the importance of the principal conclusions to be drawn from this and similar tables. *There is no escape from the fact that segregation in F_2 occurs with respect to size factors just as truly as it does with factors for other characters. There is also no getting around the plain indications derived from the F_3 progenies that F_2 size segregates may serve as the basis of permanent, true-breeding types just as in case of various other Mendelizing characters.*

Fig. 16. Height of plants of the shortest and tallest F_3 families of Tom Thumb pop × Missouri dent as grown in 1911.

It will not lessen the value of the indications noted above to call attention to the fact that the inherent differences between the F_3 fraternities were exaggerated somewhat by a peculiar combination of weather conditions with characters of the plants other than size. It happens that Tom Thumb pop is not only small but also extremely early and that Missouri dent is very late as well as large. Now as will be shown later (see Table 39), earliness segregates as does size. Whether thru genetic correlation or not, it happened that the short F_3 families were also early and the tall families late. The weather of early summer when the early families were developing rapidly was very unfavorable for growth while the weather late in summer when the late families were making their most rapid growth was very favorable. The early families were, therefore, shorter and the late families taller than they would have been had the conditions been more uniform thruout the season. This effect of the weather is readily seen by noting the various F_3 fraternities and the sizes of the F_2 plants from which they were grown. In case of each F_3 family the size class to which the F_2 parent plant belonged is indicated by blackfaced figures in the F_3 array. The

Fig. 17. Height of plants of two F_3 families of Tom Thumb × Missouri dent, as grown in 1911.—Note difference in variability.

short F_3 families are in general shorter and the tall families taller than the F_2 plants from which they were derived. Moreover, the range of variation of the F_2 plants grown in 1911 was probably increased by the weather conditions, the early plants doubtless having been made shorter and the late plants taller than they would otherwise have been. But it must also be remembered that the same influences must necessarily have been felt by the early and late parent stocks, thereby making the gap between them greater than it would otherwise have been. It is

Fig. 18. The tallest and one of the shortest F_3 families of Tom Thumb × Missouri dent as they appeared in the field September 19, 1911.

true that a comparison of heights of the Missouri dent families grown in 1911 with the heights of the 1910 plants from which they came does not show the same effect noticed in case of the late F_3 families, but this is most likely due to the repeated selfing of the parent stocks.

Early in the study of this cross it was foreseen that the great difference in earliness between the parents might introduce the complication discussed above, and an endeavor was therefore made to overcome the difficulty in later crosses by using as parents varieties that, while differing much in size, had practically the same season of growth. The varieties chosen were Missouri dent, the same one used as tall parent in the cross already described, and California Rice pop. The latter is only a little taller than Tom Thumb pop and, while it ripens somewhat earlier than Missouri dent, its time of blossoming and consequently its duration of growth are not appreciably different from Missouri dent. California pop also differs much from Tom Thumb in the number of nodes per stalk. In this respect it is very similar to Missouri dent. The difference in height between Tom Thumb pop and Missouri dent is largely a difference in number of internodes, while the difference between California pop and Missouri dent is largely one of internode length. (See Tables 31 to 34.) The one cross, therefore, supplements the other well in this study of the inheritance of height of stalks, since height is apparently merely a product of number of nodes and internode length.

In Table 30 are brought together the available data derived from the cross between Missouri dent and California pop. As in the preceding table, the progenies grown in the same garden the same year are grouped together. Of the lots grown in 1910, No. 353 is from selfed seed of a single plant of Missouri dent, 499 and 500 are from selfed seed of two plants of California pop, and Nos. 502 and 505 are from crosses of the parent plants of 499 and 500 with the parent plant of 353. In this case, therefore, the F_1 generation is fully comparable with the parent generations grown the same year. In 1911, two families, 833 and 838, were grown to represent Missouri dent. They were from selfed seed of two plants of 353. Likewise two lots, 834 and 835, grown from selfed seed of two plants of 499, represent California pop. Nos. 836 and 837 consisted of F_1 plants from the remnants of the same ears from which had been grown the previous year Nos. 505 and 502 respectively. Nos. 829, 830, 831, 832, 839, and 840 were F_2 progenies from selfed seed of six different F_1 plants.

The F_1 lots of both years were almost as tall as the tall parent, which is in marked contrast to the intermediate F_1 plants of the cross of Tom Thumb pop and Missouri dent. The difference in behavior is undoubtedly due to the fact that in the one cross number of nodes was the important consideration while in the other it was internode length and to the further fact that F_1 plants in general have an intermediate number of internodes but an internode length greater than that of the tall parent. (See Tables 31 to 34.) That the tallness of the F_1 plants of the cross under consideration was not due to dominance of long over short internodes but rather to the increased vigor accompanying heterozygosis of many characters is indicated by the fact that the mean height of the F_2 fraternities was practically the mid-parental height—partial homozygosis having occurred in F_2—and also by the fact that the F_2 frequency distributions were not so pronouncedly skewed as they would have been if great internode length were dominant. The difference between the several F_2 families is probably due to the presence of heterozygous size factors in one or more of the parent plants. No one F_2 fraternity covers the entire range between the outer extremes of the two parents and no F_2 fraternity fails to bridge the gap between the inner extremes of the parents. The larger variation coefficients of the F_2 families indicate segregation of size factors in this cross where internode length was principally concerned just the same as in the other cross where number of nodes was the main difference and with this cross, moreover, there can be no question of the influence of favorable weather at one time and unfavorable weather at another.

TABLE 25.—*Frequency distribution of heights of plants in cross (60-3x54).*

Designation	Gen.	Parent class	22	25	28	31	34	37	40	43	46	49	52	55	58	61	64	67	70	73	76	79	82	85	88	91	94	97
54	P														2	6	10	17	24	26	21	14	11	7	3	1		
60	P		—	—	—	—	—	—	—																			
(60-3x54)	F1														—	—	—	—	—	—	—	—	—	—				
(60-3x54)-1	F2							1	5	2	2	9	24	12	29	10	14	7	4	1								
(60-3x54)-5	F2							2	5	7	24	19	33	30	26	13	19	4	3	2								
(60-3x54)-6	F2							2	5	4	6	3	11	18	17	11	14	3	6	2								
(60-3x54)-total	F2							5	15	13	32	31	68	60	72	34	47	14	13	5								
(60-3x54)-1-1	F3	37		1	5	10	20	30	31	34	26	20	12	5														
(60-3x54)-1-3	F3	40				3	13	10	24	11	12	10	2															
(60-3x54)-1-7	F3	61									3	18	11	10	53	47	13	21	10	3	1	1						
(60-3x54)-5-2	F3	43				5	3	2	5	10	11	1	2															
(60-3x54)-5-3	F3	40		4	11	25	25	36	23	33	16	9	1	2														
(60-3x54)-6-5	F3	40			2	2	1	9	18	14	22	23	5	5	2													

TABLE 26.—*Frequency distribution of heights of plants in cross (60-5x54).*

Designation	Gen.	Parent class	22	25	28	31	34	37	40	43	46	49	52	55	58	61	64	67	70	73	76	79	82	85	88	91	94	97
54	P														2	6	10	17	24	26	21	14	14	7	3	1		
60	P		<	—	—	—	—	—	>																			
(60-5x54)	F1															<	—	—	—	—	—	—	—	>				
(60-5x54)-2	F2							1			1	4		3	15	31	37	18	10	12	9	1						
(60-5x54)-6	F2									1	2	6	10	16	21	27	23	19	10	3			1					
(60-5x54)-8	F2										2	2	9	8	22	19	24	12	4	3	3							
(60-5x54)-11	F2								2		2	3	3	12	20	28	24	17	7	4								
(60-5x54)-12	F2							1			1	3	1	5	17	12	18	26	28	9	10	2	2					
(60-5x54)-total	F2							2	2	1	8	18	26	44	95	117	126	92	59	31	22	3	3					
(60-5x54)-11-2	F3	43						5	9	14	32	22	20	17	3	1												

TABLE 27.—*Frequency distribution of heights of plants in cross (60-8x54).*

Designation	Gen.	Parent class	22	25	28	31	34	37	40	43	46	49	52	55	58	61	64	67	70	73	76	79	82	85	88	91	94	97
			Class centers in inches for heights of plants																									
54	P	......	......	......	......	......	......	......	......	......	......	......	......	......	2	6	10	17	24	26	21	14	14	7	3	1		
60	P	......	<	—	—	—	—	—	>																			
(60-8x54)	F1	......	......	......	......	......	......	......	......	......	......	......	<	—	—	—	—	—	—	—	—	—	—	>				
(60-8x54)-1	F2	......	......	......	......	......	......	1	......	......	4	5	8	9	23	17	21	11	13	9	5	2	1	....	1			
(60-8x54)-8	F2	......	......	......	......	......	......	......	......	1	1	1	5	4	12	12	15	16	8	11	4	4	3	1				
(60-8x54)-total	F2	......	..	......	......	......	......	1	......	1	5	6	13	13	35	29	36	27	21	20	9	6	4	1	1			
(60-8x54)-1-1	F3	43	......	......	5	6	10	17	16	16	7	10	6	3														
(60-8x54)-8-8	F3	85	......	......	......	......	......	4	4	5	3	8	10	11	14	20	16	15	17	11	8	6	4	......	......	1		

TABLE 28.*—*Frequency distribution of heights of plants in cross (5x6).*

No.	51	54	57	60	63	66	69	72	75	78	81	84	87	90	93	96	99	102	105	108	111	Mean	Coef var
	Class centers in inches for heights of plants																						
5	3	2	4	5	11	17	17	18	10	6	4	......	......	......	......	......	......	......	......	......	......	68.22 ± .41	9.51 ± .42
6	......	......	......	......	......	......	......	......	......	......	......	......	1	2	2	13	14	16	14	8	3	101.18 ± .40	5.01 ± .28
(5x6) F1	......	......	......	......	......	......	......	......	......	......	5	4	3	9	9	6	5	4	4	7	......	94.53 ± .74	8.68 ± .55
(5x6)-1 F2	2	1	3	6	3	5	10	10	22	16	10	7	11	13	6	10	8	4	4	2	......	81.00 ± .69	15.75 ± .68
(5x6)-2 F2	......	......	......	2	......	2	4	2	6	5	12	8	12	6	5	8	3	10	8	2	2	88.33 ± .80	13.34 ± .68
(5x6)-8 F2	2	......	......	......	4	4	12	7	9	13	12	15	10	8	8	1	1	1	......	......	......	79.46 ± .62	12.02 ± .56
(5x6)-14 F2	......	1	2	4	2	7	8	17	21	11	27	24	16	9	11	10	5	4	2	......	......	81.24 ± .51	12.65 ± .45

*Grown side by side in same season.

TABLE 29.—*Frequency distribution of heights of plants in cross between Tom Thumb pop and Missouri dent.*

Designation	Year	Gen.	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	Mean	Coef. var.
			Class centers in decimeters for height of plants																											
146 ..	1909	P																	<	—	—	—	—	—	>					
151 ..	1909	P				<	—	—	>																					
170 ..	1909	F1										<	—	—	—	—	—	—	>											
363 ..	1910	P																	2	4	9	9	6	11	11	2	1		24.93 ± .17	7.68 ± .49
353 ..	1910	P												1	1	2	1	3	1	6	2	4	1						21.27 ± .35	11.44 ± 1.18
357 ..	1910	P													2	1	2	6	2	4	3	3							20.91 ± .29	9.87 ± .98
354 ..	1910	P														1		4	2		2	3	3	2	2	1	2		23.86 ± .45	13.08 ± 1.35
358 ..	1910	P															2	1	1	5	4	1	3	3	1	1			23.32 ± .35	10.37 ± 1.07
508 ..	1910	P			2	6	5	2																					8.47 ± .15	10.44 ± 1.30
509 ..	1910	F1											1	3	5	7	4												17.50 ± .17	6.39 ± .68
510 ..	1910	F2			2			2	8	6	25	26	29	30	32	28	22	6	4	1	2								15.86 ± .12	16.65 ± .53
1130 ..	1911	P															2		5	3	4	2	**3**†	1					22.50 ± .29	8.49 ± .91
* 1148 ..	1911	P													1	1	..		2	2	4	4	4	1	1				23.10 ± .36	10.42 ± 1.12
1129 ..	1911	P															2	1	2	4	**11**	10	6	3	1				23.40 ± .19	7.51 ± .57
1126 ..	1911	P	2	9	7	1	**0**																						6.37 ± .12	11.66 ± 1.29
1127 ..	1911	F2					1	1	2	3	5	4	1	**6**	11	3	6	13	11	4	3	3							17.97 ± .28	19.93 ± 1.13
1128 ..	1911	F2					1	1	..	3	3	5	11	7	4	8	12	10	5	5	2	3	1	1					17.87 ± .26	19.40 ± 1.09
1132 ..	1911	F3				2	5	11	11	**5**	5	1																	10.78 ± .15	13.20 ± 1.01
1134 ..	1911	F3				1	3	10	6	13	**9**	7	5	5	4	1													12.70 ± .20	18.55 ± 1.14
* 1150 ..	1911	F3				1	3	4	7	8	**5**	6	5	5	4	2													13.12 ± .25	19.55 ± 1.37
1133 ..	1911	F3				1	4	3	15	21	9	**7**	3	1															11.98 ± .13	12.65 ± .77
1137 ..	1911	F3									3	2	**3**	9	20	12	13	3											17.22 ± .14	9.53 ± .67
1139 ..	1911	F3											**4**	2	3	13	12	20	10	16	7	2	1						20.01 ± .15	10.75 ± .55
1135 ..	1911	F3									2	4	7	**4**	13	17	16	5	9	2	3	3							18.31 ± .18	13.64 ± .72
1145 ..	1911	F3										5	2	6	**2**	12	12	12	14	8	4	3	2						19.49 ± .20	13.44 ± .72
1140 ..	1911	F3											1	3	**6**	6	5	5	15	17	16	6	3	2					21.13 ± .18	11.68 ± .61
1142 ..	1911	F3												6	**3**	17	10	22	9	11	3	1							19.60 ± .14	9.49 ± .51
1138 ..	1911	F3													3	**4**	2	12	12	16	15	15	9	3	4	1			22.39 ± .16	10.62 ± .52
1141 ..	1911	F3										1	3	5	7	6	**11**	17	17	13	3	4							19.80 ± .16	11.43 ± .59
1131 ..	1911	F3													2	3	**6**	16	22	12	5	5							20.89 ± .13	7.52 ± .43
* 1147 ..	1911	F3															**2**	5	13	19	16	16	6	1					22.53 ± .12	6.73 ± .37
1143 ..	1911	F3														8	**6**	9	5	16	17	11	9	5	1				22.21 ± .17	10.42 ± .53
1146 ..	1911	F3														1	**4**	6	16	19	17	12	7	4	5	1	1		22.77 ± .15	9.53 ± .47
1144 ..	1911	F3															**1**	1	3	16	14	21	18	6	6	1			23.86 ± .12	7.20 ± .37
1149 ..	1911	F3																	5	6	**12**	6	12	12	10	6	6	1	25.14 ± .18	9.28 ± .51

* Duplicates.

† Black type indicates the class to which the parent belonged.

TABLE 30.—*Frequency distribution of height of plants in cross between California pop and Missouri dent.*

Designation	Year	Gen.	Class centers in decimeters for height of plants																				Mean	Coef. var.
			6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25		
353	1910	P	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	1	1	2	4	3	1	6	2	1	1	21.27 ± .35	11.44 ± 1.18
499 and 500	1910	P	.....	.....	.....	1	2	7	8	1							.....	.....	.....	.....	.....	.....	11.32 ± .14	8.12 ± .89
502	1910	F1	.....	.....	.....	.....	.....						1	2	1	5	13	5	1	.....	.....	.....	19.64 ± .17	6.70 ± .60
505	1910	F1	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	1	1	4	6	8	1	1	.....	.....	20.18 ± .19	6.45 ± .66
833	1911	P	.....	.....	.....	.....	.....	.....	.....	.....	1	5	11	16	22	13	11	4	1	0	1		18.00 ± .13	9.83 ± .51
838	1911	P	.....								1	1	—	1	5	5	2	1		.....	.....	.....	18.05 ± .25	9.07 ± .99
834	1911	P	.....	.....	1	9	18	16	7	.....	.....	.....	.....	.....			.....	.....	.....	.....	.....	.....	10.37 ± .09	9.56 ± .64
835	1911	P	1	2	11	21	14	2									.....	.....	.....	.....	.....	.....	9.00 ± .09	10.69 ± .70
836	1911	F1	.....	.....	.....	.....	.....		1	0	1	2	2	6	11	12	3	.....	.....	.....	.....	.....	17.80 ± .17	9.05 ± .67
837	1911	F1	.....	.....	.....	.....	.....	.....	.....		.....	2	6	9	9	3	1	.....	.....	.....	.....	.....	17.27 ± .15	6.83 ± .60
829	1911	F2	.....	.....	.....	.....	1	1	4	10	13	11	13	20	8	8	2	1	.....	.....	.....	.....	15.75 ± .15	13.90 ± .69
830	1911	F2	.....	1		2	3	9	12	12	19	22	10	3	2	.....	.....	.....	.....	.....	.....	.....	13.65 ± .11	14.98 ± .75
831	1911	F2	.....	.....	2	0	5	6	13	14	29	11	7	6		.....	.....	.....	.....	.....	.....	.....	13.54 ± .13	14.05 ± .70
832	1911	F2	.....	.....	.....	.....	2	5	11	17	12	12	15	9	7	5	1	.....	.....	.....	.....	.....	14.69 ± .16	15.83 ± .79
839	1911	F2	.....	.....	.....	1	1	2	7	8	13	17	14	9	2		.....	.....	.....	.....	.....	.....	14.58 ± .15	12.76 ± .72
840	1911	F2	.....	.....	1	1	0	2	2	7	9	17	19	6	7	3	.....	.....	.....	.....	.....	.....	15.19 ± .16	13.77 ± .78

THE INHERITANCE OF NUMBER OF NODES PER STALK.

In case of the crosses of Missouri dent with Tom Thumb pop and of Missouri dent with California pop, discussed under the head of the inheritance of height of plant, the nodes of the main stalk of each plant were counted at the time the stalks were measured. The results of these counts are presented in Tables 31 and 32. Since the relationship of the various families was given in some detail in the discussion of height, it will not be necessary to repeat it here.

In case of Missouri dent crossed by Tom Thumb, where the number of nodes is very unlike in the parent varieties, the F_1 numbers were distinctly intermediate between the parents and scarcely bridged the gap between them. The three F_2 families showed much greater variation than the parents and the F_1 generation. They overlapped both parents but did not contain individuals with so many nodes as the largest number found in the tall parent nor individuals with so few nodes as the least number observed in the short parent. The total range of variation exhibited by the F_3 fraternities was greater by three nodes than that of the F_2 fraternities, but possibly would not have been so had an equal number of F_2 plants been grown. In no single F_3 family was the variation so great as in the F_2 lots and in some families it was no greater than in case of the parents. While the parent types were not completely recovered in the F_3 lots, the difference between the several F_3's was notable. For instance the largest number of nodes recorded in families 1132 and 1133 (Table 31) was thirteen, while the smallest number noted in families 1144, 1147, and 1149 was fifteen.

California pop, tho very short, is unlike Tom Thumb pop in that it has nearly as many nodes as Missouri dent. In the cross of these two varieties (Table 32), F_1 was intermediate between the parents. With the parents so nearly alike, it would seem that individuals like the extremes of both parents should have appeared in F_2 even with smaller numbers than were grown. Four F_2 families contained plants with as many nodes as the greatest number shown by any Missouri dent plant, but no F_2 family contained a plant with so few nodes as some of the plants of California pop. In this connection it should be noted that California pop produces numerous tillers while Missouri dent produces very few. It seems possible that the small number of nodes in California pop may in some way be associated with the large number of tillers.

TABLE 31.—*Frequency distribution of number of nodes per stalk in cross between Tom Thumb pop and Missouri dent.*

Designation	Year	Gen.	Parent class	Classes for number of nodes per stalk																				Mean	Coef. var.
				5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24		
146.	1909	P														7	10	1	3	1				18.14 ± .16	6.28 ± .64
151.	1909	P					1	4	1															9.00 ± .16	6.41 ± 1.25
170.	1909	F_1							1	3	4	6	3		1									12.61 ± .21	10.94 ± 1.32
363.	1910	P														2	6	10	15	13	8	2		20.13 ± .13	7.10 ± .45
353.	1910	P														2	6	6	4	1	...	3		19.36 ± .25	9.02 ± .92
357.	1910	P													5	2	8	5	2	...	1			18.04 ± .21	8.25 ± .82
354.	1910	P															2	3	8	6	2	1		20.27 ± .17	5.98 ± .61
358.	1910	P																2	5	7	2	5	1	21.27 ± .20	6.52 ± .66
508.	1910	P					6	5	4															8.87 ± .14	9.10 ± 1.12
509.	1910	F_1									7	8	4	1										12.95 ± .13	6.66 ± .71
510.	1910	F_2					2	7	14	30	36	45	34	28	18	6	2	1						13.04 ± .09	15.69 ± .50
* 1130.	1911	P	21															2	6	5*	6	1		20.90 ± .16	5.22 ± .56
1148.	1911	P	21														1	2	8	5	3	1		20.50 ± .18	5.67 ± .61
1129.	1911	P	18													1	5	7	15	10	2			19.85 ± .12	5.80 ± .44
1126.	1911	P	9	1	2	8	8	0																7.21 ± .13	11.51 ± 1.28
1127.	1911	F_2	13				3	4	10	13	11	9	11	9	4	3								12.42 ± .17	18.28 ± 1.03
1128.	1911	F_2	13				2	3	15	17	16	9	7	5	5	1	1	1						12.15 ± .17	18.31 ± 1.00
1132.	1911	F_3	9			2	9	10	6	8	4	1												9.63 ± .16	15.70 ± 1.21
* 1134.	1911	F_3	9			1	8	7	15	18	7	7	1											10.48 ± .13	14.99 ± .91
1150.	1911	F_3	9					2	9	10	13	8	5	1	2									11.90 ± .16	13.68 ± .94
1133.	1911	F_3	10			2	6	13	25	12	5	1												9.91 ± .10	12.31 ± .75
1145.	1911	F_3	11						1	2	3	12	20	16	18	5	5							14.72 ± .12	11.35 ± .60
1135.	1911	F_3	12					1	3	13	14	26	20	6	2									12.82 ± .10	10.96 ± .57
1142.	1911	F_3	12							4	10	22	26	13	5	2								13.70 ± .10	9.58 ± .51
1137.	1911	F_3	14						1	7	13	22	15	6	1									13.00 ± .10	9.45 ± .56
1138.	1911	F_3	14							1	5	10	20	15	20	12	8	3	2					15.32 ± .13	12.44 ± .62
1139.	1911	F_3	15								1	4	9	13	23	20	8	8	3	1				16.33 ± .13	10.96 ± .56
1143.	1911	F_3	15									2	4	10	19	25	17	6	4					16.79 ± .11	9.03 ± .46
1146.	1911	F_3	15										2	13	21	19	17	13	6	2				17.17 ± .11	9.42 ± .47
* 1131.	1911	F_3	15										5	12	31	16	6	1						16.13 ± .09	6.58 ± .37
1147.	1911	F_3	15											2	11	29	24	10	2					17.45 ± .08	5.99 ± .32
1141.	1911	F_3	16								1	4	11	12	19	17	17	5	1					16.23 ± .12	10.43 ± .54
1140.	1911	F_3	16									2	11	16	23	16	13	4						16.12 ± .11	9.04 ± .47
1144.	1911	F_3	16											3	15	35	20	13	1					17.32 ± .08	6.15 ± .31
1149.	1911	F_3	17											4	11	15	16	14	14	2				17.99 ± .12	8.63 ± .47

* Duplicates.

TABLE 32.—*Frequency distribution of number of nodes per stalk in cross between California pop and Missouri dent.*

Designation	Year	Gen.	Classes for number of nodes per stalk												Mean	Coef. var.
			13	14	15	16	17	18	19	20	21	22	23	24		
499 and 500	1910	P	...	2	8	3	5	1	...	...	...	...	...	...	15.74±.17	7.10±.78
502	1910	F$_1$	...	...	1	5	9	8	5	...	...	...	...	...	17.39±.14	6.20±.56
353	1910	P	...	...	...	...	2	6	6	4	1	...	3	...	19.36±.25	9.02±.92
505	1910	F$_1$	...	...	...	2	10	7	2	1	...	...	...	...	17.55±.14	5.35±.54
833	1911	P	...	...	...	3	10	11	27	20	8	6	...	...	19.16±.11	7.64±.40
838	1911	P	...	...	...	...	...	...	2	7	5	4	1	...	20.74±.17	5.15±.56
834	1911	P	1	6	16	7	8	4	7	2	...	...	...	...	16.27±.17	11.02±.74
835	1911	P	3	6	17	11	9	6	1	...	...	...	...	...	15.74±.13	9.00±.59
836	1911	F$_1$	...	...	...	1	12	15	11	1	1	...	...	...	18.05±.10	5.46±.41
837	1911	F$_1$	...	...	...	4	13	4	8	...	1	...	...	...	17.67±.15	6.75±.59
829	1911	F$_2$	...	...	2	9	14	16	21	12	15	5	1	...	18.81±.13	9.68±.47
830	1911	F$_2$	...	...	...	9	19	19	28	10	6	1	2	1	18.52±.11	8.82±.43
831	1911	F$_2$	...	...	2	12	14	19	20	14	6	8	1	...	18.61±.13	9.91±.48
832	1911	F$_2$	...	...	...	2	8	21	23	20	12	4	4	2	19.41±.12	8.69±.42
839	1911	F$_2$	...	...	1	6	18	20	17	6	3	3	...	...	18.23±.12	8.13±.45
840	1911	F$_2$	...	...	3	14	23	22	9	2	1	...	...	...	17.41±.09	6.88±.38

INHERITANCE OF INTERNODE LENGTH.

From the data given for height of stalks and number of nodes of the crosses of Tom Thumb pop with Missouri dent and of the latter with California pop. the average internode length per plant was determined. Tables 33 and 34 contain these calculated data.

While in 1909 the height and number of nodes were determined for each individual plant, only the frequencies of the various height and node-number classes were recorded. It is impossible, therefore, to show even the range of internode length for the lots grown that year. From the mean heights and the mean number of nodes of these lots, however, an approximation of the mean internode length can be obtained. These approximate means are indicated by crosses in Table 33. The feature of these entries is that the mean internode length of the F_1 generation is much greater than that of even the Missouri dent parent. It is evident that the increased vigor of F_1 plants is manifested almost wholly by increased internode length, since, as was shown in Table 31, number of nodes is distinctly intermediate.

In 1910, very moist conditions in early summer, when Tom Thumb pop was making its rapid growth, and very dry conditions when the later Missouri dent and F_1 plants were growing most rapidly, tended to equalize the internode lengths of the several families. Notwithstanding this the F_1 internode length was considerably in excess of that of Missouri dent. The mean internode length of the F_2 generation was somewhat less than that of F_1—doubtless because of the decreased vigor accompanying partial homozygosis of various characters—but was still in excess of the internode length of Missouri dent. The range of variation in F_2, No. 510, was somewhat remarkable when considered in connection with the ranges of the parents. The greatest internode length in F_2 was thirty millimeters greater and the shortest thirty millimeters less than the respective extremes found among the parents. This increased range may doubtless be accounted for in part, tho certainly not entirely, by the fact that more individuals were included in F_2 than in all the other lots together. It seems scarcely possible that the very weather conditions that tended to increase the internode length of the early Tom Thumb pop and to decrease that of the late Missouri dent, and thereby to restrict the combined range of variation of the parents, could have had the opposite effect on the F_2 range. There was, as a matter of fact, in this particular F_2 family, No. 510, a correlation between earliness and internode length of about .21. In other words, there was some tendency at least for the early plants to have long internodes and the late plants short internodes. This correlation is probably a mere expression of the effect of the seasonal differences in weather, accelerating the growth of the early plants and retarding that of the late ones. If, however, this correlation were genetic, or even merely physiological, so that, without respect to seasonal weather changes, the early plants tended to have longer internodes than the late plants, then the weather actually experienced in 1910 would have tended to make the long internodes longer and the short ones shorter, thus tending to extend the range of variation of F_2 while lessening that of the parents. But it is difficult to see how there can be any physiological correlation between earliness and long internodes. And it is even more difficult to believe in *genetic* correlation between *earliness* and *long internodes* when it is remembered that in case of the parents the *early* one has the *shorter* internodes. On the whole, therefore, it seems very probable that the production of internode lengths in F_2 both above the upper and below the lower extremes of the parents was due in part at least to new combinations of internode-length factors.

If the above is the correct explanation of the very long and the very short internode types that appeared in F_2 family No. 510, it should be possible to isolate strains from it that have longer or shorter internodes than do the parent varieties. The selection of F_2 plants to be tested by F_3 progenies was based upon height of stalk and upon earliness rather than upon internode length, and it happened that no F_2 plant with extremely short or extremely long internodes was chosen. The F_3 families grown in 1911 are arranged in Table 33 in order of the internode length of the F_2 plants of which they are the progenies as indicated by black-faced figures in the F_3 arrays. The exact reverse in seasonal weather conditions between 1910, when in Nebraska the early summer was favorable for growth and the late summer unfavorable, and 1911, when in Massachusetts the early summer was unfavorable and the late summer very favorable, made it impossible to determine from a single season's study of the F_3 families whether types more extreme than the parents could be isolated. By reference to Table 33 and to Table 39, which latter contains the data for earliness of these same families, it will be noted that, with the marked exception of No. 1137, there was a strong tendency for the early F_3 families to have much shorter internodes and the late families much longer internodes than the F_2 plants of which they were the progenies. In short the early F_2 plants had longer and their F_3 progenies shorter internodes while the late F_2 plants had shorter and their F_3 progenies longer internodes than would have been the case had the weather been more nearly uniform thruout the two seasons.

The parent lots grown in 1911 had, in general, without respect to earliness, shorter internodes than their progenitors of 1910. This is doubtless due to the fact that the parent stocks had been selfed until they had lost much of the vigor acquired thru their former heterozygous condition.

The F_2 families of 1911 are not readily brought into line with the behavior of the F_3 families of the same year. Since they came from different F_1 plants from the F_2 family of 1910 and, therefore, from the F_3 families of 1911, and since these different F_1 plants may have had different internode-length inheritances from the more or less heterozygous parent stocks, it is not strange that their internode length should differ from the F_2 family of the preceding year.

The cross of Missouri dent with California pop (Table 34) gave F_1 progeny with somewhat longer internodes than even the tallest parent had, but the excess was not so great as in case of the cross of Missouri dent and Tom Thumb. In 1910 the mean internode length of F_1 was about 8 mm. greater than that of Missouri dent, 40 mm. greater than that of California pop, and therefore 24 mm. greater than the average of the internode lengths of the parents. Similarly, in 1911 the F_1 families had internodes nearly 8 mm. longer than Missouri dent, nearly 38 mm. longer than California pop, and about 23 mm. longer than the average of the parents. That the long internodes of F_1 are not due to dominance of long over short internodes but rather to increased vigor of growth due to crossing is indicated by the fact that the internode lengths of the F_2 families grown in 1911 were only about 18 mm. greater than those of California pop, about 12 mm. less than those of Missouri dent, and therefore only about 3 mm. greater than the average internode lengths of the parents. Further, if dominance were concerned, the F_2 frequency distributions would be noticeably skewed.

While no F_2 family had quite the combined range of the two parents, the lower extreme of the parents, as grown in 1911, was reached by three F_2 lots and the upper extreme by two. The variation in F_2 was considerably greater than in F_1. There is, therefore, evidence of segregation of internode lengths in F_2 of this cross, as well as in the cross of Missouri dent and Tom Thumb; and here, moveover, the weather must have affected all generations practically alike since they all had about the same season of growth.

Table 33.—*Frequency distribution of internode length of plants in cross between Tom Thumb pop and Missouri dent.*

Designation	Year	Gen.	Class centers in millimeters for internode length																Mean		Coef. var.	
			54.5	64.5	74.5	84.5	94.5	104.5	114.5	124.5	134.5	144.5	154.5	164.5	174.5	184.5	194.5	204.5				
146	1909	P	......	......	......	......	......	......	......	......	× *											
151	1909	P	......	......	......	×																
170	1909	F1	......	......	......	......	......	......	......	......	......	......	......									
363	1910	P	......	......	......	1	1	2	17	22	10	2	1	......	......	......	......	......	122.54	1.04	9.43	.60
353	1910	P	......	......	......	3	4	4	5	3	......	......	......	......	......	......	......	......	105.03	2.04	12.54	1.39
357	1910	P	......	......	......	......	2	3	7	11	......	......	......	......	......	......	......	......	116.24	1.35	8.28	.82
354	1910	P	......	......	......	1	1	3	7	7	2	1	......	......	......	......	......	......	117.23	1.90	11.26	1.16
358	1910	P	......	......	......	1	3	7	6	4	1	......	......	......	......	......	......	......	109.95	1.72	10.87	1 02
508	1910	P	......	......	1	5	6	3	......	......	......	......	......	......	......	......	......	......	91.83	1.49	9.30	1.15
509	1910	F1	......	......	......	......	......	1	1	3	7	6	2	......	......	......	......	......	135.50	1.84	9.01	.96
510	1910	F2	1	1	3	9	19	30	34	33	38	28	15	8	3	1	......	......	123.47	1.03	18.40	.61
† 1130	1911	P	......	......	......	......	3	8	8	**1** ‡	......	......	......	......	......	......	......	......	108.00	1.20	7.34	.79
1148	1911	P	......	......	......	1	......	7	6	**6**	......	......	......	......	......	......	......	......	112.50	1.55	9.15	.98
1129	1911	P	......	......	......	......	......	6	17	**14**	3	......	......	......	......	......	......	......	118.00	.88	6.98	.53
1126	1911	P	......	......	5	8	......	4	**1**	1	......	......	......	......	......	......	......	......	89.76	2.27	16.30	1.83
1127	1911	F2	......	......	......	......	......	2	4	10	11	**17**	17	6	7	1	1	1	146.32	1.51	13.44	.74
1128	1911	F2	......	......	......	......	......	2	3	12	**11**	11	19	13	5	4	2	......	148.65	1.50	13.57	.73
1139	1911	F3	......	......	......	......	......	**7**	27	38	15	3	......	......	......	......	......	......	122.28	.66	7.59	.38
1140	1911	F3	......	......	......	......	......	**4**	7	28	26	15	4	1	......	......	......	......	131.20	.88	9.16	.48
1137	1911	F3	......	......	......	......	......	**0**	6	21	23	8	7	......	......	......	......	......	132.81	.92	8.31	.50
1141	1911	F3	......	......	......	......	1	7	**23**	40	12	4	......	......	......	......	......	......	122.20	.71	8.01	.41
1144	1911	F3	.. ...	......	......	......	......	......	**0**	14	36	31	6	......	......	......	......	......	137.83	.60	5.99	.31
1132	1911	F3	......	......	......	......	2	13	13	**7**	4	1	......	......	......	......	......	......	114.75	1.23	10.03	.76
† 1131	1911	F3	......	......	......	......	......	1	8	**26**	22	11	3	......	......	......	......	......	130.56	.84	8.08	.46
1147	1911	F3	......	......	......	......	......	......	9	**35**	27	7	......	......	......	......	......	......	128.60	.62	6.28	.34
1143	1911	F3	......	......	......	......	......	3	6	**22**	36	17	3	......	......	......	......	......	132.20	.77	8.00	.41
1146	1911	F3	......	......	......	......	......	2	9	**23**	32	23	3	1	......	......	......	......	132.89	.80	8.50	.42
1138	1011	F3	......	......	......	......	......	......	3	9	**17**	17	33	14	2	1	......	......	147.03	.98	9.71	.48
1135	1911	F3	......	......	......	......	......	1	5	10	**17**	18	21	12	1	......	......	......	143.56	1.11	10.56	.55
1149	1911	F3	......	......	......	......	......	......	......	6	**33**	28	8	1	......	......	......	......	139.89	.65	5.96	.33
1133	1911	F3	......	......	......	......	......	13	14	19	11	**3**	3	1	......	......	......	......	122.94	1.20	11.61	.70
1142	1911	F3	......	......	......	......	......	......	2	7	19	**29**	19	6	......	......	......	......	143.52	.85	7.97	.42
† 1150	1911	F3	......	......	......	3	8	15	11	7	5	**0**	1	......	.. ..	......	......	......	110.70	1.41	13.39	.92
1134	1911	F3	......	......	......	......	4	12	12	15	10	**7**	4	......	......	......	......	......	122.63	1.37	13.20	.80
1145	1911	F3	......	......	......	......	......	5	9	19	26	13	**9**	......	1	......	......	......	132.43	1.05	10.60	.56

* Approximate mean internode length. The range is not known—see text.
† Duplicates.
‡ Black-faced type denotes parent class.

TABLE 34.—*Frequency distribution of internode length of plants in cross between California pop and Missouri dent.*

Designation	Year	Gen.	Class centers in millimeters for internode length										Mean		Coef. var.	
			44.5	54.5	64.5	74.5	84.5	94.5	104.5	114.5	124.5	134.5				
353	1910	P	...	...		.	3	4	4	5	3	.	105.03	2.04	12.54	1.39
499 and 500	1910	P	...		4	11	1		....	....	....	....	72.92	.76	6.70	.73
502	1910	F_1		..	.	..		1	10	11	6	.....	112.36	1.04	7.28	.66
505	1910	F_1		.		..		1	6	10	1	1	113.59	1.29	7.92	.81
833	1910	P	...	.	..	5	25	24	28	3	.....	....	94.38	.73	10.59	.55
838	1910	P	.		.....	1	7	5	3	......	....	...	88.18	1.52	11.17	1.24
834	1910	P	3	13	21	11	3		.	.	....		64.11	.92	15.12	1.02
835	1910	P	4	28	18	3	.		......	..	....	.....	58.27	.66	12.12	.81
836	1910	F_1	.	..	1	1	6	7	23	3	....		98.89	1.12	10.72	.81
837	1910	F_1	...	..	...	2	3	8	14	3	..	...	98.83	1.26	10.35	.91
829	1910	F_2	....	3	8	22	37	8	17	...	.....	....	83.97	.89	15.30	.77
830	1910	F_2	1	13	19	31	22	8	1	....	.....	.	73.76	.85	16.60	.83
831	1910	F_2	2	11	17	43	17	3	3	....		....	73.15	.81	16.11	.80
832	1910	F_2	.	4	27	33	21	8	2	1	......	......	75.75	.80	15.27	.76
839	1910	F_2	1	2	7	25	27	8	1	.....	....	...	80.04	.89	14.25	.81
840	1910	F_2	....	2	5	11	27	12	14	3	......	.	87.47	1.07	15.58	.89

INHERITANCE OF NUMBER OF STALKS PER PLANT.

Missouri dent, like most large dent varieties, tillers very little, producing as a rule only one tall stalk, or at most two stalks, with occasional short tillers. Tom Thumb pop and California pop, on the other hand, produce numerous tillers. Tom Thumb pop usually has one or two principal stalks with several shorter tillers that end in ears instead of tassels. In the California pop most of the numerous tillers have both tassels and ears. In recording the data furnished by crosses of these dent and pop varieties, an attempt was made to distinguish stalks from tillers, but, since the distinction in very many cases was necessarily arbitrary, it is deemed best to count every branch from near the ground as a stalk without regard to its relative length or the presence of a tassel or an ear. Lumping everything in this way does not always bring out the real difference between different families. For instance, No. 1145 is shown in Table 36 to have contained two plants with four stalks each, whereas, as a matter of fact, there was only one plant in the whole lot that had as many as two real stalks. The almost universal condition in this family, as shown in figure 20, was one tall stalk without any tillers or with one or two very short ones. In family 1146, on the other hand, four or five almost equally tall stalks with very rarely a short tiller was the prevailing condition. In many cases, however, all gradations occurred from tall stalks with ears and tassel, tall stalks with tassel but no ear, equally tall stalks ending in an ear instead of in a tassel, etc., to very short branches with neither tassel nor ear. In these cases it was obviously impossible to group the various sorts of branches into two definite classes.

The F_1 of the cross of Missouri dent and California pop, Table 35, was intermediate between the parents in number of stalks per plant both in 1910 and 1911. The range of variation in California pop was greater than in some of the F_2 families. It is quite probable that the stock of this variety used was heterozygous. The F_2 families exhibited more variation than the F_1 families, but only one of them covered the entire range of the parents as grown the same year.

The cross of Missouri dent with Tom Thumb, Table 36, produced F_1's that were intermediate between the parents and F_2's with a range slightly greater than that of the parents combined. That this great variation in F_2 was due to segregation of factors for number of stalks per plant is shown by the F_3 families grown in 1911. These are arranged in the table according to their mean numbers of stalks. Of course, not all of the F_3's were like the F_2 plants from which they came. The modal class of family 1142 was three stalks and the mean number of its stalks a little over two and a half. Evidently its one-stalked F_2 parent was a minus variant of a two-stalked or three-stalked type. Similarly family 1146 had a modal value of five stalks and a mean of about four and a quarter, while its F_2 parent had only three stalks, and 1149 had a mean of only about two stalks, tho its F_2 parent had four. Family 1139, whose F_2 parent also had four stalks, had a mean of nearly four and a quarter stalks. The four stalks of the parent of 1149, however, consisted of one main stalk and three rather short suckers, while all the four stalks of the parent of 1139 were tall. That this difference, tho possibly significant, cannot be relied upon is shown by the fact that families 1132 and 1140, the F_2 parents of which, like that of 1149, had only one stalk and three suckers, had four stalks as their modal class and between three and four stalks as their means.

