Quantitative Relationships of Carbon, Phosphorus, and Nitrogen in Soils

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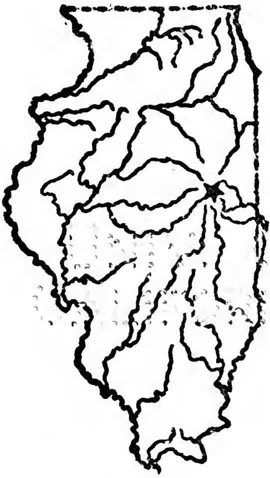

URBANA, ILLINOIS, APRIL, 1910

NOTE

Robert Stewart was born in American Fork, Utah, August 16, 1877. He secured his common school education in the public schools of Utah. In the fall of 1896 he entered the preparatory department of the Agricultural College of Utah. He graduated from this institution in June, 1902, with the degree of Bachelor of Science. He immediately received an appointment as assistant chemist in the Utah Experiment Station. While holding this position, during the years 1902-03, 1903-04, he took graduate student work in the college. During the school year 1904-05 he was a member of the Graduate School of the University of Chicago where he studied chemistry under the direction of Doctor Nef. In 1905 he was appointed assistant professor of chemistry in the Utah College and while holding this position, during the years 1905-06, 1906-07, 1907-08, he continued his graduate student work. During the summer of 1906 he was a member of the Graduate School of Agriculture held at the University of Illinois. In June, 1908, he was appointed professor of chemistry in the Utah College and was granted a leave of absence to carry on graduate work at the University of Illinois.

He is the senior author of Bulletin 103, "Milling Qualities of Utah Wheat," and Bulletin 106, "A Study of the Influence of Irrigation Waters upon the Movement and Production of Nitrates in the Soil," which has been accepted for publication by the Utah Experiment Station.

During the school year 1908-09 he held a fellowship in agronomy in the University of Illinois. He is a member of the Illinois chapter of Sigma Xi and also a charter member of the Illinois chapter of the American Society of Agronomy.

QUANTITATIVE RELATIONSHIPS OF CARBON, PHOSPHORUS AND NITROGEN IN SOILS*

By ROBERT STEWART

(A) HISTORICAL RÉSUMÉ

The literature on carbon, nitrogen and phosphorus in soils is voluminous. The résumé given herewith by no means attempts to include all that pertains to these elements in the soil, but it is confidently believed that it fairly represents the literature pertaining to this particular phase of the subject.

1. Carbon in Soils

Carbon may exist in soils as inorganic and organic carbon. The agricultural value of organic carbon, or organic matter, of soils has long been recognized by the practical husbandman, and the scientific man early recognized its value when the applications of science were made to agricultural problems.

Mülder (1) in 1844, made an elaborate study of the organic matter of the soil, and seems to have been the first one to suggest that it consisted of other elements than carbon, hydrogen and oxygen. He separated the organic matter into various supposed pure organic compounds of an acid nature, which were analyzed and studied by the usual organic method.

Wolff (2) determined the organic matter of the soil by calculation, by use of the factors 1.724 or 0.471: he multiplied the organic carbon by the former factor, or the total organic carbon dioxid by the latter. The factors were derived from the conception that "humus" contained 58 percent carbon.

Detmer (9) attempted to isolate "pure" humic acid from the soil and to study its properties. He obtained a fairly pure product which he studied and submitted to analysis.

A little later Grandeau (10) developed his well known method for determining the *matière noire* of soils, which he regarded as of great importance. He stated that soils owed their color and

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probably their fertility to its presence since it held in combination, phosphorus, nitrogen, and certain mineral elements.

Dehérain (22) determined the carbon content of soils from plots which had received different treatment. He found that the soils from the plots which had not been manured had lost over 50 percent of their carbon.

Kostytschiff (24) studied the humus obtained from substances of known origin and which were converted into humus under controlled conditions. He learned that even with the albuminous substances the carbon was lost more rapidly than the nitrogen, hence the ratio of carbon to nitrogen would be narrower in the resulting humus than in the original material.

Berthelot and André (31) found that 67.1 percent of the total carbon in soils was soluble in dilute alkalis but that over one-half of this soluble carbon, or 40 percent of the total carbon, was not precipitated from the alkalin solution by the addition of an acid.

Snyder (37) reported the results obtained by a study of the production and analysis of the humus obtained from such substances as cow manure, clover, meat scrap, etc., etc., which were converted into humus under known conditions. The carbon content of the humus varied from 41.95 percent in case of the humus produced from cow manure to 57.84 percent in case of the humus produced from cane sugar.

Hess (45) studied the effect of different systems of treatment on the humus of the soil. He found that the ratios of carbon to nitrogen and nitrogen to humus were not materially effected by the treatment applied.

André (47) studied the action of potassium hydroxid on the carbon compounds of the soil, mould, compost and peat. He determined the insoluble and soluble carbon: the latter he separated into two classes; the portion precipitated from alkalin solution by the addition of an acid, and the portion remaining in solution. The results obtained are expressed in Table 1.

TABLE 1.—PERCENTAGE OF SOLUBLE AND INSOLUBLE CARBON AND RATIO OF CARBON TO NITROGEN

		Peat		Compost		Soil		Mould	
		Ratio of C/N	Percent carbon of total	Ratio of C/N	Percent carbon of total	Ratio of C/N	Percent carbon of total	Ratio of C/N	Percent carbon of total
Insoluble portion		147.6	24.5	84.4	40.0	82.4	35.4	40.4	41.8
Soluble portion	a. Precipitated by acids	26.9	44.8	16.6	23.0	27.1	36.6	10.8	18.7
	b. Not precipitated by acids	17.7	30.7	9.8	37.0	16.7	28.0	10.1	39.5

The ratio of carbon to nitrogen in the original material was: peat 22.7; compost 15.0; soil 24.7 and mould 12.8. He concluded that the more insoluble the compound the wider was the carbon-nitrogen ratio. The potassium hydroxid showed a tendency to dissolve the compounds rich in nitrogen.

Pagnoul (51) found no fixed relation between the carbon and nitrogen of the soil but apparently the carbon, nitrogen, and humus varied in the same direction, altho irregularly.

Rimbach (53) concluded that, since the *matière noire* was readily nitrified it was the direct source of the nitrates of the soil and thus the insoluble carbon was of insignificant value.

Frear and Hess (54) found that lime caused a more rapid loss of carbon than of nitrogen in manured land, but the reverse in unmanured land.

Dyer (55) studied the carbon and nitrogen content and the relationship between carbon and nitrogen in the soil taken from 22 different plots of the Rothamsted experiment fields. These data are furnished for each individual 9-inch section to a depth of 90 inches.

The carbon and nitrogen contents of the higher depths were higher than those of the lower depths and the ratio of carbon to nitrogen is wider in the former. At the fifth to sixth depth the carbon and nitrogen contents seem to become fixed quantities and are apparently those derived from the original matter out of which the soil was formed.

A study of various clays and other material taken from great depths seemed to indicate that a nitrogen content of .04 percent was indigenous to the subsoil of the Rothamsted station.

Cameron and Breazeale (61) investigated the three general methods for determining the carbon content of the soil: namely, the "loss on ignition" method, the humus method and two forms of a combustion method. They concluded that the first two methods were unreliable: the first, because there was no apparent relationship existing between the results obtained and the true carbon content; the second, since it makes no pretense of giving the total carbon in the soil.