Fig. 19. Three F_3 families of Tom Thumb pop × Missouri dent showing marked differences in number of tillers. September 19, 1911.

The most significant fact established by these F_3 families as a whole is that from the segregates of F_2 were produced types with various numbers of stalks ranging from one parent type to the other. In forming an idea of the types represented by the parent

Fig. 20. Representative plants of two F_3 families of Tom Thumb pop × Missouri dent. The lower leaves were removed to show the number of stalks per plant.

varieties, the frequency distributions for 1909 and 1910 are perhaps more useful than those of 1911. While the 1911 stocks of these varieties were doubtless more nearly homozygous than the stocks of previous years, they probably did not possess just the factors carried by the two germ cells whose union resulted eventually in the 1911 F_3 families. Approaching homozygosis of other factors as well as of size factors rendered the 1911 parent stocks less vigorous than they were in previous years and thereby lessened the tendency to tiller, somewhat as poor or dry soil is known to do. The very few tillers produced by the 1911 stock of Tom Thumb, No. 1126, was due in part perhaps to this cause, in part to the very unfavorable weather conditions while it was developing, and doubtless in part also to the isolation of a type with comparatively few stalks.

Whether any of the F_3 lots of this cross were homozygous for all the factors that have to do with number of stalks cannot be determined without further breeding. It seems probable that some of them, like 1145 for instance, that had a small range of variation will be found to breed practically true, while from others it will perhaps be possible to isolate several types.*

Among the F_3 families of Tom Thumb × Black Mexican there was a noticeable difference in number of stalks per plant. Two families scarcely ever tillered, while one family averaged four tillers per plant.

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* While some intimation of what the future behavior of any F family will be may perhaps be obtained from an inspection of the range of variation and from the statistical constants, particularly the standard deviation, too much dependence cannot be put upon these things. It may be, as was suggested earlier for number of rows per ear, that types of plants with an inherently large number of stalks naturally fluctuate more than types with an inherently small number of stalks because each inherent stalk may furnish a basis for fluctuation. From this point of view, the coefficient of variation seems a better measure of relative fluctuation than does the standard deviation, because it expresses the deviation as a percentage of the thing that deviates. From an inspection of the arrays (Table 36) and even more from an inspection of the plants themselves, it would, however, be difficult to believe that family 1146 is less variable than family 1145, even tho the coefficients of variation are 29.86 ± 1.60 per cent for the former and 48.31 ± 3.08 per cent for the latter. The standard deviations of these families are respectively 1.27 ± .06 and 0.80 ± .04 stalks. Only further breeding experiments, rather than statistical constants, can show how much of this variation in either case is mere fluctuation and how much due to genetic differences.

Table 35.—*Frequency distribution of number of stalks per plant in cross between California pop and Missouri dent.*

Designation	Year	Gen.	Classes for number of stalks per plant											Mean	Stand. dev.	Coef. var.
			1	2	3	4	5	6	7	8	9	10	11			
353	1910	P	2	5	9	5	1	...	...	...	...	...	...	2.91 ± .14	1.00 ± .10	34.26 ± 3.84
499 and 500	1910	P	...	...	...	1	1	1	2	2	7	3	2	8.42 ± .29	1.85 ± .20	21.91 ± 2.41
502	1910	F_1	...	...	6	16	10	3	2	...	...	...	...	4.43 ± .11	1.03 ± .08	23.14 ± 1.91
505	1910	F_1	...	...	7	5	3	4	2	...	...	...	...	4.48 ± .20	1.37 ± .14	30.51 ± 3.47
833	1911	P	69	12	4	...	...	...	...	...	...	...	...	1.24 ± .04	.53 ± .03	42.42 ± 2.56
838	1911	P	3	6	4	5	1	...	...	...	...	...	...	2.74 ± .18	1.16 ± .13	42.48 ± 5.19
834	1911	P	...	...	1	5	17	13	11	3	1	...	...	5.80 ± .12	1.22 ± .08	21.05 ± 1.47
835	1911	P	1	1	2	7	7	13	13	7	2	...	...	5.94 ± .16	1.70 ± .11	28.57 ± 2.02
836	1911	F_1	1	1	5	16	16	1	1	...	...	...	...	4.27 ± .11	1.03 ± .08	24.22 ± 1.91
837	1911	F_1	...	...	9	16	4	1	...	...	...	...	...	3.90 ± .09	.74 ± .07	19.08 ± 1.71
829	1911	F_2	5	19	29	29	9	3	...	...	...	...	...	3.29 ± .08	1.14 ± .06	34.74 ± 1.90
830	1911	F_2	14	14	41	18	7	1	...	...	...	...	...	2.93 ± .08	1.14 ± .06	39.01 ± 2.18
831	1911	F_2	6	7	14	29	25	11	3	1	...	...	...	4.15 ± .10	1.46 ± .07	35.25 ± 1.92
832	1911	F_2	3	6	36	29	16	4	1	...	1	...	...	3.74 ± .09	1.24 ± .06	33.05 ± 1.78
839	1911	F_2	21	19	26	12	1	...	...	...	...	...	...	2.41 ± .08	1.07 ± .06	44.56 ± 2.83
840	1911	F_2	20	17	22	13	2	...	...	...	...	...	...	2.46 ± .09	1.14 ± .06	46.38 ± 3.08

TABLE 36.—*Frequency distribution of number of stalks per plant in cross between Tom Thumb pop and Missouri dent.*

Designation	Year	Gen.	Parent class	Classes for number of stalks per plant								Mean	Stand. dev.	Coef. var.
				1	2	3	4	5	6	7	8			
146	1909	P	......	81	9	7	1	......	......	......	......	1.27 ± .04	.63 ± .03	49.53 ± 2.91
151	1909	P	......	......	......	2	3	1	......	......	......	3.83 ± .18	.66 ± .13	17.31 ± 3.47
170	1909	F_1	......	......	16	13	5	1	......	......	......	2.74 ± .09	.81 ± .07	29.45 ± 2.56
363	1910	P	......	4	18	21	13	2	......	......	......	2.85 ± .09	.96 ± .06	33.68 ± 2.36
353	1910	P	......	2	5	9	5	1	......	......	......	2.91 ± .14	1.00 ± .10	34.26 ± 3.84
357	1910	P	......	10	6	4	2	1	......	......	......	2.04 ± .16	1.16 ± .12	56.81 ± 7.23
354	1910	P	......	7	12	6	......	......	......	......	......	1.96 ± .10	.72 ± .07	36.68 ± 3.94
358	1910	P	......	11	8	6	......	......	......	......	......	1.80 ± .11	.80 ± .08	44.44 ± 5.01
508	1910	P	......	......	......	3	10	1	1	......	......	3.67 ± .14	.80 ± .10	21.85 ± 2.83
509	1910	F_1	......	......	......	7	13	......	......	......	......	3.65 ± .07	.47 ± .05	12.99 ± 1.41
510	1910	F_2	......	20	18	59	75	46	4	......	1	3.57 ± .06	1.25 ± .04	34.87 ± 1.24
* 1130	1911	P	4	9	5	5	1	......	......	......	......	1.90 ± .14	.94 ± .10	49.63 ± 6.46
1148	1911	P	4	11	5	4	0	......	......	......	......	1.65 ± .12	.79 ± .08	47.94 ± 6.18
1129	1911	P	3	38	2	0	......	......	......	......	......	1.05 ± .07	.67 ± .05	63.90 ± 6.51
1126	1911	P	3	7	6	6	......	......	......	......	......	1.95 ± .13	.82 ± .09	42.26 ± 5.41
1127	1911	F_2	4	16	22	20	15	4	......	......	......	2.60 ± .09	1.17 ± .06	44.81 ± 2.90
1128	1911	F_2	3	11	26	16	17	8	3	1	......	2.98 ± .11	1.41 ± .07	47.15 ± 2.98
1145	1911	F_3	1	43	26	11	2	......	......	......	......	1.66 ± .06	.80 ± .04	48.31 ± 3.08
* 1131	1911	F_3	1	31	21	15	4	......	......	......	......	1.89 ± .07	.93 ± .05	49.00 ± 3.38
1147	1911	F_3	1	26	20	22	9	1	......	......	......	2.22 ± .08	1.07 ± .06	48.24 ± 3.15
1149	1911	F_3	4	31	16	24	5	......	......	......	......	2.04 ± .08	.99 ± .05	48.63 ± 3.23
1143	1911	F_3	1	28	16	33	9	1	......	......	......	2.30 ± .08	1.06 ± .05	46.22 ± 2.82
* 1150	1911	F_3	2	13	12	17	7	1	......	......	......	2.42 ± .10	1.08 ± .07	44.63 ± 3.56
1134	1911	F_3	2	17	11	24	10	2	......	......	......	2.52 ± .10	1.13 ± .07	44.88 ± 3.18
1142	1911	F_3	1	15	19	29	19	......	......	......	......	2.63 ± .08	1.03 ± .05	39.24 ± 2.34
1133	1911	F_3	5	8	11	17	23	5	......	......	......	3.09 ± .10	1.16 ± .07	37.41 ± 2.52
1141	1911	F_3	4	12	9	27	25	11	3	......	......	3.26 ± .09	1.30 ± .07	39.88 ± 2.34
1138	1911	F_3	2	7	12	29	33	13	2	......	......	3.41 ± .08	1.15 ± .06	33.78 ± 1.82
1135	1911	F_3	4	3	18	22	30	11	1	......	......	3.36 ± .08	1.09 ± .06	32.56 ± 1.85
1140	1911	F_3	4	7	11	24	27	15	......	1	......	3.42 ± .09	1.22 ± .06	35.73 ± 2.08
1137	1911	F_3	5	5	5	18	27	8	2	......	......	3.52 ± .10	1.14 ± .07	32.33 ± 2.10
1144	1911	F_3	3	4	2	28	37	14	2	......	......	3.70 ± .07	1.00 ± .05	26.92 ± 1.47
1132	1911	F_3	4	1	3	10	17	8	1	......	......	3.78 ± .11	1.01 ± .08	26.80 ± 2.16
1139	1911	F_3	4	2	7	11	30	30	10	......	......	4.21 ± .08	1.17 ± .06	27.72 ± 1.49
1146	1911	F_3	3	1	8	18	21	34	9	1	1	4.24 ± .09	1.27 ± .06	29.86 ± 1.60

* Duplicates.

INHERITANCE OF TOTAL LENGTH OF STALKS PER PLANT.

For the studies of height of plants, only the main stalk of each plant was measured and for the studies of number of stalks per plant every stalk, whether tall or short, was counted. Neither of these sets of data gives a very good idea of the total amount of stalk produced. While it certainly would have been desirable to determine the dry weight of each plant, that could not be done. Doubtless a fair notion of the amount of substance per plant could be obtained from a knowledge of the total length and average diameter of stalks per plant, but no determinations of diameters were made. The only measurements of total length of stalks were made in 1911. The data are given in Tables 37 and 38.

Table 37 affords a comparison of Missouri dent, California pop, and F_1 and F_2 generations of a cross of these varieties. While the different families of the same parent or hybrid generation differed considerably from one another, there were also fairly well marked differences between the different generations. The short-stalked California pop, Nos. 834 and 835, owing to its large number of stalks per plant, had a greater total length of stalk per plant than did the much taller Missouri dent, Nos. 833 and 838. The increased vigor of the F_1 generation, Nos. 836 and 837, resulted in a total length of stalk considerably greater than that of either parent. None of the F_2 families had an average total length of stalk equal to F_1 nor did any of them much exceed the California pop parent in this respect. Some of the F_2 families, however, equaled in range of variation the combined range of the parents and F_1. Whether a type breeding true to a total stalk length greater than that of either parent could be isolated by selection from among the F_2's can be told only by further breeding.

Similar data for the cross of Missouri dent and Tom Thumb pop are presented in Table 38. No F_1 generation of this cross was grown in 1911, but the parent and F_2 generations will serve for comparison with the various F_3 families. The unfavorable early season and previous inbreeding combined to make the total length of stalk of Tom Thumb pop, No. 1126, unusually small, only about one-fifth that of Missouri dent, Nos. 1129, 1130, and 1148. The average total length of stalks of the F_2 families, Nos. 1127 and 1128, equaled or exceeded somewhat that of the Missouri dent parent. The range of variation in F_2 was considerably greater than in the parents. In some of the F_3 families the range of variation was even greater than in F_2, while in others it was little if any greater than in the parent varieties. The mean total length of stalk of the F_3 families varied from about midway between the parents to somewhat over twice that of the larger parent. (Compare Nos. 1133 and 1145 with 1144 and 1146.) While of course the Missouri dent stock grown in 1911 did not produce as tall stalks as it doubtless would have done if it had not been previously selfed, it is quite unlikely that any cross-pollination within the original stock would have resulted in a type like family 1146. The cross of a tall, few-stalked variety with a short, many-stalked sort has resulted in the production of a tall many-stalked type quite unlike either parent.

Table 37.—*Frequency distribution of total length of stalks in cross between California pop and Missouri dent.*

Designation	Year	Gen.	Class centers in decimeters for total length of stalks											Mean	Coef. var.
				15	25	35	45	55	65	75	85	95	105		
833	1911	P	......	58	17	6	4	......	......	......	......	......	......	19.82 ± .60	41.40 ± 2.48
838	1911	P	......	3	3	7	2	2	1	1	......	......	......	37.11 ± 2.46	42.81 ± 5.06
834	1911	P	......	1	7	15	15	9	4	......	......	......	......	42.06 ± 1.13	28.36 ± 1.97
835	1911	P	1	2	7	14	16	11	2	......	......	......	......	40.66 ± 1.17	31.15 ± 2.23
836	1911	F_1	......	2	......	......	5	7	9	7	7	4	......	66.46 ± 2.00	28.49 ± 2.29
837	1911	F_1	......	......	......	2	8	7	8	3	1	1	......	58.00 ± 1.72	24.03 ± 2.22
830	1911	F_2	......	18	20	24	19	12	2	......	......	......	......	34.26 ± .94	39.78 ± 2.23
839	1911	F_2	......	25	11	14	16	8	......	......	......	......	......	31.08 ± 1.11	45.45 ± 3.00
840	1911	F_2	1	19	15	14	13	8	4	......	......	......	......	32.97 ± 1.22	47.04 ± 3.23
829	1911	F_2	......	8	14	21	26	16	7	2	1	......	......	41.53 ± 1.04	36.52 ± 2.01
831	1911	F_2	1	10	12	14	25	14	13	6	......	1	......	44.06 ± 1.24	40.91 ± 2.30
832	1911	F_2	......	4	13	25	22	15	12	3	1	......	1	44.48 ± 1.12	36.62 ± 2.01

TABLE 38.—*Frequency distribution of total length of stalks in cross between Tom Thumb pop and Missouri dent.*

Designation	Year	Gen.	Class centers in decimeters for total length of stalks														Mean	Stand. dev.	Coef. var.
			4.5	14.5	24.5	34.5	44.5	54.5	64.5	74.5	84.5	94.5	104.5	114.5	124.5	134.5			
1129	1911	P	......	2	36	......	2	......	......	......	......	......	......	......	......	......	25.00 ± .53	4.98 ± .38	19.90 ± 1.56
* { 1130	1911	P	......	1	10	2	4	2	......	1	......	......	......	......	......	......	34.50 ± 2.19	14.49 ± 1.55	42.00 ± 5.21
{ 1148	1911	P	......	1	10	2	1	1	2	......	......	......	......	......	......	......	34.50 ± 2.13	14.14 ± 1.51	40.99 ± 5.13
1126	1911	P	15	4	......	......	......	......	......	......	......	......	......	......	......	......	6.61 ± .63	4.08 ± .45	61.68 ± 8.98
1127	1911	F_2	......	12	27	18	7	6	2	3	1	1	......	......	......	......	34.11 ± 1.34	17.39 ± .95	50.98 ± 3.58
1128	1911	F_2	......	12	18	16	9	11	6	6	1	......	3	......	......	......	41.45 ± 1.65	22.12 ± 1.17	53.37 ± 3.51
1133	1911	F_3	......	28	26	10	......	......	......	......	......	......	......	......	......	......	21.69 ± .61	7.17 ± .43	33.08 ± 2.18
1145	1911	F_3	......	21	53	6	2	......	......	......	......	......	......	......	......	......	23.16 ± .48	6.39 ± .34	27.61 ± 1.56
1132	1911	F_3	......	10	14	12	3	1	......	......	......	......	......	......	......	......	27.25 ± 1.07	9.99 ± .75	36.67 ± 3.11
* { 1134	1911	F_3	......	35	14	8	4	3	......	......	......	......	......	......	......	......	22.94 ± 1.57	11.49 ± .69	50.07 ± 3.66
{ 1150	1911	F_3	2	18	16	5	6	......	2	......	......	1	......	......	......	......	26.50 ± 1.56	16.37 ± 1.10	61.78 ± 5.53
* { 1131	1911	F_3	......	5	37	10	8	7	3	......	1	......	......	......	......	......	32.95 ± 1.14	14.21 ± .80	43.11 ± 2.86
{ 1147	1911	F_3	......	1	28	16	15	9	4	3	2	......	......	......	......	......	39.24 ± 1.22	15.99 ± .86	40.75 ± 1.54
1143	1911	F_3	......	4	30	19	16	8	5	1	1	......	......	......	......	......	38.29 ± 1.21	16.70 ± .85	43.63 ± 2.63
1142	1911	F_3	......	11	15	13	14	16	10	3	......	......	......	......	......	......	40.72 ± 1.28	17.22 ± .91	42.30 ± 2.60
1149	1911	F_3	......	......	32	5	17	13	5	3	......	1	......	......	......	......	40.29 ± 1.27	16.41 ± .90	40.72 ± 2.57
1135	1911	F_3	......	4	17	23	15	13	11	1	1	......	......	......	......	......	41.32 ± 1.12	15.36 ± .79	37.16 ± 2.17
1137	1911	F_3	......	5	3	14	16	15	8	3	1	......	......	......	......	......	45.88 ± 1.31	15.68 ± .93	34.17 ± 2.24
1140	1911	F_3	......	1	11	13	25	15	10	5	3	1	......	......	......	1	48.97 ± 1.38	18.88 ± .98	38.56 ± 2.27
1141	1911	F_3	......	6	8	7	15	13	17	9	4	5	2	1	......	......	55.19 ± 1.67	23.13 ± 1.18	41.92 ± 2.49
1138	1911	F_3	......	1	10	11	15	8	15	14	13	4	4	1	......	......	60.02 ± 1.61	23.36 ± 1.14	38.92 ± 2.16
1139	1911	F_3	......	2	8	6	10	7	23	11	10	6	6	1	......	......	63.50 ± 1.68	23.67 ± 1.19	37.27 ± 2.12
1144	1911	F_3	......	......	5	1	7	13	22	16	10	7	3	2	1	......	68.52 ± 1.49	20.59 ± 1.05	30.05 ± 1.67
1146	1911	F_3	......	......	3	3	4	14	11	8	13	17	9	3	6	2	79.88 ± 1.82	26.05 ± 1.29	32.61 ± 1.78

* Duplicates.

INHERITANCE OF DURATION OF GROWTH.

While the time required for a corn plant to reach a definite stage in its development, such as blossoming or ripening, is not strictly a size character, it has nevertheless a marked influence upon the size of plants or plant parts. It has been pointed out earlier in this paper how earliness and lateness may affect the height of plants if the season is more favorable at one time than at another. Besides this effect it is reasonable to expect a physiological correlation between size and duration of growth. Obviously an extremely early plant cannot in its few weeks of growth attain a height equal to that ultimately reached by another plant whose period of growth extends thruout the entire summer. It does not follow from this, however, that there is any genetic correlation between duration of growth and ultimate size. Later in this paper we shall consider this point further.

The general average difference between times of flowering in various F_3 families of Tom Thumb × Black Mexican was very marked. Family (60-3x54)1-2AS was two weeks earlier than (60-8x54)8-8CS.

The cross of Tom Thumb pop with Missouri dent furnished favorable material for a study of the inheritance of length of growing period. Missouri dent is so late that it barely ripens at Lincoln, Nebraska, while Tom Thumb pop is one of the earliest varieties known. In 1910 both varieties were planted together on the same day and ripe ears of Tom Thumb pop were harvested the same day that some of the Missouri dent plants were pollinated.

The two dates that perhaps best measure the duration of the growing period are the date of planting and the date of ripening of the ears. It is difficult, however, to tell exactly when an ear is ripe. Hardness of kernels and dryness of the outer husks were taken to indicate ripeness, but it can never be said that the husks were green yesterday and are dry today. The date of blossoming can be fixed much more definitely than the date of ripening, but growth has not ceased at this time. The ears, in fact, make their principal growth after blossoming. The growth in height of stalks, however, is practically complete at blossoming time, so that this date and the planting date can be used to determine roughly the period of height growth. Moreover, there is a more or less definite relation between the time required by a corn plant to reach the blossoming stage and the time required by it to reach full maturity. The former is usually not far from sixty per cent of the latter. In case of a large-eared variety, the period of blossoming to maturity is likely to be relatively longer than in case of a small-eared sort. For instance, the large-eared

Missouri dent and the very small-eared California pop blossom almost together, but the latter ripens some days ahead of the former. Again, a very early corn may have its later stages shortened relatively by the heat of midsummer, while a very late variety may have its ripening stage relatively prolonged by the cooler weather of autumn.

In 1910 the date of ripening of each plant of the cross under consideration—Tom Thumb × Missouri dent—was determined as closely as practicable, while the dates of blossoming were noted for only such plants as were hand-pollinated. The various lots were examined carefully on the same day each week and all plants deemed ripe were noted. Since in 1911 this corn was grown in Massachusetts, it was foreseen that the later families could not possibly ripen, and, therefore, blossoming dates alone were recorded. The time of the exposure of the first anthers of the tassel on the main stalk was chosen as the date of blossoming. In the case of protandrous plants, which constituted the bulk of those grown, this was the only date recorded, but for the few protogynous families the date of exposure of the first silks was also noted. The entire lot of plants was examined each day and those just beginning to blossom were tagged. The available data are grouped together in Table 39. Since in 1910 the dates of ripening were taken weekly while the dates of blossoming are arranged in three-day classes, only the range of variation of the 1910 families is shown in the table. Since the blossoming dates of hand-pollinated plants only were recorded in 1910, the ranges of variation indicated in the table are not necessarily the complete ranges of blossoming dates. A large majority of the plants, however, blossomed within the dates shown in the table. The class headings indicate the number of days from planting to blossoming or ripening.

In 1910, Tom Thumb pop (family 508) was in blossom 60 to 70 days from planting time, while Missouri dent (353) was blossoming 85 to 95 days after planting. The F_1 plants (509) were distinctly intermediate, with a blossoming time of 73 to 83 days from planting. Some of the F_2 plants (510) were nearly as early as the earliest Tom Thumb pop plants, while others were almost as late as the latest Missouri dent. The F_3 blossoming time extended over a period of thirty days, 62 to 92 days after planting. The 1910 families held the same relative order in ripening as in flowering. The parent varieties were farther apart in ripening than in blossoming, Missouri dent having ripened in 144 to 151 days and Tom Thumb pop in 91 to 98 days, or while Missouri dent was still in flower. The F_1 plants were somewhat later than half way between the parents, with ripening dates 122 to 136 days after planting. The earliest F_2

plants were ripe soon after the latest F_2's were in blossom. The period of ripening extended between seven and eight weeks, or 94 to 146 days from planting.

As grown in Massachusetts in 1911, Tom Thumb pop (1126) had an average blossoming date (staminate flowers) of about 64 days from planting, or practically the same as in Nebraska the year before. Missouri dent (1129, 1130, 1148), on the other hand, was about fifteen days later than the previous year, its average flowering date having been about 105 days from planting. The comparatively cool weather of midsummer and later was doubtless responsible for the delay in case of Missouri dent, while the heat of early summer—unprecedented in Massachusetts—brought Tom Thumb pop into flower as early as in Nebraska the year before. No F_1 plants were grown in 1911. The two F_2 families (1127, 1128), tho containing fewer individuals than the F_2 family of 1910, nevertheless showed a somewhat greater range of variation. It is possible that the same conditions which made Missouri dent later than in 1910 and forced Tom Thumb pop into flower as early as in 1910 tended to extend the F_2 range considerably. Notwithstanding this, the latest F_2 plants were in flower when the earliest Missouri dent plants began blossoming.

The F_3 progenies, all grown in 1911, were of course subject to the same climatic influence as the parents and F_2. Apparently, however, no F_3 family responded to this influence quite as Missouri dent did. The parents of some F_3 families were among the latest tho not the very latest F_2 plants, as indicated in Table 39. (The blossoming time of the parent of each F_3 family is shown in the table by black-faced type in the corresponding F_3 class.) It was expected that some of the F_3 lots would, therefore, be almost if not quite as late as Missouri dent. None of them, however, had an average blossoming date later than 87.5 days from planting, while the average date of flowering of the earliest Missouri dent family was nearly 13 days later. It is probable that some of the plants chosen from among the later F_2's of 1910 were plus variates of only medium late types,—for their F_3 progenies, notwithstanding the cooler weather of the late summer of 1911, were considerably earlier than they were in 1910. This was particularly noticeable in case of families 1140, 1142, 1145, and 1146. The parent of family 1145, for instance, blossomed about 88 days after planting, while not a single plant of 1145 was quite so late as that, and the average date of flowering for the family was only a little over 73 days. Families 1149 and 1131 and the latter's duplicate 1147, on the other hand, were from F_2 plants that blossomed in 1910 about 85 days after planting, and their average date of blossoming in 1911 was almost the same, 86 days from planting.

That neither of the F_2 families and none of the F_3's had any plants that were as late as the Missouri dent plants of 1911 is perhaps not strange when it is remembered that the original parent plants were representatives of commercial strains which had not been previously self-pollinated and when it is also recalled that the lots representing the two parent varieties were not descended directly from the plants used as parents of the cross. That the parent varieties were heterozygous for time of blossoming as well as for various other characters is indicated by the fact that one of the lots grown in 1911 was considerably earlier than the other. It is possible that the single gamete from Missouri dent, which by union with a gamete from Tom Thumb pop gave rise to F_2 family 510 and to all the F_3 families, lacked some of the factors for long-continued growth that were present in the parents of the lots used later to represent Missouri dent. It might further be supposed that the 1910 representatives of Missouri dent were forced into abnormally early blossoming by the hot dry weather of that summer. It would then still remain to be explained why the weather of 1910 in Nebraska should make Missouri dent plants unusually early and at the same time cause many of the later F_2 plants to be unusually late, as was assumed before. Or perhaps it will be as necessary to explain why the weather of 1911 in Massachusetts delayed the blossoming of Missouri dent and at the same time hastened the blossoming of the later F_3 families.

In this connection it should not be forgotten that too high a temperature, particularly if associated with extreme dryness, may retard the development of plants not adapted to such conditions while hastening the development of other sorts of plants. We do not know that diverse sorts of corn actually respond in these different ways to unusual conditions, but the possibility is worth considering. Is it possible that an early corn like Tom Thumb pop, adapted to growth in the North, may develop quite as rapidly at moderate temperatures as at higher ones, *i. e.,* the optimum temperature for it is low, while a late variety, which can only be matured in the South, may be greatly retarded in its development by anything short of high temperatures? And is it possible also that this assumed adaptation to growth at somewhat low temperatures might be inherited independently of other genetic factors for earliness, so that F_2 segregates might rank quite differently in earliness in different seasons? The topic should not be dismissed without referring back to the records of height of plant of these same lots of corn (Table 29). Here we meet the suggestive facts, *first,* that the 1911 Missouri dent families were not so tall as the 1910 plants from which they came, tho it took them much longer to complete their growth, and, *second,* that certain of the 1911 F_3 families were taller than their 1910 F_2 parents, tho they required less time to complete their growth. It would seem that sometimes, at least, the conditions that hasten development are favorable to a large height growth while the conditions that prolong growth unduly are unfavorable to great height growth. Since the soil and weather conditions to which the Missouri dent and F_2 plants were subjected in 1910 were practically the same for both lots and since the same was true for the Missouri dent and F_3 families in 1911, the markedly different behavior of the two lots in the two seasons was due obviously to an inherent difference between the two lots of plants—a difference in the way they responded to like environments. Whether this difference may have been as was suggested above, a matter of different optimum temperatures for the two lots, or whether the more nearly complete homozygosis of Missouri dent than of the other lots may have been a controlling factor, cannot now be said. It is easy to see that Missouri dent may have had shorter stalks in 1911 than in 1910, notwithstanding the moister conditions of the latter season, because of repeated selfing, and that it may have been later in 1911 than the year before because of the cooler weather. The 1911 F_3 families might then have had taller stalks than their 1910 parents because of the moister conditions in 1911. It certainly could not have been due to a greater degree of heterozygosity. But why were they earlier under the cooler conditions surrounding them in 1911 than were their F_2 parents under the hotter conditions of 1910, unless the optimum temperature for them is lower than for Missouri dent?

While there are, then, numerous difficulties to be met before a satisfactory explanation can be given of some of the facts brought out in this study of the inheritance of duration of growth, they should not be permitted to obscure the simple facts that a cross of a very early variety of corn with a very late variety produced an F_1 generation strictly intermediate between the parents in earliness, F_2 generations with a range of variation from the early parent to, or nearly to, the late parent, and F_3 generations with diverse seasons of blossoming. From Table 39 it can be seen that the latest F_3 family blossomed on the average a little over three weeks later than the earliest F_3 family and that the earliest three F_3 families were all but out of flower before the latest three began blossoming. The impression of distinct difference in earliness made by the plants in the field was even more vivid than that made by the figures in the table. The photograph reproduced in this paper (figure 21) gives some indication of the differences as they appeared in the field.

There is one other feature of this study of time of blossoming that should be noted. The strain of Tom Thumb pop employed in these tests was strongly protogynous as seen from Table 39, where the frequency distribution of days to first exposure of silks in case of protogynous families is shown in italics immediately below the frequency distributions for days to first shedding of pollen for the same families. Of Tom Thumb pop (family 1126), the plants showed their silks from one to eight days earlier and on the average four days earlier than the first shedding of pollen. The Missouri dent used was evidently heterozygous for this condition, since one 1911 lot (family 1129) was protandrous thruout while the other lot (family 1130 and its duplicate 1148) was protogynous thruout, all but three plants of the latter having had their silks first exposed from one to six days and on the average just three days before any pollen was shed. Both of the F_2 families grown in 1911 (1127

Fig. 21. Two F_3 families of Tom Thumb × Missouri dent as they appeared in the field July 26, 1911. Note difference in earliness.

and 1128) were wholly protandrous. Some plants of the 1910 F_2 lot (family 510) were observed to be protogynous but no adequate records were made of that family. Only one F_3 family (1132) contained protogynous plants. Of this family twenty-eight plants were protandrous and only twelve protogynous. Of the latter the silks began to show from one to three days and on the average about two days earlier than the pollen began to shed.

Had the time of exposure of the first silks instead of the time of shedding of pollen been taken as the date of flowering for all of the families, family 1132 would have been classed as even earlier and all the other F_3 families somewhat later than they were, for in protandrous corn the silks usually appear from one to four or five days after the pollen begins to shed. If protandry is regarded as the normal condition, because it is the most common, protogyny may be thought of as delayed development of the staminate flowers, in which case the time of exposure of the silks would better be used as the date of flowering, or it may be thought of as precocious development of the pistillate flowers, in which case the time of opening of the anthers would better be taken as the blossoming date. Or in case protogyny is considered normal and protandry abnormal, the latter may be regarded as delayed development of the pistillate flowers or as the precocious development of the staminate flowers. In the first case the silks and in the second the anthers would be the better index of flowering. The fact that in protogynous types of corn, so far as they are known to us, the silks protrude from the leaf sheath before the ear-bearing shoot has appeared may perhaps be rightly regarded as an indication of precocious development of the pistillate flowers. This would place protogyny as an "abnormal" character and make the staminate flowers the better guide in fixing the date of flowering. It is probable, however, that both protogyny and protandry are quite normal.

No attempt has been made as yet to study the inheritance of protogyny or protandry as definite characters. It is possible that they are mere chance relations between times of maturity of pistillate and staminate flowers. If so, however, there must be a pronounced physiological correlation between time of maturing of the two sorts of flowers, for otherwise any lot of corn with a considerable range of variation in time of flowering would by chance show all gradations from strong protandry to strong protogyny with slight protandry or protogyny or even homogamy as the modal condition. It is also likely that there is genetic correlation between the time of maturity of the staminate and pistillate flowers and that this is in fact the only basis for considering protogyny and protandry as definite characters. If there were

TABLE 39.—*Frequency distribution of number of days from planting to flowering and ripening in cross between Tom Thumb pop and Missouri dent.*

Designation	Year	Gen.	Class centers in days to flowering and ripening																																				Mean	Coef. var.
			49	52	55	58	61	64	67	70	73	76	79	82	85	88	91	94	97	100	103	106	109	112	115	118	121	124	127	130	133	136	139	142	145	148	151		
508							<	—	—	>†							<	—	>†																				
353															<	—	—	>																<			>		
509											<	—	—	>														<		—	—	>							
510								<	—	—	—	—	—	—	—	—	>	<	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	>				
1126	1911	P					8	**6**‡	3	2																												63.84 ± .46	4.69 ± .51
1126				*1*	*3*	*4*	*7*	*3*	*1*§																														
} 1129	1911	P															**2**	5	8	9	3	2	9	2														101.35 ± .64	5.95 ± .45
* } 1130	1911	P																**0**			5	4	6	2	2	1												108.25 ± .65	4.00 ± .43
1130																				*4*	*4*	*6*	*3*	*2*	*1*														
1148	1911	P																**0**		1	3	9	5	1		1												106.90 ± .56	3.45 ± .37
1148																		*1*	*1*	*2*	*5*	*7*	*2*	*2*															
1127	1911	F2				1	7	8	14	4	6	**5**	10	7	6	4	2	3																				74.32 ± .73	12.82 ± .70
1128	1911	F2					4	14	10	15	5	13	**5**	4	7	4	1																					72.85 ± .59	10.78 ± .57
1132	1911	F3					12	**12**	4	10	1	1																										65.43 ± .43	6.15 ± .46
1132						*4*	*9*	*7*	*6*	*7*	*6*	*1*																											
1133	1911	F3				4	16	14	**12**	12	4	2																										65.50 ± .38	6.87 ± .41
} 1134	1911	F3				3	6	20	16	8	7	2	1		1																							66.86 ± .42	7.46 ± .45
* } 1150	1911	F3					2	15	**6**	10	9	3		3				1	1																			70.12 ± .70	10.44 ± .71
1135	1911	F3					10	20	**19**	15	9	9	3																									68.13 ± .36	7.21 ± .37
1137	1911	F3					8	22	14	**11**	4	4	2																									67.05 ± .38	6.77 ± .40
1142	1911	F3					1	3	12	12	13	3	17	11	**7**	3																						75.34 ± .50	8.90 ± .47
1145	1911	F3						7	12	17	9	16	15	3	3	**0**																						73.18 ± .41	7.56 ± .40
1138	1911	F3						2	3	17	13	16	13	**12**	14	5	1																					77.25 ± .43	8.08 ± .39
1139	1911	F3						2	2	13	12	14	**17**	12	13	5																						77.60 ± .42	7.63 ± .38
1140	1911	F3						1	3	7	13	12	16	17	**9**	7																						78.29 ± .42	7.33 ± .40
1141	1911	F3							6	10	6	22	17	**14**	8	4																						77.41 ± .41	7.07 ± .36
1143	1911	F3								4	6	14	20	26	**9**	6		1																				80.01 ± .34	5.79 ± .30
1144	1911	F3									1	6	26	25	24	5																						81.76 ± .24	3.98 ± .20
1146	1911	F3							1		3	4	13	15	37	**17**	3																					83.45 ± .30	5.16 ± .26
} 1131	1911	F3									1		2	9	**17**	19	14	7	2																			87.49 ± .35	5.05 ± .29
* } 1147	1911	F3										1	10	14	**29**	15	3	4	2																			85.15 ± .33	5.01 ± .27
1149	1911	F3									1		2	8	**24**	29	9	3																				86.58 ± .27	4.03 ± .22

* Duplicates.

† Broken lines indicate ranges for ripening and solid lines approximate ranges for flowering.

‡ Black-faced type indicates parent class.

§ Italics refer to exposure of silk, other figures to shedding of pollen.

no genetic correlation between time of maturity of the two sorts of flowers, crossing any very early with any very late variety of corn, both of which are protandrous, must result always in F_2 plants more strongly protogynous than any now known and also some plants more decidedly protandrous than any known.

SUMMARY AND GENERAL CONCLUSIONS.

The object of this paper, as stated in the introduction, is to discuss somewhat fully the inheritance of quantitative characters and to present data bearing upon this subject secured from experiments with maize. The results of these experiments, given in detail earlier in this paper, are here summarized.

The inheritance of number of rows per ear has been studied in eight different crosses. The parent varieties were of 8-rowed, 12-rowed, 16-rowed, and 20-rowed types. In nearly every case F_1 was intermediate between the parents, tho in case of one cross of an 8-rowed variety with an apparently 12-rowed variety, the latter condition seemed to be dominant. In most cases the F_2 generations had a wider range of variation than F_1, a range that included both parent types. Differences between the F_2 families of a single cross are believed to have been due to heterozygosis of one or other of the parent varieties. Where F_3 families were grown, the parent types were recovered in every case and intermediate types were also seen. In one case a series of F_3 lots, from a single cross, showed modal conditions of 12, 14, 16, 18, and 20 rows.

Inheritance of length of ears was studied in three crosses of distinct varieties. In each case the ear length of one parent was approximately two and one-half times that of the other parent. In the cross of Tom Thumb with Black Mexican, the F_1 ear length was distinctly intermediate between the parental ear lengths. In the cross of Missouri dent with California pop, on the other hand, the F_1 generation had ears practically as long as those of the long-eared parent, but here the extreme length of the F_1 ears was doubtless in part due to heterozygosis, for the means of the F_2 families were distinctly intermediate between the parental means. In every case the F_2 fraternities were more variable than the F_1 lots. In most cases the F_2's completely bridged the gap between the parents and in one case the F_2 range of variation was from practically the shortest ears of the short-eared parent to beyond the longest ears of the long-eared parent. The short-eared parent type has not as yet been recovered in any F_3 lot and in only a few F_3 families has the ear length been equal to that of the long-eared parent. In case of each cross, however, the ears of some F_3 families averaged nearly twice as long as the ears of other F_3

families. Nor was this difference in size the only feature differentiating the several F_3 families. The variability of some F_3 lots was no greater than that of the parents or of F_1, while other F_3 lots equaled in variability the F_2 generation.

Inheritance of diameter of ears was investigated in two crosses. The ears of F_1 were intermediate in diameter between those of the parents, but somewhat nearer the large-eared than the small-eared variety. The F_2 ranges of variation little more than filled the gap between the parent races. The parent types were not recovered in F_3, but the several F_3 lots were very distinct. The smallest ears of the largest eared F_3 families were larger than the largest ears of the smallest-eared family.

Weight of seeds was determined only for the cross Tom Thumb × Black Mexican. The seeds taken to represent the latter variety were probably too large, owing to previous mixture of strains. If allowance is made for this, the F_1 seed weight was only a little below the mid-parental weight and the ranges of variation of the F_2 fraternities well overlapped the ranges of the parents. Tho distinct types of seed weight were isolated in F_3 neither parent type was recovered. One F_3 family, however, had a seed weight nearly equal to that of Black Mexican and the great variability of several F_3 lots indicated heterozygosity sufficient to enable the isolation of the parent types on further selection.

Breadth of seeds was noted in the crosses of Missouri dent with California pop and with Tom Thumb. In both cases F_1 was distinctly intermediate between the parents in breadth of seeds. The variability of the F_1 lots was not noticeably greater than that of the parents. The F_2 generation, on the contrary, showed a markedly greater variability than F_1 or the parents. In one cross the combined range of variation of all the F_2 families together was not sufficient to overlap even the inner extremes of the parents, but in the other cross, while the F_2 ranges were not greater, the parents differed less in seed size, so that practically all the classes from below the mean of the small-seeded parent to above the mean of the large-seeded parent were occupied by F_2 individuals. Two of the F_3 lots of the latter cross, grown from small-seeded F_2 individuals, had seeds practically as small as those of the small-seeded parent. Since the total number of F_2 plants was slightly over 300 in each cross, it is thought that Missouri dent and Tom Thumb pop probably differed by not over five factors and Missouri dent and California pop by perhaps as many as six factors influencing breadth of seeds.

Four different crosses were employed in the studies of the inheritance of height of plants. A peculiarity of these crosses is that in three of them the F_1 plants were almost as tall as the tall parent and in the fourth were considerably taller than the mean of the two parents. That this increase in height of F_1 plants over the mid-parental height is in no case ascribable to dominance of tallness over shortness, but is due rather to increased vigor accompanying heterozygosis, is indicated by the fact that in every case the mean height of the F_2 plants is about half-way between the heights of the parents and is also shown by the lack of skewness in the F_2 frequency distributions. Practically all of the F_2 fraternities overlapped in height the inner extremes of their parents. Most of them had a range of from near the mean height of one parent to the mean height of the other parent and in one cross the F_2 range was from the minus extreme of the short parent to the plus extreme of the tall parent. The F_3 families were very diverse in height and in variability. Few F_3 families were as tall as the tall parent and none was quite so short as the short parent, tho the latter was approached very closely in a few cases. In case of some of the extreme F_3 lots, the variability was sufficient to make it probable that types like the parents could be isolated in the next generation. Moreover, certain F_3 families with heights variously intermediate between the parents had variabilities small enough to indicate the possibility of their breeding true to these heights.

In some of these crosses, the height of plants was separated into its components—number of nodes and internode length. In number of nodes the F_1 families were strictly intermediate between the parent varieties and the mean number of nodes in F_2 was practically the same as in F_1, both of which facts indicate that number of nodes is not appreciably affected by heterozygosis. As in all the other quantitative characters studied, the F_2 generation exhibited a wide range of variation and the several F_3 families had very different mean numbers of nodes, including types approaching those of the parents and also various intermediate types.

A study of internode lengths explains the excess in height of F_1 plants over the average of the parent heights. Tho number of nodes in F_1 is apparently always distinctly intermediate between the parent numbers, the internode length is so greatly increased by heterozygosis that F_1 plants are often nearly as tall as the tall parent, and always taller than the average of the parent heights—the former when the tall and short parents differ little and the latter when they differ much in number of nodes. In the two crosses in which internode length was calculated, the F_1 plants had internodes longer than those of the long-internode parent. In Tom Thumb × Missouri dent, in fact, the latter variety had an internode length almost exactly half way between that of the other parent variety and of the F_1 generation. One of the F_2 families of this cross had a range of variation extending from considerably below the minus extreme of the short-internode parent to much above the plus extreme of the long-internode parent and even considerably above the plus extreme of F_1. This extreme range in F_2 is thought to be due to new combinations of the internode-length factors of the two parents.

In two crosses, one parent of which produced numerous tillers and the other few tillers, F_1 was intermediate in number of stalks per plant. In both crosses F_2 was more variable than F_1 and in one cross the F_2 range was from one to eight stalks, while five was the largest number observed in the parent variety that tillered most freely. Among the F_3 families, a few were practically one-stalked types and a few others had a somewhat larger mean number of stalks than the tillering parent and a range of variation so great as to suggest the possibility of isolating by selection a type with a still larger number of stalks.

On account of the fact that in one cross the tall parent was a few-stalked type and the short parent a many-stalked type, the two parent varieties did not differ greatly in total length of stalks per plant. Owing to the intermediate number of stalks in F_1 and to the tallness of F_1 stalks in this cross, the total length of stalks in F_1 was much greater than that of either parent—in fact almost equal to the combined stalk lengths of the parents. The mean total length of stalks of F_2 was much less than of F_1 but the variability was considerably greater. In another cross, where the parents differed much more in total length of stalk, no records of the F_1 generation are available, but the mean total stalk length in F_2 (which was doubtless less than in F_1) was somewhat greater than that of either parent. Owing apparently to a lack of correlation—either physiological or genetic—between number of stalks and height of plant, some of the F_3 lots of this cross had a mean total length of stalk twice as great as that of the long-stalked parent and one F_3 family had a mean length greater than the plus extreme of that parent. This great stalk-length is thought to be due in the main to a combination of the factors influencing total stalk length, some of which come from one parent and some from the other.

In earliness F_1 plants were intermediate between their parents. The F_2 generation more than filled in the gap between the parents in all cases where exact records were made and in one case had a range from below the mean of the early parent to above the mean of the late parent. Very distinct types were obtained in F_3, some of which were practically as early in flowering as the early parent but none of which were quite so late as the late-flowering parent. In general the same relative order was maintained in ripening as in flowering. Roughly the number of days from planting to flowering was 60 per cent of the number of days from planting to ripening. The relative lengths of the two periods may, however, depend somewhat upon the season at which the type in question matures.

In general, then, it may be said that the results secured in the experiments with maize were what might well be expected if quantitative differences were due to numerous factors inherited in a strictly Mendelian manner. It is quite likely, as pointed out repeatedly in the detailed discussion of results, that genetic correlations occur between factors for distinct quantitative characters. These and the physiological correlations so frequently noted make the results more difficult of interpretation, but do not throw them out of the realm of Mendelian phenomena. Physiological correlation is a phenomenon of development, not of inheritance, and, as such, has less interest for students of genetics than for experimental morphologists. Even in practical plant breeding, correlations of this sort are of importance mainly on account of the physiological or morphological limits that they set to the perfect development of particular combinations of characters.

Real genetic correlations in the sense of gametic associations, are not antagonistic to the Mendelian doctrine. True, the fundamental principle of Mendelism is segregation of separate factors and it might, therefore, be held with some degree of plausibility that gametic coupling of factors is non-Mendelian. But where the associated factors segregate regularly from other factors or other groups of factors, they can certainly be said to Mendelize —they merely Mendalize together. Such correlations as these have distinct importance in genetics.

Numerous apparent correlations, genetic as well as physiological, have been encountered in these experiments and many of them have been noted incidentally in the detailed discussions of results. We have reason to believe, for instance, that length of ear is directly correlated with height of plant and inversely correlated with number of rows per ear. Number of rows seems also to be related in some way to the character of the endosperm, since in some crosses segregates with a large number of rows have dent grains while those with few rows have flinty grains. It is apparent also that, while height of plant and number of stalks per plant may not be directly related, number of stalks and diameter of stalks are negatively correlated. Our records indicate that there is little if any correlation, either genetic or physiological, between duration of growth and internode length. There is also little or no physiological correlation between duration of growth and number of nodes, but apparently a distinct genetic correlation between these two characters. In 1911, families of corn, which from previous selfing were approaching a condition of homozygosis of factors for size and duration of growth, showed a slight negative correlation between number of nodes and duration of growth, while F_2 families, heterozygous for both number of nodes and for duration of growth, showed a marked positive correlation between these characters. If height of stalk alone had been considered instead of number of nodes and internode length, it might have been thought, in case of these F_2 families, that the unfavorable weather of early summer, by checking the growth of the early plants, and the favorable weather of late summer, by increasing the growth of the late plants, had brought about an apparent rather than a real correlation. But since the number of nodes is determined before the plant has made much growth, this character could not have been influenced by the differences between the weather of early and that of late summer. It is not that one set of conditions made the early plants have few nodes and another set made the late plants have many nodes, but rather that some of the genetic factors that caused the plants to have a large number of nodes were associated with the factors (or were themselves the factors) that caused prolonged growth. It is expected that the detailed evidence upon which this conclusion is based will be prepared for publication in the near future.

The interrelations of such characters as number of rows per ear, circumference of ear, and breadth of seeds are obvious. An ear, of course, could not have many rows and a small circumference without having very narrow seeds. But this and similar relations are in the main mere mechanical adjustments rather than physiological—to say nothing of genetic—correlations.

It is often impossible to tell in a particular case whether distinct factors are coupled in inheritance or whether a single factor plays a part in the development of what are regarded as distinct characters. In the latter case the problem is fundamentally one of the physiology of development rather than of genetics.

Certain quantitative relations in maize illustrate the influence of a single factor in the development of two or more characters as well as the influence of several factors in the development of a single character. It is obvious that any factor that affects say internode length will also have an influence upon length of stalk and therefore, in addition, upon stalk weight and even upon total weight of plant. Likewise any factors influencing number of nodes, any with an effect upon amount of tillering, any factor for diameter of stalk, size of ears, number of ears, etc., will all be concerned in the development of the one character complex—total weight of plant. If one argue that total weight is not a simple character, he must also admit that neither is length of stalk, which of course is determined by the number of nodes and the internode length, and must not forget that even the length of one internode is the product of the number and the length of the cells contained in it. Tho total weight can thus be analyzed into numerous sub-characters, it is none the less a "character" of the plant or type in question.

It was shown earlier in this paper that the multiple-factor hypothesis furnishes a satisfactory and simple interpretation not only of all of the results secured from these maize experiments but also of the results from experiments previously reported for other plants and for animals. We are familiar with no hypothesis not based upon the Mendelian principle of segregation and recombination of factors which furnishes a plausible explanation of many of the facts regarding the inheritance of quantitative characters. We are aware of the suggestion (Castle 1912) that a heterogeneous distribution of growth-inducing substances in the cells of an organism would result in variability in its progeny, that this heterogeneity of the protoplasm would be increased by crossing, and that, therefore, the variability would be increased in generations beyond F_1. This hypothesis cannot, however, be said to interpret satisfactorily the facts of size inheritance, until it is shown, for instance, how one F_2 plant of a size intermediate between the parent varieties can produce an F_3 progeny of great variability while another F_2 plant of the same intermediate size yields an F_3 progeny scarcely more variable than the parents or the F_1 generation. In other words, it must first be shown how some F_2 plants come to have the size of F_1 plants and at the same time have the comparatively homogeneous distribution of growth-inducing substances characteristic of the parent races while other F_2 plants are like F_1 plants in both size and heterogeneity of these substances. And even if increased protoplasmic heterogeneity of general growth-inducing substances can be made to account for the increased variability ordinarily observed in F_2, it is still not quite clear just how such a condition can bring about an F_2 range of variation away beyond the outer extremes of the parents in respect to one size character, while the range of variation of some other size character of the same F_2 individuals is scarcely enough to bridge the gap between the inner extremes of the parents.

THE MANIPULATION OF QUANTITATIVE CHARACTERS IN PLANT BREEDING.

The principal use of hybridization in plant breeding is to secure new combinations of the characters present in the parents—to combine in one race the desirable characters of the parent races and to eliminate the undesirable characters. In some cases a condition intermediate between the parents with respect to a certain character may be desired. Crossing may also be of value for bringing out characters that were latent in the parents, tho, since such possibilities are usually unforeseen, this is seldom the main object of any cross. The increased vigor induced by heterozygosis, tho often of value in plant breeding, is aside from the purpose of this discussion.

The method of procedure to be followed in combining the more simple independent Mendelian characters is well known. All that is necessary to do is to cross appropriate types and to grow a sufficient number of F_2 individuals to be sure of getting the desired combination of characters in the homozygous condition. When this has been done it remains merely to propagate the new types. If it is known beforehand upon what factors the desired characters depend, it can even be told how many F_2 individuals must be grown to afford an even chance of getting the combinations wanted. For instance, if it were desired to combine in one variety of corn the two characters sugary endosperm and a peculiar, erect leaf-habit, due to the absence of auricles from the base of the leaves (Emerson 1912), a type that was first found in cultures of dent corn, and if all the other characters of this non-auriculate dent corn were sufficiently like those of the sweet corn chosen as the other parent so that they could be disregarded, or if it were immaterial what these other characters were, then the two varieties could be crossed with considerable assurance that about one in every sixteen F_2 plants would be homozygous for both erect leaves and sugary endosperm. Or better yet, since segregation in endosperm characters occurs on F_1 ears, only sugary grains need be planted, with the assurance that all the progeny will be sweet corn and about one in four will have erect, non-auriculate leaves.

The procedure when dealing with quantitative characters differs in no way from that outlined above, except that, since more factors are concerned, more individuals must be grown in F_2 or else the F_3 and perhaps even the F_4 generation must be awaited for the desired combination. As a matter of fact even the more simple qualitative characters are rarely ever so easily handled in actual practice as is indicated by the example given above. It almost never happens that both of the types chosen for crossing are alike in all but one or two characters. A breeder does not usually want merely to combine one desirable character of a certain variety with one other equally desirable character of another variety. He perhaps has a variety that for certain uses is almost ideal but it lacks conspicuously some one or two very desirable characters that he sees in some other variety. He wants to keep his variety as it is but transfer bodily to it the coveted character from the other variety. Very likely this second variety is only mediocre in numerous respects. As a matter of fact, the two varieties, instead of differing by merely this one prominent character, actually differ in very many characters. In transferring, therefore, the much desired character to the otherwise ideal variety, one is apt to lose some of the highly-prized characters of the latter, unless very large numbers are grown in F_2 or selection practiced for two or three more generations. The only difference in manipulation between quantitative and qualitative characters is, then, one of degree. And in practical breeding even this distinction is of little importance, since practically every cross between commercial varieties must necessarily include both qualitative and quantitative characters.

The large number of factors by which distinct commercial sorts of crop plants are differentiated is almost staggering. Take as an illustration an extremely simple case. We will suppose that it is desired to produce a better silage corn than that afforded by the tall few-stalked dent varieties of the West and South or the short many-stalked flint varieties of the Northeast—in short a type of corn combining the tallness of one variety with the many-stalked condition of the other. For sake of simplicity, we will disregard differences in season of ripening and various other characters, which, as a matter of fact, could not ordinarily be disregarded in actual practice. We will assume that the varieties in question differ by five Mendelian factors influencing height of plant and by five factors affecting number of stalks per plant and that each of these factors is independent of the others in inheritance—assumptions which, tho they may not represent accurately the actual facts, are at least not unreasonable and will at any rate serve for purpose of illustration. From a cross of these varieties, an F_2 of only about 1,000 plants, or say one-tenth acre, would be required to give an even chance of recovering a many-stalked type like the one parent or a tall-stalked type like the other parent. Perhaps three or four times that many plants should be grown to be at all sure of recovering the parent types. But with this number of individuals there could be no hope that any of the very tall plants would also have very many stalks. With the parents differing by ten factors, five for height and five for number of stalks, as was assumed, over a million F_2 plants, or in other words over one hundred acres, would be required for an even chance of getting the desired combination of characters, and at least two or three times that number should be grown to make sure of results. Now, while perhaps breeding work on such a scale is not impossible, it is certainly out of the question under ordinary conditions. What method then can be used in such cases? Or are the proposed results impossible of accomplishment?

Under the conditions assumed here, the method of back crossing with one parent type (Castle 1911), cannot be used, for, while insuring the ultimate recovery of say the many-stalked parent type, it would at the same time make impossible the recovery of the tall-stalked type of the other parent. The only method available in such cases is to grow as many F_2 plants as is practicable, select such plants as combine most nearly the desired combination of height and number of stalks, and from selfed seed of these grow F_3 progenies, from which in turn further selections must be made, and so on for as many generations as are necessary. In this way, results can be secured with a much smaller total number of plants but of course at the expense of considerably more time.

It may not be out of place here to show more definitely just how the element of time can be made to take the place of numbers in solving a breeding problem like that discussed above. For sake of simplicity, let us now assume that the parents differ in only four factors for height and four for number of stalks, eight in all, thus requiring something over 65,000 F_2 plants for an even chance of getting the desired combination of many stalks and tall stalks. By referring back to Table 1 (page 19), we can see at a glance what behavior will result both in F_2 and in F_3 following a cross the parents of which differ in four factors A, B, C, D. If the class headings are read as decimeters, they will represent well actual heights of short and tall corn plants. Each single factor will then be assumed to add two decimeters to the initial height of 10 decimeters. If now we call the factors A', B', C', D' and suppose each to add one stalk to an initial 1-stalked condition, the same table will serve to illustrate the inheritance of number of stalks per plant where the parents are 1-stalked and 9-stalked respectively. Now from the cross of a 1-stalked, 26-decimeter type with a 9-stalked, 10-decimeter type, we see from Table 1 that, out of 256 F_2 individuals, one should have the formula $AABBCCDD$ and also that one should have the formula $A'A'B'B'C'C'D'D'$. In other words, there should be one 26-decimeter plant and also one 9-stalked plant. But the 26-decimeter plant would almost surely not have 9 stalks and the 9-stalked plant would not be 26 decimeters tall, unless about 65,000 F_2 plants, instead of 256, were grown. To be sure of getting one 9-stalked plant, let us grow four times the number indicated in Table 1, or approximately 1,000 F_2 plants. Since over 70 per cent of all the F_2 plants are from 16 to 20 decimeters tall, the chances are that the one 9-stalked plant will be from 16 to 20 centimeters tall—if taller so much the better. Let us suppose that it is 18 decimeters tall like the F_1 plants.

On selfing this one 9-stalked, 18-decimeter plant, what will be obtained in F_2? We can be sure that the progeny will all be of the 9-stalked type. Table 1 shows that out of the 70 18-decimeter plants, 6 (group III) will breed true, 48 (group XI) will have an F_3 range from 14 to 22 decimeters, and 16 (group XV) will have an F_3 range from 10 to 26 decimeters. If then the 9-stalked F_2 plant belonged in group XV for height, we would have an even chance of getting one 9-stalked, 26-decimeter plant by growing only 256 F_3 individuals and would be fairly sure of getting it by growing 1,000 F_3 plants. In short, we would accomplish with 2,000 F_2 and F_3 plants what we would have had only an even chance of accomplishing with 65,000 F_2 plants or what would have required perhaps 250,000 F_2 plants to make its accomplishment fairly sure. If, however, as is much more likely, the 9-stalked, 18-decimeter F_2 plant belonged in group XI in height, its F_3 progeny, while breeding true to the 9-stalked condition, would have a range from 14 to 22 decimeters. Since all the individuals of group XI are heterozygous in only two factors, an F_3 generation of 16 plants would afford an even chance—and 50 plants make fairly sure—of getting one 9-stalked, 22-decimeter plant. But also, since all the F_2 individuals of group XI lack one of the four height factors, the one 22-decimeter F_3 plant must be homozygous for three factors and lack the fourth, like the F_2 plants of group IV, and will, therefore, breed true to this height. There will then be no possibility of producing a 26-decimeter type from it directly any more than from an F_2 18-decimeter plant belonging to group III, which would of course breed true to that height. Crossing between different F_3 types must then be resorted to.

We have assumed that a 9-stalked, 22-decimeter plant has been produced as described above by selection from 1,000 F_2 and 50 F_3 plants. From the same 1,000 F_2 plants a 26-decimeter, 5-stalked plant could also have been selected and would of course have bred true to the 26-decimeter height. It would possibly also have bred true to the 5-stalked condition (if belonging to group III for number of stalks), or possibly would have produced an F_5 range from 1 to 9 stalks (if belonging to group XV for stalk number), in which case the desired 26-decimeter, 9-stalked type would have been secured in F_3. But since the chance is greater, let us suppose that the 26-decimeter, 5-stalked F_2 plant belonged to group XI for number of stalks. Then by growing 50 F_3 plants from it, we would be fairly certain of getting at least one 26-decimeter, 7-stalked plant which would breed true in both height and number of stalks. The next step, obviously, is to cross this homozygous 26-decimeter, 7-stalked F_3 plant, or any of its F_4 progeny, with the 9-stalked, 22-decimeter F_3 plant obtained from the other selection, or with any of its F_4 progeny. The two F_3 plants would have zygotic formula similar to the following:

(1) 26-decimeter, 7-stalked plant—$AABBCCDD$—$A'A'B'B'C'C'd'd'$.

(2) 22-decimeter, 9-stalked plant—$AABBCCdd$—$A'A'B'B'C'C'D'D'$.

It is clear, therefore, that since the two F_3 plants, (1) and (2), differ only in the factors D and D', only 16 F_4 plants—or say 50 to make more sure—need be grown to give one plant that is homozygous in all eight factors. We have then grown 1,000 F_2 plants, 100 F_3 plants, and 50 F_4 plants to accomplish what would perhaps have required a quarter-million F_2 plants alone. And it would have taken no more plants and no more time had we crossed the 9-stalked, 22-decimeter F_2 plant with the 7-stalked 26-decimeter one instead of having first grown their F_3 progenies.

In the same way it can be shown that the desired combination of quantitative characters could be secured in comparatively few generations even if a sufficient number of plants could not be grown in F_2 to recover the exact parent type of either character in any individual. But the theoretical possibilities have been sufficiently illustrated.

In the detailed account of our experiments, several examples have been pointed out that illustrate the possibility of securing parent types in F_3 from F_2 lots that contained no individuals as extreme as the parents. For instance, in certain crosses, no F_2 plants had seeds quite like those of the parent varieties, but in F_3 the parent sizes were recovered in several cases. (See Tables 20, 22, 24.) Perhaps the most striking case of this sort observed in our experiments is that of height of stalk. While in some crosses the parent heights were recovered in F_2, in other crosses the most extreme F_2 heights lacked much of reaching the outer extremes of the parent ranges. In Tom Thumb × Black Mexican (Table 25), this was true of the F_2 families, but two of the F_3 lots—progenies of approximately the smallest F_2 plants—

contained individuals very near the minus extreme of the small parent.

In actual practice, of course, one must use larger numbers than those declared, a few paragraphs back, to be sufficient. Following one assumption, it was shown that 1,000 F_2 and 1.000 F_3 plants—2,000 in all—would be as likely to give the desired results as about 250,000 F_2 plants, while following another assumption, it was seen that 1,000 F_2, 100 F_3, and 50 F_4 plants—only 1,150 in all—would be required to secure the same result. And the assumptions in these two cases were that the F_2 plants selected differed not in size but merely in zygotic formulae. Since in practice, however, there is no way of distinguishing between the several F_2 plants all belonging to the same size class but having different zygotic formulae, as many F_3 individuals will of course have to be grown from the F_2 plant that is heterozygous in only two factors as from the one that is heterozygous in four factors. Furthermore—and even more important than this—it cannot be known whether two plants of the same size actually belong in the same size class. Not only may plants that belong in the same size class have different zygotic formulae, but those that belong in very different classes may be forced together and made to appear to belong in the same class by the specially favorable conditions surrounding one and the unfavorable conditions surrounding another, or those not only belonging in the same class but having identical zygotic formulae may similarly be made to appear in different classes.

Numerous examples of confusion of this sort arising thru our inability to distinguish at sight between genetic variations and mere fluctuations have been encountered in our experiments. One will be sufficient for illustration. Of all the F_3 families in one cross (Table 36), No. 1146 had the largest number of stalks per plant, its mean being 4.24 stalks, but its F_2 parent had only three stalks—less than the mean of the F_2 family (510) to which it belonged. F_3 family 1149, on the other hand, had a mean number of stalks of only 2.04 notwithstanding that its F_2 parent had four stalks—above the F_2 mean. One of these F_2 plants had been forced below and the other above its true class. In height of plant (Table 29), family 1149 exceeded family 1146 by only 2.37 decimeters, their respective mean heights being 25.14 and 22.77 decimeters, tho the F_2 parents of 1149 belonged to the highest F_2 class and was about four decimeters taller than the F_2 parent of 1146. If one had been selecting a single F_2 plant for both height and number of stalks, the 23-decimeter, 4-stalked parent of 1149 would doubtless have been chosen in preference to the 19-decimeter, 3-stalked parent of 1146, but 1149 actually had a mean total stalk length of only 40.29 decimeters while that of 1146 was 79.88 decimeters—almost twice as great. Evidently in actual practice one must select a number of promising F_2 plants to stand a fair chance of getting an F_3 type to his liking.

There is very little doubt that the striking results secured in the well known selection experiments with corn carried out at the Illinois Experiment Station (Smith 1908) are to be explained on the same basis as that used in this discussion to show how types can be isolated by the proper use of small numbers of plants in F_2, F_3, F_4, or later generations—types that could not be expected to appear in F_2 unless excessively large numbers were grown. If oil and protein content in corn are inherited in the same way that other quantitative characters are, we should not expect to find very high or very low oil or protein content in any 100 or 200 ears taken at random from a lot of open-pollinated, unselected plants, most of which are almost certainly heterozygous in oil and protein factors—just as they are known to be for other characters—and have, therefore, an intermediate development of these characters. It is not necessary to resort to the somewhat mystic idea that selection (the mere act of choosing) has profoundly changed the oil and protein characters of this corn nor to call into service the possibility that mutations occurred repeatedly during the progress of the experiment. There is as much reason for assuming that the height-of-plant character as exhibited in the F_2 ranges (Table 25) was changed into a much smaller height character in F_3 by selection or that a mutation occurred opportunely. Between the isolation of a short-stalked type in F_3 from a largely heterozygous F_2 and the isolation of a low or high oil type from an open-fertilized lot of corn—also largely heterozygous—there is apparently no foundamental difference.

A word should be said of the practical possibilities of producing sizes greater or less than the parent sizes. A noticeable example of this from our own experiments is the production of a type with a total length of stalks more than twice that of the larger parent. (See Table 38.) This was accomplished by combining the many-stalked condition of one parent with the tall stalked condition of the other parent. Visibly different parent characters were combined to form a new character complex. The production of a type with internodes much longer than those of the parents (Table 33) differs from the above only in that here the parent types were apparently very similar and it could not be foreseen that the similar internode lengths of the two varieties were due to different factors that could be combined into a new character complex by crossing. It is of course unlikely that many crosses between varieties having similar quantitative characters will be made with the avowed object of securing combinations of factors unlike the combinations responsible for the parent characters, but the idea is well worth keeping in mind. When the factors that have to do with the development of the principal crop plants have all been sorted out and tagged—as they will be some day—breeders can proceed in their undertakings with much more assurance than at present.

By way of summary it can be said that a breeder dealing with quantitative characters should proceed just as he would if dealing with such qualitative characters as color where the parent types differ in respect to numerous characters. The principal differences between the two cases are that quantitative crosses are likely to differ in more factors than qualitative ones and that environmental influences cause more confusion. He should in short grow as many F_2 plants as his facilities allow. From these he should select *numerous* promising individuals, self pollinate them and grow their progenies on as large a scale as practicable. If any of these F_3 lots are of the desired type he will of course propagate them and discard the others, or if not he can at least discard some of them and continue his selection from the most promising ones. Or if no F_3 family shows by its range of variation any tendency to produce ultimately a type better than itself, resort should be had to intercrossing between the several more promising F_3 lots or between their F_4 progenies, with the hope (we might almost say assurance) that among these several types, tho they are similar in appearance, there exist sufficiently different sets of factors to insure the desired combination in crossing. When dealing with plants like corn that are much more vigorous when many characters are heterozygous, the breeder will find it necessary to practice cross breeding between different isolated types or to provide for natural crossing. He should not, therefore, be content with the production of a single strain of the desired type—say a single high-oil type—but should isolate two or more such types, both of which are high in oil content, but which differ by enough minor characters, quantitative or qualitative, to insure a vigorous development on crossing them or on allowing them to intercross naturally when grown in mixtures.

These methods apply just as well to improvement by selection from the complex hybrid mixtures, which, because of some outstanding features, have been assigned varietal names, as they do to the isolation of types from the similar, tho perhaps somewhat more complex, hybrid mixtures resulting from the crossing of two named varieties. The difference at most is only a matter of degree. The breeder who selects for high-oil content from open-pollinated plants will, if he persists long enough, succeed in producing a type with enough factors for oil production to rank as a high-oil type and with these factors sufficiently homozygous to insure comparative constancy, while at the same time a sufficient number of factors for other minor characters are heterozygous to insure a comparatively vigorous strain. But by self-pollination, together with the same sort of selection, several practically homozygous high-oil strains could almost surely have been produced in much less time. And these strains would doubtless have been sufficiently unlike in factors for minor characters to insure abundant vigor of growth on crossing them. While a few years' time may not be an important consideration where the character in question can be determined at sight or by mere weighing or measuring, in breeding work requiring costly chemical analysis it is extremely important that the desired results be obtained in as few years and therefore with as few analyses as possible.

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A STUDY OF HYBRIDS BETWEEN NICOTIANA BIGELOVII AND N. QUADRIVALVIS

(WITH FOUR FIGURES)

E. M. EAST

Reprinted for private circulation from
The Botanical Gazette, Vol. LIII, No. 3, March 1912

A STUDY OF HYBRIDS BETWEEN NICOTIANA BIGELOVII AND N. QUADRIVALVIS[1]

E. M. EAST

(WITH FOUR FIGURES)

The genus *Nicotiana* was divided by G. DON into four sections: TABACUM, RUSTICA, PETUNIODES, and POLIDICLIA. This classification has been followed in all *Nicotiana* monographs down to the present day, although several species have been shifted back and forth. The section POLIDICLIA is based upon *Nicotiana quadrivalvis* Pursh (Lehm. *Gen. Nic. Hist. pl. 4*) and its variety *multivalvis* Gray (*Syn. Fl. N. Amer.* 2¹: p. 253). (See COMES' *Monographie du genre Nicotiana.* Naples. 1899, p. 54.) The experiments on *N. Bigelovii* Watson and *N. quadrivalvis* Pursh reported in this paper show that such a section is unwarranted.

The writer began an extended series of genetic investigations upon the species of the genus *Nicotiana* in 1907 at the Connecticut Agricultural Experiment Station. Seed of several species was very generously given by Professor O. COMES of Naples, Italy, through Dr. D. G. FAIRCHILD of the United States Department of Agriculture; by Dr. A. SPLENDORE of Scafati, Italy; and by Professor W. A. SETCHELL[2] of the University of California. The source of the seed from Italy is unknown to me, but several of the species obtained from Professor SETCHELL were only one or two generations removed from the wild. The following description of *N. quadrivalvis* Pursh is taken from Gray's *Synoptical flora of North America.*

N. QUADRIVALVIS Pursh. A foot high, rather stout, more or less viscid pubescent, low-branching: leaves oblong or the uppermost lanceolate and the lower ovate-lanceolate, acute at both ends, mostly sessile (3–5 in. long); the

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[1] Contribution from the Laboratory of Genetics, Bussey Institution of Harvard University. No. 14.

[2] I had the pleasure of describing some of my experiments with *Nicotiana* hybrids to Professor SETCHELL, during his visit to Boston last winter. He said at that time that he had reached conclusions similar to mine in regard to *N. Bigelovii* and *N. quadrivalvis*, although what experiments he has made I am unable to state.

lowest larger and petioled: flowers few; calyx teeth much shorter than the tube, about equalling the 4-celled (or sometimes 3-celled?) capsule: tube of the corolla barely an inch long, the 5-lobed limb an inch and a half or more in diameter; its lobes ovate and obtusish, veiny.

Oregon, and cultivated by the Indians from Oregon to Missouri; their most prized tobacco plant. Perhaps a derivative of the preceding species.[3]

Three sets of seed, purporting to be this species, two from Italy and one from California, were grown. The plants obtained were alike in every detail within the limits of fluctuating variation. One selection has bred true for four generations. They differed from the above description in only one character. The lower leaves could hardly be called petioled, although they tapered almost to a petiole. The plants when grown in a normal fertile soil always had a large number of capsules with four cells. There were individual capsules, however, with three and sometimes even two cells on the same plants. This feature is evidently a physiological variation, for when grown in small pots in the greenhouse

FIG. 1.—At left, *Nicotiana quadrivalvis* Pursh; at right, *N. Bigelovii* Watson; young plants.

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[3] This statement, overlooked by me until the conclusion of the experiments, refers to *N. Bigelovii*.

and partially starved, the percentage of two-celled and three-celled capsules is much increased. The progeny of the starved plants gave only normal plants.

GRAY'S description of *N. Bigelovii* Watson is as follows:

> N. BIGELOVII Watson. A foot or two high: leaves oblong-lanceolate, sessile or nearly so; the lower (5–7 in. long) with tapering base; the upper (3 to 4½ in. long) more acuminate, with either acute or some with broader and partly clasping base; inflorescence loosely racemiform, with all the upper flowers bractless: calyx teeth unequal, linear-subulate, about equalling the tube, surpassing the capsule: tube of the corolla 1¼ to 2 in. long, narrow, with a gradually expanded throat; the 5-angulate-lobed limb 12–18 lines in diameter.

FIG. 2.—At left, *Nicotiana quadrivalvis* Pursh; at right, *N. Bigelovii* var. *quadrivalvis;* mature plants.

Seed from Italy and from California gave plants agreeing perfectly with this description. What was not so noticeable in the published descriptions of the two species was the remarkable similarity of living plants of the two species *N. Bigelovii* and *N. quadrivalvis*. The latter differs from the former only in its smaller size and the number of cells in the capsule. Even the viscid odor, which is stronger than in other species of the genus with which I am familiar, is the same in both. It naturally occurred to me that they might both be the same species, a thought simply a little more radical than the one that had already occurred to GRAY.

The species were crossed, therefore, and gave perfectly fertile hybrids intermediate in character, with partial dominance of the four-celled capsule. Unfortunately the cross between the normal two-celled *N. Bigelovii* and *N. quadrivalvis* has been lost. It is interesting from the standpoint of the transmission of that character, and will be remade.

The similarity of the two plants and the fact that they give a cross that is fertile in the F_1 generation is sufficient evidence to convince me that *N. quadrivalvis* Pursh is really *N. Bigelovii* var. *quadrivalvis.* There is further evidence in the fact that *N. Bigelovii* has produced a *quadrivalvis* variety while under observation.

FIG. 3.—At left, *Nicotiana Bigelovii* var. *quadrivalvis;* at right, *N. quadrivalvis* Pursh; in center, F_1 generation of reciprocal crosses.

Several plants from the different selections of *N. Bigelovii* with a single capsule having three cells were observed. Seeds from these plants were selected with the object of producing a race having three-celled capsules. Selection had absolutely no effect. Among the progeny an occasional three-celled capsule was found, but the percentage could not be increased. In this strain of *N. Bigelovii* there was evidently no ability to transmit the three-celled character. It simply gave an occasional zygotic variation of this kind, just as do many other species of *Nicotiana.*

On the other hand, several other plants, typically *N. Bigelovii* in size, produced several three-celled capsules. It is quite probable that they were all progeny of one plant of the preceding generation. One of these plants was selfed and the resulting seed planted on rich ground the following year.[4] Each plant among the progeny had numerous three-celled capsules, together with occasional two-celled and four-celled capsules. For two generations the strain has bred true to this condition. The only other abnormality observed is the occurrence of a greater number of flowers with six sepals and six petals than is common in the normal *N. Bigelovii* or the *N. quadrivalvis*. About 1 per cent of the flowers from the normal species have the extra petal and sepal, but on individual plants in the aberrant strain of *N. Bigelovii* from 2 to 5 per cent of the flowers vary in this manner.

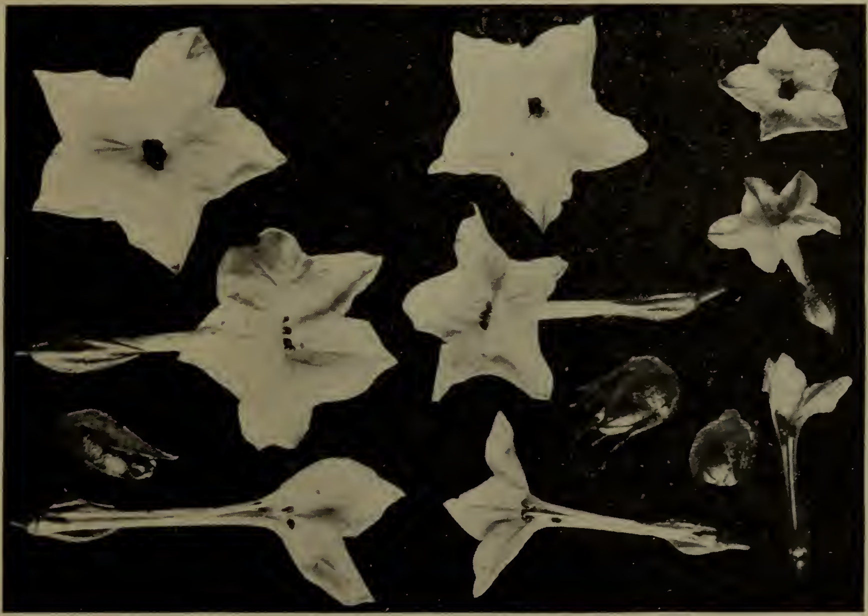

FIG. 4.—At left, *Nicotiana Bigelovii* var. *quadrivalvis;* at right, *N. quadrivalvis* Pursh; in center, F_1 generation of cross.

The aberrant *N. Bigelovii* was crossed reciprocally with *N. quadrivalvis*. The F_1 plants were alike in each case. They were intermediate in stature and in size of flower. The earlier capsules were four-celled; later in the season both three cells and two cells were produced. The F_1 plants were fully as fertile as the parent species. Each plant produced hundreds of well filled capsules.

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[4] Seeds are always started in sterilized soil and seedlings set in the open.

One plant each from the cross and its reciprocal were selfed. About 70 plants were grown from each mother plant. No difference was noticed in the two F_2 generations. The entire lot can therefore be considered together. The plants varied in height from one foot to two feet. The flowers were in general intermediate in size, but varied to the extremes that characterize each parent. No plants having only two-celled capsules were found; 71 had a large number of four-celled capsules; 40 had only a few four-celled capsules; while 19 had no four-celled capsules. Every plant had large numbers of three-celled and two-celled capsules, whether or not four-celled capsules were present.

Summary

1. Two elementary species of *N. Bigelovii* Pursh have been found. In one the capsules are two-celled and selection of individuals having an occasional three-celled capsule does not increase the tendency; in the other the tendency to have a greater number of cells than two in the capsule is always transmitted.

2. *N. quadrivalvis* Pursh and normal *N. Bigelovii* Watson are alike in all specific characters except the number of cells in the capsule, and since they give fertile hybrids when crossed it is thought that *N. Bigelovii* gave rise to *N. quadrivalvis.*

3. It is proposed that the section POLIDICLIA in the genus *Nicotiana* be dropped, and *N. quadrivalvis* Pursh be called *N. Bigelovii* var. *quadrivalvis.*

HARVARD UNIVERSITY.

THE BEARING OF TERATOLOGICAL DEVELOPMENT IN NICOTIANA ON THEORIES OF HEREDITY

ORLAND E. WHITE

NEW YORK
1913

[Reprinted without change of paging, from the AMERICAN NATURALIST, 1913.]

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ORLAND E. WHITE

[Reprinted from THE AMERICAN NATURALIST, Vol. XLVII., April, 1913.]

THE BEARING OF TERATOLOGICAL DEVELOPMENT IN NICOTIANA ON THEORIES OF HEREDITY[1]

ORLAND E. WHITE

BUSSEY INSTITUTION, HARVARD UNIVERSITY

IT is desirable, though difficult, to atack genetic problems by both pedigree-culture and cytological methods. It is desirable because the problems are viewed from different standpoints; it is difficult because few forms are especially favorable for either kind of work. The present paper is a preliminary report upon certain characters in a species fairly desirable from each point of attack.

Among plants teratological phenomena are common, especially those known as fasciations, Masters[2] citing, in 1869, 120 genera in which they were not infrequent.

The term fasciation is a broad one and includes, from a genetic standpoint, some very different phenomena. At least two distinct kinds of variation are now emphasized in genetic work, somatic and germinal, although often it is impossible to distinguish between them except by experimental cultures. Fasciation is a phenomenon of variation in which both types occur, though the evidence on this point is not all that could be desired. All observers agree that the fasciated character is constant and heritable in such races as *Celosia cristata*[3] (cockscomb), *Pisum sativum umbellatum*,[4] *Sedum reflexum cristata*,[5] some races of *Zea mays* and *Nicotiana*

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[1] Contribution from the Laboratory of Genetics, Bussey Institution of Harvard University.

[2] Masters, M. T., ''Vegetable Teratology,'' pp. 9–21, London, 1869.

[3] De Vries, H., ''The Mutation Theory,'' **2**: 68, 516–519, 1910; also Lynch, Irwin, ''Evolution of Plants,'' *Journ. Roy. Hort. Soc.*, **25**: 17–31, 1900.

[4] De Vries, H., *ibid.*, p. 513, 1910. See also Lynch, I., *ibid.*

[5] Masters, M. T., *ibid.*, pp. 18–19, 1869.

[6] East, E. M., and H. K. Hayes, ''Inheritance in Maize,'' *Conn. Agr.*

tabacum fasciata. On the other hand, many examples of fasciation are slight or severe somatic modifications, no more permanent than a swollen limb due to a bruise in our own bodies, or a bone spavin in a horse's foot, though the tissue proliferation may remain as a lasting scar. Examples of this form may be found in *Œnothera,*[7] *Nasturtium,*[8] *Picris hieracioides* and *Raphanus raphanistrum.*[9] Such modifications are imperfectly understood, but may be brought about directly or indirectly by external agencies such as bruises, culture methods and insect injuries to the initial meristem.

Aside from the work of Mendel[10] and De Vries,[11] the phenomena of fasciation have not been dealt with in the light of modern genetics. Mendel's investigations were made on a fasciated strain of pea (*Pisum sativum umbellatum*). When crossed with a non-fasciated strain the teratological character was recessive and segregated in F_2 in a simple 3:1 ratio. This result was essentially confirmed by Lock and Bateson, although environmental conditions were found by them to affect the character more than is usual in such phenomena.

De Vries failed to distinguish between fasciations strictly heritable and those non-heritable. The only con-

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Exp. Sta. Bull., No. 167, and Contrib. from Lab. of Genetics, Bussey Inst. of Harvard Univ., No. 9, p. 133, Pl. XXII (*a*) and (*b*), 1911; also Emerson, R. A., personal communication, 1911.

[7] Knox, A. A., "Induction, Development and Heritability of Fasciations," Carnegie Inst. of Wash. Pub. **98**: 1–21, Pls. I–V, 1908.

[8] Knox, A. A., *ibid.*, p. 14.

[9] Molliard, M., "Cas de virescence et de fasciation d'origine parasitaire," *Rev. Gén. de Botanique*, **12**: 323–327, 1900; also Godron, A., "Mélanges de teratologie végétale," *Mem. Soc. d. Sc. Nat. d. Cherbourg*, **16**: 81–127, pp. 96–97, 1871–1872.

[10] Mendel, G. J., "Versuche über Pflanzen-Hybriden," *Verh. Naturf. Ver. in Brünn*, **10** Abh., p. I. See Bateson, W., "Mendel's Principles of Heredity," pp. 322, 328, 330, Cambridge Univ. Press, 1909.