It is interesting to note that they reported that the ammoniacal extract contained so much suspended material that it was found undesirable to work with, until it was passed through a Chamberland-Pasteur filter, when a perfectly clear solution was obtained.

König (67) recently studied the influence of hydrogen peroxid on the organic matter of the soil. He found that it consisted of two parts, one easily oxidized by hydrogen peroxid, the other not oxidized by this reagent.

Hopkins and Pettit (68) reported the total carbon, nitrogen and phosphorus contents of a great number of samples of the soils of Illinois. This work is, thus, made the basis of calculating the relationship of carbon, nitrogen and phosphorus reported in part (B) of this thesis.

2. NITROGEN IN SOILS

The nitrogen in soils exists chiefly as organic nitrogen with a very small amount of inorganic nitrogen. The organic nitrogen may exist in some known and probably some unknown forms.

Mülder (1) believed the nitrogen, found in the humus, to be associated with the organic matter in the form of the ammoniacal salts of the various organic acids obtained by him.

Müller (4) thought he detected a tendency for the nitrogen to vary inversely as the carbon.

Detmer (9) believed that the nitrogen formed a definite compound with the organic carbon of the soil since the nitrogen could be liberated only with great difficulty and by the use of the most drastic chemical agents.

Simon (11) believed that the organic matter of the soil possessed the property of absorbing the free nitrogen of the atmosphere and of converting it into ammonia which in turn united with the organic acids in the form of their ammoniacal salts. Sostegni (19) a little later discussed the work of Simon and reported a series of experiments to prove that Simon's assumption was untenable.

Berthelot (19), in 1886, reported the carbon and nitrogen contents of calcareous clayey soil, originally very deficient in organic carbon and nitrogen but which was gradually increasing in carbon and nitrogen content owing to the action of diatoms.

Berthelot and André (20, 74, 75, 76) later carried on a series of experiments for the purpose of separating the organic nitrogenous material into its various compounds. They reported the amount of total, nitric, amido and ammoniacal nitrogen present in the soil.

Eggertz (21) differed very materially from Mülder. He concluded that Mülder's contention, that the nitrogen associated with the organic matter of the soil existed only as the ammoniacal salts of the various organic acids, was untenable. If the nitrogen existed simply as the ammoniacal salt of the humic acid, treatment with hydrochloric acid should liberate *all* the nitrogen as ammonia, which, experimental evidence showed, was not the case.

Furthermore artificial humic acid, treated with ammonia, did form ammonium humate which, however, was readily decomposed by treatment with a mineral acid; yet, if this artificial humic acid be *heated* in a current of ammonia gas, combination took place and the resulting compound could not be decomposed by treatment with mineral acids. He, therefore, concluded that the nitrogen formed an integral part of the humic acid radical.

Berthelot and André (26) studied artificial humic acid prepared out of sugar. This acid formed salts with various bases, which were easily decomposed again by treatment with an acid, *except* in the case of the *ammonium salt,* the nitrogen of which could not be entirely liberated by this treatment. They concluded that the nitrogen in part at least, formed an integral part of the humic acid radical.

Hilgard and Jaffa (30), in 1892, propounded their well known view regarding the importance of the nitrogen associated with the extracted *matière noire.*

Berthelot and André (31) regarded the organic matter of the soil as of great importance since it prevented the loss of nitrogen thru drainage since the nitrogen was held in insoluble combination in the organic matter.

Fulmer (38) determined the humic nitrogen in 53 samples of Washington soil and attempted to work out the relationship between carbon and nitrogen by means of the formula $c = \frac{b \text{ x } 55}{a}$, where c= the percentage of nitrogen in the *matière noire;* b= the percentage of the total soil nitrogen; a= the percentage of humus. By means of this formula the 53 samples of soil were separated into three classes; the first class contained 19 samples in which the variation in the humic nitrogen calculated by means of the formula was within one percent of the analytical result; the second class contained 10 samples and the variation was from one to two percent; the third class contained 24 samples and the variation was anywhere over two percent. These results furnished good evidence that no one given relation would hold for all soils.

Wheeler (48) found that lime or gypsum caused a decrease in the amount of humus but that the percentage of humic nitrogen was increased. Similar results were obtained by Frear and Hess (54) on manured land.

Dojarenko (56) recently studied the "humic" nitrogen of soils. He determined the total, humic, amid, ammoniacal and amido nitrogen in seven samples of black Russian soils. The results are reported in Table 2.

TABLE 2.—PERCENTAGE OF TOTAL HUMIC, AMIDO, AMID AND AMMONIACAL NITROGEN IN HUMUS

	Percent in dry substance				Percent of total quantity of nitrogen		
No.	Total humic nitrogen	Amido nitrogen	Amid nitrogen	Ammoniacal nitrogen	Amido nitrogen	Amid nitrogen	Ammoniacal nitrogen
1	2.735	1.34	0.31	0.04	49.09	11.38	1.46
2	3.38	1.81	0.41	0.08	53.55	12.13	2.36
3	2.64	1.30	0.29	0.02	49.20	10.99	0.80
4	3.33	2.34	0.32	0.03	70.27	9.61	0.90
5	4.58	1.01	0.48	0.06	22.01	10.46	1.31
6	3.65	1.26	0.27	0.07	34.52	7.40	1.90
7	4.02	1.96	0.22	0.03	48.75	5.47	0.78

This did not account for all of the nitrogen present and so the question arises in what form does the remainder exist?

D'Utra (70) found that the humic nitrogen showed wide variations.

Hilgard (71) reported the average humic nitrogen of 466 samples of soil from the humid regions as 5.45 percent, while the average of 313 samples of soil from the arid section was 15.87 percent. Later (73) he found that the average humic nitrogen for 696 samples of humid soil was 5.00 percent, while that of 573 samples of arid soil was 15.23 percent. It must be remembered, however, that the total quantity of nitrogen of the two regions is in the inverse order. The total nitrogen of the uplands and lowlands of California for example, is 0.101 percent and 0.101 percent respectively, while the total nitrogen of the ordinary brown silt loam soils of the corn belt in Illinois varies from 0.218 percent to 0.337 percent.

3. PHOSPHORUS IN SOILS

The phosphorus of the soil may exist in the inorganic and organic condition. The greater part is in the inorganic form with an unknown amount in the organic state. The form and amount of the organic phosphorus is uncertain, and, indeed it has been questioned, especially during recent years, whether or not organic phosphorus occurred in the soil to any appreciable extent.

Mülder (1), as early as 1844, noted that the organic material was not readily freed from phosphorus.

The work of Thénard, Schutzenber (5, 6, 7) showed that union may take place between various forms of artificial humus and phosphates under certain conditions and indicated that combination may possibly take place in the soil between organic carbon and inorganic phosphorus.

Detmer (9) in the preparation of his "pure humic" acid, noted that the material could be freed from phosphorus only with great difficulty.

Grandeau (10) regarded the phosphorus associated with the extracted *matière noire* as being of the greatest importance, and, was probably in special combination with the organic matter. He regarded it as an index of the fertility of the soil.

Simon (11) believed that he had demonstrated that union took place between organic matter and phosphates. When freshly precipitated humic acid was suspended in water and digested with calcium phosphate and then filtered, the filtrate showed an excess of phosphoric acid: this excess, he concluded must be in union with the organic matter in solution. He thought that a double compound of ammonia and phosphorus existed in the soil.

Schultz (12) showed that the addition of humus to "Basaltböden" increased the absorption ability of the soil for phosphates.

Eichhorn (13) repeated some of Simon's work and concluded that organic combination did not take place as indicated by Simon but that the humus had decomposed the tri-calcium phosphate with the formation of acid phosphate.

Pitsch (14) determined the solubility of the various mineral phosphates, including iron and aluminium phosphates, in a solution of *ammonium humate itself.* He concluded, that, since this solution exerted a solvent action on mineral phosphates, the ammonia extract of the soil contained phosphorus, other than that originally associated with the organic matter in the soil and probably part, at least, of the ammonia soluble phosphorus was derived from the iron and aluminium phosphates.

M. P. DeGasparin (15) found in calcareous clay soil five percent of the total phosphorus in organic combination. He noted, furthermore, that the mosses and lichens contained from 5 to 6 times as much phosphorus as the rocks on which they grew; the soil formed therefore, from the debris of these plants should be relatively richer in phosphorus and should have a part of its phosphorus in combination with carbon in the organic material.

Eggertz (21) found that the ammoniacal extract of the soil, when treated with an acid, formed a precipitate of organic matter which always contained phosphorus. He concluded, therefore, that part of the phosphorus of the soil was united to the carbon in organic combination.

Later, Eggertz and Nilson demonstrated that the amount of phosphorus soluble in dilute mineral acids showed a marked increase after ignition of the soil. Ignition rendered 10 times as much phosphorus soluble in 2 percent hydrochloric acid. They attributed this to the destruction of the organic matter which had held the phosphorus in combination which would not yield up its phosphorus to acids.

Van Bemmelen (23) believed that the iron, calcium, silica, phosphoric acid, etc., found in the ash of the *matière noire* by Eggertz, were not originally chemically combined to carbon in the organic matter of the soil but were absorbed by the precipitated gelatinous *matière noire.* According to Van Bemmelen the phosphorus existed in the soil principally as calcium phosphate with a very small quantity occurring in the absorbed state in the form of a *colloidalen Humate-Silicat-Komplex.*

Two questions seemed to be of paramount importance to Wiklund (25) regarding the work of Eggertz: first, was the amount of the ammonia-soluble phosphorus obtained from different soils constant? Second, did the phosphorus exist in the mullkörpers (*matière noire* of Grandeau) in chemical combination with carbon, or simply as absorbed phosphorus? He concluded that there was a tendency for the ammonia-soluble phosphorus to be constant in different soils. He showed, further, that one digestion with 12 percent hydrochloric acid did not completely remove all of the acid soluble phosphorus, but a second and even a third digestion still removed some phosphorus. Now, he reasoned, if the phosphorus removed by the second and third digestion was simply extracted from the absorbed phosphorus, extraction of the soil with ammonia after the first digestion with hydrochloric acid, should yield a solution of *matière noire* containing a higher phosphorus content than when the soil was completely extracted with the hydrochloric acid. Such, however, was not the case, therefore, the phosphorus did not exist as absorbed phosphorus and must be in combination with carbon in the organic matter.

Snyder (34) noted that some phosphorus, iron, etc., were extracted with the *matière noire* but he did not seem to think at this time that there was any evidence of combination with carbon. About the same time he observed the rapid loss of phosphorus associated with the humus in continuous cultivated soil.

According to Berthelot and André (27) phosphorus may be found in the soil (a) in inorganic or mineral phosphates, (b) in organic ethers and (c) in organic or mineral compounds not readily decomposed.

Schmoeger (29) reviewed the rival claims of Eggertz and Nilson, and Wiklund on the one hand, and Van Bemmelen on the other, regarding the phenomenon of ignition rendering the phosphorus of peaty soil more readily soluble in acids.

It seemed possible to Schmoeger that the soil might possess such a tenacious absorbent power for phosphorus that it would not yield up its phosphorus to acid treatment before ignition. But he deduced experimental evidence to show that such was not the case.

Digestion of the soil itself and also the extracted *matière noire* with a solution of potassium hydrogen phosphate failed to add any phosphorus which was not again recovered by treatment with hydrochloric acid. This was conclusive evidence to Schmoeger that the phosphorus did not exist as absorbed phosphorus and must, therefore, exist in organic combination. Two possibilities suggested themselves to Schmoeger: first, the phosphorus existed in the form of lecithin; second, it existed as nuclein. Lecithin was found to be present only in traces. The characteristic property of nuclein to "split-off" its phosphorus in the form of phosphoric acid, when heated, under pressure in the presence of water, to a temperature of 150°-160° was utilized by Schmoeger. The soil under examination, treated in this way, yielded as much soluble phosphorus as did the ignited soil. This experimental evidence led him to conclude that nuclein or some closely allied bodies were present in the soil.

Later Schmoeger (39) confirmed his previous work and produced additional evidence in favor of his view that nuclein or similar bodies existed in the soil. Table 3 shows some of the results obtained.

TABLE 3.—PERCENTAGE OF SULFURIC ACID AND PHOSPHORUS SOLUBLE IN DILUTE ACID

	Percent	
	Sulfuric Acid	Phosphorus
Soil in original state	0.122	0.043
Evaporated soil	0 290	0.083
Ignited soil	0.939	0.095

Since sulfur is regarded by many authors as being a constituent of plant nuclein, the increased solubility of this substance together with the phosphorus when the soil was treated as indicated above, was regarded as evidence in favor of his assumption.

In a later article (40) he showed, by similar treatment, that analogous bodies existed in the moor grass out of which the moor soil was formed. This was regarded as additional evidence in favor of his view.

Tacke (33) observed that the drying out of soil rendered the phosphorus available. There were three possible explanations suggested to him: first, the phosphorus existed in the soil in organic combination which was destroyed by the process of drying; second, it existed in the soil in the colloidal form as suggested by Van Bemmelen; third, the drying out of the soil gave rise to substances of a strong acid nature which acted upon the insoluble phosphorus compounds rendering the phosphorus soluble.

In a later article (42) he showed that very little water soluble phosphorous existed in the soil under consideration, but that drying at 70°-80° rendered over 50 percent of the total phosphorus soluble in water.

Snyder (36) reported results of a confirmative nature regarding the phosphorus associated with the humus in virgin and cultivated soils.

Later he (37,41) studied the product obtained by the conversion of known substances, under known condition, into humus. The ash of the *matière noire* obtained from this material contained phosphorus, among other substances, and according to Snyder: "There is every indication that these elements are in organic combination with the carbon, hydrogen and oxygen of the humus." As regards the question whether or not the humus united with the inorganic phosphorus of the soil, he concluded that his experimental evidence showed that such union did take place.

Nannes (49) found that a well decomposed peat soil contained 0.166 percent phosphorus. He found that 0.057 percent of phosphorus was extracted with the *matière noire.* When the ammoniacal solution of the *matière noire* was treated with hydrochloric acid, 0.039 percent of the phosphorus was found in the organic precipitate. He also attempted to isolate a definite organic phosphorus compound and he believed that he detected the presence of lecithin and chlorophyllan.

Ladd (43) found in a study of eight samples of different soil that an average of 41 percent of the phosphorus was associated with the extracted *matière noire;* the variation, however, was from 10 percent to 90 percent.

In a later article (44) he showed that as the humus of the soil increased the phosphorus associated with the extracted *matière noire* also increased. From the fact that the organic precipitate, formed by neutralizing the ammoniacal extract, contained the phosphorus he concluded that it existed in the soil in organic combination, but just what the relationship was not clear.

Emmerling (52) believed that there were four forms of phosphorus in the soil, one of which was phosphorus in organic combination.