[11] De Vries H., *ibid.*, III, "The Inconstancy of Fasciated Races," pp. 488–526, 1910; "Monstruosités héréditaires offertes en échange aux Jardins Botaniques," *Bot. Jaarboek*, **9**: 62–93, 1897; "Over de erfelykheid der fasciatiën. Avec un résumé en langue française," *Bot. Jaarboek Dodonaea*, **6**: 72, 1894; "Sur la culture des monstruosités," *Comptes Rendus*, **128**: 125, 1899; "Sur la culture des fasciations des espèces annuelles et biannuelles," *Rev. Gén. de Bot.*, **2**: 136, 1899.

stant fasciated race[12] with which he worked is the cockscomb and his experimental researches on this plant led him to conclude that "complete atavists," or normal plants carrying the fasciated character in a latent state, are very rare, and even under repeated selection are to be obtained in very small numbers. Further, the normal plants thus obtained do not breed true, but revert very soon to the abnormal condition. While investigations have not been made showing definitely that many of De Vries's fasciations were not heritable, but were simply somatic modifications, enough evidence is at hand from numerous sources to justify at least the expression of a strong doubt of their heritable character. According to the observations of Knox, fasciated stems in Œnotheras are not germinal in origin, but traceable directly, in most cases, to insect injuries. Observations by Molliard on *Raphanus* and *Picris* support this conclusion, while Godron was unable to secure fasciated individuals from the seeds of a *Picris* plant thus affected. The fact that fasciation appears in every generation of *Œnothera* plants in varying percentages, in certain cultures, especially those of a biennial nature, is best explainable on a re-infection basis. Spiral torsion races such as *Dipsacus sylvestris torsus* in De Vries's cultures behaved, from a genetic standpoint, in the same manner as his fasciated races. Races of *Dipsacus* species are rich in torsions in Holland and Denmark, but, according to Johannsen,[13] the seeds of torsus strains when grown in England produced normal progeny. This would indicate an environmental rather than a germinal basis as a causal factor.

In all of De Vries's experimental cultures of fasciated races (with the exception of *Celosia*) only a certain per cent. (averaging in most races 50 per cent. or less) of the individuals in each generation possessed the abnormality, and he was never able to breed a constant and

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[12] Possibly *Geranium molle fasciatum* may be an exception in which more than one unit factor is responsible for the anomaly. Otherwise it should have bred true at least by the sixth generation if the seed sown each year was from carefully guarded plants.

[13] Johannsen, W., public lecture IV, Boston, 1911.

genetically pure race. Even though he had been able to do this, doubt could still be cast upon the belief that he was dealing with a strictly heritable character, because the only method that seems to preclude doubt is crossing with the normal and securing the F_2 ratio. This method would eliminate the suspicion that minute bacterial or even ultra-microscopic organisms were acting as causal agents.

Emerson and East in their maize studies have obtained races breeding constant for fasciated ears. I have myself examined such a race in Emerson's cultures. Hus, on the other hand, with the same sort of an abnormality in the same plant species, *Zea mays,* secured results similar to those of De Vries.[14] Is the difference in results due to methods or to the nature of the plant abnormality itself? I shall consider the results of De Vries and others holding similar opinions in greater detail in a later paper, as such results entail an extended review.

THE PROBLEM AND THE MATERIAL

The problem to be discussed briefly in this paper is the relation of the cytological phenomena in the reduction divisions to certain segregating Mendelian characters, and the nature of these characters in development and inheritance.

The material upon which the study is largely based is a fasciated variety of *Nicotiana tabacum.* Although fasciations are very common in many genera and not infrequent in others, they have never been recorded (so far as I can determine) in *Nicotiana.* The present race was obtained from the selfed seed of a mutant found growing in a field of Cuban tobacco in the district of Partidos, near the town of Alquiza, Cuba, in 1907. I am indebted to Dr. E. M. East and to Mr. J. S. Dewey[15] for

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[14] Hus, H., and Murdock, A. W., "Inheritance of Fasciation in *Zea mays,*" *Plant World,* **14**: 88–96, 1911.

[15] Mr. J. S. Dewey is superintendent of the United States tobacco plantation belonging to the same company that controls the Cuban plantation near Alquiza.

data on this race, prior to the summer of 1910. The sport is characterized by a flattened, fasciated condition of the stem and floral structures, and a consequent increase in the number of leaves. The original mutant is described by Dewey as possessing 152 leaves on the main stalk, flowers abnormal, stem fasciated. When the abnormal plants were studied in more detail, many smaller teratological features were found, and these were especially plentiful in connection with the floral structures. The pistil frequently was incapable of functioning, because of various forms of tissue proliferation in the region of the stigma. The style was often shortened, coiled or fused near its base with an anther (staminody of the pistil). The ovary locules were very much increased in number, ranging from two (extremely rare) to as high

Fig. 1. ½ Stems and flowers from the abnormal and normal strains of N. tabacum.

as twenty. Very often two or even three distinct pistils were formed in the same flower, all of which in some cases could function. The stamens were affected in both filament and anther. The filaments were often coiled, twisted, shortened or fused to the corolla. In rare cases, they were petaliferous. The anther deformities consisted of split anthers, anthers with small pistils growing from them—two or three to an anther being present in one case (pistillody of the anthers). The number of pollen sacs varied from the normal four to six.

The corolla and calyx were often split, and the lobes of the calyx and not uncommonly of the corolla, were irregular in size and shape. Occasionally the calyx and corolla merged into each other by a spiral twist. Two flowers sometimes were enclosed by the same calyx. Once or twice flowers have been found consisting of only a corolla and a few stamens, growing on the side of the normal corolla and partly fused with it. The corollas never show a doubling phenomenon to accommodate the increase in petal number but the circumference of the flower is extended, and very often these flowers are as regular and symmetrical as those of the normal. Two cases of leaves fused at the base have been found and the phyllotaxy is altered and irregular. The fasciated plants when young are practically indistinguishable from the normal. The anatomical features have not been investigated sufficiently for a report upon them at this time, and it is possible that differences between the normal and abnormal seedlings will be found when this part of the study is completed.

Five generations of the abnormal strain have been grown, amounting in all to over a thousand plants, and each individual plant has possessed the unmistakable characters of the original mutant. The monstrous character is, however, a variable one, since the stems may be extremely flattened throughout the greater part of their length or only flattened and fasciated toward their apical ends. Other characters, as already implied, fluctuate between extremes, depending in part on environment

A Comparison of Certain Characters of the F_1 and F_2 Generation of

303–1–12 is a type of extreme abnormalness. 301–1–2 represents the extreme *tabacum*. (304 × 402)–1–10 = normal F_2 segregate. (304 × 402)–1–12 = abnormal heterozygous F_2 segregate similar to the F_1 (304 × 402)–30.

		Stem	No. of Leaves	Height, In.	Flower	1	2	3	4	5	6	7	8
♀P	303–1–12	V. abn.	69	76	Sepals . .	7	6	6	7	6	8	9	7
					Petals . .	12	9	6	10	11	12	11	13
					Stamens	12	12	7	12	8	13	11	12
					Ovary locules.	3–3–3	3–3	4	3–3–2	4–3	4–4	3–3	6–3
	301–1–2	N.(?)	33	68	Sepals . .	6	6	7	6	7	6	6	6
					Petals . .	7	6	7	7	8	8	7	7
					Stamens	7	6	6	7	7	7	7	7
					Ovary locules.	3	3	3	3	3	3	3	4
♂P	402	N.	24	72	Normal as in (304 × 402)–1–10								
F_1	(304 × 402) –30	N.(?)	26	87	Sepals . .	6	6	7	6	6	6	5	6
					Petals . .	7	6	7	6	5	6	5	7
					Stamens	7	6	7	6	6	6	5	7
					Ovary locules.	3	3	3	2	2	3	2	2
F_2	(304 × 402) –1–10	N.	24	75	Sepals . .	5	5	5	5	5	5	5	5
					Petals . .	5	5	5	5	5	5	5	5
					Stamens	5	5	5	5	5	5	5	5
					Ovary locules.	2	2	2	2	2	2	2	2
	(304 × 402) –1–12	V. abn.	80	75	Sepals . .	10	12	6	8	10	7	8	9
					Petals . .	12	20	8	12	15	8	21	12
					Stamens	16	15	7	8	12	7	18	12
					Ovary locules.	6–10	6–3–4	5	3–3	3–4	4	6–4–2–2	4–3
	(304 × 402) –1–34	S. abn.	63	75	Sepals . .	8	7	7	7	7	6	6	6
					Petals . .	8	8	7	7	7	7	7	8
					Stamens	7	8	7	7	8	7	7	8
					Ovary locules.	3	3	3	3	4	3	3	3
	(304 × 402) –1–6	N.(?)	32	75	Sepals . .	5	6	6	6	7	5	6	6
					Petals . .	5	6	6	6	7	5	7	6
					Stamens	5	6	6	6	7	5	7	6
					Ovary locules.	2	3	2	3	3	3	3	3

[16] Double flower.

ABNORMAL × NORMAL *Nicotiana* (304×402) WITH THOSE OF THE PARENT STRAINS

variation of the abnormal characters toward the normal. 402 is the normal *N.* F₂ segregate. (304 × 402)–1–34 = heterozygous F₂ segregate. (304 × 402)–1–6 =

9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26
8	7	7	6	7	7	6	6	6	8	8	7	6	6	7	7	7	
11	12	11	10	8	8	7	9	9	16	8	10	7	7	11	7	11	
11	14	11	12	9	8	7	11	8	17	9	12	7	6	11	7	11	
3–3	4–6–4	5–3	3–4	3–3	5	4	3–2	4–4	8–5	5	2–4	3	3	3–1	3	3–3	
8	7	6	7	8	7	6	6	6	6	6	7	7	6	6	6	6	8[16]
8	7	8	7	8	8	9	8	7	7	7	7	8	7	8	8	5	7
10	7	6	7	7	7	8	9	7	6	6	7	7	6	8	7	5	7
4–2	4	3	3	4	3	3	2–3	4	4	3	3	3	3	4	3	3	3

Normal as in (304×402)–1–10

9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26
5	7	7	6	6	6	6	6	6	7	6	5	5	5	5	7	5	
5	7	6	6	7	6	6	7	6	7	6	6	6	5	6	7	7	
5	7	6	6	6	6	6	7	6	7	6	6	6	5	5	7	7	
3	2	3	2	3	3	3	3	3	3	3	3	3	2	3	3	2	
5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	
5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	
5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	
2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	
7	8	8	12	7	10	6	7	8	8	7	7	7	7	8	7	8	
10	9	18	24	12	15	8	8	10	15	12	15	8	8	9	12	8	
7	9	16	24	8	18	7	7	6	15	8	10	7	8	8	10	8	
3	4–3	4–4–4–3	7–6–4–4	4	4–4–2	3	4	3	5–3	3–2	4–3	4	5	4	3–4	3–2	
8	7	8	9	8	7	8	7	8	7	12	9	8	8	8	12	11	
8	7	7	11	8	7	8	7	9	7	16	12	11	14	8	12	18	
7	7	8	10	7	7	7	7	9	7	16	12	11	14	7	12	15	
4	3	4	4–4	3	3	3	3	4	4	4–6–4	4–3	4–4	4–4	2	2–2	4–5–4	
5	5	5	5	6	6	6	6	6	5	5	5	5	6	6	5	6	
6	6	5	6	7	7	6	6	6	6	7	6	7	7	7	5	7	
6	6	5	6	6	7	6	6	6	6	6	5	7	6	6	5	7	
2	3	2	3	3	3	4	3	3	3	3	3	3	3	3	3	3	

and in part on the *innate* nature of the character itself. One may confuse this fluctuation to the arc made by a swinging pendulum. The arc through which the pendulum can swing is limited, but within those limits the arc may be medium, large or small, depending on the agencies that set the pendulum in motion. Agencies very different in nature may produce the same result. The pendulum is the material body which makes the arc recognizable as an entity, and in this simile may be compared to the gene for fasciation. When the pendulum is motionless, there is no arc, and there would be no fasciation if the gene remained potential.

A more definite idea of the characters of the plant and their variability may be secured by consulting the table on page 212. It should be stated that plants have been grown under many environments and with many variations in culture. But so far as our present interest goes, no very great changes have resulted. The race has always been clearly distinguishable in the adult state from the normal, whether grown under cramped greenhouse conditions, or out-of-doors; whether surrounded by a Cuban or a New England environment. No especial care, such as De Vries prescribes, regarding culture and transplantation has been given, and yet the anomaly has always bred absolutely true and no "atavists" have appeared.

The normal Cuban variety from which the fasciated strain arose is characterized by a normal round stem, regular phyllotaxy, flowers with five petals, sepals, stamens and a two-loculed ovary. The number of commercial leaves varies between 20 and 25, all leaf counts in the present investigation, being made by the commercial method.[17] Fertility is practically 100 per cent. Occasionally among hundreds of flowers examined a flower is found with an extra sepal or petal, otherwise abnormalities are unknown in our cultures of the normal variety.

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[17] All leaves were recorded up to the first leafless branch ("bald sucker"), exclusive of the first three basal leaves.

In the cultures at the Bussey Institution of Harvard University the normal Cuban is known as 402: the fasciated strain as 300–309, the range in numbers representing an attempt at selection.

METHODS

Data were collected on each plant and tabulated separately. The characters noted were, extent of fasciation in the stem; number of leaves, petals, sepals, stamens and ovary locules. Twenty-five flowers were taken from each plant and the parts of each flower recorded separately. In all crosses made the flowers were castrated in the bud and bagged. Pollen was taken only from anthers still in the closed bud and 95 per cent. alcohol was liberally used after each operation on hands and instruments. The Webber system of recording the plants by number was used. All seed was sown in sterilized soil and all possible care taken to avoid mixtures.

EXPERIMENTAL WORK

Numerous crosses were made between distinct species and the abnormal race, but all of the progeny were sterile, though the abnormal character was visible in their flowers and in the increased number of leaves which they bore.

Four crosses were made between normal *N. tabacum* varieties and the abnormal, all of which produced fertile F_1 plants. The most interesting of these is a cross of the abnormal with the normal Cuban variety from which it mutated. Three generations of this cross (304 × 402) have been grown. The F_1 generation consisted of 39 plants, the F_2 of 97 and the F_3 of 647, totaling 783 individuals. The F_1 was intermediate in character between the two parents, as the table will show. The F_2 gave the three expected types in the ratio of 1:2:1, the actual numbers being:

	Normal	Heterozygote	Abnormal
Actual............................	28	52	17
Expected........................	24	48	24

The F_2 selections gave the results expected in F_3. Counting the total progeny (248) from F_1 and F_2 heterozygotes, the figures are:

	Normal	Heterozygote	Abnormal
Actual	68	124	56
Expected	62	124	62

The F_2 heterozygotes were in appearance duplicates of the F_1 individuals and after a little experience could be easily distinguished from the abnormal homozygotes.

Clean segregates were obtained from the heterozygous

FIG. 2. F_2 segregates from the abnormal × normal (304 × 402) Nicotiana. Abnormal homozygote, heterozygote and normal homozygote. The grandparents in appearance are duplicates of the two homozygotes

plants in both F_2 and F_3 and the homozygous normals and abnormals obtained in this manner bred true in F_3 and F_4. Not being satisfied that only one factor represented the difference between the normal and the abnormal, I thought that it might be possible, through selection, to secure a normal strain from the abnormal, or at least to modify the unit character, as Castle and his students appear to have done with the hooded pattern in rats.

Selection work was started by selecting from the cultures the most abnormal and the least abnormal plants as seed producers. The work was carried through two generations with no prospect of success and there it remains at present. Progeny of the least abnormal plants were as much fasciated and otherwise abnormal as the original parent strain growing beside it. And one could not distinguish the least abnormal from the most abnormal strain except by the label. So far as the work has progressed, this fasciated strain seems no more amenable to selection than the cockscomb with which De Vries worked, and of which he said "at present at least there seems not to be any prospect of obtaining a pure atavistic strain."[18]

From a comparison between the drawing in Gerarde's Herball of 1597[19] and certain woodcuts from old horticultural magazines with the plants as they are to-day, it does not appear that much change has taken place in the cockscomb fasciation since its introduction into Europe in 1570.

The changes in the expression of the comb that gardeners and florists will maintain have taken place as a result of selection can all be accounted for by the influence of the environmental factor.

Lock planted seeds of very slightly fasciated individuals of the F_2 generation of normal × fasciated stem in *Pisum*. The F_3 plants were almost, if not as much fasciated, as the original grandparent strain.[20]

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[18] De Vries, H., "The Mutation Theory," 2: 519, 1910.

[19] Gerarde, John, "Herball or Generall Historie of Plantes," 1st ed., pp. 323–325, Fig. on p. 323, 1597.

[20] Lock, R. H., *loc. cit.*, p. 106.

This fasciated strain of pea (Mummy Pea or *Pisum sativum umbellatum*) would appear to have been a very constant race, at least since 1597, when it was figured in Gerarde's "Herball."[21]

From the results of hybridization and selection, one may draw the conclusion that the fasciated mutant differed from the normal parent strain by only one factor and that it represents a mutation upon the variability of which selection has no modifying effect. The character itself appears to be due to one underlying cause and its variableness is only the external manifestation of the capricious working of that cause.

After completing a satisfactory study of the gross aspects of this character, a cytological investigation was made, with the hope that here might be found a clue to the cause or causes underlying the appearance of the anomaly.

CYTOLOGY

Much trouble in fixing material was caused through the presence of resinous substances in the tissues. Flemming's medium and strong solutions were finally found to be the most successful, although prolonged bleaching of the sections with H_2O_2 was necessary to eliminate the blackening. Care had to be exercised to secure quick penetration, as poor fixation and shrinkage were likely to result after a bath of over 24 hours. The preparations were stained in Heidenhain's iron hæmatoxylin and counterstained with clove oil saturated with erythrosin. This combination usually gave the best results —a deep black chromatin stain against a brilliant red background. Preparations were also stained with the safranin-gentianviolet-orange G combination of Flemming and restained with iron hæmatoxylin. This method gave very sharp outlines, not easily obtainable in some phases, when the ordinary hæmatoxylin method was used. Another combination which was found valuable in cases where the chromosomes were closely crowded

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[21] Compare with photograph in Darbishire, A. D., "Breeding and Mendelian Discovery," p. 22, Fig. 8, 1911.

together, as in certain metaphases, is safranin, magdala red and azure II. Large quantities of the fresh material of the anthers in various stages of maturity were stained with methyl green and microscopically examined. In most cases one anther of a bud to be fixed was inspected in this manner. This precaution was necessary as a check on the occurrence of artifacts from fixation.

Briefly, conditions in the normal Cuban variety (402) are as follows. The ordinary maturation processes are those cytologists have so often described for plants, and need no recapitulation here. The spireme in prophase is single and, just preceding diakinesis, breaks up into segments which take the form of twisted and horseshoe-shaped loops. The latter resemble Davis's figures for *O. grandiflora.*[22] Each loop consists of two spireme segments joined at one end, which in the later heterotypic phases separate and go to opposite poles. Each segment is interpreted as a somatic chromosome, and the members of a pair are homologues. The other phases present nothing peculiar. The homotypic chromosomes appear as entities first in very late anaphase of the first division. The reduced chromosome number, as determined by very numerous counts of heterotypic metaphases and anaphases, and homotypic telophases is 24, the $2n$ being 48. The somatic number ($2n$) was determined by adding together the homotypic telophase chromosomes of a tetrad and dividing the entire number by two. Polar views of the metaphase of the first division in sections of 10μ have repeatedly shown the 24 gemini, each geminal chromosome consisting of a diakinetic pair. Variation of chromosome number in these normal (402) anthers is very rarely, if ever, to be found. None was found in the present investigation. Irregular divisions are not common, although occasionally one sees lagging chromosomes. Usually the phases of a single pollen sac

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[22] Davis, B. M., "Cytological Studies on *Œnothera.* I. Pollen Development of *Œnothera grandiflora,*" *Ann. Bot.*, **23**: 551–571, Pl. XLI, Figs. 31, 34, 36, 1909.

are more advanced at one end than at the other and the maturation processes are at a similar stage in the different sacs of the same anther. Cursory examination of reduction phenomena in the ovule confirmed these observations.

Many anthers of the abnormal (300–309) when examined cytologically, were entirely normal in all their phases of maturation. Others showed evidences of almost total sterility through premature breaking down of the archesporial tissue, while still others were only partially sterile. Anthers of this strain were mentioned earlier as sometimes having more than four pollen sacs. In such cases the maturation phases were in very different stages in the different sacs. In one sac the archesporium might be in early prophase, while in other compartments there would be almost mature pollen. This extreme variation in maturation was not confined to anthers with an abnormal sac number, but was often true of those normal in this respect. Conditions in the anthers of the abnormal strain are similar as regards the normal cytological phenomena, but various abnormalities are not uncommon. These manifest themselves in such a manner that one can not avoid believing that some subtle agent is at work here, too, distorting the internal as well as the larger so-called external characters. In both reduction divisions in all the strains examined, various abnormal phenomena are to be found which are not due to fixation or other technical operations. Contrasted with the normal (402) the maturation phases in different sacs of the same anther may be far apart. Nearly mature pollen is present in some sacs. while others in the same anther may not have progressed farther than diakinesis. Pollen tetrads are often rare in nearly mature anthers. This is true of at least five per cent. of those examined. Mother-cells may break down during early prophase, diakinesis or any of the later phases. In early prophase, the nuclear membrane may disappear and the whole archesporium disintegrate.

Again, I have found that in some sacs most of the archesporium has broken down, but some few cells seem to have escaped destruction and matured. The metaphase, so far as I have observed, is not so likely to be disturbed. An occasional premature splitting of the chromosomes takes place, increasing the number to be seen in the polar view of the nuclear plate. These are rare, but they have been observed in both the abnormal strain and the abnormal segregates. This feature has been referred to as a premature splitting,[23] but it may be interpreted as an actual increase in number such as Wilson found in *Metapodius,*[24] Stevens in *Diabrotica*[25] and Strasburger in *Wikstroemia.*[26]

In one pollen mother-cell, 51 chromosomes were clearly distinguishable, but disintegration had already commenced. In another case 30 were counted, the mother-cell appearing perfectly normal, although in the anther containing it irregular divisions were taking place. The nuclear metaphases in which such an increase in number can be seen are rare, but so far as I have observed, and I have counted many nuclear plates ideal for such work, they only occur in the abnormal or in the abnormal segregates. The heterotypic anaphases of the abnormal often show the chromosomes lagging or distributed promiscuously over the spindle. In only one case so far have I found irregular conditions in the telophase and this only in the case of one mother-cell. Counting is unsatisfactory in the anaphase of the first division, as the nucleus is small and the chromosomes are many.

Irregular divisions are present in the homotypic, but most of the abnormalities occur during the heterotypic mitosis.

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[23] Preparatory for the homotypic division.

[24] Wilson, E. B., ''Studies on Chromosomes. V. The Chromosomes of *Metapodius,* a Contribution to the Hypothesis of the Genetic Continuity of Chromosomes,'' *Journ. of Exp. Zool.,* **6**: 147–205, 1909, 1 plate and 13 text figures.

[25] Stevens, N. M., ''Further Observations on Supernumerary Chromosomes, and Sex Ratios in *Diabrotica soror,*'' *Biol. Bull.,* **22**: 231–238, figs. 1–13, 1912.

[26] Strasburger, E., ''Chromomenzahl,'' *Flora,* **100**: 1910.

It is impossible to say whether pollen grains capable of functioning ever result from those divisions where the chromosomes are irregularly distributed. The irregularities in reduction do not produce supernumerary pollen grains, such as have been described by Juel and Strasburger for *Hemerocallis,* for in all tetrads mature enough to show the separation of the pollen grains I have always counted four. One might expect an increase in number of pollen grains formed by one mother-cell, judging from the grosser manifestations of this abnormal factor. Functioning pollen is formed in quantity and no trouble at all is found in securing plenty of selfed seed of the abnormal strain.

Reduction phenomena in the ovule of the abnormal have so far been given only a superficial examination and the observations are not complete enough to report. Observations on the ripe capsules of selfed plants would lead one to believe that here, as in the case of the pollen, partial sterility is present, due to the abortive development of the ovules, but the latter is only a supposition, which further cytological study may or may not support.

Cytological examination of the anthers of the three classes of plants obtained from the abnormal × normal (304 × 402) was made. The conditions in the reduction divisions of the normal and homozygous abnormal segregates are identical with those present in the two grandparents. The heterozygote differs from the pure abnormal in degree only, having fewer sterile anthers and other abnormalities. Otherwise what has been said of the pure abnormal (300–309) applies also to the heterozygote.

In connection with this cross, it is interesting to note what bearing the nature of the reduction divisions in the ovule (300–309) might have upon the F_2 ratio. The abnormal class, although within the probable error, is always deficient. This is true also in the fasciated peas with which Mendel[27] worked and in one race of peas hav-

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[27] *Loc. cit.*

ing sterile anthers with which Bateson[28] experimented. While the number of plants has not been large in any of these cases, one wonders why it is always the abnormal (pure) class which is deficient. If the reduction phenomena in the ovules of the abnormal *Nicotiana* agree with the conditions present in the anthers, it seems not unreasonable to believe that there may be a relation between the mortality of the gametes carrying the factor for abnormalness and the deficiency in the ratio. Increased mortality of this class of gametes over the normal class would reduce the chances for combinations of the abnormal gametes, and as a consequence the normal and heterozygote combinations would be increased.

Summary of Observations

Concluding, one must bear in mind that the facts so far obtained seem to warrant the belief that some agent is at work on the internal structure as well as on the so-called external, and is of such a nature as to produce abnormalities in cell structure as well as in cell complexes or plant organs. The data, as a whole, raise a question as to the significance of chromosomes in inheritance.

Two strains of *Nicotiana tabacum* have been investigated, one being a sport from the other. The sport has been shown to differ from the normal in the possession of a unit character due to one Mendelian factor. When it is crossed with the normal, there results in F_2 a simple Mendelian ratio of 3:1 as regards normal and abnormal characters. The heterozygote is, with a little practise, distinguishable, making the ratio 1:2:1 with abnormalness partially dominant. The F_3 generation has proved these segregates to breed true. *Absolutely* clean normal segregates appear in F_2 and breed true. The abnormal character has been described in detail, and shown to affect practically all the structural parts of the plant individual, even to the germ cells. Both strains have the same chromosome number, 48 and 24, as a definite mode.

[28] Bateson, W., and others, Reports to the Evol. Com., II, p. 91, 1905.

CHROMOSOMES IN RELATION TO MENDELIAN FACTORS AND A PHYSICAL BASIS OF INHERITANCE

Suppose we maintain the factor for the abnormal condition to be a particle of one chromosome. Gametes of the abnormal strain all contain the factor for abnormalness, as reciprocal crosses with the normal give the same results. In a cross a pollen grain of the abnormal strain unites with an egg of normal (402) parentage, and an intermediate is produced in F_1. The chromosome containing the factor for abnormalness is partly neutralized by pairing with a normal homologue. Gametes of two kinds are formed in approximately equal numbers in F_1, those containing the factor for abnormalness and those without it. But on a chromosome hypothesis, how are these gametes formed? There are two reduction divisions and 48 chromosomes, 24 of abnormal parentage and 24 of normal. According to current cytological investigation and interpretation, each chromosome separates from its homologue in its entirety during the first reduction division, so that, eventually, two kinds of gametes are formed as regards chromosomes. The factor for abnormalness or fasciation is in one chromosome, and chromosomes are believed to be in homologous pairs —one maternal with one paternal. The chromosomes of a homologous pair separate during the heterotypic anaphase, one going to each pole, it being contrary to current interpretation to believe that both members of a pair may go to the same pole. On this basis, according to the law of chance, approximately half the nuclei at the end of the heterotypic division will contain the chromosome carrying the factor for abnormalness and from half it will be absent.

Experimentally it has been shown that we have been dealing with only one pair of unit characters and that no complications are present. The various crosses have always given uniform results in F_1, even between species, and the fertile cross has given a close 1:2:1 ratio in F_2. Logically, then, one is led to believe that one out of the

24 chromosomes of abnormal parentage, and only one can contain the factor for abnormalness and produce the experimental results. If more than one contained it, the ratio in F_2 would be changed. For example, if it were present in two chromosomes, the ratio (as suggested by Emerson)[29] must be 15:1 or in this particular case where the heterozygote is distinguishable, 7:8:1. We might postulate its presence in all 24 chromosomes and believe, as Cannon[30] did, that parental chromosomes separate as a phalanx in the F_1 reduction division, each group going to one pole and thereby bringing about the formation of pure parental gametes. But the cytological investigations of Sutton, Rosenberg, Strasburger and others have brought to light evidence which precludes such a supposition. The experimental data from genetic researches are also opposed to this hypothesis, if one attempts to show a relation between the reduction division and Mendelian segregation. On a chromosome hypothesis, then, one *must* believe the factor for abnormalness to be present in only one chromosome out of the 48 concerned in the F_1 reduction phenomena, in order to be in agreement with the experimental results. This being the case, how is one to account for the abnormalities which occur during the reduction divisions in the anthers of the F_1 heterozygote? For they affect, not alone one chromosome, but all the nuclear and cell material concerned in the formation of the pollen grains. Can one postulate the influence of one chromosome to be so great, at times, as to bring destruction to its 23 associates of abnormal parentage, its 24 associates of normal parentage, as well as all the other organized contents of the mother-cell? Why, it may well be asked, if this destruction is the result of the activity of one chromosome does not it take place in the case of every anther and of every pollen mother-cell? Why should it affect only two or three

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[29] Emerson, R. A., "Genetic Correlation and Spurious Allelomorphism in Maize," 24th Ann. Rpt. Nebr. Agr. Exp. Sta., pp. 59–90, 1911.

[30] Cannon, W. A., "A Cytological Basis for the Mendelian Laws," *Bull. Torr. Bot. Club,* **29**: 657–661, 1902.

anthers in a flower containing 8 or 10? Not because it is absent from the other anthers, because the pollen from these anthers transmits the character. It is not a question of segregation then, but one of environment.

Evidently the gene is inactive or latent, for we know there is something present which for convenience we call a gene, and yet we can not see any of the visible signs of its presence, such as we see in the affected anthers. On a morphological conception it must be there; physiologically for the time being, so far as we can determine, it is non-existent. The inactivity we may suppose is due to a lack of a properly adjusted environment. This proper adjustment is only true of a few anthers in the F_1 plants. We believe this scarcity to be due to two kinds of latency —inactivity of the gene as in the pure abnormal and inactivity of the gene because of association with the cell materials that trace their lineage back to the sperm of the normal father. But latency is a vague term. In genetics, it is used to describe the period between the disappearance of a character and its reappearance. By pushing this conception to its logical conclusion it is clear that one can practically never prove the origin of a new character. Fasciation, while new to *Nicotiana,* is phylogenetically an old character. The production of purple fruits in *Rosa* would mean, phylogenetically, the reappearance of a latent character, for purple fruits are common to the *Amelanchiers* and to a species of *Pyrus.*[31]

The characters of the whole plant kingdom would be in a state of latency and patency, of inactivity and activity. To determine whether a character were new or not would involve a canvass of that part of the plant kingdom phylogenetically older than the family under investigation. Of course, we speak of segregation in phylogenetical lines, but the term has a different meaning in such cases. My F_2 normal segregates are pure and will breed true for absence of abnormalness, I believe, for any number of generations unless a new muta-

[31] *Pyrus Niedwetzkyana.*

tion occurs. These recurrent mutations, Johannsen says, are rare in his experience, but they are admitted to occur in almost any long-continued pedigree line, and if fasciation should appear as a repeated mutation after 20 generations of plants involving 2,000,000 individuals had been grown, is one to infer that the gene was present all this time, but latent or inactive? Or is this a new gene produced by the same condition that brought about the original fasciation? Logically, if one defends the latency conception, he must believe that the original gene for fasciation was inactive in all these millions of plants, which in our present stage of knowledge is a ridiculous assumption, since the term is used to describe a somatic appearance. Applied to genetic problems in general, hopeless chaos would result. But on the supposition that a portion of a chromosome is responsible for the abnormality, it seems to me necessary to assume the chromosome to be capable of becoming active or latent without cause. For it seems probable that the anthers are all alike from a constitutional standpoint. How else can one account for the normal anthers and the abnormal ones, the normal pollen mother-cells and those affected by the abnormality?

The conception of latency is not necessary in the case of complete or incomplete dominance in F_1 hybrids, for in such cases there is evidence that a gene from one parent may be partially or completely inhibited in its expression by factors from the other parent, and this is probably what happens when we bring a line of chromosomes and cell materials from the normal (402) plants and associate them (by fertilization) with a line of cell materials from the abnormal (300–309).

While the phenomena of segregation described in the preceding pages may be capable of interpretation on a morphological basis, the gene for fasciation appears to me to lie deeper in sporogenesis than chromosomes. The abnormal character development appears most easily interpreted from a physiological standpoint. In

F_1 there is no break in the continuity of its manifestations between sporophyte and gametophyte, even though reduction and probably segregation have occurred. And should we not expect to see such a break if segregation by chromosomes took place in sporogenesis?

The evidence as a whole I think, warrants one in the suggestion that chromosomes are characters of the zygote and gametophyte, on the same footing in development with other plant characters. It is more difficult to comprehend this conception of these bodies, because they appear as characters in the development of the cell, rather than in the development of the larger unit, the individual organism. They are characters in the sense that they disappear and reappear at a place and time in the life history of the organism which we can predict. They can be transferred from one race of organisms to another provided fertile F_1 hybrids are possible. They are influenced in as definite a manner, by the underlying cause represented by the term factor for abnormalness, as are the zygotic expressions included in the word fasciation.

Concluding, I realize these speculations are largely negative in character, but they are in accord with a steadily growing skepticism among students of genetics as to the importance of chromosomes in inheritance, and their relation to segregating Mendelian characters. The impression has been distinctly gained from a study of this abnormal strain and its crosses with the normal that chromosomes are not the omnipotent creators of destiny, but characters on the same footing with other structures. The same dynamic forces, whatever they are, are changing and modifying these chromosome characters in the same capricious manner as those of a grosser nature. One would be inclined to ascribe these changes to an ultra-microscopic parasitic organism were it not for the experimental evidence in F_2, which precludes such a belief.

My warmest thanks are due Dr. E. M. East for suggestions and criticisms while engaged in this investigation.

July, 1912.

INHERITANCE OF FLOWER SIZE IN CROSSES BETWEEN SPECIES OF NICOTIANA

(WITH PLATES VI–X)

E. M. EAST

Reprinted for private circulation from
The Botanical Gazette, Vol. LV, No. 3, March 1913

VOLUME LV NUMBER 3

THE

BOTANICAL GAZETTE

MARCH 1913

INHERITANCE OF FLOWER SIZE IN CROSSES BETWEEN SPECIES OF NICOTIANA[1]

E. M. EAST

(WITH PLATES VI–X)

Since the independent investigations of NILSSON-EHLE and of the writer demonstrated the feasibility of using the Mendelian notation to describe the inheritance of size characters that blend in the first hybrid generation, a number of botanical papers have appeared that supported this interpretation. These papers have considered the behavior in crosses of such characters as height of plant, size of leaf, number of leaves, time of flowering, and size of fruit. If the number of leaves in certain plants is excluded, this type of character is one particularly affected during development by external conditions. Since fluctuations produced in this manner are not transmitted, if the conclusions drawn from the sum total of our limited experimental cultures are to be given weight, the validity of the evidence in these investigations is not disturbed. At the same time, one must admit that these fluctuations obscure an analysis of the crude data. For example, plant *B* may be six inches higher than plant *A* when both are grown in the same environment, owing to a different heritage, but plant *A* may grow considerably higher than plant *B* if the environment of *A* is the best possible for maximum growth and the environment of *B* is poor.

In this paper, therefore, I propose to consider the inheritance of

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[1] Contribution from the Laboratory of Genetics, Bussey Institution of Harvard University.

a character-complex which I believe to be the least affected by external conditions of any character that shows marked varietal differences. This character-complex is corolla size. The fact that corolla size is so comparatively constant under all conditions attending development has such a definite bearing on some broad questions of organography that it merits separate discussion. On this account, the liberty of asserting the truth of the statement with only the following data in its support is requested.

During the past four years, I have grown about 20 species of *Nicotiana* in considerable numbers. They have been grown under very diverse conditions. Some have been starved in four-inch pots, others have had the best of greenhouse treatment; some have had poor field conditions, others have had all field conditions practically at their best. The height of the plants, the size of the leaves, and similar size complexes have varied enormously, but the size of the corollas has scarcely varied at all. For example, plants of *Nicotiana silvestris* Speg. and Comes grown to maturity in four-inch pots produced no leaves longer than 7 in. On the other hand, sister plants of the same pure line produced leaves 30 in. long in the field. Both series, however, produced flowers with the same length and spread of corolla. Furthermore, cuttings from 20 of the field plants reported in this study were rooted and grown in small pots in the greenhouse. Their blossoms were the same size as those of the field grown plants from which they came.

The material used in this particular experiment consisted of pure lines of two *Nicotiana* types that are generally treated as distinct species (pl. VI). The male parent was *Nicotiana alata grandiflora* Comes, it being the plant called *Nicotiana affinis* by horticulturists. Three lots of it were under observation; one was obtained from Italy and the other two from the United States, but the original sources of the strains are unknown. These three lots were alike, and in successive generations were constant in their characters. They accorded perfectly with COMES' description and were remarkably narrow in their variability. The female parent I have called *Nicotiana forgetiana*, Hort. Sand., and thereby hangs a tale. I found in the Gray Herbarium of Harvard University a sheet from near Los Angeles, California, marked *Nicotiana clevelandii* Gray.

This designation was manifestly incorrect, as the plant was exactly like SANDER'S figure of *N. forgetiana* in the *Botanical Magazine* (No. 8006). As it had been collected only a few years, I took seed from one of the capsules and planted it. It grew and again produced plants like *N. forgetiana.* Miss DAY, the librarian of the Gray Herbarium, then looked up the correspondence regarding the specimen and found that it was evidently a garden specimen grown by a Californian botanist, since deceased, from seed furnished by Sander & Sons and called *N. Sandarae.* The plants have not the mixed colors and the variability of the specimens now sold as *Sandarae* hybrids, but are constant in their characters and are identical with *Nicotiana forgetiana.* I have come to the conclusion, therefore, that I have obtained (as LOCK[2] probably did) seeds of the real *N. forgetiana* that had been mixed with the *Sandarae*[3] hybrids by Sander & Sons.

In view of the fact that *N. alata grandiflora* and *N. forgetiana* do not differ essentially in their foliage and habit of growth, but only in flower size and color—the one being white, the other red—perhaps one should not call them two species. I hold no brief either way. I simply accept the taxonomic ruling. At least, there existed here two strains very different from each other and very constant in their characters. Both were self-fertile, and in fact were usually self-pollinated naturally. They were crossed. There was no trouble about this, as every cross attempted was successful, and the capsules were filled with seeds.

This, then, seemed to be an excellent opportunity for studying size inheritance: two strains, uniform in pure lines, one with a corolla three times the length of the other, could be crossed easily. All was not plain sailing, however, for the plants of the F_1 generation (pl. VII) were absolutely self-sterile. This fact would have cut off the experiment in the flower of a promising youth but for the further fact that each plant was perfectly cross-fertile with every other plant. It did indeed reduce my interest in the inheritance of corolla size, for it precluded the study of an F_3 generation, but this was offset by the more fascinating problem of self-sterility.

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[2] Ann. Roy. Bot. Gard. Peradeniya 4:195–227. 1909.

[3] The *Sandarae* hybrids were supposed to have been produced by the cross *N. forgetiana*×*N. alata grandiflora.*

It was impossible to study the F_3 generation because crosses between two F_2 individuals alike somatically would be without meaning, since nothing could be known of the gametic potentiality of each. Crosses between F_1 individuals, on the other hand, meant something, because they were alike gametically. Six F_1 crosses were made therefore, and from them were grown 828 plants.

TABLE I

FREQUENCY DISTRIBUTIONS FOR LENGTH OF COROLLA IN A CROSS BETWEEN *Nicotiana forgetiana* AND *N. alata grandiflora*

Designation	Class centers in millimeters														
	20	25	30	35	40	45	50	55	60	65	70	75	80	85	90
N. forgetiana, 314	9	133	28	..	...	...	...	...	...	..	..	..	..	..	..
N. alata gr. 321	..	...	..	..	...	...	...	...	...	1	19	50	56	32	9
F_1 (314×321)	..	...	..	3	30	58	20	...	...	..	..	..	..	..	..
F_2 (314×321) 1–6	..	5	27	79	136	125	132	102	105	64	30	15	6	2	..

Table I shows the frequency distribution for length of corolla of *N. forgetiana* (314) inbred, *N. alata grandiflora* (321) inbred, the F_1 generation (314×321), and the F_2 generation (314×321) 1–6. The measurement was taken from the end of the pedicel to the center of the contraction commonly known as the corolla throat. The classes have a magnitude of 5 mm. and are centered at the even centimeters and half-centimeters. A glance at the distributions themselves is sufficient to show the small variability of the parent types and of the F_1 generation, and the great variability of the F_2 generation. The F_1 generation is strictly intermediate, as is the mean of the F_2 generation. *Among the individuals of the F_2 generation, however, are flowers identical with each parent.* This last fact is perhaps more clearly shown in the figures of pls. IX and X.