Rimbach (53) found 6.15 percent P_2O_5 in the ash of the *matière noire* which was precipitated from the ammoniacal solution by the addition of gypsum and magnesium sulfate.

Nagaoka (57) found that ignition of the soil for fifteen minutes at a faint red heat materially increased the availability of the phosphorus. He attributed this action to the destruction of the humophosphates.

Asō (58) confirmed, in a general way, the results obtained by Schmoeger. He also found 0.049 percent of lecithin in the soil. He drew the following conclusions:

1. Phosphorus existed in the soil as inorganic and organic compounds.
2. The organic phosphorus material was principally nuclein with a very small part of lecithin.
3. Ignition rendered the phosphorus in organic combination available.

Hartwell and Kellogg (60) found that an average of one-half of the phosphorus was associated with the organic matter in the soil taken from four plots which had received different treatment.

Dumont (62) studied a complete manure, the composition of which was as follows: soluble matter (in dilute alkali) 50.4 percent; insoluble matter 49.6 percent; total nitrogen 1.6 percent; total phosphorus 1.27 percent.

The soluble portion contained 35 percent of the nitrogen and 46 percent of the phosphorus. In order to obtain data upon the state of combination of the phosphorus, the ammoniacal solution of *matière noire* was treated with various reagents with the result (recalculated to the element basis) shown in Table 4.

Table 4.—Distribution of Phosphorus When Matiere Noire is Precipitated

Precipitating agent	Phosphorus	
	In precipitate	In filtrate
Citric acid	0.383	0 203
Hydrochloric acid	0.386	0.199
Ferric chlorid	0.532	0.053
Aluminium sulfate	0.566	0.019
Calcium chlorid	0.584	0.0009

These results furnished conclusive proof to Dumont that a part of the phosphorus of the soil was in organic combination.

Later (64) he obtained better cultural results from application of humic phosphatic manures than from mineral phosphatic manures and better even than from barnyard manure, which he attributed to the phosphorus in organic combination.

In a still later article (65) he said that the organic phosphorus was derived from two sources: first, from the nuclein and lecithin of the decaying vegetable and animal debris; second from the union of the humus with the water soluble phosphates of the soil.

Evidence of the latter contention was obtained by precipitating the *matière noire* in the presence of potassium hydrogen phosphate by different reagents as indicated in Table 5.

TABLE 5.—AMOUNT OF PHOSPHORUS IN PRECIPITATED HUMUS AND FILTRATE

Precipitating agent	Series A Phosphorus introduced=.087		Series B Phosphorus introduced=.218		Series C Phosphorus introduced=.437	
	Phosphorus in humus	Phosphorus in filtrate	Phosphorus in humus	Phosphorus in filtrate	Phosphorus in humus	Phosphorus in filtrate
Acetic acid.......	0.056	0.031	0.057	0.160	0.058	0.374
Citric acid........	0.054	0.033	0.055	0.163	0.054	0.381
Hydrochloric acid	0.057	0.030	0.059	0.158	0.059	0.378
Sulfuric acid.....	0.057	0.030	0.061	0.156	0.062	0.372

Altho the amount of dipotassium phosphate added to the solution had increased, the amount of phosphorus absorbed by the humus was practically constant, due to the formation of definite "composés phospho-humique."

König (67) found that hydrogen peroxid oxidized from 40 percent to 70 percent of the humus present in the soil and that much more of the phosphorus was soluble in pure and carbonated water after oxidation than before, due, he believed, to the destruction of the organic phosphorus compounds.

Fraps (69), quite recently, made a study of the phosphorus extracted from the soil by 4 percent ammonia in the usual determination of humus. He confirmed Pitsch's results regarding the possibility of some of the ammonia-soluble phosphorus being of inorganic origin. He separates the ammonia-soluble phosphorus into three classes as follows:

1. The phosphorus associated with the clay held in suspension in the liquid.
2. The phosphorus precipitated with the organic matter when the solution was neutralized with an acid.
3. The phosphorus which remained in solution after the precipitation of the organic matter.

With the soils under consideration he found that 1/9 of the ammonia-soluble phosphorus was in the first class, 1/3 was in the second class and 5/9 was in the third class.

The phosphorus found in the first class was assumed to be associated with the clay particles as iron and aluminium phosphates. He concluded that the phosphorus precipitated with the organic matter from the ammonical solution by the addition of acids was in organic combination. The phosphorus remaining in the mother liquor was assumed to be derived from the iron and aluminium phosphates of the soil.

Mooers and Hampton (77) recently proposed a method for obviating the error introduced in the humus determinations by the suspended clay. They claimed that filtration through the Chamberlain-Pasteur filter, as suggested by Cameron, introduced a serious error inasmuch as the filter absorbed some organic matter. They proposed an evaporation method: by evaporation of the ammoniacal extract to dryness re-dissolving in ammonia and filtration, several times, a perfectly clear solution was obtained. Determination of the humus in this filtrate gave very concordant results.

Hopkins and Pettit (68) found that in certain soils the mineral composition had a tendency to be constant in the surface, subsurface, and subsoil. This was indicated by the uniform potassium content of the surface, subsurface, and subsoil and by the fact that different samples of surface soil of the same type showed a wide variation in the phosphorus content but that this variation largely disappeared in the subsoil. The potassium exists in the soil in the inorganic form, the nitrogen exists chiefly in the organic form while the phosphorus may exist in the inorganic and organic state. They suggested, therefore, a method for calculating the phosphorus in the organic state in the surface soil. The difference in amount of nitrogen in the surface soil and subsoil, and the difference in the amount of phosphorus in the surface soil and subsoil gave apparently the amount of nitrogen and phosphorus associated together in organic combination. By means of this ratio and the total amount of nitrogen in the surface soil the total amount of organic phosphorus present in the surface soil could be calculated.

4. CARBON AND NITROGEN CONTENT OF FUNDAMENTAL ROCKS

The fundamental rocks out of which soils have been formed contain an appreciable amount of carbon and nitrogen which is indigenous to them.

Dellese (3) discovered that mineral matter, crystalline, sedimentary and eruptive contained carbon associated with nitrogen. This mineral matter, which was formed under similar conditions of temperature, pressure, etc., had a tendency to contain a constant amount of carbon and nitrogen.

The work of Lawes and Gilbert (16), Dyer (55) and Hall and Miller (66) on the clays and other fundamental rock material taken from various great depths indicated that an appreciable amount of carbon and nitrogen was indigenous to the underlying soil material.

(B) EXPERIMENTAL PART

The starting point of such an investigation consists of a consideration from a mathematical point of view of the existing data regarding the relationships of carbon, phosphorus and nitrogen of the soil.

I. MATHEMATICAL

(a) INFLUENCE OF AGE UPON THE NITROGEN-CARBON RATIO

From the data available in the literature it is possible to determine within certain limits the influence of age upon the nitrogen-carbon ratio in soils. From the average results of a number of nitrogen determinations (68) and the carbon content obtained by calculation from the proximate analysis, it is possible to determine the approximate nitrogen-carbon ratio in the more common humus producing materials. The results obtained in this way will be found in Table 6. The materials naturally fall into two groups: in the first group the ratio varies from 1:52.2 for corn stover to 1:84.1 for wheat straw; in the second group the variation is from 1:16.7 for alfalfa hay to 1:35.4 for timothy hay.