The statistical constants for each frequency distribution are shown in table II.

The spread of the corolla, measured to the tips of alternate lobes, behaved in the same way. Corolla breadth in *N. forgetiana* varied from 25–35 mm., with a sharp mode at 30 mm. The corolla spread of *N. alata grandiflora* was somewhat more variable in terms of the arithmetical standard, ranging from 55 mm. to 80 mm. The range of the F_1 individuals extended from 45 mm. to 55 mm.

In the F_2 generation plants were produced with a corolla breadth identical with each parent. In fact, there were four individuals as small as the smallest specimens of N. *forgetiana*, and there was one individual with flowers within 3 mm. of the size of those of the largest *N. alata grandiflora* growing in my cultures. The mean of this distribution was 48.57±0.19 mm., the standard deviation 8.07±0.13 mm., and the coefficient of variation 16.62±0.28 per cent.

TABLE II

STATISTICAL CONSTANTS FOR FREQUENCY DISTRIBUTION OF TABLE I

Designation	Mean	Standard deviation	Coefficient of variation
N. forgetiana, 314	25.6±0.12	2.27±0.08	8.86±0.33
N. alata gr., 321	78.8±0.28	5.38±0.20	6.82±0.25
F_1 (314×321)	44.3±0.23	3.67±0.17	8.28±0.38
F_2 (314×321) 1–6	49.9±0.26	11.26±0.19	22.57±0.39

Examination of the F_2 generation of this cross indicated a correlation between the length of the corollas and the lengths of the filaments and the styles that for all practical purposes was perfect. By this statement I mean that the anthers were invariably just above the stigmas and the stigmas were invariably at the throat of the corolla. Of course absolute measurements would not show an integral coefficient of correlation, nevertheless one may assume, I think, that the fact is simply obscured by slight fluctuations. It seems as if the numbers were adequate from which to conclude either that the determiner or determiners of corolla length are also determiners of the length of the style and the filament or that these factor complexes are perfectly coupled in inheritance.

Corolla spread is also correlated with corolla length. It is by no means uncommon to find a sharp break in the correlation, as is witnessed by the individual with the very broad corolla and comparatively short tube pictured in pl. X, fig. 8; but one never finds inverse extremes in the same individual. Just what the correlation coefficient 0.610±.015 would prove to mean if individual analysis of later generations were made, is doubtful. We cannot go back of the gross statement that such a correlation exists in the general population. Perfect coupling of certain factors together with independent combination of others may be possible; partial coupling arising from a peculiar gametic distribution may be equally possible.

TABLE III

CORRELATION BETWEEN LENGTH AND SPREAD OF COROLLA IN F_2 GENERATION OF CROSS BETWEEN *N. forgetiana* AND *N. alata grandiflora*

Breadth of corolla in millimeters

Length of corolla in millimeters	25	30	35	40	45	50	55	60	65	70	75	
25	1	...	4	...	...	...	...	...	...	...	...	5
30	2	5	4	10	3	3	...	...	...	...	...	27
35	1	5	17	26	21	6	3	...	...	...	...	79
40	...	6	12	38	41	31	7	1	...	...	...	136
45	...	...	8	20	29	31	26	8	2	1	...	125
50	...	...	3	22	30	44	25	7	1	...	...	132
55	...	...	1	2	22	27	37	9	4	...	...	102
60	...	...	1	3	11	32	29	19	9	1	...	105
65	...	...	...	...	6	17	21	12	7	1	...	64
70	...	...	...	1	1	5	15	4	4	...	...	30
75	...	...	...	...	...	7	3	4	...	1	...	15
80	...	...	...	...	...	1	1	3	...	...	1	6
85	...	...	...	...	...	1	...	...	1	...	...	2
	4	16	50	122	164	205	167	67	28	4	1	828

Coef. cor. 0.610±0.015

These are the principal facts collected regarding this cross, if the small leaf differences and other minor variations are left out of consideration. How unimportant the latter are, can be seen by a reference to pl. VIII. There are several suggestions that may be made regarding the simple facts obtained, however, that may be helpful in further Mendelian interpretations of size complexes.

Elsewhere[4] it has been shown that the behavior of such characters in crosses is adequately represented by the segregation and

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[4] Amer. Nat. **44**:65–82. 1910.

recombination of cumulative unit factors that do not show the phenomenon of dominance. The frequency distribution of the F_2 generation in these cases is not $(\frac{3}{4}+\frac{1}{4})^n$, as it is where dominance is complete, but is $(\frac{1}{2}+\frac{1}{2})^{2n}$, because a factor in the heterozygous condition is to be regarded as producing one-half the effect that it produces when in the homozygous condition.

Regarding this expression as proper for the moment, let us examine the F_2 frequency distribution for length of corolla with the idea of assigning a definite number for n in the expression $(\frac{1}{2}+\frac{1}{2})^{2n}$.

If n is made equal to 3, then by the theory the F_2 distribution should have seven classes with the frequencies

1–6–15–20–15–6–1

per 64 individuals. For 828 individuals, the grandparental sizes should each be recovered $(828 \div 64) = 13.0-$) nearly thirteen times. This was not the case in the actual distribution.

If n is made equal to 5, the F_2 distribution should have eleven classes with the frequencies

1–10–45–120–210–252–210–120–45–10–1

per 1024 individuals. With 828 individuals the grandparental classes should each be recovered only 0.8 times; in fact, a majority of populations of this size would not show the grandparental classes at all. This also is not the condition that was actually found.

There is left only the possibility of making n equal to 4. When this is done the F_2 distribution for 256 individuals—the smallest number in which a representative of each class may be found—and for 828 individuals is as follows:

1 – 8 – 28– 56– 70– 56– 28– 8– 1
3.2–25.9–90.6–181.1–226.4–181.1–90.6–25.9–3.2

This calculation points to the recovery of each grandparent about 3 times in the F_2 population under observation. Reference to table I shows that the figures actually obtained agree rather closely with this observation. But table I also shows another important fact. The arbitrary classes used had a range of 5 mm., which makes 13 classes necessary to express the F_2 generation.

This class size was adopted in accordance with the usual biometrical procedure, the variations in the small parent (314) being included in only 3 classes. But when this is done, the F_2 distribution is decidedly skew. The theoretical mode is along about the fourth or fifth class instead of the central class. What is the reason for the production of this type of curve? There must be a reason, and it seems to me that this reason must be biological and not a mathematical transnomination, as have been all the biometrical analyses of skew curves. The matter appears clear in the light of the following interpretation.

In ordinary statistical work, one produces a frequency distribution by throwing his tabular entries into arbitrary classes of equal size. By this procedure he has in all probability distorted their relationship. This fact is partially recognized by using the coefficient of variability instead of the standard deviation as a measure of variation. Unfortunately, it is usually said that the coefficient of variability is used instead of the standard deviation because it is an abstract measure and pounds can be compared with inches, etc. Standard deviations in the same concrete terms are usually thought comparable with each other. But is this true? Apply the rule to the data in tables I and II. The range of length of corolla of *N. forgetiana* (314) is 3 classes, the standard deviation is 2.27 ± 0.08 mm., and the coefficient of variability is 8.86 ± 0.33 per cent. The range of *N. alata grandiflora* (321) is 6 classes, its standard deviation is 5.38 ± 0.20 mm., and its coefficient of variability is 6.82 ± 0.25 per cent. Comparing standard deviations, *N. alata grandiflora* is twice as variable as *N. forgetiana*. Comparing coefficients of variability, which being functions of the mean give weight to the size of the mean, the large-flowered type (321) is less variable than the small-flowered type (314).

Let us now look at the matter from an ordinary common-sense biological standpoint. These pure line populations may be considered as composed of near-homozygous individuals. The range of variability shown is therefore almost wholly due to environment. In general, *N. alata grandiflora* has a corolla more than twice as long as *N. forgetiana*. Is it not reasonable to suppose that the unit change effected by environment and expressed as a fluctuation is proportional to the size of the individual? Is it not true that favorable circumstances which force the corollas of *N. forgetiana* to become 5 mm. longer than usual will produce a 10 mm. change in *N. alata grandiflora?*

If this is the correct way of looking at these two cases, then it is assuredly an error to plot the F_2 distribution—which includes both grandparental sizes—in classes of equal size. Assuming that our hypothetical size factors affect the individual as growth forces, it seems probable that they are not only cumulative but accelerative. Roughly one might imagine the effect on the individual to be something like a constant percentage. I do not believe these cases of size inheritance can be analyzed into their component factors and these factors given their proper weight (using the word factor in the general sense of elements or causes that produce a result) sufficiently well to give a precise value to the character determiners themselves. On the other hand, it is interesting to see just what is necessary in the way of class range to bring our F_2 corolla distribution to the normal distribution for four factors ($n=4$). Fortunately the corolla sizes were taken by millimeters, so this can be done. First I have smoothed the figures according to the regular method. The distribution in one-millimeter classes is then as follows (table IV, p. 186).

Suppose now we begin at 24 mm. and take for this class a range of 4 mm. Then let us increase our class range 1 mm. each time. This gives a simple arithmetical progression with an advancing difference of the second order, that is, the differences between the class ranges are constant. Compare the frequency distribution thus obtained with the expansion of $(\frac{1}{2}+\frac{1}{2})^{2n}$ where n is equal to 4. This is done in table V, with an agreement among the figures that is very remarkable. If I were a biometrician, I probably could show that this agreement could not be due to chance—since by chance it could only occur once in some hundreds of thousands of times—and must therefore have some great significance. I should prefer to believe that I happened by chance upon a series of class ranges that fitted the normal frequency theory. But it must be emphasized that it was a *constant* increase in class range that produced the normal curve from the distorted skew curve.

Perhaps no two actual frequency distributions would be alike in thus yielding to a simple arithmetical correction. Such a correction is probably fallacious in its simplicity. It serves our purpose, however, if it calls attention to the manifest error of expressing a wide range of biological variation by a frequency polygon of equal size classes.

TABLE IV

MEASUREMENTS OF LENGTH OF COROLLA IN F_2 POPULATION OF CROSS BETWEEN *Nicotiana forgetiana* AND *N. alata grandiflora*

Size mm.	freq.	Size mm.	freq.	Size mm.	freq.
24	0	45	25	66	12
25	2	46	28	67	10
26	2	47	29	68	7
27	2	48	28	69	7
28	1	49	28	70	6
29	3	50	26	71	6
30	5	51	28	72	5
31	8	52	23	73	6
32	10	53	20	74	4
33	11	54	20	75	3
34	15	55	19	76	2
35	13	56	22	77	2
36	18	57	22	78	0
37	18	58	22	79	1
38	19	59	26	80	2
39	26	60	21	81	2
40	28	61	22	82	1
41	33	62	15	83	0
42	24	63	15	84	1
43	23	64	14	85	0
44	24	65	12	86	1

TABLE V

COMPARISON BETWEEN THE THEORETICAL FREQUENCY DISTRIBUTION FOR FOUR FACTORS AND THE ACTUAL FREQUENCY DISTRIBUTION RESULTING WHEN CLASSES WITH A CERTAIN CONSTANTLY INCREASING RANGE ARE USED

Class limits	24–27	28–32	33–38	39–45	46–53	54–62	63–72	73–83	84–95
Class range......	4	5	6	7	8	9	10	11	12
Frequency.......	6	27	94	183	210	189	94	23	2
Calculated frequency for 828 individuals for $(\frac{1}{2}+\frac{1}{2})^8$........	3.2	25.9	90.6	181.1	264.4	181.1	90.6	25.9	3.2

Summary

Concluding, the following points may be again emphasized:

1. The inheritance of size complexes is so intricate that it is necessary to simplify an experiment upon them in every possible manner. The material used in this investigation, *Nicotiana forgetiana* Hort. Sand. and *N. alata grandiflora* Comes, lacks three of the complicating features that usually ensnarl such work. They are almost always naturally self-fertilized, and through numerous generations of self-fertilization have become automatically as homozygous in their characters as may be expected in plants that reproduce sexually. Their fecundity is so great that practically any quantity of F_2 individuals can be produced from a single F_1 plant. A plant character was investigated upon which the effect of environment is so small as to be negligible, namely corolla size.

2. These self-fertile species, which are perfectly fertile *inter se*, gave self-sterile progeny. This fact did not affect the production of an F_2 generation, as the F_1 plants from homozygous parents are alike in gametic constitution, and these were perfectly fertile *inter se*.

3. *N. forgetiana* with a mean corolla length of 25.6 mm. crossed with *N. alata grandiflora* with a mean corolla length of 78.8 mm. resulted in an intermediate F_1 generation with a mean variability of 44.3 mm.

4. The variability of the F_1 generation was very small, being about the same as that of the remarkably constant parental species. The F_2 generation, on the contrary, was very variable and both grandparental types were reproduced.

5. It is shown that the F_2 generation is what would be expected if the difference in corolla length shown by these two species were represented by the segregation and recombination of four cumulative but independent pairs of unit factors, dominance being absent.

6. The coincidence of theory and result is as great in this case as it is in qualitative characters of like complexity. If the Mendelian notation is useful to describe complex qualitative inheritance, it is similarly useful in describing the inheritance of quantitative characters.

7. Length of style and of filament are perfectly correlated with corolla length.

8. Breadth of corolla shows an average correlation with length of corolla equal to 61 per cent.

9. The frequency distribution of corolla length for the F_2 generation is positively skew. It is pointed out that the range of fluctuations of corolla length in the two pure species is twice as great in the one of larger size than in the other. Classes of equal size in frequency distributions of great variability appear to be arbitrary and improper, if size factors are assumed to be dynamic factors with fluctuations roughly expressed by the term growth force. To show this accelerative action, the class ranges must gradually increase as the size (that is, the number of factors) increases. It is shown that the distribution under discussion will be changed from skew to normal if a simple arithmetical increase in the size of the classes is made.

Bussey Institution
Boston, Mass.

EXPLANATION OF PLATES

PLATE VI

At the left, a young flowering plant of *Nicotiana alata* Link and Otto, var. *grandiflora* Comes; at the right, a young flowering plant of *N. forgetiana* Hort. Sand.

PLATE VII

A mature plant of the first hybrid generation of a cross between *N. forgetiana* and *N. alata grandiflora.*

PLATE VIII

Figs. 1, 2, and 3, upper, median, and lower leaves of a mature plant of *N. alata grandiflora;* figs. 4, 5, and 6, upper, median, and lower leaves of the first generation of a cross between *N. forgetiana* and *N. alata grandiflora;* figs. 7, 8, and 9, upper, median, and lower leaves of a mature plant of *N. forgetiana.*

PLATE IX

At the left, a flower of *N. alata grandiflora;* at the right, a flower of *N. forgetiana;* between them are extreme F_2 segregates in length and spread of corolla; taken on the same plate, three-fourths natural size.

PLATE X

Fig. 1, *N. alata grandiflora;* fig. 2, *N. forgetiana;* fig. 3, cross between *N. forgetiana* and *N. alata grandiflora,* F_1 generation; the remaining figures are F_2 segregates; all figures are three-fourths natural size.

PLATE VI

EAST on NICOTIANA

EAST on NICOTIANA

EAST on NICOTIANA

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EAST on NICOTIANA

STUDIES OF NATURAL AND ARTIFICIAL PARTHENOGENESIS IN THE GENUS NICOTIANA.

RICHARD WELLINGTON

NEW YORK
1913

[Reprinted without change of paging, from the AMERICAN NATURALIST, 1913.]

[Reprinted from THE AMERICAN NATURALIST, Vol. XLVII., May, 1913.]

STUDIES OF NATURAL AND ARTIFICIAL PARTHENOGENESIS IN THE GENUS NICOTIANA.[1]

RICHARD WELLINGTON

ASSOCIATE HORTICULTURIST, NEW YORK AGRICULTURAL EXPERIMENT STATION

PARTHENOGENESIS is a phenomenon that is known to exist in many widely separated genera of the higher plants. In but few cases does it seem likely that the regular reduction of gametogenesis with the subsequent nuclear fusion of fertilization never occurs, yet it is probable—from the frequent discovery of new examples—that it will ultimately be found that the ability to dispense with typical sexual reproduction is comparatively common. Should this prove to be the case, one would be forced to conclude that sexual reproduction was developed for reasons other than protoplasmic necessity, as Maupas and his followers would have biologists believe.

This is the fundamental problem toward the solution of which all data on parthenogenesis contribute, but pending the time when it can be discussed intelligently, there are sub-questions that are not without their interest. Loeb's researches have shown that the stimulus to development which is an attendant result of fertilization, is physico-chemical. Observations on several genera of parthenogenetic insects have shown that the presence or absence of sexual reproduction is largely dependent upon external conditions such as food, light, temperature, etc. Little is known of the rôle played by such stimuli in parthenogenesis in plants, however, although knowledge on the subject is of some import aside

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[1] Contribution from the Laboratory of Genetics, Bussey Institution of Harvard University.

from theory. For example, the geneticist is concerned, if, under any of the conditions likely to obtain in his experiments, plants ordinarily reproducing sexually should be incited to reproduce parthenogenetically.

This paper describes some facts on the subject obtained by experiments on the genus *Nicotiana*.

The Material.

The material used in the investigation was turned over to me by Professor E. M. East, who had received it from various parts of the world. Each species had been cultivated in pure lines for at least three generations, so that it may be considered to be fairly well known. The specific names used are those accepted by Comes in his "Monographie du genre Nicotiana," Naples, 1899. To his descriptions, and to such figures as are published in the *Botanical Magazine,* the plants corresponded perfectly. To all intents and purposes, therefore, the plants may be considered wild, although they have been under cultivation several years.

The writer desires to express his thanks to Professor East, under whose direction the investigation was carried out, for the use of the pedigreed material and for much valuable advice. Certain unpublished data obtained in his own researches on *Nicotiana* are incorporated with his consent.

Historical.

For historical purposes it is only necessary to give a brief review of Hans Winkler's paper, "Über Parthenogenesis und Apogamie im Pflanzenreiche," published in 1908; and the less comprehensive paper, "Parthénogénèse des Végétaux Supérieurs," of L. Blaringhem, published in 1909. Blaringhem in his historical account of this subject, states:

> Déja Camerarius dans sa lettre célèbre sur le sexe des plantes (*De sexu plantarum epistola,* 1694) reconnaît que dans ses essais de castration du Maïs il obtient, malgré l'absence de pollen, le développement de graines fertiles sur les épis latéreaux femelles.

Among the early observers of parthenogenetic[2] qualities in plants are given Spallanzani (1767–1779), Henschel (1817–1818), Lecoq (1827), Girou de Buzareinques (1827–1833), Ramisch (1833–1838), Bernhardi (1834–1839), Tenore (1854), Gasparini (1846) and Naudin (1856). As a few of the plants cited by these authors are at the present time the object of research, Blaringhem gives a list of the observed plants with an indication of the more doubtful.

The list is given as follows:

(*a*) Plantes Dioiques.
- *Bryonia dioïca* d'après Naudin (confirmé en 1904 par Bitter),
- *Cannabis sativa* d'après Camerarius, Spallanzani, Henschel, Girou de Buzareinques, Berhardi et Naudin,
- *Datisca cannabina* d'après Wenderoth et Fresenius (très douteux),
- *Lychnis dioïca* d'après Henschel et Girou de Buzareinques,
- *Mercurialis annua* d'après Lecoq, Ramisch, Naudin et Thuret,
- *Pistacia narbonensis* d'après Bocconi et Tenore,
- *Spinacia oleracea* d'après Spallanzani, Lecoq et Girou de Buzareinques.

(*b*) Plantes Monoiques.
- *Cucurbita Melopepo, C. Citrullus* et autres espèces d'après Spallanzani, Sageret et Henschel,
- *Ficus Carica* d'après Gasparini,
- *Urtica pilulifera* d'après Henschel (très douteux).

Winkler, in his introduction, cites *Cœlebogyne ilicifolia* J. Smith, a diœcius member of the Euphorbiaceæ native to eastern Australia, which had been cultivated since 1829 at Kew in three "weiblichen Stocken," as the first mentioned case of seed production without the assistance of pollen grains. This observation led Smith to believe

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[2] No doubt many of these observations were incorrect, owing to imperfect control.

that pollen is not essential for the perfecting of Euphorbiaceæ seed. In 1857 A. Braun described *Chara crinita* Wallr. as a true case of parthenogenesis. In 1877, Strasburger with the aid of modern technique found that the embryos in *Cælebogyne ilicifolia* were formed without fertilization, but that parthenogenesis was absent, as the embryos came not from unfertilized eggs, but from adventitious growths (Sprossungen) of the nucellus tissue. In 1900, Juel definitely proved its existence in *Antennaria,* thus establishing its presence in the higher plants.

As botanical investigators do not always agree in the use of the terms parthenogenesis and apogamy, Winkler divides all reproductive phenomena into three divisions, namely: Amphimixis, Pseudomixis, and Apomixis.

1. Amphimixis, which designates the normal sexual process.

2. Pseudomixis, which means the replacement of true sex-cell fusions by a false sexual process. Pseudomixis thus differs from amphimixis, essentially, only in the circumstance that the fusing cells are not differentiated as gametes. As an example of the pseudomictic (pseudomiktische) method of reproduction is cited *Lastrea pseudomas* var. *polydactyla* Wills, in which the sporophyte arises from a prothallium cell, its primordial nucleus fusing with a nucleus from a neighboring cell. Farmer and Digby (1907, p. 191) name this procedure "pseudoapogamy." All non-sexual nuclear or cell fusions must not be considered as pseudosexual, however, for there is an asexual cell fusion in addition to the sexual and the pseudosexual, as, for example, the nucleus fusion described by Nemec (1902, 1903) in chloralized roots of *Vicia,* and also the frequently mentioned nucleus fusion in the young ascus of the Ascomycetes.

3. Apomixis, which is the replacement of sexual reproduction by another, an asexual process, which is not bound up with nuclear fusions. For it, there is already another term, namely that of apogamy. This latter term was applied by de Bary (1878, p. 479) for the fact, "dass

einer Species (oder Varietät) die sexuelle Zeugung verloren geht und durch einen anderen Reproduktionsprocess ersetzt wird.'' The word apogamy used with the meaning intended by de Bary covers the term apomixis of Winkler; but as all the recent authors use the expression apogamy in a new sense, the introduction of a new term seems justifiable.

Apomixis is subdivided into vegetative propagation, apogamy, and parthenogenesis:

(*A*) Vegetative propagation consists of the replacement of fertilization by vegetative formations (Ausläuferbildungen), arising of leafy (blattbürtiger) shoots, vivipary and similar examples of simple vegetative division and the adventitious embryo formation from nucellus cells.

(*B*) Apogamy, the origin through apomixis of a sporophyte out of vegetative cells of the gametophyte, is subdivided into (*a*) somatic apogamy, if the cell or the cell complex which produces the sporophyte possesses the diploid chromosome number, and (*b*) generative apogamy, if the mother cells of the sporophyte carry only the haploid chromosome number.

(*C*) Parthenogenesis, the apomictic origin of a sporophyte from an egg, is subdivided into (*a*) somatic parthenogenesis, if the egg possesses a nucleus with the diploid or unreduced chromosome number, and (*b*) generative parthenogenesis, if the nucleus of the egg is provided with only the haploid number of chromosomes.

Winkler remarks, it is probable that the relations between somatic apogamy and apospory are very close, as the former without the latter is surely not thinkable, while the latter (the primary proceeding) may exist without somatic apogamy. Examples of somatic apogamy are given, but no certain cases of generative apogamy are known; nevertheless, Winkler is very certain that their existence is possible.

Somatic parthenogenesis can be obtained in two ways: first, it can combine with apospory, that is, a normal sporophyte cell with the diploid number of chromosomes can grow directly into the gametophyte; second, the gametophyte can arise from the spores in the usual manner, except that the reduction division is discontinued. Examples are known for both cases. After discussing the cell division of the more interesting cases of somatic parthenogenesis, he sums up the families in which it occurs, as follows:

1. Polypodiaceæ (*Athyrium Filix-fœmina* var. *clarissima* Bolton and var. *unco-glomeratum* Stansfield; *Scolopendrium vulgare* var. *crispum Drummondæ*).
2. Marsiliaceæ (*Marsilia Drummondii* R. Br.).
3. Ranunculaceæ (*Thalictrum purpurascens, Th. Fendleri*).
4. Rosaceæ (*Alchimilla* § *Eualchimilla*).
5. Thymelæaceæ (*Wikstroemia indica*).
6. Compositæ (*Antennaria alpina, A. fallax, A. neodioica; Taraxacum; Hieracium* § *Archieracium* and § *Pilosella*, almost completely).

According to Juel (1900, 1904), Murbeck (1901), Guérin (1904) and Strasburger (1904, 1907), somatic parthenogenesis is simply a vegetative process, the egg being merely an ovate-shaped body cell of the sporophyte. Winkler disagrees with this opinion, for if it be true, the female individuals of parthenogenetic plants could produce only female offspring. But this is not the case, for from parthenogenetic seed of *Thalictrum Fendleri,* Day obtained seeds which yielded abundantly staminate and pistillate plants. Thus, it is conclusively proven that cells are not always equivalent, even though they are physiologically and morphologically alike.

Two theoretical cases of generative parthenogenesis are given as thinkable; first, the whole cycle of development could occur without a change in the number of chromosomes, that is, the haploid number is retained throughout; second, a regenerative doubling of the chromosomes could appear in the development of an egg with the haploid number into the sporophyte. No examples of the latter are known to occur in the plant kingdom.

Merogany (Merogonie) is given a brief notice. This expression was first used by Delage (1899), for the successful fertilization of a denucleated fragment of an egg by a spermatozoon. It was established in animals by O. and R. Hertwig (1887) and Boveri (1889) and in the plant *Cystosira barbata* by Winkler (1901).

Parthenocarpy is more fully discussed, as it has much in common with both parthenogenesis and apogamy, and is a great source of danger in investigations made to determine their presence or absence. Noll (1902) introduced the term, and defined it as the capacity of many plants, under exclusion of pollen, to form fruits outwardly normal, but in which seeds are absent or aborted. This condition was discovered by the elder Gärtner (1788) who named it "*frutificatio spuria*" and was for the first time critically investigated by the younger Gärtner (1844), who called it "Fruchtungsvermögen." Winkler thinks that it might be possible to separate the cases of stimulative parthenocarpy, in which the seedless fruits are produced only after pollination with their own or foreign pollen or in consequence of an insect prick or some other irritation; and the cases of vegetative parthenocarpy, in which the seedless fruits are formed without any pollination or other outer irritation. The latter phenomenon is thought to occur less frequently than the former. Noll in 1902 described it in the cucumber (Gurke) and mentioned the then known cases, the fig and the seedless medlar. Ewert[3] has found that several kinds of fruit can develop without the assistance of pollen. The best results were obtained when all the blossoms of an individual plant were protected from fertilization, as otherwise the fertilized flowers were so markedly favored in their development when compared with the remaining unfertilized ones, that the latter dropped while immature.

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[3] Ewert (1909, 1911) has noted the presence of parthenocarpy in the apple, pear, grape and gooseberry, and Kirschner (1900) has noted the same in the quince.

The relation between parthenocarpy and parthenogenesis of higher plants is very close, as all the known cases of parthenogenesis are associated with parthenocarpy, for not only embryos and seeds, but fruits develop at the same time without fertilization. Since both fruits and seeds which appear perfectly normal will develop, although they are without embryos, one can not be positive about parthenogenesis unless the presence of the embryo is ascertained.

In the discussion on the causes of parthenogenesis and apogamy, Winkler suggests the possibility of physico-chemical changes operating in a flower in consequence of non-pollination, and causing the parthenogenetic development of the ovules. Also, similar changes might be induced by the entrance of the pollen tube, even though fertilization did not take place, as when parthenocarpic fruits appear. If mutations occur which can supply the proper conditions for these physico-chemical changes, then it is possible to explain the inheritance of the parthenogenetic character after it has once appeared. Physical changes in the cytoplasm surrounding the egg, as well as changes in the osmotic pressure, are considered as only theoretical explanations for parthenogenesis. If they should be a cause, Winkler asks, why should these changes occur in some flowers and not in others; and if they appear in all flowers, why should not parthenogenetic embryo formations occur in all?

One who is not acquainted with Winkler's and Blaringhem's papers should refer to the originals, as it is impossible to give all the subject matter proper treatment in a brief review. The complete bibliographies appended to these papers are also well worthy of reference.

TESTS FOR THE PRESENCE OF NATURAL PARTHENOGENESIS IN THE GENUS NICOTIANA

The writer obtained no viable seed in his numerous castration experiments with the exception of one doubtful case of *N. plumbaginifolia.* The seed of this exception was secured in a field experiment conducted on the heavy clay loam of western New York, used as a check on the experiments of Professor East made on the light sandy loam of eastern Massachusetts. Since the seed from this one capsule of *N. plumbaginifolia* was all that was obtained from ninety-eight emasculated blossoms of this species, it is reasonable to treat it as the result of an experimental error.

The method of testing for parthenogenesis in these field experiments consisted simply in emasculating and covering the flowers. Both paper sacks and cotton batting were used to protect the stigmas from self or cross-pollination. When the latter covering was used the anthers were removed with the assistance of a small wire hook which minimized the injury to the corolla and the cotton wad was then fastened over the end of the corolla tube with the aid of a rubber band. The supposed advantage of the cotton batting was that it would interfere less with the photosynthesis processes, than the paper sack, as it excludes much less air and sunlight. The seed of *N. plumbaginifolia* was obtained from a capsule covered with the cotton batting; otherwise, no definite results were noted in favor of either covering. As the heavy rains and strong winds will break off the capsules covered with cotton, it is advisable to enclose them with netting sacks.

The extent of these emasculation experiments of Professor East and myself in which not a single seed was produced outside of the capsule of *N. plumbaginifolia* already noted, is already seen by referring to Table I.

Mrs. R. H. Thomas was much more fortunate in her emasculation work, as she obtained fertile seed with no apparent difficulty. Why parthenogenetic tobacco seed should develop so readily in England and so rarely, if ever, in the eastern part of the United States is difficult to understand. The explanation may be found in the differences of the soils and the climatic conditions of the two places, but this assumption is improbable. It seems more likely that new buds which escaped notice were developed in the course of her experiments. This explanation of these divergent results is very probable, as adventitious buds appear for several weeks after the formation of the first buds. Both self-fertilized and parthenogenetic blossoms produce offspring true to the mother species; and consequently an error, if it did occur, could not be detected.

TABLE I

Field Castration Experiment

Species	Mass.			N. Y.		
	No. Buds	Treatment	No. Seed	No. Buds	Treatment	No. Seed
N. alata var. *grandiflora*	14	Emas. and covered	0		Emas. and covered	
N. attenuata				17	Emas. and covered	0
N. Bigelovii	12	Emas. and covered	0			
N. Forgetiana	20	Emas. and covered	0			
N. glutinosa	8	Emas. and covered	0			
N. Langsdorffii	12	Emas. and covered	0	54	Emas. and covered	0
N. Langsdorffii var. *grandiflora*	8	Emas. and covered	0	87	Emas. and covered	0
N. longiflora	16	Emas. and covered	0	45	Emas. and covered	0
N. paniculata	21	Emas. and covered	0			
N. plumbaginifolia	11	Emas. and covered	0	98	Emas. and covered	Several (1 capsule)
N. quadrivalvis	14	Emas. and covered	0			
N. rustica var. *brazilica*	14	Emas. and covered	0	16	Emas. and covered	0
N. rustica var. *humilis*	20	Emas. and covered	0	113	Emas. and covered	0
N. rustica var. *texana*	12	Emas. and covered	0			
N. suaveolens	10	Emas. and covered	0	13	Emas. and covered	0
N. tabacum (broadleaf)	10	Emas. and covered	0	11	Emas. and covered	0
N. tabacum (calyciflora)				83	Emas. and covered	0
N. tabacum (fasciated)	14	Emas. and covered	0			
N. tabacum (Havana)	28	Emas. and covered	0			
N. tabacum (Sumatra)	16	Emas. and covered	0	74	Emas. and covered	0
N. tabacum var. *fruticosa*				77	Emas. and covered	0
N. tabacum var. *macrophylla purpurea*				33		

Experiments on the Artificial Production of Apomictic Seed in the Genus Nicotiana

For the simplification of the following subject matter, the experimental procedures used in the attempted production of parthenogenetic seed have been divided into four classes, namely, the effects of foreign pollen, of mutilation, of fumigation, and of injections.

THE EFFECTS OF FOREIGN POLLEN

Gärtner (Burbidge, 1877), while making species crosses, obtained seed in a few cases which produced plants true to the maternal species and also true hybrids. Mrs. R. H. Thomas (1909) and Professor E. M. East have also observed the same phenomenon in their work. Professor East's results were as follows:

Seed was obtained which produced plants like the mother species and also true hybrids, from crosses *N. paniculata*[4] × *N. alata* var. *grandiflora, N. rustica* × *N. tabacum,* and *N. tabacum* × *N. Bigelovii;* seed which produced plants like the mother species and no true hybrids, from crosses *N. paniculata* × *N. Langsdorffii, N. paniculata* × *N. longiflora, N. paniculata* × *N. Forgetiana,* and *N. Bigelovii* × *N. sylvestris;* and seed which produced no true hybrids on one occasion but did produce true hybrids on other occasions, from cross *N. tabacum* var. *lancifolia* × *N. alata* var. *grandiflora.* These crosses gave per capsule from one to twenty-five good seeds that produced plants true to the mother parent, and many angular and undeveloped seed that produced very few hybrids. In the cases where no hybrids were produced, abortive seeds—probably hybrid in character—were present.

These seeds, true to the mother species, are thought by Professor East to be due to adventitious embryos arising from the tissue of the nucellus, for no case of seed formation after simple castration occurred in some hundreds of experiments, nor did seed giving maternal plants arise in any but wide species crosses giving sterile or nearly sterile progeny. If such be the case, parthenogenesis did not occur in these crosses.

Pollen grains of certain species in the plant kingdom are known to be capable of instigating the development of parthenocarpic fruits and of polyembryonic seed of foreign species, but whether they can cause the parthenogenetic development of ovules is still a question; even

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[4] The authorities for the specific names of the *Nicotiana* species used in these experiments are given on p. 23.

though varieties of *Vitis vinifera* have been noted by Millardet (1901) as giving only *Vitis vinifera* progeny, when pollinated by *Ampelopsis hederacea.* Examples of the parthenocarpic fruits, however, are common. The writer, while attempting to cross the tomato with the Jerusalem Cherry (*Solanum Pseudo-capsicum*) obtained parthenocarpic tomato fruits, but no fruit of any kind developed when the reciprocal cross was made. Parthenocarpic Seckel pear fruits were also produced by the application of Yellow Transparent apple pollen. In the crosses between *Nicotiana* species already mentioned, seed true to the mother parent was produced; but as in the case of the *Vitis vinifera,* there is no positive proof of a parthenogenetic development. What stimulatory effect is imparted by the pollen grain must be due either to an irritation caused by the entrance of the pollen tube or to the exudation of a "growth enzyme."

THE EFFECTS OF MUTILATION

The floral and axial organs of the plants were mutilated by emasculation, by the removal of the anthers, by decapitation, by the removal of both the stigmas and anthers, and by burning various portions of young buds, with the object of upsetting the normal functional processes in such a way as to incite the parthenogenetic development of seed. To simple emasculation and decapitation were added several modifications. Emasculated buds were covered with both paper bags and celluloid covers, but no advantages in favor of either covering could be detected. The decapitated buds were covered with the same two coverings, and in addition buds were left uncovered, but no differences in the results of these three methods were noted. Theoretically, the buds provided with the greatest amount of light and air should be favored in their development, but in this particular case, the results did not permit one to draw conclusions, since only negative results were obtained. Since capsules of *N. tabacum* were found to develop from pollinated flowers as well under the paper bags as under the celluloid covers, the latter covering was soon discarded. The advantages of the paper bags are, first, they cover a great many buds and, second, they are put on and removed very easily.

Clusters of buds that had been emasculated as well as those that had been decapitated were also ringed a few inches below the buds. The operation was performed with the hope that the food stored above the injury would upset the natural equilibrium of nutrition in such a way as to cause the development of the ovules. In these ringing experiments only negative results were obtained, although Ewert found that injuries to gooseberry branches favored the development of parthenocarpic fruits.

Neither the tickling of *N. tabacum* buds, varying in size from small to large, with a camel's hair brush every half hour for five consecutive hours, nor the cutting of the bases of *N. suaveolens* and *N. commutata* buds, with the point of a scalpel, gave results. Professor East has, however, produced a slight swelling in the capsules, but no seeds, by occasionally tickling the buds of the following species with a sharp-pointed instrument—*N. tabacum* (vars. fasciated, Sumatra, broadleaf, and Havana), *N. alata, N. Bigelovii, N. Forgetiana, N. glutinosa, N. Langsdorffii, N. Langsdorffii* var. *grandiflora, N. longiflora, N. paniculata, N. plumbaginifolia, N. quadrivalvis, N. rustica* (vars. *humilis, brazilica,* and *texana*).

Stimulation was also attempted, as already noted, by burning or rather singeing buds varying in development from very young to nearly mature, with a heated platinum wire. The hot wire was applied to various portions of the buds, namely, to the base, to the top of the ovary, the stigma, and to both the stigma and the ovary. When the pistils were not injured, the blossoms were covered with bags, but covering was not considered essential when the pistils were made functionless. *N. Langsdorffi* var. *grandiflora* and *N. plumbaginifolia* gave no results, but one capsule of *N. tabacum* produced fifty-six apparently normal seeds—none of which germinated after a period of several months' rest. The stage of maturity and the parts burned of each bud were not recorded and therefore the condition and exact treatment of this particular bud are unknown.

As a check on the uncovered decapitated pistils,[5] pollen from the same and other varieties was applied directly to the cut surface of the styles; in addition to pollen, cane-sugar solutions varying from 25 per cent. to 50 per cent. in strength,[6] stigmatic fluids, and in one instance nectar taken from the base of buds, were also applied. If the shortened pistils could be fertilized, it was thought that certain impossible crosses, as *N. alata* × *N. Forgetiana* and *Mirabilis Jalapa* × *M. longiflora* might be made, providing the difficulty existed in the extreme length of the styles. In one case, the applied stigmatic fluid and the pollen grains were taken from the same species. This precaution was used, as it was thought that the stigmatic fluid of one species might contain an enzyme or an inhibiting substance which would prevent the germination of foreign pollen grains. This supposition was supported by the growth of pollen grains in stigmatic fluids placed within Van Tieghem cells. For instance, the *N. glauca* pollen grains germinated and made good growth in the stigmatic fluid taken from *N. glauca* plants, while *N. suaveolens* pollen grains did not extend their pollen tubes in the stigmatic fluid taken from *N. Forgetiana.* If the tissue of the style contains an inhibiting agent, also, the germination of pollen grains on the cut style would be of no benefit. (This supposition may explain the negative results.[7])

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[5] Ewert (1909) quotes Gärtner who states that Henschel obtained seven ripe fertile seed from six blossoms of *Salvia sclarea* whose pistils had been destroyed, and four abortive seeds from three capsules of *Polemonium gracile* whose pistils had also been destroyed.

[6] A 33⅓ per cent. strength was used in the later work, as the pollen grains of *N. glauca*, *N. longiflora* and *N. tabacum* germinated readily and made good growth in this solution.

[7] The presence of one or more inhibiting agents might be used to explain the failure of grafts between plant species, for they may act like the antibodies, produced in animals by the transference of the blood of one species to that of another, and cause death.

Whether the pollen tubes in these experiments reached the ovules is not known, but probably not, since no fertile seed was produced. The production of numerous seed normal in appearance indicates, however, either that the pollen tubes must have stimulated the nucellus tissue in some way, or that normal seed development was started but not finished, for no seed of any kind was produced in the decapitated blossoms where pollen grains were not applied.

The total abortive seed produced by the pollination of the decapitated styles included two from *N. tabacum* where the stubs were covered with 50 per cent. cane sugar solution and self-pollinated, four from the same species where the stubs were covered with stigmatic fluid and self-pollinated, twenty-seven from *N. paniculata* where the stubs were covered with stigmatic fluid and self-pollinated, and fourteen from *N. tabacum* where *N. Forgetiana* pollen and no fluid was applied.

In connection with the decapitation experiments, an experiment on the grafting of pistils[8] was conducted. One hypothesis for the non-crossing of certain species, as has already been mentioned, is the extraordinary length of the style. By removing a portion of the style and grafting the stigma end of a pistil of either the same or another species to the stub, the style was shortened from one to one and a half inches. Immediately after grafting, the stigmas were pollinated. From one of the five grafted *N. tabacum* blossoms was produced one abortive seed. The development of this one seed may or may not have been due to the penetration of one or more pollen tubes, as in the cases where pollen grains were applied directly to the decapitated pistils.

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[8] The grafting technique is simple, nevertheless, the operation is difficult, owing to the small size of the styles. A light splinter was first attached to the base of the style by means of collodion, then the upper portion of the style was removed with a sharp knife. The end of the pistil to be grafted on the stub was cut off at the same angle and placed on the stub and made fast with the collodion.