TABLE 6.—APPROXIMATE NITROGEN-CARBON RATIO IN THE MORE COMMON HUMUS PRODUCING MATERIALS

Kind of material	Carbon to 1 of nitrogen
Corn stover	52.2
Oat straw	67.8
Wheat straw	84.1
Timothy hay	35.4
Clover hay	21.3
Cowpea hay	19.5
Alfalfa hay	16.7
Albumin	3 2
Zein	3.4
Nuclein	2.8

In Table 6 will also be found the nitrogen-carbon ratio in some of the compounds which might be expected to be found in humus. The ratio is very narrow and does not vary much from 1:3.

The next step in the study in the influence of age upon the nitrogen-carbon ratio would be to determine the ratio in as fresh humus as possible from known materials. Snyder (35) in his study of the production of humus from known materials placed a weighed quantity of the material together with a weighed quantity of soil having a low humus content in a box and set aside for one year. At the end of the experiment the humus was extracted and the carbon and nitrogen determined in the *matière noire.* At first thought this would appear to furnish the desired information, but unfortunately no check was run with the untreated soil, so no correction can be made for the carbon and nitrogen which may have been converted into humus from the unhumified material of the soil. This is evidently not a quantity which can be ignored since the humus content of the original soil is .06 percent while the total nitrogen of the soil is .02 percent showing that considerable unhumified organic matter was present, otherwise the humic nitrogen would be 33.33 percent while it has been shown (71) that the humic nitrogen would more probably be nearer 5 percent. In addition, the fact that the humus obtained from sugar contains some nitrogen is evidence that some of the unhumified organic matter of the soil has been converted into humus, since sugar does not contain nitrogen. The results, however, will be found in Table 7. In the first five substances the variation is from 1:21.7 for oat straw to 1:4.5 for meat scraps.

TABLE 7.—MINNESOTA SOIL STUDIES: HUMUS PRODUCTION FROM KNOWN MATERIALS

Material used	Percent			Ratio of carbon to 1 of nitrogen
	Humus	Carbon in humus	Nitrogen in humus	
Original soil	0.06	?	?	?
Cow manure	0.58	41.95	6.16	6.8
Clover	0.37	54.22	8.24	6.6
Meat scraps	0.31	48.77	10.96	4.5
Oat straw	0.46	54.30	2.50	21.7
Flour	0.47	51.02	5.02	10.2
Saw dust	0.59	49.28	0.32	153.8
Sugar	0.32	57.84	0.08	741.0

The large number of carbon and nitrogen determinations made in the soils of Illinois (68) rendered it possible to determine the nitrogen-carbon ratio, not only for the surface soil, but also for the subsurface and subsoil. The average of 19 determinations for the soil type, gray silt loam on tight clay, gave the ratios 1:10.4, 1:8.8, and 1:7.6 for the surface, subsurface, and subsoil respectively. The ordinary brown silt loam soils as an average of 68 determinations gave a nitrogen-carbon ratio of 1:12.1, 1:11.5 and 1:8.9 for the surface, subsurface, and subsoil respectively. The black clay loam soils as an average of 25 determinations gave 1:11.7, 1:11.9 and 1:9 respectively in the surface, subsurface, and subsoil. The peat soil as the average result of 5 determinations gave 1:11.8 and 1:12.9 for the surface and subsoil respectively.

TABLE 8.—RATIOS OF CARBON AND NITROGEN IN ILLINOIS SOILS

Soil type No.	Soil types	No. of analysis on which calculations are based	Carbon to 1 of nitrogen		
			Surface	Sub-surface	Subsoil
330	Gray silt loam on tight clay........	19	10.4	8.8	7.6
426	Brown silt loam.....	11	12.5	11.7	9.6
526	Brown silt loam.....	8	13.2	12.9	8.7
626	Brown silt loam.....	6	11.4	10.5	8.7
726	Brown silt loam.....	4	11.9	11.1	8.8
1126	Brown silt loam.....	30	11.9	11.5	8.6
1026	Brown silt loam.....	9	12.0	11.5	9.3
	Averages	68	12.1	11.5	8.9
420	Black clay loam.....	7	12.2	12.2	8.9
520	Black clay loam.....	5	12.4	12.2	11.4
1120	Black clay loam.....	11	11.1	11.1	8.3
1220	Black clay loam....	2	11.2	12.2	7.4
	Averages	25	11.7	11.9	9.0
1401	Deep peat........ ..	5	11.8	12.9	12.9

The Rothamsted work furnished information regarding the nitrogen-carbon ratio of the soil in nine inch sections to a depth of 90 inches. These results will be found in Tables 9 and 10. The ratio for the Broadbalk wheat fields varies from 1:9.5 to 4.8 for the surface and ninth 9 inches respectively. After the fifth 9 inches there is very little change in the ratio. In the Hoosfield barley soils the ratio varies from 1:10.6 to 1:8.8 for the surface and subsoil respectively.

TABLE 9.—BROADBALK WHEAT SOILS: RATIO OF CARBON TO NITROGEN

Depth	Percent		Carbon to 1 of nitrogen
	Carbon	Total nitrogen	
First 9 inches (all plats)................	1.155	.1222	9.5
Second 9 inches........................	.640	.0784	8.2
Third 9 inches........................	.492	.0666	7.4
Fourth 9 inches	.339	.0511	6.6
Fifth 9 inches..........................	.279	.0472	5.9
Sixth 9 inches..	.256	.0430	5.9
Seventh 9 inches......................	.248	.0420	5.9
Eighth 9 inches........................	.215	.0396	5.4
Ninth 9 inches...................... ..	.189	.0391	4.8
Tenth 9 inches.................	.188	.0375	5.0

TABLE 10.—HOOSFIELD BARLEY SOILS: RATIO OF CARBON TO NITROGEN

Depth	Carbon to 1 of nitrogen
First 9 inches......	10.6
Second 9 inches......................	8.8
Third 9 inches.	8.8

Hall and Miller (66) reported the carbon and nitrogen content, and ratio of carbon to nitrogen in samples of various materials taken from such great depths as to preclude all possibility of weathering. Since the nitrogen was always found to be associated with carbon it was regarded as being of organic origin and as being derived in part from the organic matter present in the clay at the time of its deposit. These results are shown in Table 11.

TABLE 11.—CARBON AND NITROGEN IN UNWEATHERED ROCKS

No. of soil	Formation	Depth at which sample was taken, ft.	Percent		C / N Ratio
			Organic carbon	Nitrogen	
1	Lower Bagshot Sand, Weybridge..	18—20	0.02	.00384	5.4
2	Upper Greensand, Farnham, Surrey	30	0.032	.00718	4.5
3	Folkestone Beds, Brabourne, Kent.	20	0 019	.00453	4.2
4	Lower Greensand, Sevenoaks......	30	0.076	.00881	8.7
5	London Clay, London..............	130	0 391	.041	9.5
6	Gault Clay, Nackholt, Kent........	18	0 427	.0415	10.3
7	Weald Clay, Pluckley. Kent.......	30	0 135	.0647	2.1
8	Carboniferous Shale, Barnsley....	1236	1.938	.137	14.1
9	Lower Gault, Dover...............	280—400	0.172	.0325	5.3
10	Oxford Clay, Dover...............	920	0.548	.0528	10.4
11	Kimmeridge Clay, Welton Lincs...	246	2.139	.107	20.0
12	Kimmeridge Clay, Dover	570	0.387	.0455	8.5
13	Lower Lias, Micketon, Glos.......	700	1.120	.0803	13.9
14	Clay with Flints, Harpenden	10	0 577	.0294	19.6

NOTES

1. A grey coarse sand.
2. Pale grey fine sandy rock.
3. Coarse yellowish sand.
4. Fine yellowish sand.
5. Solid grey clay.
6. Solid dark green clay.
7. Close grey and red mottled clay.
8. Hard grey shale.
9, 10, 12. Hard grey clays from the coal pit shafts at Dover.
11. Hard grey clay.
13. Hard grey clay.
14. Reddish sandy brick earth.

It will be seen from a study of the above tables that under normal conditions the nitrogen-carbon ratio of the soil has a tendency to become narrower as the age of the organic material increases. The ratio, however, never becomes narrower or ever equal to the ratio of the more common proteins contained in the humus producing materials.

(b) CARBON, NITROGEN AND PHOSPHROUS IN ILLINOIS SOILS

Before discussing the phosphorus-carbon and phosphorus-nitrogen ratios in the soil it seemed desirable to determine as closely as possible these ratios in fresh material out of which humus might be formed.

TABLE 12.—APPROXIMATE CARBON-PHOSPHORUS AND NITROGEN-PHOSPHORUS RATIOS IN THE MORE COMMON HUMUS PRODUCING MATERIALS

Kind of material	Carbon to 1 of phosphorus.	Nitrogen to 1 of phosphorus
Corn stover	417	8.0
Oat straw	420	6.2
Wheat straw	525	6.2
Timothy hay	283	8.0
Clover hay	171	8.0
Cowpea hay	181	9.3
Alfalfa hay	186	11.1
Nuclein	3.7	1.4

The ratios in the more common humus producing materials calculated from the average of a number of analysis for nitrogen and phosphorus (68), will be found in Table 12. In the coarser material the phosphorus-carbon ratio varies from 1:417 to 1:525: the phosphorus-nitrogen ratio is more constant being 1:6.2 and 1:8. In the hays, the phosphorus-carbon ratio varies from 1:186 to 1:283 while again the phosphorus-nitrogen ratio is more constant, the variation being 1:11.1 to 1:8. In nuclein the ratios are 1:3.7 and 1:1.4 respectively.

In Table 13 will be found the phosphorus-carbon and phosphorus-nitrogen ratios obtained by calculation from the Minnesota Soil Studies. The phosphorus determinations which were reported as phosphoric anhydrid were first recalculated to the element basis. Both ratios, it will be observed, are very narrow.

In Table 14 will be found the phosphorus-carbon and phosphorus-nitrogen ratios in Illinois soils calculated from the data reported by Hopkins and Pettit (68). The average of 7 calculations of the gray silt loam on tight clay gave the phosphorus-carbon and phosphorus-nitrogen ratios as 1:42.6 and 1:13.8 respectively.

TABLE 13.—MINNESOTA SOIL STUDIES: CARBON-PHOSPHORUS AND NITROGEN-PHOSPHORUS RATIOS

Kind of material	Carbon to 1 of phosphorus.	Nitrogen to 1 of phosphorus
Original soil	?	?
Cow manure	9.8	1.4
Clover	11.2	1.7
Meat scraps	9.7	2.2
Oat straw	38.2	1.8
Flour	22.0	2.2
Saw dust	37.0	0.24
Sugar	42.4	0.06

TABLE 14.—ILLINOIS SOILS: ORGANIC PHOSPHORUS; RATIOS OF CARBON TO PHOSPHORUS, NITROGEN TO PHOSPHORUS AND CARBON TO NITROGEN, FACTORS FOR CALCULATING THE ORGANIC PHOSPHORUS.

Soil type No.	Number of analysis on which calculations are based	Organic phosphorus as per cent of total phosphorus	Ratios of		Factors for calculating the organic phosphorus from the organic carbon
			Carbon to 1 of organic phosphorus	Nitrogen to 1 of organic phosphorus	
330	7	24.4	142.	13.8	0.007012
426	9	34.4	132.	10.6	0.007570
526	8	29.5	185.	14.0	0.005393
626	5	13.9	298.	25.1	0.003348
726	4	38.1	133.	10.5	0.007491
1126	24	40.9	116.	9.9	0.008583
1026	9	44.9	125.	10.9	0.007988
Average	59	33.5	165.	13.5	0.006053
420	7	36.0	134.	10.3	0.007441
520	5	33.8	260.	12.0	0.003836
1120	11	46.2	90.	8.3	0.005907
1220	2	32.4	169.	15.2	0.002956
Average	25	37.1	163.	11.4	0.006113
1401	4	100.0	230.	19.6	0.004311
1401	(b) 5	100.0	338.	26.5	0.002956

(b) Subsoil (7″—40″)

The ordinary brown silt loam soils, as an average of 59 determinations, gave the ratios of 1 :165.2 and 1 :13.5 respectively. The black clay loam soils, as an average of 25 calculations, gave the ratios of 1 :163 and 1 :11.4 respectively. The ratios in the surface peaty soil, assuming all the phosphorus to be in the organic state, are 1 :230 and 1 :19.6. The ratios in the subsoil of the peaty soil are somewhat wider, being 1 :338 and 1 :26.5 respectively.

(c) FACTORS FOR CALCULATING ORGANIC PHOSPHORUS

By means of the carbon-phosphorus ratios established as indicated above, it is possible to develop factors for calculating the amount of the organic phosphorus in the surface soil from the total organic carbon. For example the carbon-phosphorus ratio, 1 :163, in the black clay loam soils means that for every part of organic phosphorus there are 163 parts of carbon or for each part of organic carbon there are 0.006113 parts of organic phosphorus. Hence by multiplying the amount of organic carbon by the latter number the amount of organic phosphorus may be obtained.

The factors as developed will be found in the last column of Table 14. It is hoped that they will be of value in drawing broad general conclusions regarding organic phosphorus of the soil from a number of analyses. The variations in the various samples considered in any single type are too great to permit the utilization of the factors in isolated cases.

It will be observed that from 1/4 to 2/5 of the total phosphorus of the several soil types considered is in organic combination. These results indicate that a larger amount of phosphorus is in organic combination than the work of some American investigators would lead us to believe.

2. CHEMICAL

(a) ANALYTICAL RESULTS OF SOIL FROM ILLINOIS SOUTH EXPERIMENTAL FARM

A sample of soil for a study of the organic phosphorus, by the available methods for the determination of the organic phosphorus of the soil, was obtained from the Illinois South Experimental Farm. This soil is the ordinary brown silt loam soil of the corn belt.

The total potassium, carbon, nitrogen, and phosphorus in the surface and subsoil were determined by the methods adopted by the Illinois Experiment Station. The results, expressed as pounds per two million pounds of dry soil, will be found in Table 15. The average potassium content of 36,700 pounds, and 37,070 pounds in the surface and subsoil indicate a constant mineral composition, hence the calculation method may be safely applied for the determination of the organic phosphorus. The amount of organic phosphorus, the various ratios and the developed factor will be found in Table 16. It will be seen that 46 percent of the total phosphorus in this soil is in organic combination.

TABLE 15.—ANALYTICAL RESULTS OF SOIL FROM ILLINOIS SOUTH EXPERIMENTAL FARM: RESULTS EXPRESSED AS POUNDS IN TWO MILLION POUNDS OF DRY SOIL

Soil No.	Soil stratum	Potassium	Carbon	Nitrogen	Phosphorus*
1A	Surface	36280	41800	3760	938
1B	Surface	37120	42180	3844	901
Average	Surface	36700	41990	3802	919*
3A	Subsoil	36600	.....	1735	702
3B	Subsoil	37540	......	1680	670
Average	Subsoil	37070	......	1708	686

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*Later inspection of the strip of land from which this sample was taken led to the conclusion that it may have been modified by a previous disturbance of the soil in putting in tile drainage; and a composite sample was subsequently collected at points a short distance from the tile drain which showed 985 pounds of phosphorus instead of 919 pounds. The possible influence of this difference should be kept in mind. Thus the percent of total phosphorus in organic form would be reduced from 46 percent to 43 percent.—C. G. Hopkins.