MUTILATION EXPERIMENTS—FORCING HOUSE

TABLE II

Species	No. Clusters	No. Buds	Treatment	Stig. Fluid	Pollen	No. Seed	Remarks
N. alata var. *grandiflora*...	2		Decap.	Self	*N. Forgetiana*	0	
N. alata var. *grandiflora*...	1		Decap.	Self	*N. Sandaræ*	0	
N. alata var. *grandiflora*...	2		Decap.	Self	Self	0	
N. alata var. *grandiflora*...	2		Decap.	$33\frac{1}{3}$ per cent. sugar	*N. Forgetiana*	0	
N. alata var. *grandiflora*...	2		Decap.	$33\frac{1}{3}$ per cent. sugar	*N. Langsdorffii*	0	
N. alata var. *grandiflora*...	1		Decap.	$33\frac{1}{3}$ per cent. sugar	*N. Sandaræ*	0	
N. alata var. *grandiflora*...	1		Decap.	$33\frac{1}{3}$ per cent. sugar	Self	0	
N. Forgetiana............	1		Emas. and covered			0	
N. Forgetiana............	1		Decap.	*N. Langsdorffii*	*N. Langsdorffii*	0	
N. glauca...............	3	5	Emas. and covered			0	
N. glauca...............	2		Decap.			0	
N. glauca...............	1		Decap. and bagged			0	
N. glauca...............		3	Hot pt. wire and emas.			0	Singed stigmas.
N. Langsdorffii...........		5	Decap.	Self	*N. Forgetiana*	0	
N. Langsdorffii...........	1		Decap.	$33\frac{1}{3}$ per cent. sugar	*N. Forgetiana*	0	
N. Langsdorffii...........	2		Hot pt. wire and emas.			0	Singed base of buds.
N. Langsdorffii...........	1		Cut and emas.			0	Cut base of buds with scalpel point.
N. paniculata............	4	12	Emas. and bagged			1	Seed abortive.
N. paniculata............	5	5	Decap. and bagged			0	
N. paniculata............		3	Decap.	Self	Self	27	Seed abortive.
N. paniculata............	2		Decap.	25 per cent. sugar	*N. Tabacum*	0	
N. paniculata............	4		Hot pt. wire and emas.			0	Singed stigmas.
N. plumbaginifolia.......	2		Hot pt. wire and emas.			0	Singed base of buds.

TABLE II (*Continued*)

Species	No. Clusters	No. Buds	Treatment	Stig. Fluid	Pollen	No. Seed	Remarks
N. Sandarae	1		Decap.			0	
N. Sandarae	1		Decap.	Self	*N. Forgetiana*	0	
N. suaveolens		3	Decap.	Self	*N. Forgetiana*	0	
N. suaveolens	1		Cut and emas.			0	Cut base of buds with scalpel point.
N. tabacum	5	12	Decap. and covered			0	
N. tabacum	2		Decap. and bagged	0	*N. Forgetiana*	14	Seed contained in one locule.
N. tabacum	3	28	Decap. and bagged	0	Self	6	Three seeds in one capsule and one seed in other capsules.
N. tabacum	14	10	Emas. and covered			0	
N. tabacum	1	15	Emas. and ringed			0	
N. tabacum		8	Emas. and tickled			0	
N. tabacum	1		Decap. and grafted		Self	1	
N. tabacum	2	2	Decap. and ringed			0	
N. tabacum	1	1	Decap.	$33\frac{1}{3}$ per cent. sugar	*N. Forgetiana*	0	
N. tabacum	1	1	Decap.	50 per cent. sugar	*N. glauca*	0	
N. tabacum	1	2	Decap.	50 per cent. sugar	Self	2	Seed found in one capsule.
N. tabacum	1	1	Decap.	Nectar	Self	0	
N. tabacum		14	Hot pt. wire and emas.			56	Singed stigmas. Seed borne in one capsule.
N. tabacum	1	4	Hot pt. wire and emas.			0	Singed both stigmas and ovaries.

The mutilation experiments all proved to be valueless in the production of fertile seed; nevertheless, they were interesting, since they were the only methods, except where actual crosses were made and where chloroform gas was used, which caused any seed development.

(See Table II—Mutilation Experiments.)

Effects of Fumigation

Several species, the names of which are listed in Table III, were exposed before the plants had reached the flowering stage to gases given off by acetone, carbon tetrachloride, chloroform, ether, ethyl acetate, ethyl bromide, ethyl chloride, ethyl iodide, and formaldehyde. As in the previous experiments, the buds were emasculated and bagged. The object of this experiment, as of the one on mutilation of the plants, was to endeavor to upset the normal development of the floral organs in such a way as to cause the production of seed without the aid of fertilization.

The methods used in conducting this experiment were simple. Plants, growing in six-inch pots, were fumigated approximately one seventh of a cubic foot when drawn in at the top. The bags were closed either around the stems which had been previously surrounded with cotton batting or about the top of the pots, the method of treatment depending on the height of the plants. When everything was in readiness for fumigation, the gas was set free by the opening of the vial which was glued to the interior of the bag. Though the seams and the bottoms of the bags were sealed by melted paraffine, the retention of all the gas was not expected. A sufficient quantity was present, however, when acetone and formaldehyde were used, for the foliage of the plants, treated with these gases, to become noticeably injured.

TABLE III

FUMIGATION EXPERIMENTS—FORCING HOUSE

Species	Ht. Plant, In.	Liquid	C.c per Cu. Ft.	Hrs. Exposed	No. Seed	Remarks
N. alata var. *grandiflora*...	3	Ethyl acetate	8	24	0	Leaves drooped, three capsules developed.
N. alata var. *grandiflora*...	$1\frac{1}{2}$	Ethyl acetate	12	24	0	Leaves drooped.
N. Bigelovii............	4	Acetone	12	40	0	Several leaves injured.
N. Bigelovii............	$2\frac{1}{4}$	CCl_4	12	40	0	Slight injury.
Dewey's Sport No. 1......	5	Ethyl acetate	4	43	0	No injury.
Dewey's Sport No. 1......	$9\frac{3}{4}$	Ethyl acetate	4	72	0	No injury, six capsules developed.
Dewey's Sport No. 1......	$4\frac{1}{4}$	Ethyl acetate	6	43	0	No injury.
Dewey's Sport No. 1......	$9\frac{3}{4}$	Ethyl acetate	6	72	0	No injury, three capsules developed.
Dewey's Sport No. 1......	5	Chloroform	2	43	0	No injury.
Dewey's Sport No. 1......	$12\frac{1}{4}$	Chloroform	2	72	0	One capsule had six locules, six capsules developed.
Dewey's Sport No. 1......	4	Chloroform	4	43	0	No injury.
Dewey's Sport No. 1......	$9\frac{5}{8}$	Chloroform	4	72	0	No injury, one capsule developed.
Dewey's Sport No. 1......	6	Formaldehyde	4	43	0	
Dewey's Sport No. 1......	$13\frac{1}{2}$	Formaldehyde	4	72	0	Five capsules developed.
Dewey's Sport No. 1......	$3\frac{3}{4}$	CCl_4	4	43	0	No injury.
Dewey's Sport No. 1......	$9\frac{1}{8}$	CCl_4	4	72	0	No injury, four capsules developed.
Dewey's Sport No. 1......	$5\frac{1}{2}$	CCl_4	6	43	0	No injury.
Dewey's Sport No. 1......	$11\frac{1}{4}$	CCl_4	6	72	0	No injury, five capsules developed.
N. Forgetiana...........	$1\frac{1}{2}$	Formaldehyde	8	72	0	
N. longiflora............		Ethyl chloride	12	24	0	Four capsules developed.
N. longiflora............	Sessile	Ethyl iodide	12	24	0	
N. longiflora............	Sessile	Ethyl iodide	12	24	0	Two capsules developed.
N. longiflora............	$2\frac{3}{4}$	Ethyl acetate	12	24	0	Two capsules developed.
N. longiflora............	Sessile	Ethyl acetate	12	24	0	Four capsules developed.
N. paniculata...........	$4\frac{1}{2}$	Ethyl bromide	2	22	0	No injury.
N. paniculata...........	5	Ethyl bromide	4	22	0	No injury.
N. paniculata...........		Ethyl bromide	6	20	0	Cover a bell-jar—not a paper bag.
N. paniculata...........	$6\frac{3}{4}$	Ethyl bromide	8	72	0	No injury.
N. paniculata...........	$6\frac{1}{4}$	Ethyl bromide	12	72	0	No injury, seven capsules developed.
N. paniculata...........	6	Formaldehyde	4	20	0	
N. paniculata...........	7	Formaldehyde	2	20	0	
N. paniculata...........	$5\frac{1}{2}$	Acetone	2	22	0	Slight injury to foliage.
N. paniculata...........	7	Acetone	4	22	0	No injury to foliage.
N. paniculata...........	$10\frac{1}{2}$	Chloroform	1	22	2	No injury, one capsule developed.
N. paniculata...........	10	Chloroform	2	22	0	No injury.
N. paniculata...........	11	Ether	4	22	0	No injury.
N. plumbaginifolia........	Short	Ether	12	24	0	
N. plumbaginifolia........	Short	Ether	12	24	0	
N. plumbaginifolia........	$10\frac{1}{2}$	Ethyl iodide	8	24	0	
N. plumbaginifolia........	$14\frac{1}{2}$	Ethyl iodide	8	24	0	
N. plumbaginifolia........	$1\frac{1}{2}$	Ethyl bromide	12	40	0	One capsule developed.
N. quadrivalvis..........	$8\frac{1}{4}$	Ethyl bromide	12	24	0	No injury, one capsule developed.

TABLE III (*Continued*)

Species	Ht. Plant, In.	Liquid	C.c. per Cu. Ft.	Hrs. Exposed	No. Seed	Remarks
N. quadrivalvis	9½	Ethyl bromide	12	24	0	No injury, two capsules developed.
N. quadrivalvis	8	Ethyl bromide	8	24	0	No injury, two capsules developed.
N. quadrivalvis	10	Ethyl bromide	8	24	0	No injury.
N. quadrivalvis	7	Ethyl chloride	12	24	0	Three capsules developed.
N. quadrivalvis	6	Ethyl chloride	12	24	0	Three capsules developed.
N. quadrivalvis	9	CCl_4	4	22	0	No injury.
N. quadrivalvis	12	CCl_4	2	22	0	No injury.
N. rustica var. *texana*	6¼	Acetone	4	43	0	No apparent injury.
N. rustica var. *texana*	20⅛	Acetone	4	72	0	Terminal growth injured, four capsules developed.
N. rustica var. *texana*	9	Acetone	6	43	0	Terminal leaves, slightly injured.
N. rustica var. *texana*	21¼	Acetone	6	72	0	One leaf injured, four capsules developed.
N. rustica var. *texana*	5	Ethyl bromide	4	43	0	No injury.
N. rustica var. *texana*	17	Ethyl bromide	4	72	0	No injury, seven capsules developed.
N. rustica var. *texana*	5¾	Ethyl bromide	6	43	0	One leaf slightly injured.
N. rustica var. *texana*	17½	Ethyl bromide	6	72	0	One terminal blossom leafy, six capsules developed.
N. rustica var. *texana*	7	Ether	4	43	0	No injury.
N. rustica var. *texana*	15¾	Ether	4	72	0	Terminal bud injured and produced leafy blossom. Eight capsules developed.
N. rustica var. *texana*	6¾	Ether	6	43	0	No injury.
N. rustica var. *texana*	18	Ether	6	72	0	No injury, thirteen capsules developed.
N. Sandaræ		Chloroform	8	24	0	No injury, three capsules developed.
N. Sandaræ		Chloroform	12	24	0	No injury, one capsule developed.
N. sylvestris	Sessile	Chloroform	12	24	0	Leaves (outer) turned yellowish after two days.
N. sylvestris	Sessile	Chloroform	12	24	0	
N. sylvestris	Sessile	Ether	8	24	0	
N. sylvestris	Sessile	Ether	8	24	0	
N. sylvestris	Sessile	CCl_4	12	24	0	No injury.
N. sylvestris	Sessile	CCl_4	12	24	0	No injury.
N. tabacum var. *fruticosa*	11	Ether	8	72	0	No injury.
N. tabacum var. *fruticosa*	5½	Ether	12	72	0	No injury.
N. tabacum var. *fruticosa*	7¼	CCl_4	8	72	0	No injury.
N. tabacum var. *fruticosa*	5⅜	CCl_4	12	72	0	No injury.
N. trigonophylla	5	Ethyl bromide	12	40	0	
N. trigonophylla	1½	Acetone	12	40	0	Leaves injured.
N. trigonophylla	3½	CCl_4	12	40	0	No injury.

Untreated plants of all of the above species were held as checks on the results of each test. A *N. rustica* var. *texana* plant produced one leafy terminal blossom, otherwise, all the plants were normal.

A *N. paniculata* plant treated with chloroform gave two abortive seeds, but none of the other species produced a seed. *N. rustica* var. *texana,* however, after two exposures to acetone vapor underwent very marked morphological changes in the structure of the terminal blossoms, both of the main and of the lateral stems. In other words, the most exposed buds suffered the greatest injury. As the ether, ethyl bromide, and check treatments produced one blossom apiece which was similarly affected, and no other species, even though treated with acetone, was injured in the same way, indicates that the *N. rustica* var. *texana* floral parts are somewhat unstable. This opinion is substantiated by Penzig who in his *Pflanzen-Teratologie* cites observations where *N. rustica* blossoms have been modified to such a degree that the petals have turned green and where five blossoms have been compressed into a common calyx. Perhaps the presence of a small amount of chlorophyll in the greenish yellow corollas is an indication of a close relationship of the petalous to the leafy condition. Even though the *N. rustica* blossoms are easily modified, it is very evident that the acetone vapor caused a disturbance in the natural development of the floral organs, for the two treated plants were affected in the same way and degree. The calyxes, corollas and stamens were modified markedly, while the carpels and pistils and most of the stamens were usually normal in appearance. For instance, in some cases the calyxes were fused together and enlarged to such a size that they resembled distorted and crinkled leaves. One blossom had three sepals fused to the corolla and two sepals located one half inch below the base of the blossom. The lower two had a node as distinct as any leaf on the branch, and within their axis were borne two small buds, which lacked calyxes. One of the upper three sepals also bore a similar naked bud in its axis. It may be that in this case the acetone vapor stunted the branches in such a way that many latent buds were present in a very small space. The corollas in some cases were entirely replaced by small green leaves—smaller than the sepals—and in other cases they were partially replaced by leafy tissue. A few stamens had their filaments flattened and their anthers replaced by a small green leaf. These changes might be advanced, as an evidence of the evolutionary development of the floral organs, if the theory that these organs are simply modified leaves and that reversions are frequently caused by injuries were not already so well substantiated. Whether any mutations might have occurred in the progeny produced from these blossoms is unknown, as fertilization of the ovules was not attempted. No parthenogenetic seed was obtained from these injured blossoms, and this might have been expected, since leafy formations in the blossoms are generally accompanied by sterility.

Effects of Liquid Injections

The forcing of liquids into the plants was performed with the same object in view as in the preceding experiments, viz., to endeavor to stimulate cell division and thus possibly produce unfertilized seed. To certain liquids has been ascribed the power of being able to cause mutations when injected into the buds of certain plants, but in this experiment all the injections were made directly into stems of plants, eight to twenty inches in height.

The apparatus used was simple. Glass capillary tubes were connected by rubber tubing to glass tubes, about 30 inches long and about one quarter inch in diameter, which contained the liquids. The rubber tubing permitted the stems to lengthen without disturbing the operations. The end of the capillary tube was inserted from one eighth inch to one quarter inch into the stem, the distance depending upon the diameter of the stems and 1 inch to 15 inches below the terminal bud. An application of collodion held the capillary tube in place and stopped all leakage. After having supported the tube, the pinchcock—previously fastened to the rubber tubing—was released and the liquid flowed into the stem as rapidly as it could be used by the plant. The injection was assisted by the weight of its own column, and, in the most cases, by the addition of a short column of mercury, suspended by the surface tension of the liquid. The use of the mercury required considerable care, for when the surface tension was overcome by a jar, the mercury sank to the bottom and plugged the capillary tubes.

The treated species were: *N. tabacum* var. *fruticosa, N. paniculata, N. Langsdorffi* var. *grandiflora, N. Langsdorffii, N. alata* var. *grandiflora, N. attenuata,* and *N. Sandaræ.*

The materials used for the injections are: Sodium phosphate, butyric and valeric acids, ethyl acetate, acetone, benzol, chloroform, formaldehyde, methyl blue, saffranin and thiazin. The last three are simply stains and were used to trace the course of the liquids. The coloring matter was found to follow the vascular bundles of the stems and the leaves for several inches, and yet the slightest trace was not discovered in the buds. Acetone, butyric, and valeric acids of .5 per cent. strength caused severe injury, formaldyhyde at 2 per cent. caused a slight injury to the foliage, but no other liquid caused a noticeable disturbance.

All the treated plants, as in the previous experiments, had at least one cluster of buds emasculated and bagged, but all to no purpose, since not even one abortive seed developed.

SUMMARY

1. Seed giving plants true to the maternal species in the F_1 generation accompanied by aborted seed probably hybrid in nature, was found when certain *Nicotiana* species were cross-fertilized. Hybrid plants and plants purely maternal were obtained from the same capsules in other crosses.

2. The capsules of several *Nicotiana* species were

caused to swell slightly by merely tickling them with a sharp-pointed instrument, but no seeds were produced.

3. Abortive seed probably without embryos was produced by singeing young buds with a hot platinum wire, by the exposure of young plants to chloroform gas, and by cutting away a portion of the pistil and pollinating the stub both with and without the accompaniment of a germinative fluid.

4. Abortive seed was produced by shortening the pistils of a flower and grafting the stigma end of another pistil on to the stub and pollinating the same.

5. The ringing of the branches below a cluster of buds did not assist in the production of seed.

6. No seed was produced by the simple methods of emasculation and decapitation of blossoms, except in one doubtful case of *N. plumbaginifolia*.

7. It is likely that an agent inhibitory to the growth of pollen grains is present in the stigmatic fluids of certain species of the genus *Nicotiana;* at least, the pollen grains of *N. suaveolens* did not germinate in *N. Forgetiana* stigmatic fluid when placed within a Van Tieghem cell.

8. The exposure of young *N. rustica* var. *texana* plants to acetone gas caused the transformation of the corollas and the stamens of most of the terminal flowers into leafy tissue; otherwise, except in the mentioned case of the chloroform, no results were secured by the use of anaesthetic and toxic gases.

9. The injection of chemicals into the stems of tobacco plants was valueless in the production of seed.

10. As no unquestionable case of parthenogenetic seed was produced in the several hundred trials, it seems very improbable that parthenogenesis exists in the genus *Nicotiana*—at least in the species tested. The seed obtained in the crosses which came true to the mother species is probably polyembryonic—the stimulus of development being imparted either by the penetrating pollen tubes or by a substance exuded from the same.

Nicotiana Species Used in the Experiments

N. alata Lk. & Otto var. *grandiflora* Comes.
N. attenuata Torr.
N. Bigelovii Wats.
Dewey's Sport No. 1.
N. Forgetiana Sand.
N. glauca Grah.
N. glutinosa L.
N. Langsdorffii Weinm.
N. Langsdorffii Weinm. var. *grandiflora* Comes.
N. longiflora Cav.
N. paniculata L.
N. plumbaginifolia Viv.
N. quadrivalvis Pursh.
N. rustica L. var. *brazilica* Schrank.
N. rustica L. var. *humilis* Schrank.
N. rustica L. var. *texana* Comes.
N. Sandaræ Hort. (hybrid).
N. suaveolens Lehm.
N. sylvestris Speg & Comes.
N. tabacum (broadleaf).
N. tabacum (calyciflora).
N. tabacum (fasciated).
N. tabacum (Havana).
N. tabacum L. var. *fruticosa* Comes.
N. tabacum var. *lancifolia* (W.) Comes.
N. tabacum L. var. *macrophylla purpurea.*
N. tabacum L. (Sumatra).
N. trigonophylla Dun.

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THE CONNECTICUT

AGRICULTURAL EXPERIMENT STATION

NEW HAVEN, CONN.

BULLETIN 176, MAY, 1913.

TOBACCO BREEDING IN CONNECTICUT.

BY H. K. HAYES, E. M. EAST AND E. G. BEINHART.

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TOBACCO BREEDING IN CONNECTICUT.

By collaboration of H. K. Hayes, *Plant Breeder, Connecticut Agricultural Station,* E. M. East, *Bussey Institution, Harvard University,* and E. G. Beinhart, *Assistant, Office of Tobacco Investigations, Bureau of Plant Industry, U. S. Department of Agriculture.*

INTRODUCTION.

The investigations, with which this paper deals, were commenced in the year 1908, and since that time have been carried on in co-operative agreement between the Office of Tobacco Investigations of the Bureau of Plant Industry, United States Department of Agriculture, Laboratory of Genetics of Harvard University, and The Connecticut Agricultural Experiment Station.

The primary object of the work has been to study some of the fundamental principles involved in tobacco breeding, with the belief that a knowledge of these principles is absolutely necessary if one is to build up a system of both practical and scientific breeding.

It is self evident that the complex nature of the problems involved makes it impossible to reach anything like a final solution at present; this paper, therefore, is to be considered in the nature of a report of progress. In it are described the results obtained during the past four years.

EFFECTS OF INBREEDING IN A CLOSE-FERTILIZED SPECIES.

Tobacco is a naturally close pollinated plant, although intercrossing through the agency of insects is probably somewhat frequent. Observations on the earlier blossoms of the flower head have convinced the writers that in many cases, at least, fertilization of the pistil has taken place before the blossom opens. In the later flowers the chances of intercrossing are much greater, as the blossom often opens before fertilization has been accomplished. It is evident that, as tobacco is a naturally close-fertilized plant, it must be vigorous under self fertilization, but some data on actual controlled inbreeding are given to further substantiate this belief.

Darwin, in his classical experiments on inbreeding and crossbreeding, found some types which were very vigorous when continually self-fertilized.

Garner (1912) reports that a number of types have been inbred under bags for six or eight years by the United States Department of Agriculture without any observable change in vigor or growth habit. A certain strain of our present Connecticut Cuban shade type, now grown on one of our large plantations, was inbred for a period of five years (1903-1908) by saving seed from individual plants under a paper bag. Since that time seed has been saved from desirable plants under cloth tent, the chances, however, seeming very small that seed so produced will be cross-fertilized. Instead of showing a loss of vigor due to self-fertilization, this type seems more vigorous than in the early years of its introduction.

The Sumatra type, which has been used as one of our parent varieties, has been inbred for a period of seven years, without giving any evidence of accumulated evil effects of inbreeding.

In a large series of generic crosses of *Nicotiana* the writers have observed a wide range of variation as to increased vigor due to crossing. In some cases the first hybrid generation was very vigorous while other species crosses were non-vigorous.

In a previous paper (Hayes, 1912) on variety crosses within the species, five characters were measured in F_1 and were compared with the average of their parents for three sets of crosses. These characters were height of plant, length, breadth and size of leaf, and number of leaves per plant. All showed an increase over the average of the parents, except in the number of leaves per plant, which was almost exactly intermediate.

To quote from a previous paper (East and Hayes, 1912):

"We believe it to be established that:

"1. The decrease in vigor due to inbreeding naturally cross-fertilized species, and the increase in vigor due to crossing naturally self-fertilized species, are manifestations of the same phenomenon. This phenomenon is heterozygosis.* Crossing produces heterozygosis in all characters by which the parent plants differ. Inbreeding tends to produce homozygosis automatically.

"2. Inbreeding is not injurious in itself, but weak types, kept in existence in a cross-fertilized species through heterozygosis, may be isolated by its means. Weak types appear in self-fertilized species, but they must stand or fall by their own merits."

The matter has been mentioned here because of its bearing on the subject in hand. Houser (1911) has advocated the system of growing first generation hybrid tobacco as a commercial proposition. This was suggested for the heavy filler types

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*Owing to the rediscovery of Mendel's law of inheritance, we now know that many characters are separately inherited, and by the use of descriptive factorial formulas the breeding facts are made clear. If a certain character breeds true it is in a homozygous condition and each male or female reproductive cell is supposed to bear some substance or factor for the development of the character. If a cross is made between two races which differ in a certain character we know that of the two uniting reproductive cells, the one contains the factor for the contrasted character and the other does not. The resulting plants of this cross will not breed true in the next generation and they are said to be in a heterozygous condition for the character involved. The amount of heterozygosis produced by any cross depends on the number of gametic factorial differences of the parent plants.

of tobacco which are grown in Ohio. While it is doubtless true that by this method the yield could be somewhat increased, the yield factor, for cigar wrapper types at least, is only of secondary importance compared with quality. Because of the great importance of quality it seems much more reasonable to suppose that further advance can be made by the production of fixed types which in themselves contain desirable growth factors, such as size, shape, position, uniformity, venation, and number of leaves, together with that complex of conditions which goes to make up quality, than by any other method.

PREVIOUS WORK ON EFFECTS OF SELECTION.

It is a well-recognized fact that among both plants and animals no two individuals are exactly alike. This diversity is due to two main kinds of variation:

1. Fluctuating Variations, such as size, shape, and number of various plant organs, which are due to different conditions of fertility, or to better positions for development. Such variations are not inherited.

2. Inherited Variations, which may be either large or small, but are caused by some differences in the factors of inheritance and are entirely independent of their surrounding conditions for their transmission, although favorable environment is often needed for their full development.

The real basis of the Mendelian conception of heredity is a recognition of the fact that the appearance of a plant is not a correct criterion of that particular plant's possibilities of transmitting any particular quality, but that the breeding test is the only real means of determining the plant's hereditary value.

By the universal adoption of Vilmorin's "isolation principle," in which the average condition of a plant's progeny is used as the index of that particular plant's breeding capacity, breeders have recognized these classes of variation.

A practical example demonstrating the truth of this classification is the work of Dr. H. Nilsson and his associates at Svalöf, Sweden. In 1891 a large number of heads from autumn wheat varieties were collected and were separated into their respective botanical and morphological groups, about 200 groups in all being thus selected. In several cases certain forms were found which had no duplicates, and in these cases the individual form represented a group in itself. The following season each group was given a separate plot and careful records were made of the number of heads and plants which were the ancestors of each plot.

A careful study of the resulting harvest showed that, of all the cultures under observation, only those which originally came from a single plant produced a uniform progeny (Newman, 1912).

The theoretical interpretation of this class of results was given by Johannsen (1909) through his work with beans and barley. This investigator found that a commercial variety was in reality composed of different and distinct types which could be separated from each other by self-pollinating the individual plants and studying their progeny. For example, he investigated the character weight as applied to individual beans and found that progress could be made when larger beans were selected from the mixed commercial crop for several seasons. On the other hand, after types comparatively homozygous had been isolated by inbreeding, the same results were obtained in each isolated line when large beans were planted as when the smaller ones were used for seed — although fluctuation due to external conditions still continued. This he explained as due to the fact that environmental influences were not inherited but that a plant simply transmits its inherent germinal qualities.

Certain corroborative results which show that fluctuating variations are not inherited and that characters in a homozygous condition are reproduced in practically the same degree generation after generation have been obtained by Barber (1907) with yeasts; Pearl and Surface (1909) and Pearl (1912) with poultry; East (1910) with potatoes; Hanel (1907) with *Hydra:* Jennings (1908, 1910) with *Paramaecium;* Love (1910) with peas, and Shull (1911a) with maize.

It is true that Castle (1911, 1912 a. b.) reports experiments with a variable black and white coat color of the rat, in which he shows that selection progressively modifies a character which, in crossing with other types, behaves as a simple Mendelian unit. These results can be interpreted and, we believe, interpreted in a manner more helpful to practical breeding by assuming that although the coat pattern is transmitted as a single unit, its development is affected by several other unit characters independent of the general color pattern in their transmission. It may be that a few characters are so unstable that they may be modified by selection after reaching a homozygous condition, but so many thousand characters have been shown to Mendelize and to breed true in successive generations when in the homozygous state that for all practical purposes these laws may be assumed to be universal in sexual reproduction. Further reasons for this conclusion are given in the next few pages.

PREVIOUS WORK ON INHERITANCE OF SIZE CHARACTERS.

Since different degrees of expression of quantitative characters are inherited, as has been shown by Johannsen, and since within an inbred line homozygous for a character, change can seldom if ever be effected by selection, there seems good reason — as stated before — for believing that size characters are inherited in the same manner as qualitative or color characters.

The discovery of Nilsson-Ehle (1909) that certain hybrids are heterozygous for several inherited factors, either of which alone is capable of producing the character, laid the foundation for the proof of the generality of the Mendelian interpretation of inheritance in sexual reproduction.

It was from similar facts that East (1910a) made the first Mendelian interpretation of the inheritance of quantitative characters by assuming absence of dominance and a multiplicity of factors each inherited independently and capable of adding to the character, the heterozygous condition of any character being half the homozygous.

In the last few years a number of investigations have been made which show that linear or quantitative characters show segregation. Some of the investigations which show segregation in quantitative characters are as follows: Emerson (1910) for shapes and sizes in maize, beans and gourds; Shull (1910, 1911b) for row classes of maize and for *Bursa* characters; East (1911) and East and Hayes (1911) for height of plants, length of ears, weight of seeds, and row classes in maize; Tammes (1911) for certain characters of *Linum* forms; Tschermak

(1911, 1912) for time of flowering in peas and for weight of seeds; Hayes (1912) for height of plants, area of leaves, and leaf number of tobacco; Davis (1912) for *Oenothera* characters; Webber (1912) for plant characters of peppers; Belling (1912) for plant characters of beans; McLendon (1912) for cotton characters; Gilbert (1912) for characters of tomatoes; Heribert-Nilsson (1912) for *Oenothera* characters; Phillips (1912) for body size in ducks; Pearl (1912) for fecundity in fowls; and Emerson and East (1913) for other characters of maize.

A few investigations which also comprise the F_3 generation show that in some cases forms breed true giving no greater variability than the parent types. These results are of value in any system of breeding which, in a large measure, deals with size characters. Thus, by crossing two types which differ in quantitative characters we may expect to obtain a segregation in F_2 and in F_3, some forms breeding true for some characters and others again recombining the characters in which they are heterozygous.

The possibilities of obtaining pure forms in F_3 will, then, largely depend on the number of character differences of the parental types. A complete exposition of both theory and practice when dealing with quantitative characters is given in Research Bulletin No. 2 of the Nebraska Agricultural Experiment Station entitled "The Inheritance of Quantitative Characters in Maize" by collaboration of Emerson and East (1913).

PREVIOUS WORK ON TOBACCO BREEDING.

There are two factors which must be reckoned with in any system of breeding. These are heredity and environment.

Previous tobacco investigations have shown the great importance of environmental conditions for both quality and productivity. For example, Jenkins (1896) shows that on similar land there are large variations in quality and yield due to different systems of fertilization.

Selby and Houser (1912) have shown that the time of harvesting, after topping, has a great effect on both quality and yield.

It has been stated by Frear and Hibsham (1910) that the climate of Pennsylvania has a much greater effect on the character of tobacco produced than either hereditary varietal differences or soil.

It is a well-known fact that tobacco harvested by the priming method (picking individual leaves) has a different character than when harvested by cutting the whole stalk. These few illustrations, while in no way complete, indicate the great importance of the environmental factor in tobacco breeding.

One of the earliest experiments on inheritance of tobacco characters ever recorded was made by Naudin (Focke, 1881). This careful experimenter crossed one variety which had lanceolate leaves with a type which produced broadly oval leaves. The plants, resulting from this cross were alike in all essential features. In the second generation the differences were more marked and many individuals were found which resembled the parent types. Godron received two types of these hybrid forms from Naudin, the one with small leaves and the other with broad leaves. Both forms bred true in later generations.

Since the year 1900 many attempts have been made to improve the present types of tobacco by selection and crossbreeding. Shamel and his co-workers have done an important work by pointing out the value of selecting good type individuals for seed plants, and the production of inbred seed by bagging the seed head. Such methods have accomplished much by tending to produce uniform and better races.

In regard to the benefits which may be obtained from hybridization and subsequent selection, our knowledge is very meagre. On this subject Shamel and Cobey (1906) say:

"The best plan which can be followed in the case of crosses is to grow 100 plants of each cross and carefully note the characteristics of the hybrid plants. It will be found that there will be considerable variation in the plants the first season. Seed should be saved from those plants which are the most desirable and which show the greatest improvement over the native varieties. The next season a larger area can be planted from this seed; and if the crop is uniformly of the type desired, enough seed can then be selected the second season, to plant the entire crop the third year."

This quotation certainly shows a lack of belief in the uniformity of the first hybrid generation, and on the other hand, no conception of segregation in F_2.

Shamel (1910) also says:

"The writer believes that the two efficient means of inducing variability as a source of new types are change of environment and crossing. So far as the writer is concerned, the change of environment — usually the growing of southern grown seed in the north — is the most effective means of inducing variability."

Hasselbring (1912), however, gives experimental evidence from a number of pure lines of tobacco which he grew both in Cuba and in Michigan, and comes to the conclusion that there is no breaking up in type due to changes of environment, and that whatever changes take place affect all individuals of a strain in a similar manner.

Some observations of the writers on the appearance of several types grown in the Connecticut Valley from foreign seed serve to corroborate Hasselbring's conclusions.

These few citations from previous investigators show that there is no very definite knowledge of the manner of inheritance of tobacco characters, and the writers hope that the present paper may clear up some of the more important phases of this subject.

THE MATERIAL USED.

Four different types of commercial tobaccos furnished the starting point for these investigations. They consisted of two imported varieties tested for shade purposes, which prior to 1908, had been grown for a number of years in row selections from selfed seed, and the two standard Connecticut types — Broadleaf and Havana — which have been grown in Connecticut since the early history of the tobacco industry. The following descriptions give some of the more important features of these types.

No. 401 Broadleaf.

The Broadleaf variety produces long, pointed, drooping leaves, averaging in length a little over twice the breadth, with an average leaf area of about 9 sq. dcms. The number of leaves per plant ranges from 16 to 23 and averages from 19 to 20, The average height of plant is about 56 inches. This variety sells for slightly more per pound than the Havana, and when used as a wrapper or binder is generally considered to give a little better flavor to a cigar than the Havana type.

No. 402 Havana.

Havana produces medium length leaves, standing nearly erect though drooping slightly at the tip. The average length of the leaves is a little over twice the breadth. The number of leaves per plant ranges from 16 to 25 and averages from 19 to 20. The average height of the plant is about the same as the Broadleaf. This variety is well known as a wrapper and binder tobacco.

No. 403 Sumatra.

This variety produces short, round pointed, erect leaves, a little over half as broad as long, with an average leaf area of about 3 sq. dcms. The upper leaves of this type are generally narrow and pointed. The number of leaves ranges from 21 to 32 and averages from 26 to 28. The average height, when grown under shade, is about 6½ feet. This variety produces a larger percentage of wrappers than the Cuban type but the quality is very inferior, being of a light, papery texture.

No. 405 Cuban.

The leaf of this variety averages about the same width as the Havana, but is shorter and rounder. The position of the leaves is nearly erect. The leaf number ranges from 16 to 25 and averages about 20 per plant. The leaves are somewhat larger than those of Sumatra. This type is grown widely in the Connecticut Valley under shade covering, and produces wrapper tobacco of high quality.

THE METHODS USED.

As far as possible every precaution was taken to prevent experimental errors. With the exception of a very few cases the parental varieties have been grown from inbred seed, and if, for various reasons, other seed has been used, the fact is noted. Selfed seed has been obtained by covering the seed head with a Manila paper bag, and crosses have been made in the manner explained in previous papers (Hayes, 1912).

Much efficient aid has been given by Mr. C. D. Hubbell of The Connecticut Agricultural Experiment Station, who has materially helped in taking data, shelling and filing seed, and in the calculations. In the summer of 1912 Mr. A. F. Schulze, of the Connecticut Agricultural College, assisted in the field work.

We also wish to express our thanks to the Windsor Tobacco Growers' Corporation and its manager, Mr. J. B. Stewart, for so faithfully carrying out their part of the agreement by which means we were enabled to obtain the accurate data reported here.

As in previous work, each parental type has been given a number. A cross between No. 402 Havana and 403 Sumatra has been written (402×403), the female parent appearing first. Later generations have been designated (402×403)−1, (402×403)−1−1, and 403−1−2, which denote respectively the second and third generations of a cross between Havana and Sumatra, and the third parental generation of Sumatra.

The seedlings have been grown in sterilized soil. The sterilization of the beds has been accomplished by the use of steam at a pressure of at least 70 pounds, as explained by Hinson and Jenkins (1910). The actual sowing of the seed has always been done by one of the authors.

The different families and selections have been marked in the field by heavy stakes, to which wired tree labels were attached, and a planting plan has always been kept on file showing the exact location of the different selections. With this brief description of methods used, we will take up the consideration of the results obtained, and for convenience each family will be discussed separately.

FAMILY (402×403) HAVANA×SUMATRA.

A large number of crosses between tobacco varieties were made by Shamel in 1903, and among these was one between Havana as female and a small-leaved Sumatra type as male. Shamel (1905) states that the male parent, which was descended from Florida Sumatra seed, had been grown in Connecticut for two seasons and was partially acclimated. The Havana parent was a type which had been grown for a number of years by Mr. D. P. Cooley of Granby, Conn. The cross was grown at the Cooley farm in 1904 and 1905.

According to Shamel the first hybrid generation grew somewhat more vigorously than the parent types and was rather uniform in its habit of development. The second generation was thought to be no more variable than the first. Selected plants of this generation were grown at the farm of Edmund Halladay in Suffield in 1906.

It was the custom of the tobacco experts of The United States Department of Agriculture, who at this time conducted the work of tobacco breeding in Connecticut, to select desirable field types, harvest the leaves from each seed plant separately, and to base their judgment on the combined data from the growing plants and the cured leaves.

After examining the data on the F_3 generation collected in this manner, Mr. Halladay and Mr. J. B. Stewart concluded that one particular plant, bearing 26 short, round, pointed leaves with short internodes between them, gave great promise of becoming a desirable commercial type. Accordingly, Mr. Halladay added one row of plants from inbred seed of this individual to the two acres of experimental tobacco grown by him in pursuance of a co-operative agreement with the Department of Agriculture.

The plants in this row, numbered 2h-29 in accordance with the Department nomenclature, grew comparatively uniformly and several were inbred. In Mr. Halladay's absence, however, Mr. Shamel and an employee of Mr. Halladay's, in reducing the number of seed plants saved, topped all the plants except a late one, which was afterwards inbred.

In view of Mr. Halladay's high opinion of this type, the seed of this plant and that remaining from its parent were used for planting in 1908, each generation being given a separate number.

The field in 1908 presented a fairly uniform appearance and gave promise of producing a valuable wrapper tobacco. The new type was named "Halladay Havana," in honor of Mr. Halladay, who, in a large measure, was responsible for its production. It averaged about twenty-six leaves per plant and grew to about the height of Havana. The leaves were of medium length, averaging slightly shorter than Havana; they were fairly uniform in shape, with somewhat rounded tips.

The crop, when cured, lacked uniformity. Some leaves of exceptionally fine quality were produced, but the general fault of the crop was a lack of grain and too large a proportion of the heavy leaves known to the trade as "tops."

From this 1908 crop one hundred seed plants were saved, the leaves of each being carefully harvested, cured and fermented. Mr. J. B. Stewart and one of the writers made careful notes on the quality of these individuals, especial attention being paid to the feature known as "grain." The plants showed great variability; some of them had produced a fairly high grade of wrapper tobacco, others exhibited rather poor quality.

In 1909, seed from twelve of the best of these plants was used to continue our own experiments, but small amounts were also distributed to a number of Connecticut farmers. In addition, three acres were grown in Massachusetts. Some of these results were very promising. At the Arnold farm in Southwick, Mass., for example, a measured acre produced 3,000 pounds and brought the grower over $700. Other results were less favorable, but on the whole the experiment seemed worth repeating on a larger scale.

Accordingly, about 125 acres of Halladay Havana were grown in the Valley the following year and, while some men sold their crops at a good price, the results, in the main, were not encouraging. The chief faults mentioned by the buyers were lack of grain, too large proportion of dark and heavy leaves, and poor burn, although, in some cases, the burn was satisfactory.

This was the status of the work on the Havana × Sumatra cross when the data collected previously were turned over to the writers in 1908. Shamel, who had been in charge of the work up to this time, had come to the conclusion that the Halladay type was the result of a mutation. Apparently, he did not lend his approval to certain biological beliefs current at this time which indicated an alternative theory as an interpretation of its origin. For example, he believed that in general there was no greater variation in the second generation of a cross than in the first, and that considerable progress could be made by selecting good F_1 plants, some of which would breed true and give uniform progeny in F_2.

The writers did not take this view of the problem. It was contrary to all modern ideas of breeding to expect a cross between two self-fertilized varieties to be variable in F_1. High variability should occur in F_2, due to the recombination of Mendelian factors. New types should be produced in F_2 which could be reduced to an homozygous condition by selection and thereby fixed.

It was not impossible that the many-leaved type could have originated by mutation, but it appeared much more probable that it had been produced by recombination of parental characters. The type had the number of leaves and leaf shape of the Sumatra parent, combined with the habit of growth of Havana, and a close approach to the Havana leaf size. Other characters were in a somewhat intermediate condition; for example, the crinkling of the leaf was apparently a blend of the smooth Havana leaf with the much crumpled Sumatra leaf.

Family (403×402) Sumatra×Havana.