TABLE 16.—ORGANIC PHOSPHORUS: RATIOS OF CARBON TO ORGANIC PHOSPHORUS, NITROGEN TO 1 ORGANIC PHOSPHORUS, CARBON TO NITROGEN IN SURFACE SOIL

Pounds of organic phosphorus in two million pounds of soil	Organic phosphorus as percent of total phosphorus	Ratios of			Factor for calculating the organic phosphorus from organic carbon
		Nitrogen to 1 organic phosphorus	Carbon to 1 nitrogen	Carbon to 1 organic phosphorus	
423	46.0	9.0	11.1	99.2	0.01008

(b) PHOSPHORUS ASSOCIATED WITH THE MATIÈRE NOIRE

Of the two available methods for determining the organic phosphorus, the one, the determination of the phosphorus associated with the *matière noire* extracted from the soil by 4 percent ammonia, has given rise to some confusion. Grandeau (10) regarded the phosphorus extracted with the *matière noire* as being probably in organic combination. Eggertz (21), Nilson (79), Wiklund (25), Dumont (65), Ladd (43) and Snyder (41) also regarded it as organic in form. Pitsch (14) and Van Bemmelen (23) took the opposite view. Pitsch thought that part of the extracted phosphorus may have been derived from the inorganic phosphates of the soil. Van Bemmelen regarded the phosphorus precipitated with the *matière noire* as absorbed phosphorus. Quite recently, Fraps (69) concluded that only 1/3 of the phosphorus extracted by ammonia was in organic combination, while still later Stoddart (80) concluded that only 1/5 of the extracted phosphorus was in organic combination. As a result of this conflicting evidence there is considerable confusion regarding the nature of the phosphorus extracted by ammonia. One cause of this confusion, no doubt, is the difficulty of getting rid of the suspended clay,—ordinary filtration will not remove it. Obviously all of the phosphorus associated with the suspended clay should not be included with the organic phosphorus, altho part of it may be organic in form. Fraps removed the clay by precipitation with ammonium sulfate. There is no evidence, however, that this reagent does not also precipitate some organic matter either chemically or mechanically. Ammonium sulfate is used by physiological chemists to precipitate the proteins in order to make certain group separations, while some preliminary work here showed that complete saturation of the ammoniacal extract of the soil with ammonium sulfate, after the removal of the suspended clay, produced a heavier qualitative precipitate of organic matter than did the addition of hydrochloric acid. It would seem, therefore, that the evaporation method of Hampton and Mooers (77) is more desirable for the removal of the suspended clay. Unless otherwise stated the latter method was used for the removal of the suspended clay in all the work reported in this paper.

Owing to the conflicting evidence regarding the phosphorus associated with the extracted *matière noire,* it seemed desirable to do some work with this material other than the simple determination of the phosphorus.

The soil, without previous treatment with hydrochloric acid to remove the calcium, was extracted with 4 percent ammonia in the ratio of 1 part of soil to 50 parts of ammonia water for 36 hours as in the usual humus determinations. The clay was removed by evaporation and the *matière noire* was obtained in quantity for study. Conditions here are such that the maximum quantity of inorganic phosphorus should be found in the ammoniacal extract since none has previously been removed by treatment with a mineral acid as in the usual humus determinations.

The suspended clay removed by evaporation was analyzed for carbon and phosphorus with results as follows: Carbon 3.73 percent and 3.61 percent, or an average of 3.67 per cent; phosphorus 0.118 percent and 0.109 percent, or an average of 0.113 percent. Since the carbon in the original soil was only 2.09 percent while the phosphorus was .046 percent, the relative increase of carbon and phosphorus in the suspended clay indicates undoubtedly the accumulation of organic matter with the "clay." It would appear probable that the grinding of the sample of soil, while preparing it for analysis, would convert the organic matter into an impalpable powder which would have a tendency to remain suspended in the liquid together with the fine clay particles when the soil was extracted with ammonia.

The amount of the extracted *matière noire* was determined. It was then analyzed for carbon, nitrogen, and phosphorus. The carbon was determined by the method suggested by Pettit and Schaub (59). The total nitrogen was determined by the regular Kjeldahl method; correction was then made for the absorbed ammoniacal nitrogen by determining the latter in a separate sample by distillation with magnesium oxide. The phosphorus was determined by igniting a sample of the *matière noire* and treating the ash with aqua regia; the silica was removed by evaporation and the phosphorus determined by the usual volumetric method. A confirmatory test made by determining phosphorus by fusion with sodium peroxid gave 0.835 percent and 0.815 percent phosphorus in the *matière noire* while the method adopted gave 0.860 percent and 0.830 percent.

The results obtained expressed as pounds per two million pounds of soil are recorded in Table 17.

TABLE 17.—*Matière Noire*, CARBON, NITROGEN AND PHOSPHORUS IN THE MATIERE NOIRE; RESULTS EXPRESSED AS POUNDS PER TWO MILLION POUNDS OF DRY SOIL.

(Soil not acid-extracted before treatment with ammonia)

Number	*Matière Noire*	Carbon in the *Matière Noire*	Nitrogen in the *Matière Noire*	Phosphorus in the *Matière Noire*
A	27600	10870	1642	233
B	28600	10850	1662	242
Average	28100	10860	1652	238

Phosphorus-nitrogen ratio = 1 :6.9
Nitrogen-carbon ratio = 1 :6.6
Phosphorus-carbon ratio = 1 :45.6.

The *matière noire* was now redissolved in dilute ammonia, an excess of 1 percent hydrochloric acid added and set aside over night. The precipitate of organic matter was brought on to a filter paper, which had previously been dried at 110°C and weighed. The precipitate was washed with 1 percent hydrochloric acid, dried at 110°C and weighed. The precipitated *matière noire* was obtained in quantity and analyzed for carbon, nitrogen and phosphorus. The results obtained are recorded in Table 18.

TABLE 18.—PERCIPITATED *Matière Noire*, CARBON, NITROGEN AND PHOSPHORUS IN PRECIPITATED *Matière Noire:* RESULTS EXPRESSED AS POUNDS PER TWO MILLION POUNDS OF DRY SOIL

(Soil not acid-extracted before treatment with ammonia)

Number	percipitated *Matière Noire*	Carbon in precipated *Matière Noire*	Nitrogen in precipitated *Matière Noire*	Phosphorus in precipitated *Matière Noire*
A	9174	4262	604	20
B	9203	4303	628	18
Average	9189	4282	616	19

Phosphorus-nitrogen ratio = 1 :32.4
Nitrogen-carbon ratio = 1 :7
Phosphorus-carbon ratio = 1 :225

The results recorded in Tables 17 and 18 are very significant as can be more readily seen by glancing at Table 19 which summarizes the above data.

TABLE 19.—*Matière Noire*, CARBON, NITROGEN AND PHOSPHORUS PRECIPITATED FROM AMMONIACAL SOLUTION BY HYDROCHLORIC ACID: RESULTS EXPRESSED AS PERCENT OF TOTAL SOLUBLE IN AMMONIA.