To test the hypothesis that the Halladay is a result of the recombination of parental characters and can be reproduced whenever desired, a cross was made in 1910 between Sumatra female and Havana male. The Sumatra was a direct descendant of the type used by Shamel in 1903 and had been grown from inbred seed for a number of generations. The Havana was the commercial variety grown at the Windsor Tobacco Growers' Corporation in Bloomfield. Although this variety of Havana was not exactly the same as that used by Shamel, it was the same in all essential features, the probability being very large that both types originally came from the same source.

The data on number of leaves per plant in this cross are given in Table I. The Sumatra and F_1 generation were grown at New Haven in 1911; the Havana was grown at Bloomfield from commercial seed of the same type as that used for the male parent of the cross. The F_1 generation was intermediate for leaf number and leaf size and was as uniform as the parental types. The variability of the F_2 generation for leaf number, size, shape and height of plants was very large. Some types were produced which could not be distinguished from pure Sumatra; others had Sumatra leaf characters and Havana leaf number; others resembled Havana in all features; and

TABLE I.

FREQUENCY DISTRIBUTION OF NUMBER OF LEAVES PER PLANT IN CROSS BETWEEN SUMATRA AND HAVANA.

No.	Leaf Classes.															Total.	A.	C. V.
	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31			
Havana	3	22	44	42	22	10	6	1	..	..	..	..	..	..	..	150	19.8 ±.08	6.98 ±.27
Sumatra	..	..	..	..	..	..	3	13	27	25	21	16	15	4	1	125	26.5 ±.11	6.64 ±.28
Sumatra × Havana, F_1	..	..	..	2	4	9	14	15	5	2	1	..	..	..	..	52	23.3 ±.14	6.24 ±.41
Sumatra × Havana, F_2	..	1	9	21	33	48	66	71	71	59	49	25	18	7	2	480	24.4 ±.08	10.29 ±.23

still others had the leaf size and growth habit of the Havana, combined with the leaf number of the Sumatra. These results, illustrated in Plates I-IV, give conclusive evidence that the Halladay type can be reproduced whenever desired.

Effects of Selection on the "Havana×Sumatra" Cross.

Let us now consider the effects of three years of selection on the Halladay strains of Shamel's cross. The purely genetic results of selecting for high and low leaf number are described in another paper. The work is considered briefly at this point, however, as the results have an important bearing on practical tobacco breeding. They show why the type lacked uniformity in 1908 and 1909, and hence the reason for its failure as a commercial proposition. Further, they go far toward indicating the proper procedure in obtaining results of economic value after hybridization.

In brief, the method pursued in this selection experiment was as follows:

Of the nine families with which the experiment was started (Table II), eight were grown at the Krohn Tobacco Company, in Bloomfield, in 1909, and the other (No. K) at a farm nearby. These nine families were selected from the 100 seed plants of Shamel's cross which were grown at the farm of Edmund Halladay, in Suffield, in 1908. From each of these families an inbred plant was saved which bore a high leaf number, and another with a low leaf number. These were made the basis of *plus* and *minus* selections, which were grown the following year, and from this time on seed plants with a high leaf number have been saved from the high or *plus* selection, and seed plants bearing a low leaf number from the low or *minus* selection.

These results, given in Table II, include the selection number, year grown, generation, number of leaves of parent, range of variation for leaf number, total plants, and biometrical constants, consisting of the mean for leaf number (A), and coefficient of variability (C. V.).

A consideration of these data shows that in one family, No. 27, no appreciable shift of the mean has been obtained, the mean of the low selection for 1912 being 25.9 ± .07, and that of the high selection being 25.0 ± .06.

TABLE II.

Results of Selection For High and Low Leaf Number in Cross Between Havana × Sumatra.

No.	Year Grown.	Generation.	Leaves of Parent.	Range of Variation.	Total.	A.	C. V.
(5 – 1) – 1	1911	F_8	24	23–32	239	26.5 ± .06	5.13 ± .16
(5 – 1)	1910	F_7	25	21–33	211	26.6 ± .09	7.18 ± .24
5	1909	F_6	28	23–33	294	28.1 ± .06	5.34 ± .15
(5–2)	1910	F_7	31	25–34	226	29.2 ± .08	5.75 ± .18
(5 – 2) – 1	1911	F_8	32	24–34	243	28.6 ± .08	6.05 ± .19
(5 – 2) – 1 – 3	1912	F_9	33	23–34	282	29.0 ± .07	6.34 ± .18
(6 – 1) – 2	1911	F_8	23	21–32	235	25.1 ± .07	6.05 ± .19
(6 – 1)	1910	F_7	25	21–33	181	25.2 ± .08	6.07 ± .22
6	1909	F_6	28	22–31	307	25.8 ± .06	6.16 ± .17
(6 – 2)	1910	F_7	30	25–36	205	30.7 ± .09	6.18 ± .21
(6 – 2) – 1	1911	F_8	32	26–34	239	29.1 ± .05	4.17 ± .13
(6 – 2) – 1 – 4	1912	F_9	32	18–36	243	30.5 ± .13	9.93 ± .30
(12 – 1) – 1	1912	F_8	29	24–35	296	29.1 ± .08	6.67 ± .19
(12 – 1) – 1	1911	F_8	29	24–34	210	28.7 ± .09	6.72 ± .22
(12 – 1)	1910	F_7	23	29–31	6	....	....
12	1909	F_6	24	21–28	113	24.5 ± .10	6.53 ± .29
(12 – 2)	1910	F_7	28	21–32	215	26.1 ± .10	8.12 ± .26
(12 – 2) – 1	1911	F_8	30	23–32	188	26.8 ± .07	5.61 ± .20
(12 – 2) – 1	1912	F_9	30	23–36	231	29.0 ± .08	6.52 ± .21
(27 – 1) – 1	1912	F_8	22	20–31	316	25.9 ± .07	6.76 ± .18
(27 – 1) – 1	1911	F_8	22	22–31	214	26.3 ± .06	5.01 ± .16
(27 – 1)	1910	F_7	25	21–31	254	26.5 ± .08	7.32 ± .22
27	1909	F_6	29	22–31	300	27.8 ± .06	5.04 ± .14
(27 – 2)	1910	F_7	28	21–29	222	25.2 ± .07	6.47 ± .21
(27 – 2) – 1	1911	F_8	28	21–29	222	26.0 ± .07	5.78 ± .19
(27 – 2) – 1	1912	F_8	28	21–32	305	25.0 ± .06	6.68 ± .18

TABLE II — CONTINUED.

No.	Year Grown.	Generation.	Leaves of Parent.	Range of Variation.	Total.	A.	C. V.
(41 – 1) – 2	1912	F_8	25	22–32	288	27.4 ± .07	6.42 ± .18
(41 – 1) – 2	1911	F_8	25	24–32	225	28.1 ± .07	5.85 ± .19
(41 – 1)	1910	F_7	24	21–30	224	26.3 ± .08	6.65 ± .21
41	1909	F_6	26	19–29	296	23.9 ± .07	7.25 ± .20
*(41 – 2)	1910	F_7	28	12–30	234	25.7 ± .09	7.47 ± .23
†(41 – 2) – 1	1911	F_8	30	18–32	142	25.6 ± .14	9.71 ± .39
(41 – 2) – 3	1911	F_8	12	8–30	126	19.8 ± .28	23.50 ± .11
(41 – 2) – 1	1912	F_8	30	20–33	310	26.9 ± .08	7.70 ± .21
(41 – 2) – 3 – 2	1912	F_9	10	11–27	278	17.8 ± .08	11.24 ± .33
(73 – 1) – 2 – 1	1912	F_8	24	21–30	221	25.7 ± .08	7.12 ± .23
(73 – 1) – 2	1911	F_7	23	21–30	217	25.6 ± .07	5.71 ± .19
(73 – 1)	1910	F_6	25	20–33	216	26.3 ± .09	7.64 ± .25
73	1909	F_5	28	23–32	300	26.9 ± .06	5.39 ± .15
(73 – 2)	1910	F_6	29	19–30	201	26.3 ± .08	6.58 ± .22
(73 – 2) – 3	1911	F_7	31	24–34	195	28.2 ± .09	6.37 ± .22
(73 – 2) – 3 – 3	1912	F_8	32	22–30	76	26.7 ± .13	6.18 ± .34
(76 – 1) – 1	1911	F_7	23	20–28	310	23.9 ± .05	5.28 ± .14
(76 – 1)	1910	F_6	22	20–27	31	25.2 ± .19	6.07 ± .52
76	1909	F_5	23	20–28	89	24.1 ± .11	6.64 ± .34
(76 – 2)	1910	F_6	27	21–30	209	24.4 ± .07	6.27 ± .21
(76 – 2) – 1	1911	F_7	28	22–32	230	26.1 ± .08	6.88 ± .22
(76 – 2) – 1 – 1	1912	F_8	29	22–32	275	26.9 ± .07	6.28 ± .18
(77 – 1) – 1 – 2	1912	F_8	20	14–25	236	18.4 ± .07	8.37 ± .26
(77 – 1) – 1	1911	F_7	20	18–26	306	21.3 ± .05	6.60 ± .18
(77 – 1)	1910	F_6	20	17–28	266	21.9 ± .08	9.27 ± .27

* One 12-leaved plant appeared not used in computations.

† End row poorly fertilized.

TABLE II — CONTINUED.

No.	Year Grown.	Generation.	Leaves of Parent.	Range of Variation.	Total.	A.	C. V.
77	1909	F_5	23	20–27	85	22.4 ± .11	6.38 ± .33
(77 – 2)	1910	F_6	27	20–32	224	24.9 ± .11	9.32 ± .30
(77 – 2) – 1	1911	F_7	30	22–33	281	26.6 ± .07	6.92 ± .20
(77 – 2) – 1	1912	F_7	30	21–33	280	25.8 ± .08	7.48 ± .21
(K – 1) – 1 – 2	1912	F_8	24	17–28	209	21.5 ± .08	7.77 ± .26
(K – 1) – 1	1911	F_7	22	20–29	202	23.5 ± .07	6.52 ± .22
(K – 1)	1910	F_6	. .	21–29	148	24.4 ± .09	6.93 ± .27
K	1909	F_5	26	20–24	31	22.3 ± .13	4.62 ± .40
(K – 2)	1910	F_6	. .	19–29	130	22.8 ± .11	7.98 ± .33
(K – 2) – 1	1911	F_7	26	17–29	204	24.0 ± .07	6.03 ± .20
(K – 2) – 1 – 6	1912	F_8	26	18–30	223	22.8 ± .07	7.15 ± .23

All other *plus* selections except (73 − 2) − 3 − 3 and (K − 2) − 1 − 6 have given a change toward the high leaf condition. These selections gave about the same average leaf number as in 1909. In some strains the mean has been gradually shifted, as in the *plus* selection of family 76, which gave progressive changes from a mean leaf value in F_5 of 24.1 ± .11 to 24.4 ± .07 in F_6, then to 26.1 ± .08 in F_7, and finally in F_8 to 26.9 ± .07. Other families, as Nos. 5 and 6, gave a large change in mean due to the first year of selection but in later generations have given no further changes due to continued selection. In general, the results have been what one would expect if selection simply isolated homozygous types from a heterozygous population.

Selection for low leaf number has caused decreases in (5 − 1) − 1, (K − 1) − 1 − 2 and (77 − 1) − 1 − 2, and slight decreases in (6 − 1) − 2, (73 − 1) − 2 − 1 and (76 − 1) − 1, but of such a small nature that little dependence can be placed upon them. A negative effect is shown in case (41 − 1) − 2.

In previous papers we have shown that the number of leaves per plant is a very stable character and, as such, little affected by environment. That selection has made various degrees of change in the mean of some types and no change in others, we believe to be due to the fact that some selections, as for example No. 27, were in a pure or nearly homozygous condition in 1909, while others were heterozygous for different numbers of factors for leaf number.

General field notes on the Halladay types, which were grown in 1912, are given in Table III. Three different observations on these types were made: general vigor, shape of leaf, and leaf character, whether smooth or crinkled. Of the fourteen selections given in this table, three were classed as very vigorous, seven as having good vigor, three as of fair vigor, and one as non-vigorous. As to shape, eleven have broad round tipped leaves, one has broad leaves with a pointed tip, and two from family No. 77 have leaves which resemble the Havana in shape. Considering fullness between the veins, one selection has very crinkled leaves, eight have crinkled leaves, two have slightly crinkled leaves, and three are classed as smooth-leaved types.

It is of interest to know that the leaf character and also the length of internodes of No. 77 closely approach the type of the Havana parent.

TABLE III.

GENERAL CHARACTERISTICS OF SELECTED HALLADAY STRAINS GROWN AT BLOOMFIELD IN 1912.

No.	General Vigor	Shape of Leaf	Type of Leaf
5−2−1−3	Very good	Broad, round tip	Slightly crinkled
6−2−1−4	Good	“ pointed“	“ “
12−1−1	Very good	“ round “	Smooth
12−2−1	“ “	“ “ “	Very crinkled
27−1−1	Good	“ “ “	Crinkled
27−2−1	“	“ “ “	“
41−1−2	Fair	“ “ “	“
41−2−1	“	“ “ “	“
73−2−3−3	Good	“ “ “	“
76−2−1−1	“	“ “ “	“
77−1−1−2	Fair	Fairly broad, pointed tip	Smooth
77−2−1	Good	“ “ “ “	“
K−1−1−2	Poor	Broad, round tip	Crinkled
K−2−1−6	Good	“ “ “	“

Some data obtained on comparative leaf length of these Halladay types are given in Table IV. This table gives the average number of leaves per plant, by actual count, the total yield of cured tobacco on an acre basis, and the number of pounds of tobacco in each leaf length class. This, of course, does not give the number of leaves of each length, as it naturally takes more 12-inch leaves than 20-inch leaves to weigh a pound. However, a general idea of the average length of leaves of a selection can be obtained by this means.

This table shows that leaf length is not very closely correlated with number of leaves per plant. For example, selection (73−2)−3−3, which averaged 26.7 leaves per plant, produced only 256 pounds of 18-inch tobacco, while selection (12−1)−1, which averaged 29.1 leaves per plant, produced 1,162 pounds of 18-inch tobacco. (K−1)−1−2, which averaged 21.5 leave , produced only 113 pounds of 20-inch length, while (K−2)−1−6, which originally, in 1908, came from the same plant as (K−1)−1−2, and which averaged 22.8 leaves per plant, gave a production of 944 pounds of 20-inch length.

TABLE IV.

COMPARATIVE LENGTH OF LEAVES OF THE HALLADAY STRAINS IN 1912.

No.	Average No. of Leaves per Plant	Yield per Acre	Yield in Pounds for Leaf Length Classes in Inches							
			12	13	14	15	16	17	18	20
5−2−1−3	29.0	2813	87	126	199	349	538	709	730	75
6−2−1−4	30.5	2822	82	109	181	245	402	588	872	343
12−1−1	29.1	3370	38	131	142	254	363	812	1162	467
12−2−1	29.0	3085	143	152	253	423	629	755	677	52
27−1−1	25.9	2766	86	138	146	300	483	628	833	150
27−2−1	25.0	2736	95	65	93	180	356	495	909	543
41−1−2	27.4	2196	72	93	125	175	271	430	800	234
41−2−1	26.9	2694	101	112	160	220	351	523	971	257
41−2−3−2	17.8	1936	115	122	199	263	323	355	462	97
73−2−3−3	26.7	2645	229	229	379	512	617	423	256	...
76−2−1−1	26.9	2721	126	119	204	356	566	672	634	44
77−1−1−2	18.4	2271	35	40	97	137	185	316	638	823
77−2−1	25.8	2341	64	47	88	138	219	359	942	483
K−1−1−2	21.5	2332	84	65	142	239	343	524	822	113
K−2−1−6	22.8	2740	62	49	86	216	210	392	781	944
Havana	*20.0	2119	56	57	86	128	191	284	570	747

*Estimated.

The largest amount of tobacco by weight was produced in the 18-inch class by ten of the selections, in the 17-inch class by two, in the 16-inch class by one, and in the 20-inch class by two selections. The Havana grown for comparison also produced the greatest amount of tobacco in the 20-inch class.

Quality of Cured Leaves.

The data already submitted have shown that by 1912 several types markedly different in leaf number have been produced. Though it is less easy to demonstrate by concrete figures, these types also differ in vigor, shape of leaf, plant height, etc. This fact is of practical importance and gives conclusive evidence for believing that the Halladay type, as grown commercially in 1908-1910, was not the uniform type which it was, in general, considered to be. May not these facts explain the reason for the commercial failure of the Halladay by showing that the type, as a whole, was in a heterozygous condition and, therefore, could not give tobacco uniform in quality. That some growers were favorably impressed and others less so may then be entirely due to the fact that some grew favorable types, and others types which, from a commercial standpoint, were very inferior. It was for this reason, justifiable from the commercial point of view, that the culture of the Halladay was dropped.

From 1909 to 1911 inclusive, no data were taken on the cured leaf of the Halladay, as our sole aim was to study the effects of selection on the field habit. In 1912, however, the tobacco was harvested, cured, fermented, and assorted, to determine if certain selections had come to be better than the others and if any gave promise of commercial value. Because the season of 1912 was a dry one and not very favorable for tobacco, the crop, as a whole, was of inferior quality. A small plot of commercial Havana of the same type as that grown by the Windsor Tobacco Growers' Corporation was grown on the same field, however, and was cured, fermented, and assorted in the same manner as the experimental tobacco. By this method we were able to obtain some idea of the comparative value of our selections, using Havana as the standard.

However, it should be noted that on account of practical difficulties the time of harvesting the various pickings was not always at the proper degree of ripeness. For example, the first and third pickings should probably have been made a few days earlier, but for unavoidable reasons this was impossible. Further, some selections were a few days earlier in maturity than others, and as all selections were harvested on the same day, some may have received more favorable treatment. This was partly corrected by making a larger picking, that is, by taking more leaves from the very mature types at an early picking than were taken from the later maturing types at the same picking.

The method of harvesting tobacco by the "priming" method is well known (see Stewart, 1908) and will be mentioned only briefly here. Four pickings were made of our experimental tobacco, as follows: About 5 leaves were harvested at the first picking, 5 to 8 at the second picking, 7 to 12 at the third picking, and all remaining leaves of commercial size at the last picking. The leaves of each picking were then tagged with the selection number and carried to the barn, where they were strung and hung on laths, from 36 to 40 leaves to the lath, with a tag containing the selection number attached to each lath.

The curing season was somewhat wet and at two different times it was necessary to dry out the tobacco by firing, which was accomplished by building charcoal fires in small stoves.

After the tobacco was cured it was taken down when in "kase," that is, when just damp enough to be pressed in the hands without breaking the leaves. The leaves from each lath, with tag attached, were tied into hands, and the tobacco then placed in a "bulk" to go through a period of fermentation. The experimental tobacco was not fermented sufficiently for commercial use, but the fermentation tended to even up the colors so that the tobacco could be assorted with better judgment.

After the tobacco had remained in the bulk for about four weeks it was removed and all of each selection placed together, the different pickings being kept separate. Four hands of the first three pickings of the different selections were drawn at random and were examined for quality by three tobacco judges. The same hands were carefully examined by the writers for "grain" and "texture."

The total crop of tobacco was then sized by the usual method. This consists in separating the leaves into different lengths, from 12 to 20-inch classes being made. This work was done by girls under our supervision.

After the tobacco was sized it was assorted into grades as in commercial practice. The actual work of assorting was done by experienced sorters, and the different lengths and grades were weighed in pounds and ounces.

"Grain" in Tobacco Leaves.

The presence of small pimple-like projections scattered over the cured leaf of tobacco is called "grain." It is a well-known fact that all tobacco does not exhibit this tendency in the same degree. In some cases the grain is large and easily seen, and in other cases small and scarcely visible to the naked eye.

One of the tobacco experts who kindly examined our Halladay selections made the criticism that the "grain" was over-developed, and another expert expressed the opinion that the selections, as a whole, were lacking in grain. This fact is mentioned to show that the ideals of some of the best growers differ on this matter. Both men desired grain in the leaves, but one preferred large pimply grains, easily seen, and the other a fine grain, scarcely distinguishable.

Sturgis (1899) found by microscopical examination that the grain of tobacco leaves was due to a crystalline deposit of some material, the compound being, in his opinion, calcium oxalate. Contrary to expectations, he found no increased deposit due to heavy liming of the soil but he did find that the thinner leaves which were produced under shade apparently contained it in smaller amounts.

If grain is calcium oxalate and as such of no value for burning qualities, it is very probable that it does not deserve the importance that it generally receives, although, as Connecticut growers generally consider the presence of grain to be an indication of quality and as tobacco buyers as a rule make it a factor in their judgment of the crop, it becomes necessary to consider its production. From the writers' standpoint a fine-grained wrapper leaf presents a more handsome appearance than leaf with larger grains, although the final test of any quality depends upon the demand of the consumer.

As has already been mentioned, some of the parent plants of our 1909 selections were examined for grain because it was believed that the Halladay Havana, as a whole, lacked in this particular. We have therefore considered this character in our experimental work in 1912.

Before the tobacco was sized and after fermentation had taken place, four hands containing approximately forty leaves each were drawn at random from the first three pickings of each selection and were examined for grain. The method followed was an arbitrary one. Seven general classes were made; those leaves which had a maximum amount of grain were placed in Class 1, and those in which no grain could be distinguished were placed in Class 7. Obviously the remaining classes ranged in value from maximum to minimum grain production. The results are given in Table V.

TABLE V.

VARIATION IN GRAIN OF HALLADAY STRAINS IN 1912.

No.	Leaves per Plant	Picking	Grain Classes							Mean Class
			1	2	3	4	5	6	7	
5-2-1-3 From good grained plant in 1908	30.5	1	15	20	45	46	19	..	..	3.23
		2	2	7	27	49	22	4	..	3.85
		3	1	4	15	26	67	37	..	4.77
		Total	18	31	87	121	108	41	..	3.97
6-2-1-4 From fair grained plant in 1908	29.1	1	16	28	52	27	23	..	..	3.09
		2	6	25	27	68	22	..	..	3.50
		3	2	6	21	46	54	21	..	4.38
		Total	24	59	100	141	99	21	..	3.66
12-1-1	29.1	1	2	5	24	59	40	16	..	4.21
		2	5	18	31	54	37	10	..	3.84
		3	..	2	7	30	63	52	2	5.04
		Total	7	25	62	143	140	78	2	4.37
12-2-1	29.0	1	13	24	41	33	18	2	..	3.19
		2	7	22	46	56	16	..	..	3.35
		3	1	11	25	13	63	27	..	4.48
		Total	21	57	112	102	97	29	..	3.68
27-1-1 From fair grained plant in 1908	25.9	1	4	18	23	49	39	9	1	3.92
		2	..	5	20	52	47	23	..	4.43
		3	..	..	10	40	64	41	..	4.88
		Total	4	23	53	141	150	73	1	4.42
27-2-1 as 27-1-1	25.0	1	10	18	48	47	25	..	..	3.40
		2	5	13	53	61	16	..	..	3.47
		3	1	8	28	68	36	18	..	4.16
		Total	16	39	129	176	77	18	..	3.69
41-1-2 From good grained plant in 1908	27.4	1	..	18	27	47	34	19	1	4.08
		2	3	15	30	52	38	9	..	3.91
		3	..	..	..	..	..	..	..	....
		Total	..	..	..	..	..	..	..	....
41-2-1 as 41-1-2	26.9	1	8	17	31	55	27	4	..	3.62
		2	8	19	33	57	30	4	..	3.54
		3	..	..	4	41	92	15	..	4.78
		Total	16	36	68	153	149	23	..	4.02
73-2-3-3 From good grained plant in 1908	26.7	1	4	11	22	28	21	6	..	3.75
		2	8	15	36	49	18	9	..	3.60
		3	..	2	16	45	60	31	..	4.66
		Total	12	28	74	122	99	46	..	4.07
76-2-1-1	26.9	1	6	26	43	44	54	20	..	3.90
		2	5	15	31	46	42	10	..	3.91
		3	..	1	25	60	46	20	..	4.39
		Total	11	42	99	150	142	50	..	4.05

TABLE V—CONTINUED.

No.	Leaves per Plant	Picking	Grain Classes							Mean Class
			1	2	3	4	5	6	7	
77–1–1–2	18.4	1	8	28	52	51	13	..	..	3.22
		2	13	34	41	43	4	..	..	2.93
		3	..	6	40	74	23	..	..	3.87
		Total	21	68	133	168	40	..	..	3.32
77–2–1	25.8	1	25	36	37	40	4	..	..	3.44
		2	5	20	52	57	19	..	..	3.42
		3	..	..	4	37	64	10	..	4.70
		Total	30	56	93	134	87	10	..	3.54
K–1–1–2	21.5	1	13	31	42	42	16	..	..	3.12
		2	9	15	35	54	23	9	..	3.65
		3	..	..	..	26	65	59	2	5.24
		Total	22	46	77	122	104	68	2	4.02
K–2–1–6	22.8	1	4	17	38	45	33	3	..	3.68
		2	5	12	44	35	15	3	..	3.46
		3	1	11	35	69	36	7	..	3.94
		Total	10	40	117	149	84	13	..	3.72
Havana	20.0	1	36	37	37	24	10	1	..	2.57
		2	36	53	37	17	2	..	..	2.28
		3	8	29	29	37	28	12	4	3.68
		Total	80	119	103	78	40	23	4	2.92
82–2–1 From poor grained plant in 1908	26.7	1	..	6	19	55	52	15	..	4.35
		2	..	3	20	41	51	32	1	4.62
		3	..	..	2	21	70	58	5	5.28
		Total	..	9	41	117	173	105	6	4.76

A consideration of this table brings some interesting facts to light. It will be seen that in general there is less grain in the upper leaves—that is, the later pickings—than in the lower leaves. On comparing the results obtained from the experimental selections with the Havana selection grown on the same field, we observe that although the Havana was variable in this character it had a larger amount of grain than the other selections. This, however, we know is due to the fact that each individual "grain" of the Havana was larger than in the other selections, our classes representing total grain production and not closeness of grain.

In the first column of the table, under the selection numbers, the "grain" condition of the 1908 ancestral parent plant is given when known. Of the sixteen selections given in the table only eight can be considered under this head, and in one of the eight no third picking was examined, so only seven cases remain for discussion. Of these seven, three descended from plants classed as having good grain, three from fair-grained plants, and one from a poor-grained plant. Those descending from good-grained plants have means of 4.02, 4.07 and 3.97; those from fair-grained plants have means of 3.66, 4.42 and 3.69; and the selection descending from the poor-grained plant has a mean of 4.76.

Of course it would not be fair to lay very much stress on these results, it being probable that all tobacco has the ability to produce some grain. Our results simply indicate that some types, under favorable conditions, produce more grain than others. As such is the case, it seems only fair to conclude that different degrees of grain production are inherited.

Texture Observations.

The same leaves which were examined for grain were also classed as to texture. In this work grain received no weight, and the following brief descriptions give an idea of the characteristics of each class.

Class I — Included those leaves having a dry nature, lacking in oils and gums, with a body so thick as to render it too heavy for the best wrapper leaf.

Class II — Included those leaves of a semi-dry nature, apparently having no more oil than those of Class I, but more gum. The body stiff but sufficiently elastic as to allow its use for wrapper purposes.

Class III — Included those leaves most desirable for wrapper purposes, the oils and gums being present in sufficient quantity and accompanying a medium body, resulting in a leaf of good elasticity, soft but firm handling qualities.

Class IV — Included those leaves of medium body and the gum content, but with excessive amount of oils, giving the leaf a coarse appearance with a tendency to a "rubbery" nature.

Class V — Included those leaves of excessive oil and gum content with a medium to heavy body, resulting in a texture of a decided "rubbery" nature.

Of the classes here given Class III is most desirable from a wrapper standpoint and Classes I and V least desirable.

The results given in Table VI show that many of the selections have a much greater percentage of leaves in Class III than Havana, while other selections have a smaller percentage of leaves of good texture than Havana.

TABLE VI.

Variations in Texture of Halladay Strains in 1912.

No.	Texture Classes									
	I		II		III		IV		V	
	No. Leaves	Per cent.	No. Leaves	Per cent.	No. Leaves	Per cent.	No. Leaves	Per cent.	No. Leaves	Per cent.
5–2–1–3	..	...	40	9.9	201	49.5	137	33.7	28	6.9
6–2–1–4	..	...	42	9.3	222	49.2	179	39.7	8	1.8
12–1–1	2	.4	32	7.0	132	28.9	225	49.1	67	14.6
12–2–1	..	...	21	5.0	252	59.7	141	33.4	8	1.9
27–1–1	36	8.1	73	16.5	93	21.0	174	39.3	67	15.1
27–2–1	..	...	43	9.5	185	40.8	183	40.4	42	9.3
41–1–2	17	5.8	57	19.6	56	19.2	129	44.4	32	11.0
41–2–1	..	...	40	8.9	185	41.2	159	35.4	65	14.5
73–2–3–3	..	...	21	5.5	176	46.0	171	44.6	15	3.9
76–2–1–1	10	2.2	49	10.7	169	36.8	201	43.8	30	6.5
77–1–1–2	..	...	63	14.6	182	42.3	168	39.1	17	4.0
77–2–1	..	...	43	10.3	170	40.6	179	42.7	27	6.4
K–1–1–2	31	7.1	44	10.1	114	26.1	149	34.1	99	22.6
K–2–1–6	..	...	31	7.5	186	44.8	192	46.3	6	1.4
Havana	26	5.9	56	12.8	104	23.8	180	41.1	72	16.4

These data were taken in such a manner that any possible correlation with the grain classes of the previous discussion could be determined, and while no correlation coefficients have been figured we feel justified in concluding from inspection that there is no correlation between grain and the characters here discussed.

While there was no great difference between the selections in texture, there is no question but that some selections were better than others, and several of them gave a somewhat larger percentage of better leaves than the Havana.

Results of Sorting Test.

The results of the actual sorting test are given in Table VIII. For convenience they are calculated to an acre basis, since by this means one can easily compare the value of one selection with another. During the actual sorting, the various lengths of each picking were kept separate, but for convenience they are grouped in the table.

The tobacco was sorted into five different grades: Light Wrappers, Medium Wrappers, Dark Wrappers, Binders and Tops. The Light Wrappers comprise those leaves which have a light even color and thin texture with good body and good vein. Medium Wrappers are a little darker and heavier than the Light Wrappers but must also have good texture and vein. Dark Wrappers are heavier than Medium Wrappers and of a darker color. A great many leaves, which under ordinary circumstances would have been classed as Mediums, are placed in the Dark Wrapper class because of white veins. Binders are thin leaves which are either off-colored, have white veins, or have a tear in them, such faults not permitting them to be graded as Light Wrappers. Tops are heavy, dark, oily leaves.

Table VII gives the prices used in computing the comparative values. These figures were obtained by consulting tobacco men who handled primed sun-grown tobacco in 1911 and 1912, and taking the averages of the prices so obtained. These prices refer to the packed value after fermentation.

The computations for actual packed value were made as follows: First, the yield per acre for a perfect stand of plants was calculated from the healthy plants in a measured row.

Second, the total amount and percentage of each grade was figured to this basis by utilizing the actual sorting data. It was then assumed that these grades could be sold at the prices quoted in Table VII.

TABLE VII.

PRICES PER POUND USED IN COMPUTING VALUES.

Grade	Prices per Pound for Leaf Lengths and Grades		
	12 in.	13–14 in.	15–20 in.
Light wrappers	20 cents	30 cents	80 cents
Medium "	10 "	18 "	50 "
Seconds	8 "	10 "	22 "
Dark wrappers	8 "	10 "	25 "
Tops	5 "	7 "	12 "

Deductions were made for harvesting an extra number of leaves, as many of the selections produced a larger number of leaves per plant than Havana. These deductions were made as follows:

Taking an actual case, for example (5−2)−1−3 averages 29 leaves per plant, by count, and our standard Havana averages about 20 leaves. If we assume that all leaves have an equal weight, 9/29 of 2,813 pounds of tobacco, or 873 pounds must be handled because of the nine extra leaves. One of our best-known growers said that it actually cost him 28 cents per pound to put primed Havana into bales. Thus, the extra cost of handling nine leaves, after growing, and fertilizing the land, would be about 20 cents a pound, and for 873 pounds would amount to $174.60.

If we take the Havana, which averages about 20 leaves per plant, as the standard, and compare its relative value with that of (5−2)−1−3, we must first deduct $174.60 from the packed value of (5−2)−1−3. Assuming the value of Havana as 100, we can then obtain relative values of our other selections by dividing their packed value, after deducting the extra cost for larger leaf number, by the calculated packed value of Havana. Relative values so computed appear in the last column of Table VIII.

TABLE VIII.

SORTING TEST AND RELATIVE VALUES OF THE HALLADAY TYPES.

No.	Mean Leaf Production	Yield in Pounds per Acre	Light Wrappers		Medium Wrappers		Binders		Dark Wrappers		Tops		Relative Value
			Pounds	Per cent.	Pounds	Per cent.	Pounds	Per cent.	Pounds	Per cent.	Pounds	Per cent.	
5—2—1—3	29.0	2813	271	9.7	176	6.2	718	25.5	990	35.2	658	23.4	108.3
6—2—1—4	30.5	2822	582	20.7	266	9.4	771	27.0	497	17.6	704	25.0	137.1
12—1—1	29.1	3370	575	17.0	87	2.6	893	26.5	935	27.8	880	26.1	147.3
12—2—1	29.0	3085	365	11.9	28	0.9	785	25.4	805	26.1	1102	35.7	104.7
27—1—1	25.9	2766	414	15.0	22	0.8	935	33.8	643	23.2	752	27.2	115.1
27—2—1	25.0	2736	677	24.7	100	3.6	889	32.5	573	20.9	497	18.2	157.3
41—1—2	27.4	2196	240	11.0	44	2.0	710	32.3	509	23.2	693	31.5	83.3
41—2—1	26.9	2694	553	20.5	54	2.0	1034	38.4	415	15.4	637	23.7	132.8
41—2—3—2	17.8	1936	362	18.7	9	0.5	612	31.6	203	10.5	750	38.7	111.3
73—2—3—3	26.7	2645	308	11.7	0	0.0	785	29.6	256	9.7	1296	49.0	74.2
76—2—1—1	26.9	2721	456	16.8	126	4.6	557	20.5	714	26.2	868	31.9	114.1
77—1—1—2	18.4	2271	481	21.2	40	1.7	521	22.9	494	21.8	735	32.4	144.7
77—2—1	25.8	2341	445	19.0	39	1.7	380	16.2	689	29.4	788	33.7	111.7
K—1—1—2	21.5	2332	508	21.8	45	1.9	540	23.2	621	26.6	618	26.5	131.5
K—2—1—6	22.8	2740	617	22.5	65	2.3	787	28.7	737	26.9	534	19.5	162.6
Havana	20.0	2119	201	9.5	80	3.9	485	22.8	612	28.8	740	35.0	100.0

A glance at the percentages of Light Wrappers received shows in no case a very favorable result. Selection (27 – 2) – 1, which gave a relative value of 157.3, leads all the selections by producing a total of 24.7% of Light Wrappers. As the Havana which was grown on the same field produced only 9.5% Light Wrappers, the results seem more favorable. There is certainly a wide range of value for these Halladay selections. The poorest, (73 – 2) – 3 – 3, which also was the selection which produced the shortest leaves of the lot, had a relative value of 74.2, while the most favorable, (K – 2) – 1 – 6, gave a relative value of 162.6 as compared with Havana.

It has already been mentioned that before the tobacco was sorted it was examined by three tobacco men. These three men examined the same hands which had been used for the grain and texture results, each working independently and without prejudice of any kind other than some diversity of opinion as to what constitutes an ideal tobacco. None of the three men were very favorably impressed, the general criticism of each being that the tobacco lacked a bright finish. The different selections, however, were given relative placings, at our request. After the placings had been roughly made, each man was then given the second picking of the six selections which, in preliminary judgment, were rated the highest. With these second pickings final placings were then made, and the results are given in the table below, the gradings being placed in sequence with the better type at the top.

Judge 1	Judge 2	Judge 3
(77 – 1) – 1 – 2	(K – 2) – 1 – 6	(K – 2) – 1 – 6
Havana	(73 – 2) – 3 – 3	Havana
(77 – 2) – 1	Havana	(27 – 2) – 1
(76 – 2) – 1 – 1	(77 – 2) – 1	(77 – 2) – 1
(K – 1) – 1 – 2	(77 – 1) – 1 – 2	(41 – 2) – 1
(6 – 2) – 1 – 4	(27 – 2) – 1	(76 – 2) – 1 – 1

It will be noted that (K – 2) – 1 – 6 appears first twice and it is also of interest to know that this selection gave a high relative value by the sorting test. Commercial Havana ranks second twice and third once. The only other selection which appears three times in the judges' table is (77 – 1) – 1 – 2. (27 – 2) – 1, which gave the second highest relative value, appears twice in this table.

As the crop was of such an inferior nature no hard and fast conclusions can be drawn as to the commercial value of the selections. It is encouraging that under similar conditions several types gave much higher relative values than Havana.

Conclusions.

Our results show conclusively that the Halladay Havana was not a mutation or sport, but that it resulted from a recombination of parental characters, in which the number of leaves and leaf shape of the Sumatra were united with the leaf size and habit of growth of the Havana. That the general Halladay Havana type as it appears in the field can be reproduced whenever desired is an undoubted fact.

The apparent uniformity of the Halladay type in 1908 has proved to be of only superficial nature. By selection we have been able to produce several strains which differ very widely in number of leaves, leaf size and vigor. In other families of this cross, selection has as yet given no results of appreciable value. It seems only fair to conclude that by selection we have been able simply to isolate different lines that approach a homozygous condition, and that in those cases where selection has given no results the lines were already in a nearly homozygous condition.

Quality of cured leaf is, without a doubt, due to both external and internal factors. Environment, of which we may mention physical characters of soil, moisture, temperature and soil fertility, and methods of handling, such as time of harvesting, are of great importance. These may be roughly classed as external factors.

In our experiments we have eliminated, as far as possible, unfavorable external factors, but the total elimination of unfavorable conditions is a physical impossibility. All that we have been able to do is to give all selections as nearly an equal chance under as favorable conditions as possible. The relative values of the experimental selections were compared with Havana grown under similar conditions. Assuming the value of Havana as 100, the experimental types have ranged in value from 74.2 to 162.6.

Previous experiments have shown that the Sumatra parent lacks wrapper quality when grown in Connecticut. It has, however, a large leaf number and a good leaf shape. The Havana parent, while widely grown, is not an ideal type. The leaf is too pointed in shape and there are also possibilities of improving its quality. A leaf which is of intermediate weight between Sumatra and Havana and which shows the bright appearance and elasticity of the Havana parent would be of commercial value. Nearly all of our Halladay strains have good leaf size and an improved leaf shape. Some of the types are very inferior in quality, others are of intermediate value, and a few closely resemble Havana. The better selections will be further tested as they show promise of being of commercial value.

FAMILY (403×401), SUMATRA×BROADLEAF.

In 1909 a cross was made between Sumatra (403) and the Connecticut out-door type of tobacco known as Broadleaf (401). The Sumatra had been grown under tent from inbred seed for four years and appeared uniform. The Broadleaf parent was a commercial variety, and as seed of the same type has proved very uniform we feel justified in saying that this cross was made between types which, as to external characters, were in a nearly homozygous condition.

The objects of this cross were to study the inheritance of certain characters as a check on the Halladay Havana results, and to produce a type of tobacco which had the desirable quality of the Broadleaf parent together with more desirable morphological characters, and it was thought that a recombination of factors from both the Broadleaf and the Sumatra might furnish such a variety. The leaves of the Broadleaf are long and drooping, and for this reason the tobacco is hard to cultivate and harvest. The shape of the leaf, with its narrow pointed tip, is such that considerable waste is made in cutting wrappers. A shorter, rounder, more erect leaf of as good quality as the Broadleaf would be of material value. It has not been produced as yet but the results are of interest as some facts of importance have been obtained.

The first generation of the cross together with its parents was grown in New Haven in 1910, though a few plants of the F_1 generation were also grown in Bloomfield.

In 1911 the parents and two F_2 generations were grown in New Haven and large cultures of three F_2 generations were grown in Bloomfield. It was our intention to harvest the Bloomfield selections and to examine them for quality, but there was a heavy hail storm a few weeks before harvesting time, and as only about half the leaves were worth harvesting, the tobacco was sold in the bundle and no actual sorting data were taken. However, some leaves were of good quality.

A number of F_3 generations were grown in Bloomfield in 1912, and others, together with further generations of the parents, were grown in New Haven. The Bloomfield selections were assorted in the same manner as the Halladay Havana types.

Inheritance of Leaf Number.

The inheritance of number of leaves per plant for this family has been considered in a previous paper (Hayes, 1912) and the F_1 and F_2 hybrid generation results were then given.

Table IX gives the results of three generations of the parents, the first generation of the cross which was grown in New Haven, two F_2 generations, and nine F_3 generations, which were grown in Bloomfield. This table gives the number of leaves of the parent, the total number of variates, the means, and the coefficients of variability.

The Broadleaf parent (401) has shown little variation in mean leaf number in the three years grown, the means being 19.2 ± .05 leaves in 1910 and 19.9 ± .07 in 1912. The coefficient of variability is slightly higher in 1912 than in 1910.

The mean leaf number of the Sumatra variety was 28.2 ± .08 in 1910, 26.5 ± .11 in 1911, and 26.2 ± .12 in 1912. The duplication of the results in the last two years indicates an error of counting in 1910, since such an error might arise by not discarding the three basal leaves uniformly as was done in the later years.

The coefficient of variability for the Sumatra parent was 5.27 ± .21 in 1910, 6.64 ± .28 in 1911, and 8.28 ± .32 in 1912. The cause of this rise in variability in 1912 is not clear. It may be due to a small mutation in one of the germ cells of the 1910 plant that gave rise to the 1911 population. The population in 1912 would then be the F_2 generation of the

TABLE IX.

INHERITANCE OF LEAF NUMBER IN CROSS (403×401) SUMATRA×BROADLEAF.

No.	Year Grown	Generation	Leaves of Parent	Range of Variation	Total	A.	C. V.
403, Sumatra	1910	P_1	. .	24–31	150	28.2 ± .08	5.27 ± .21
403 – 1	1911	P_2	29	23–31	125	26.5 ± .11	6.64 ± .28
403 – 1 – 2	1912	P_3	29	21–32	151	26.2 ± .12	8.28 ± .32
401, Broadleaf	1910	P_1	. .	17–22	150	19.2 ± .05	5.00 ± .19
401 – 1	1911	P_2	20	16–22	108	19.1 ± .08	6.54 ± .30
401 – 1 – 1	1912	P_3	22	17–23	145	19.9 ± .07	6.03 ± .24
(403×401) = B	1910	F_1	. .	19–26	150	23.6 ± .07	5.51 ± .21
B – 1	1911	F_2	25	17–32	2402	22.7 ± .03	8.99 ± .11
B – 3	1911	F_2	24	17–35	1632	22.5 ± .03	9.51 ± .10
B – 1 – 4	1912	F_3	25	16–29	179	22.5 ± .12	10.84 ± .39
B – 1 – 7	1912	F_3	22	17–28	207	21.5 ± .10	10.14 ± .34
B – 1 – 8	1912	F_3	28	19–33	82	26.3 ± .20	10.38 ± .55
B – 1 – 10	1912	F_3	26	19–27	151	23.1 ± .10	7.75 ± .30
B – 1 – 12	1912	F_3	25	18–30	209	23.7 ± .14	10.51 ± .41
*B – 1 – 14	1912	F_3	25	19–29	56	21.8 ± .14	7.18 ± .46
B – 3 – 5	1912	F_3	27	17–28	159	21.7 ± .11	9.45 ± .36
B – 3 – 6	1912	F_3	28	16–27	229	22.5 ± .09	8.71 ± .27
B – 3 – 8	1912	F_3	25	17–23	85	20.6 ± .12	8.25 ± .43

* 131 plants were grown at New Haven, the calculated coefficient of variability being 6.44 ± .27.

mutating germ cell with a normal cell. On the other hand, though, we have data on another cross that indicate that the field environment has but little effect in determining the number of leaves, it may be that this effect is somewhat greater on the Sumatra variety with its different habit of growth.

Cross (403×401) has been designated as B in Table IX, and as such it will be described in the text. An inspection of the table will show that the first generation of the cross is no more variable than the parents, although intermediate in leaf number, whereas the F_2 generations, B – 1 and B – 3, of which large cultures were grown, are extremely variable, giving coefficients of variability of $8.99 \pm .11$ and $9.51 \pm .10$, and ranging in value from the leaf number of the Broadleaf to that of Sumatra.

Of the nine F_3 generations, B – 1 – 8 has a mean for leaf number of $26.3 \pm .20$, which is about the same as Sumatra, while the remainder show means of intermediate value, although that of B – 3 – 8, $20.6 \pm .12$, is only slightly greater than the Broadleaf parent.

B – 1 – 14 shows a coefficient of variability of $7.18 \pm .46$, which is only slightly higher than the parents. This same selection was also grown in New Haven and gave a coefficient of variability of $6.44 \pm .27$. For this reason, if one is to attach any value to this biometrical constant, it seems only fair to conclude that this type is in a homozygous condition for leaf number. B – 1 – 10 also proved rather uniform since it had a variability coefficient of only $7.75 \pm .30$. These two types were both of intermediate value for leaf number.

On the other hand, five of the remaining populations have coefficients of variability of practically the same value as the F_2 generation, and two show an intermediate value. This difference in the variability of F_3 populations grown from individuals from various F_2 classes is exactly what should be expected if several Mendelian factors have recombined in the F_2 generation.

Shape and Size of Leaf.

In the data on inheritance of leaf size in cross B, which were given in an earlier paper, there were no F_2 plants with as large an average leaf area as the extreme variates of the Broadleaf.

TABLE X.

BREADTH INDEX OF F_1, F_2 GENERATIONS AND PARENTS OF CROSS (403×401), SUMATRA×BROADLEAF.

No.	Year grown	Generation	Breadth Index Classes												Total	A.	C. V.
			36	39	42	45	48	51	54	57	60	63	66	69			
403, Sumatra	1910	P_1	..	..	..	2	15	47	43	30	12	1	..	..	150	53.5 ±.19	6.60 ±.26
401, Broadleaf	1910	P_1	..	2	13	41	46	32	13	3	..	..	..	..	150	47.9 ±.20	7.62 ±.30
(403×401) = B	1910	F_1	..	..	..	5	15	34	62	28	6	..	..	..	150	53.2 ±.18	6.18 ±.24
B – 1	1911	F_2	1	2	9	21	30	15	16	9	2	1	..	1	107	49.3 ±.35	10.81 ±.50
B – 2	1911	F_2	2	17	40	65	57	40	15	3	2	..	..	..	241	46.5 ±.19	9.40 ±.30

This was explained by the fact that the environmental conditions for F_2 were poorer than the parents or F_1 had enjoyed. While no statistical records were taken, the large size of leaves of numerous plants of several of our F_3 generations grown at Bloomfield in 1912 has shown this explanation to be the correct one.

Size of leaf, as perhaps should be expected, is greatly influenced by environment, which made proper analysis of our breeding results a difficult task; but shape of leaf, which is the basis of our next study, is fortunately less subject to such modification.

The method of determining leaf shape which has been used is called breadth index. It is obtained by dividing the breadth by the length and expressing the result in per cent.

The same variates which showed no distinct segregation in leaf size have been considered, the results of this method of treatment appearing in Table X. The middle leaf of each plant was used in computing breadth index.

The table shows that the average breadth index of the Sumatra is $53.5 \pm .19$, which means that, on the average, the breadth of leaf of the Sumatra is a little more than half the length. The Broadleaf gave an index of $47.9 \pm .20$, and the F_1 generation an index of $53.2 \pm .18$. The indexes of the two F_2 generations are shown by the table to be $49.3 \pm .35$ and $46.5 \pm .19$. The conditions for the F_2 generations were very unfavorable and the indexes are smaller than one would expect. That there is some sort of segregation of leaf shape seems very evident, as the coefficients of variability of the F_2 are much larger than those of the parents, or F_1.

Table XI gives comparative results for length of leaf of the F_3 selections grown at Bloomfield in 1912. This table gives the average number of leaves per plant, by actual count, the yield of cured tobacco per acre, and the number of pounds of cured tobacco of leaf length classes, which range from 12 to 20 inches. It is regretted that no Broadleaf selection was grown to compare with the hybrids.

TABLE XI.

Comparative Length of Leaves of the F_3 Generations of Cross (403 × 401), Sumatra × Broadleaf.

No.	Mean Leaf Production	Yield in Pounds per Acre	Yield in Pounds for Leaf Length Classes in Inches							
			12	13	14	15	16	17	18	20
B−1−4	22.0	2030	130	220	295	350	350	330	299	55
B−1−7	21.5	2476	63	126	213	281	352	399	567	475
B−1−8	26.3	2579	305	291	410	388	298	276	410	201
B−1−10	23.1	2517	41	133	233	388	484	443	653	142
B−1−12	23.7	2405	46	101	150	261	362	421	545	519
B−1−14	21.8	2629	..	159	265	361	520	392	583	350
B−3−5	21.7	3206	..	152	190	262	410	512	982	698
B−3−6	22.5	2927	58	173	203	275	323	405	643	845
B−3−8	20.6	2566	36	154	190	298	361	425	669	434

In considering these results it is important to note that only medium size and large leaved plants were used as parents of the F_3 generations. There is considerable variation in leaf lengths, as shown by this table. Thus, B−1−4 produced a large number of leaves on classes 15 and 16. B−1−8 and B−1−14, while producing the greater weight of leaves on class 18, also produced a large number of leaves on classes 15 and 16. B−3−6 is the only selection which produced the most leaves by weight in class 20. The selections, then, show considerable variation in leaf length when compared with each other and show that there are probably a number of factors affecting leaf size.

Some general notes on the leaf conditions of these F_3 generations of cross B are given in Table XII. Three general features — uniformity, color of leaves and type of leaf — were considered. Uniformity refers to the leaf characters of the selection as a whole. Those marked "good" in the table were uniform in all characters, while the remainder showed considerable variation. These facts are mentioned here, as our results point to the conclusion that the different characters, such as leaf number, shape of leaf and type of leaf, in which the parents differ, are in a large measure inherited independently. One other purpose was to determine if any single external character could be correlated with quality.

TABLE XII.

General Notes on the Leaf Condition of thf F_3 Generations of Cross (403×401), Sumatra ×Broadleaf.

No.	Uniformity	Color of Leaves	Type of Leaf
B−1−4	Good	Light green	Moderately crinkled
B−1−7	Fair	Medium green	Smooth to crinkled
B−1−8	Good	Light green	Very crinkled
B−1−10	Fair	Medium green to bluish	Slightly crinkled
B−1−12	Fair	Somewhat bluish	Leaves mostly smooth
B−1−14	Good	Medium green	....
B−3−5	Fair	Light to medium green	Slightly crinkled
B−3−6	Fair	Medium to dark green	Moderately crinkled
B−3−8	Fair	Medium green	Moderately crinkled

Quality of the F_3 Selections.

Data on texture and grain were not taken for the F_3 Sumatra ×Broadleaf crosses, with the exception of two selections which were examined for grain, the leaves being classified into seven grain classes as for the Halladay types. The selections used were B−1−10, which proved uniform for number of leaves per plant, giving a variability coefficient of 7.75±.30, and B−1−7 which was not uniform for leaf number and which gave a variability coefficient of 10.14±.34.

If there were a correlation between grain and leaf number we should expect the classes for B−1−10 to be more uniform than those for B−1−7. A glance at Table XIII indicates that such is not the case, since both selections were about equally variable and both have a large amount of grain. At the same time it is realized that the method of determining grain is exceedingly arbitrary.

TABLE XIII.

Comparison of Grain of B−1−7 and B−1−10.

No.	Leaves per Plant	Picking	Grain Classes					
			1	2	3	4	5	6
B−1−7	21.5	1	37	41	42	25	13	4
		2	32	51	40	26	11	..
		3	32	39	53	23	10	..
		Total	101	131	135	74	34	4
B−1−10	23.1	1	..	35	40	31	10	1
		2	30	40	46	26	9	1
		3	29	44	44	34	5	..
		Total	59	119	130	91	24	2

TABLE XIV.

SORTING TEST AND RELATIVE VALUES OF F_3 BROADLEAF SELECTIONS.

No.	Mean Leaf Production	Yield in Pounds per Acre	Light Wrappers		Medium Wrappers		Binders		Dark Wrappers		Tops		Relative Value
			Pounds	Per cent.	Pounds	Per cent.	Pounds	Per cent.	Pounds	Per cent.	Pounds	Per cent.	
B–1–4	22.0	2030	468	23.0	4	0.2	287	14.2	622	30.6	649	32.0	100.0
B–1–7	21.5	2476	583	23.5	5	0.2	457	18.5	819	33.1	612	24.7	141.8
B–1–8	26.3	2579	469	....	..	...	335	....	224	....	715	....	.....
B–1–10	23.1	2517	502	20.0	41	1.6	407	16.2	690	27.4	877	34.8	122.6
B–1–12	23.7	2405	587	24.4	16	0.7	428	17.8	633	26.3	741	30.8	113.6
B–1–14	21.8	2629	297	11.3	21	0.8	329	12.5	1028	39.1	954	36.3	102.7
B–3–5	21.7	3206	558	17.4	30	0.9	812	25.3	998	31.3	808	25.2	162.6
B–3–6	22.5	2927	699	23.9	13	0.4	491	16.8	966	33.0	758	25.9	159.5
B–3–8	20.6	2566	416	16.2	..	...	659	25.7	750	29.2	741	28.9	127.4

Relation of Havana to B–1–4 = 100:105.1.

Table XIV gives the sorting test and relative values of the F_3 selections. The yield ranged from 2,030 pounds per acre in B−1−4 to 3,206 pounds in B−3−5. This seems to be good evidence that a selection can be produced which would give a much higher yield per acre than the commercial Broadleaf now grown. The success of our experiment does not depend so largely on yield factors as it does on quality values, however, and on this subject no very definite conclusions can be drawn until the selections are more uniform for external plant characters and have been tested for quality another season.

B−1−4 has about the same relative value as the Havana type given in Table VIII, the relation of B−1−4 to Havana being 105.1 to 100. For the relative values given in the last column of Table XIV, B−1−4 has been used as the standard (100), the actual prices for grades being assumed to be the same as for the Halladay types which were given in Table VII. B−1−14 gave about the same relative value as B−1−4, although it gave a yield of 2,629 pounds per acre while B−1−4 only gave a yield of 2,030 pounds. B−3−5 gave the highest yield, and also the highest relative value of any of the selections.

The attempt to discover some external character or characters which are correlated with quality has not, as yet, proved successful. It seems very probable that, although it may be necessary to have all characters in a nearly homozygous condition in order to produce tobacco that is of uniform quality, this is not because there is a close relation between quality and any one external character. If the type is in a complex hybrid condition, variation in time of maturity, venation, etc., will be the rule. Such conditions will not be favorable to producing a uniform quality of tobacco.

Conclusions.

The results obtained from the Broadleaf×Sumatra cross show that, as a rule each character, such as leaf size, leaf shape, number of leaves and type of leaf, are inherited independently. Hence the difficulty of producing a uniform strain after crossing will depend largely on the gametic condition of the parents. If the parents differ in a large number of factors the difficulties will be much greater than if there are but a small number with which to deal.

The really important feature is that there is a segregation of quantitative characters in the F_2 generation of tobacco crosses and that some segregates will breed true in F_3. As this is the case, there seems to be no need of using a different method when working with quantitative characters than for qualitative or color characters.

Since quality of cured leaf depends on many factors, external as well as internal, it is probably unreasonable to expect a single external character to be closely correlated with quality, but as homozygosis produces uniformity in both quantitative and qualitative characters it must tend to produce uniform quality.

The important matter in practice is simply to grow a sufficient number of F_3 and later generations to run a fair chance of testing out all the combinations of factors possible to the parental varieties used.

FAMILY (402×405), HAVANA×CUBAN.

This cross was made in 1909 between strains of Havana and Cuban which had been grown for several years from inbred seed. The P_1 generation of the Cuban parental type given in the tables was not grown from inbred seed of a single plant, but from commercial seed saved under tent covering. The plants from which this seed was saved were grown from seed of direct descendants of the inbred Cuban type used as the male parent. The P_1 generation of Havana given in our tables was also grown from commercial seed.

This cross has been designated as C in our discussion. The parents and different generations of this cross have been grown under shade covering at the Windsor Tobacco Growers' Corporation in Bloomfield, with the exception of C—1—5 and C—1—6, which were grown outdoors on the same field as the Halladay and F_3 Broadleaf selections. The conditions for this cross grown under cloth shade are more uniform than for the previous experimental selections which were grown in the open, due to the protection the covering affords from heavy winds and storms.

The parents and F_1 were grown in 1910, further generations of the parents and F_2 in 1911, and the third generation of parents and five F_3 generation families in 1912.

TABLE XV.

INHERITANCE OF LEAF NUMBER IN CROSS BETWEEN (402×405), HAVANA×CUBAN

No.	Year grown	Generation	Leaves of Parent	Range of Variation	Total	Mean	C. V.
405, Cuban	1910	P_1	..	16–25	150	19.9 ± .08	7.53 ± .28
405–1	1911	P_2	21	18–23	124	20.6 ± .07	5.29 ± .23
405–1–1	1912	P_3	23	17–25	150	20.9 ± .07	6.17 ± .24
402, Havana	1910	P_1	..	17–24	143	19.8 ± .07	6.98 ± .27
402–1	1911	P_2	20	16–25	150	20.3 ± .10	8.87 ± .35
402–1–1	1912	P_3	20	15–25	150	19.4 ± .05	4.59 ± .18
(402×405) = C	1910	F_1	..	15–25	150	19.8 ± .07	6.10 ± .24
C–1	1911	F_2	..	14–33	192	20.9 ± .16	15.84 ± .54
C–1–2	1912	F_3	20	13–29	112	19.7 ± .18	14.67 ± .67
C–1–3	1912	F_3	20	15–22	142	18.4 ± .09	9.02 ± .36
C–1–4	1912	F_3	28	20–35	148	26.6 ± .16	11.20 ± .44
C–1–5	1912	F_3	30	22–34	45	28.0 ± .28	10.00 ± .72
C–1–6	1912	F_3	22	13–29	201	20.1 ± .15	16.17 ± .56

Inheritance of Leaf Number.

The inheritance of number of leaves per plant is given in Table XV. The Cuban selection gave a range of variation of 16 to 25 leaves in 1910 and from 17 to 25 in 1912. The mean number of leaves per plant was 19.9±.08 in 1910, 20.6±.07 in 1911, and 20.9±.07 in 1912. There has been a slight progressive change in leaf number for the three years, but whether this is due to an actual germinal change or to unavoidable errors in our leaf counts is impossible to say. No wide changes are shown by the coefficients of variability, which were 7.53±.28 in 1910, 5.29±.23 in 1911, and 6.17±.24 in 1912.

The Havana selection gave a mean of 19.8±.07 leaves in 1910, 20.3±.10 in 1911, and 19.4±.05 in 1912. This selection shows no great change for leaf number. The coefficient of variability shows considerable variation, as it was 6.98±.27 in 1910, 8.87±.35 in 1911, and 4.59±.18 in 1912.

The F_1 gave about the same mean and variability coefficient as the parent types, the mean being 19.8±.07 and the coefficient of variability 6.10±.24.

If the parents both contained the same inherited factors for leaf number, which one might expect from their having about the same average number of leaves per plant, no increased variability over F_1 should be obtained in F_2. The range of variation, 14 to 33 leaves, and the coefficient of variability of the F_2 generation, 15.84±.54, both show that such is not the case. Plants appeared which bore a higher and also a lower number of leaves than in F_1.

The counts of leaf number for the five F_3 generations show conclusively that the increased variability in F_2 was a germinal one. These five F_3 selections were grown from F_2 plants which bore 20, 20, 22, 28 and 30 leaves respectively. Progeny from one of the 20-leaved F_2 plants, C−1−3, gave rather uniform results in F_3, the mean being 18.4±.09 and the coefficient of variability 9.02±.36. Progeny from the other 20-leaved parent plant, C−1−2, and also the 22-leaved plant, C−1−6, gave means of about 20 leaves per plant and large variability coefficients, 14.67±.67 and 16.17±.56 respectively.

The two remaining selections, C−1−4 and C−1−5, with coefficient of variability values of 11.20±.44 and 10.00±.72 were more variable than the F_1 and less variable than the F_2.

TABLE XVI.

BREADTH INDEX OF F_1, F_2, F_3 GENERATIONS AND PARENTS OF CROSS (402×405), HAVANA×CUBAN.

No.	Year grown	Generation	Breadth Index Classes													Total	A.	C. V.
			36	39	42	45	48	51	54	57	60	63	66	69	72			
402, Havana	1910	P_1	..	4	29	69	38	9	1	..	..	..	..	..	..	150	45.4 ± .15	6.04 ± .24
402 – 1 – 1	1912	P_3	..	3	21	74	48	4	..	..	..	..	..	..	..	150	45.6 ± .13	5.13 ± .20
405, Cuban	1910	P_1	..	..	..	..	..	3	19	56	56	14	1	..	1	150	58.3 ± .16	5.09 ± .20
405 – 1 – 1	1912	P_3	..	..	..	..	..	..	1	30	76	34	9	..	..	150	60.4 ± .14	4.09 ± .16
(402×405) = C	1910	F_1	..	..	1	13	38	69	25	3	1	..	..	..	..	150	50.3 ± .16	5.73 ± .22
C – 1	1911	F_2	1	1	10	15	37	57	34	26	10	1	..	..	..	192	51.2 ± .23	9.18 ± .32
C – 1 – 2	1912	F_3	..	..	..	..	3	8	19	35	38	8	1	1	..	113	57.5 ± .23	6.40 ± .29
C – 1 – 3	1912	F_3	..	6	25	56	35	19	1	..	..	..	..	..	..	142	45.8 ± .18	6.92 ± .28
C – 1 – 4	1912	F_3	..	6	10	50	55	24	3	2	..	..	..	..	..	150	47.0 ± .18	7.00 ± .27

TABLE XVII.

INHERITANCE OF LEAF SIZE IN CROSS BETWEEN (402×405), HAVANA×CUBAN.

No.	Year grown	Generation	Classes in Sq. Dcms.										Total	A.	C. V
			3	4	5	6	7	8	9	10	11	12			
402, Havana	1910	P_1	..	1	23	42	41	35	8	..	..	..	150	6.7 ±.06	17.1 ±.68
402–1–1	1912	P_3	2	47	65	33	3	..	..	..	..	..	150	4.9 ±.05	16.6 ±.67
405, Cuban	1910	P_1	3	26	58	54	9	..	..	..	..	..	150	5.3 ±.05	16.7 ±.67
405–1–1	1912	P_3	4	41	87	18	..	..	..	..	..	..	150	4.8 ±.04	14.1 ±.56
(402×405) = C	1910	F_1	..	6	27	46	39	27	5	..	..	..	150	6.5 ±.07	18.4 ±.74
C–1	1911	F_2	3	10	32	32	45	33	24	8	4	1	192	7.0 ±.09	25.3 ±.92
C–1–2	1912	F_3	5	22	43	31	8	1	..	..	..	..	110	5.2 ±.07	19.3 ±.91
C–1–3	1912	F_3	..	3	45	46	34	13	..	..	..	..	141	6.1 ±.06	16.5 ±.68
C–1–4	1912	F_3	4	33	58	31	12	10	2	..	..	..	150	5.3 ±.07	23.4 ±.96

The means for leaf number were 26.6 ± .16 and 28.0 ± .28. Thus, from crossing two types bearing an average of about 20 leaves per plant, a new type has been produced with a larger leaf number.

Size and Shape of Leaf.

It was pointed out in an earlier paper that Cuban and Havana have about the same average leaf width but that Havana has somewhat longer leaves than Cuban. The breadth indexes of the parental varieties and crosses are given in Table XVI. As in the other cross, the middle leaf of each plant was used for these computations. The Havana leaf is shown to be proportionally much narrower for its length than the Cuban. The F_1 was of intermediate value for breadth index, and in F_2 there was an increase of variability. The F_3 strain, C − 1 − 2, bred comparatively uniformly for the Cuban shape of leaf, giving a mean breadth index of 57.5 ± .23. This is slightly lower than the index of the 1910 Cuban selection, which is 58.3 ± .16, but the difference between these values is slightly less than four times the probable error. The parent F_2 plant of C − 1 − 3 resembled Havana in all particulars and the progeny was of Havana type in both leaf size and breadth index value. The breadth index of C − 1 − 4 was also of Havana type, and the coefficient of variability showed this selection to be uniform in leaf shape.

Table XVII gives the inheritance of leaf size for this cross. For this work, the areas of the fourth leaf from the bottom, the middle leaf, and the last leaf at the top below the bald sucker were taken. The area of leaf used in the table is the average of these three measurements.

The table shows that in 1910 the average Havana leaf area was greater than the Cuban and that the F_1 generation had nearly as large an average leaf area as Havana. The average leaf area of the F_2 generation was slightly greater than in F_1 and the variability was also much greater.

It is true that none of the shade selections grew as vigorously in 1912 as in previous years, but this does not explain the proportionally greater decrease in leaf size of the Havana as compared with the Cuban. It is of interest to know that selection C − 1 − 3, which was not very variable for leaf number and which was of uniform leaf shape, gave a variability coefficient of about the same value as the parental selections. The coefficient of variability of C−1−2 was only slightly greater than that of the parents, while C−1−4 seemed to be more variable.

It should be mentioned that the coefficient of variability is not a very safe criterion by which to judge when dealing with a character such as area of leaves. It is to be expected that a selection which is heterozygous in other plant characters will be more variable in a character such as leaf area than a completely homozygous selection, as stimulus to development is greater in a heterozygous than in a homozygous state, and when segregation is taking place some plants of a generation are homozygous and others complex hybrids.

The comparative length of leaves of the parents and F_3 generations is given in Table XVIII. As in previous tables of this kind, one must remember that these computations are made on the acre basis and that the figures in the table under the heading "leaf classes in inches" refer to pounds and not to number of leaves.

TABLE XVIII.

COMPARATIVE LENGTH OF LEAVES OF THE PARENTS AND F_3 GENERATIONS OF CROSS (402×405), HAVANA×CUBAN.

No.	Mean Leaf Production	Yield in Pounds per Acre	Leaf Classes in Inches							
			12	13	14	15	16	17	18	20
405−1−1*	20.9	1493	186	193	142	350	328	218	76	..
402−1−1†	19.4	1508	51	29	208	113	164	273	499	171
C−1−2	19.7	1635	102	137	183	218	295	355	279	66
C−1−3	18.4	1369	44	36	33	120	153	127	517	339
C−1−4	26.6	2036	..	6	51	93	93	206	556	1032
C−1−5	28.0	1709	..	100	168	200	302	369	469	101
C−1−6	20.1	2206	98	151	214	351	292	411	538	151

*Cuban. † Havana.

This table shows that the Cuban produces a larger percentage of short leaves than the Havana. C−1−2, which it will be remembered was of Cuban shape except that is leaves average slightly larger, shows a population similar to 405−1−1. C−1−

3, the F_3 Havana type, shows a population more nearly like Havana. Selection C-1-4 is of interest as it produced a much larger number of leaves per plant than the other shade selections. It also produced a large proportion of leaves of 20 inch length, averaging 1032 pounds per acre. The results given for C-1-5 and C-1-6 should not be given much weight in the discussion of comparative leaf lengths as they were grown out of doors. The interesting feature of these results is that one of the five F_3 generations closely resembled the Havana parent in leaf size and shape while another F_3 generation produced leaves that were of the shape and size of the Cuban parent.

Inheritance of Quality.

The results of a sorting test for quality are given in Table XIX, and the prices per pound which were used in computing relative values are given in Table VII. It is, of course, true that the selections which were grown under shade are worth more per pound than the prices used indicate; however, for our purposes these prices are probably as valuable as any other. No corrections were made for leaf number except for C-1-4, which produced 26.6 leaves per plant, this being reduced to a 20 leaf basis. The fourth picking of C-1-5 was lost, so the figures given for this selection represent the first three pickings only. Selection C-1-6 was weighed before sizing and the yield given in the table is correct. During the warehouse work the third picking of C-1-6 was mixed with a Broadleaf selection. The Broadleaf selection was discarded, but in the case of the C-1-6 the value per pound of the third picking was estimated, as we knew the actual value of the first, second, and fourth pickings.

The results of this sorting test throw some light on the problem of quality inheritance. Both parental varieties in this cross are tobaccos which produced a good quality of wrapper leaf. The percentages of light wrappers are 31.9 for 405-1-1, Cuban, and 39.8 for 402-1-1, Havana. For the computation of the relative values, Havana is again taken as the standard and the ratio of the shade selection 402-1-1 to the out-door Havana given in Table IX is 118.3:100.

TABLE XIX.

SORTING TESTS AND RELATIVE VALUES OF THE PARENTS AND F_2 GENERATIONS OF CROSS (402×405), HAVANA×CUBAN.

No.	Mean Leaf Production	Yield in Pounds per Acre	Light Wrappers		Binders		Dark Wrappers		Tops		Relative Value
			Pounds	Per cent.	Pounds	Per cent.	Pounds	Per cent.	Pounds	Per cent.	
405–1–1	20.9	1493	477	31.9	382	25.6	470	31.5	164	12.0	81.0
402–1–1	19.4	1508	601	39.8	317	21.0	452	30.0	138	9.2	100.0
C–1–2	19.7	1635	640	39.1	325	19.9	477	29.2	193	11.8	106.5
C–1–3	18.4	1428	601	42.0	390	27.3	324	22.7	113	7.9	104.7
C–1–4	26.6	2036	905	44.5	553	27.1	414	20.4	164	8.0	142.9
C–1–5	28.0	1709	837	49.0	486	28.4	336	19.6	50	3.0	133.0
C–1–6	20.1	2206	189	15.2	523	41.8	183	15.0	355	28.4	86.1

C = cross between Havana and Cuban, 405–1–1 being Cuban and 402–1–1 being Havana.
The comparative value of the shade Havana to the outdoor Havana of Table IX is 118.3:100.

That the increase of leaf number does not cause an increase of dark and top leaves is clearly shown by selections C—1—4 and C—1—5. These selections both produced a high percentage of light wrappers and gave a high relative value.

The yields of the shade tobacco are much less than they would be if they were grown in the open, as the shade covering produces a thin leaf. A sample of Havana shade-grown light wrappers was shown to a well-known buyer who was in the warehouse when the experimental tobacco was being assorted and he was asked what they were. He immediately replied, "A fine quality of Havana." On the other hand, an out-door Cuban selection retained its distinctive character, although the percentage of dark leaves was greater and the leaves were heavier in the out-door tobacco. Thus we must come to the conclusion that quality, while decidedly affected by environment, is nevertheless greatly dependent on heredity.

The relative value of C—1—6 is only 86.1 although this selection gave a yield per acre of 2,206 pounds. This seems most easily explained by the fact that this selection was in a heterozygous condition for many characters. The variation in leaf number per plant was very high, as is shown by Table XV, and we know from observation that the variation in leaf shape and size was also very large. Hence, though some leaves of this selection were of high quality, the percentage was very low, and a large percentage of off-colored and dark leaves was produced. These results show that uniformly high quality cannot be expected if many characters are in a heterozygous condition.

Conclusions.

The results obtained from this cross show clearly that an external similarity of size characters in tobacco varieties does not necessarily mean a genetic similarity. Havana and Cuban both produced about the same average number of leaves per plant, yet when they were crossed together an increased variability occurred in F_2. The five F_3 generation selections show that this increased variability was germinal, two of the five F_3 selections giving a much higher leaf average than the parents.

Similar results have been obtained frequently in inheritance of qualitative characters. The general basis of the Mendelian conception of heredity depends on the fact that the somatic appearance of a plant is not a correct expression of its breeding nature. Of two red-flowered plants in the second generation of a cross between white and red-flowered races in which complete dominance is the rule, the one may breed true for the red color, giving only red progeny, and the other may give both red and white progeny. Advances may be disguised and may appear in crosses as well as simple recessives, although advances due to crossing are as a rule less frequent than simple recessives. In such cases as the purple aleurone color of maize, which depends on the presence of at least two color factors we may receive purple aleurone seeds on crossing white races if one white race contains one of the necessary color factors and the other white race contains the other. That similar results are obtained when dealing with size characters and that in both quantitative and qualitative characters it is impossible to know the germinal characters except by a breeding test seems further proof of the belief that both are inherited in a similar manner.

The results of the sorting test of the parents and third generation crosses show that heterozygosis affects quality and that uniformity of external characters tends to produce uniformity of quality in the cured leaves. Some of the hybrids gave increased yields and good quality and look promising from a commercial standpoint. It will be necessary, however, to continue the selections in row cultures until all characters are in a homozygous condition or nearly so.

Interpretation of Results.

In a previous paper (Hayes, 1912) the data obtained from the first and second hybrid generations of size studies of tobacco were given a strict Mendelian interpretation by assuming a multiplicity of factors, each inherited independently and capable of adding to the character, the effect of the heterozygous condition of each factor being half the homozygous. The data on the third generations and on the Halladay reported in this paper show no need of a change of interpretation.

In order that the above interpretation may be justified, certain results must be obtained. The first generation of a cross between two homozygous varieties which differ in a quantitative character, such as number of leaves per plant, must be of intermediate value and no more variable than the parents; the F_2 generation should give an increase in variability and, when sufficient individuals are studied, should give a range of variability equal to the combined range of the parents. Certain selected F_2 plants should breed true giving no greater variability than the parents; others should give a variation as great as the F_2 generation, and others should give variabilities intermediate between the value of the F_1 and F_2. All of these conditions are fulfilled in our crosses.

The exact number of factors involved in any cross is difficult of determination, due to the obscuring effects of fluctuating variability. It might be possible to determine the number accurately by growing the parents, the F_1 and F_2 generations and a large number of F_3 generations under as uniform environmental conditions as possible. But even when only a limited number of F_3 generations are grown, it is possible to obtain an approximate idea of the factorial condition.

For the sake of illustration, let us first consider the inheritance of leaf number in the cross between Sumatra and Broadleaf given in Table IX. In this cross the parents differ by about six leaves per plant, the Broadleaf producing an average of about 20 leaves and the Sumatra an average of about 26 leaves. The F_1 generation was of intermediate value and no more variable as determined by the coefficient of variability than the parents, while the F_2 generation gave a range of variability equal to the combined range of the parents.

Of the nine F_3 generations, B – 1 – 14 is comparatively uniform. Only 56 variates of B – 1 – 14 were grown at Bloomfield, the calculated coefficient of variability being 7.18 ± .46, but 131 variates of this same selection were grown in New Haven and a variability coefficient of 6.44 ± .27 was obtained. Considering the large probable errors of these determinations it seems only fair to conclude that the coefficients of variability are really identical and that B – 1 – 14 is in a homozygous condition for leaf number. B – 1 – 10 is also rather uniform giving a variability coefficient of 7.75 ± .30. Of the remaining selections, four show coefficients of variability slightly greater than in F_2, one has about the same coefficient value as F_2 and two are of intermediate variability.

The results of this cross can be explained by supposing that the parental varieties are each pure for the same basal factorial formula for 20 leaves and that in addition the Sumatra has three independently inherited factors, each adding two leaves when homozygous and one when heterozygous.

Our gametic conditions for Broadleaf will be 20 aabbcc and for Sumatra 20 AABBCC. The F_1 formula will be 20 AaBbCc or 23 leaves, and in F_2 there will be a germinal variation from 20 to 26 leaves. With these gametic formulas we should expect one out of every eight F_3 generations to breed true. Of the nine F_3 generations given in Table IX, one gave a coefficient of variability of about the same value as the parents. That the F_3 generations gave different averages for leaf number may be seen by consulting our results.

All crosses cannot be explained in as simple manner as this one. In the case of inheritance of leaf number of cross (402×405) Havana×Cuban, the conditions are apparently more complex. Here both parents and F_1 gave an average of about 20 leaves per plant and about the same coefficients of variability. The F_2 generation was very variable, and of the five F_3 generations grown two proved as variable as the F_2, two were of intermediate variability, and one showed a coefficient of variability slightly larger than the parents or F_1. As selections were grown in F_3 which gave higher and lower leaf averages than the parents, the variability of F_2 must have been germinal. As only about 150 variates were counted and only five F_3 generations grown it is impossible to say definitely how many factors are involved.

If we suppose our parental formulas for leaf number to be 14 AABBCC and 14 DDEEFF, we will obtain a condition in F_1 of 14 AaBbCcDdEeFf or 20 leaves, and a germinal variation of 14 to 26 leaves in F_2. While this hypothesis may not be correct, the results can be explained by some such means.

In the inheritance of leaf shape of the cross between Havana and Cuban, the conditions are very simple. The data from this cross are given in Table XVI. The F_1 generation is shown to be intermediate in leaf shape and in F_2 there is segregation. Of the three F_3 generations given in the table, all are comparatively uniform, two having the Havana leaf shape and one the Cuban leaf shape. Two other F_3 generations were grown and although no statistical results can be given we know by observation that one selection had the Cuban leaf shape and the other had a variable leaf shape. These results can probably be explained by the use of a single factor.

It is not assumed that the factorial formulas here given are necessarily correct, as the conditions may be of a more complex nature, but we wish to show that some such mathematical description simplifies the breeding results in a manner that is helpful in actual practice.

GENERAL CONCLUSIONS.

Our results show that the F_1 generations of size crosses in tobacco are as uniform as the parents and of an intermediate value; that there is an increase of variability in F_2 and where sufficient variates are studied, a range of variation equal to the combined range of the parents; that certain F_2 individuals breed true in F_3, and that others give variabilities ranging in value from the parents to that of the F_2 generation.

These results can be explained in essentially the Mendelian manner — by the segregation of potential characters in the germ cells and their chance recombination — therefore, from the plant breeding standpoint there seems good reason for believing that quantitative characters are inherited in the same manner as qualitative characters.

The production of fixed forms which contain certain desirable plant characters is not, however, a simple problem, due to the large number of factors in which plants of different races differ and because a superficial resemblance does not necessarily mean a genetical resemblance. It is necessary to grow large F_2 generations and to save seed from those plants which most nearly conform to the desired type. Progeny of these F_2 plants should be grown in row tests in F_3 and selection continued in later generations until the desired form has been obtained. The length of time which it takes to produce a uniform type will depend largely on the number of variates which can be grown in F_2 and the number of row tests which can be grown in F_3.

Quality of cured leaf is a complex character and due to many conditions, environmental as well as inherited. There is also the added difficulty that the quality of leaf must conform to the trade ideals. The experiments here reported indicate that a good quality of leaf can more generally be expected in a hybrid, if the parents are both of high quality, than if one parent is a good variety and the other somewhat lacking.

It should be realized that the production of improved cigar wrapper types is not an easy problem and that desirable results cannot be obtained without the outlay of considerable time and money.

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PLATE I.

a. 403-1-1, Sumatra, averaged 26.2 leaves per plant.
New Haven, 1912.

b. Havana type plant, averages about 20 leaves per plant.

c. (403 x 402)-1, F_2 of cross between Sumatra and Havana. This plant could not be distinguished from pure Sumatra.
New Haven, 1912.

PLATE II.

a. (403 x 402)-1, F_2 of cross between Sumatra and Havana. Plant of Havana type.
New Haven, 1912.

b. (403 x 402)-1, F_2 of cross between Sumatra and Havana. Plant resembling F_1 generation.
New Haven, 1912.

c. (403 x 402)-1, a tall vigorous plant of the F_2 of a cross between Sumatra and Havana bearing 30 leaves.
New Haven, 1912.

PLATE III.

a. (403 x 402)-1, F_2 of cross between Sumatra and Havana. Plant bearing 28 leaves and short internodes. New Haven, 1912.

b. (403 x 402) -1, F_2 of cross between Sumatra and Havana. Plant bearing 19 leaves of Havana character. New Haven, 1912.

PLATE IV.

a. (403 **x** 402)-1, an F_2 plant of the cross between Sumatra and Havana, bearing 27 leaves and short internodes. A ''Halladay Havana'' type.
New Haven, 1912.

b. (403 x 402)-1, an F_2 plant of the cross between Sumatra and Havana, bearing 26 leaves. Another ''Halladay'' type.
New Haven, 1912.

At left, 12-1-1, a vigorous strain and at right, K-1-1-2, non-vigorous strain of Halladay Havana. Bloomfield, 1912.

PLATE VI.

At right (12-2)-1 which has erect, crinkly leaves, and at left (12-1)-1 which bears smooth, drooping leaves. These strains were produced from family No. 12 and are absolutely distinct. Bloomfield, 1912.

PLATE VII.

a. No. 401, Broadleaf. Averages about 19 leaves per plant. Leaves drooping in habit. Centerville, 1911.

b. (403 x 401)-1-5, an F_3 generation of a cross between Sumatra and Broadleaf which bred true for an intermediate leaf size and for the drooping leaf habit of the Broadleaf, and gave a mean leaf number of 22.0 ± .08 and a C.V. of 7.54 ± .27. New Haven, 1912.

PLATE VIII.

(403 x 401)-1-6, an F_3 generation of a cross between Sumatra and Broadleaf which gave a mean leaf number of 23.9 ± .08 and a C.V. of 6.61 ± .23. The size of leaf is as yet very variable. New Haven, 1912.

PLATE IX.

Average middle leaf of No. 403, Sumatra at left, of No. 401, Broadleaf at right and F_1 in center.

a. 402-1-1, Havana. Averages about 20 leaves per plant. Grown under cloth shade. Bloomfield, 1912.

b. (402 x 405)-1-3. An F_3 generation of a cross between Havana and Cuban which gave a mean leaf number of 18.4 and which bred true for the Havana leaf characters. Grown under cloth shade. Bloomfield, 1912.

PLATE XI.

a. 405-1-1, Cuban. Averages about 20 leaves per plant. Grown under cloth shade. Bloomfield, 1912.

b. (402 x 405)-1-2, an F_3 generation of a cross between Havana and Cuban which bred true for the Cuban leaf size and shape but was very variable for leaf number. Grown under cloth shade. Bloomfield, 1912.

c. (402 x 405)-1-5, a 30 leaved plant of the F_2 of a cross between Havana and Cuban. Grown under cloth shade at Bloomfield, 1911. The progeny of this plant gave a mean leaf number of 28.0 ± .28 leaves in 1912.

PLATE XII.

Middle leaves of the F_3 generation plants shown in plates X and XI. From left to right (402 x 405)-1-3, Havana segregate; 402-1-1, Havana; 405-1-1, Cuban; and (402 x 405)-1-2, which bred true for the Cuban leaf size and shape.

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