(Soil not acid-extracted before treatment with ammonia)

Matière Noire	Carbon	Nitrogen	Phosphorus
32.7	39.4	37.3	8.0

Of the total *matière noire* obtained, only 32.7 percent has been precipitated from the alkaline solution by hydrochloric acid. The portion remaining in solution does not consist of inorganic salts, as might be supposed, as is readily shown by the fact that only 39.4 percent of the carbon and 37.3 percent of the nitrogen has been precipitated. This shows conclusively that only about 1/3 of the dissolved organic matter has been precipitated.

Only 8.0 percent of the total soluble phosphorus, or 19 pounds out of 238 pounds, has been precipitated from alkaline solution by hydrochloric acid. Has the phosphorus remaining in solution in the mother liquor been derived from organic or inorganic sources? The fact that 60.6 percent of the carbon and 62.7 percent of the nitrogen also remain dissolved in the mother liquor would appear to be significant.

Having made a study of the *matière noire* obtained from the original soil it seemed desirable to investigate the *matière noire* obtained in the usual way after the soil had been extracted with 1 percent hydrochloric acid to remove the calcium and magnesium. It seemed reasonable to suppose that the acid extraction would remove also a considerable quantity, if not all, of the inorganic phosphorus, which may have previously passed into the ammonia solution.

As before, the amount of *matière noire* was determined and then extracted in quantity for the determination of carbon, nitrogen and phosphorus. The results obtained will be found in Table 20.

TABLE 20.—*Matière Noire*, CARBON, NITROGEN AND PHOSPHORUS IN THE *Matière Noire:* RESULTS EXPRESSED AS POUNDS PER TWO MILLION POUNDS OF DRY SOIL

(Soil acid extracted before treatment with ammonia)

Number	*Matière Noire*	Carbon in the *Matière Noire*	Nitrogen in the *Matière Noire*	Phosphorus in the *Matière Noire*
A	60840	25860	2805	524
B	61660	25790	2885	508
Average	61250	25825	2845	516

Phosphorus-nitrogen ratio = 1 :5.5
Nitrogen-carbon ratio = 1 :9.1
Phosphorus-carbon ratio = 1 :50

The *matière noire* was redissolved in dilute ammonia and an excess of 1 percent hydrochloric acid added. The amount of the precipitate and the carbon, nitrogen and phosphorus in the precipitate were determined as before. The results obtained are recorded in Table 21.

TABLE 21.—PRECIPITATED *Matière Noire;* CARBON, NITROGEN AND PHOSPHORUS IN THE PRECIPITATED *Matière Noire:* RESULTS EXPRESSED AS POUNDS IN TWO MILLION POUNDS OF DRY SOIL

(Soil first acid-extracted before treatment with ammonia)

Number	Precipitated *Matière Noire*	Carbon in precipitated *Matière Noire*	Nitrogen in precipitated *Matière Noire*	Phosphorus in precipitated *Matière Noire*
A	30110	11410	1242	57
B	31140	11555	1198	56
Average	30625	11482	1220	56

Phosphorus-nitrogen ratio = 1 :21.7
Nitrogen-carbon ratio = 1 :9.5
Phosphorus-carbon ratio = 1 :205

Table 22 summarizes the results reported in Tables 20 and 21. Of the total dissolved *matière noire* only 50 percent was precipitated. Again the greater part of the carbon and nitrogen remain in the mother liquor. Only 44.5 percent of the carbon and 42.9 percent of the nitrogen were precipitated while but 8.7 percent of the phosphorus was precipitated.

TABLE 22.—*Matière Noire;* CARBON, NITROGEN AND PHOSPHORUS PRECIPITATED FROM AMMONIACAL SOLUTION BY HYDROCHLORIC ACID: RESULTS EXPRESSED AS PERCENT OF TOTAL SOLUBLE IN AMMONIA

(Soil first acid-extracted before treatment with ammonia)

Matière Noire	Carbon	Nitrogen	Phosphorus
50.0	44.5	42.9	8.7

Again, the question regarding the source of the phosphorus remaining in solution arises. It will be seen that 55.6 percent of the soluble carbon and 58.1 percent of the soluble nitrogen also remain in solution. Attention should be called to the fact that when the original soil was treated direct with ammonia, without previous extraction with hydrochloric acid, under conditions where the maximum amount of inorganic phosphorus should be dissolved, only 238 pounds of phosphorus per two million pounds of soil were obtained: yet, after the soil had been treated with hydrochloric acid to remove the calcium, under conditions where the minimum amount of inorganic phosphorus would be dissolved by ammonia, 516 pounds of phosphorus per two million pounds of soil were obtained. The difference between these two numbers, 278 pounds, unquestionably represents phosphorus which must have been derived from organic sources. Now, since only 55 pounds of phosphorus is precipitated with the *matière noire* by hydrochloric acid, it would appear that the organic phosphorus associated with the precipitated *matière noire* is only a very small part of the organic phosphorus present in the soil.

Schmoeger (39) has demonstrated that the organic phosphorus compounds of the soil are decomposed by heating under pressure. It would appear probable, therefore, that the simple evaporation of the ammoniacal solution on the water bath in the preparation of the *matière noire* in quantity for analysis would cause a decomposition of the phosphorus compounds; hence when the *matière noire* is redissolved and precipitated by hydrochloric acid, less phosphorus would be obtained in the precipitate than would be the case if the material had not been heated. This idea was confirmed by experimental evidence as is shown in Table 23. The precipitated *matière noire* obtained from the original soil, which had not been extracted with hydrochloric acid, showed 19 pounds of phosphorus per two million pounds of soil. A portion of the ammoniacal extract of this soil was freed from clay by Frap's method; an aliquot part of the extract was then neutralized with hydrochloric acid: the precipitate obtained showed 68 pounds of phosphorus per two million pounds of soil, or over three times as much as did the precipitate obtained from the evaporated material.

TABLE 23.—PHOSPHORUS IN PRECIPITATED *Matière Noire:* RESULTS EXPRESSED AS POUNDS PER TWO MILLION POUNDS OF DRY SOIL

	Soil not acid-extracted		Soil acid-extracted		
Number	Phosphorus in heated *Matière Noire*	Phosphorus in unheated *Matière Noire*	Phosphorus in heated *Matière Noire*	Phosphorus in unheated *Matière Noire*	Phosphorus in barium precipitate
A	20	61	56	133	133
B	18	77	57	164	138
Average	19	69	56	149	135

The acid extracted soil gave similar results: the precipitated *matière noire* which had been subjected to heat gave only 55 pounds of phosphorus per two million pounds of soil, while the precipitated *matière noire* which had not been subjected to heat gave 149 pounds or nearly three times as much. The latter result was again confirmed. When the ammoniacal extract, freed from clay by precipitation with ammonium sulfate, is treated with barium chloride, the organic matter is quantitatively precipitated as is indicated by the decolorizing of the supernatent liquid and by the fact that evaporation of the filtrate and ignition of the residue gives only a very faint charring. But unfortunately the barium chloride also precipitates the inorganic phosphorus as barium phosphate under these conditions, and when the liquid is rendered acid it becomes colored, showing that organic matter as well as inorganic phosphorus has been dissolved. However, the precipitate was separated by filtration, washed with hydrochloric acid until free from barium and the phosphorus determined. This phosphorus must have been derived from organic sources. The results are recorded in the last column of Table 23 and compare very well with those previously obtained.

According to Schmoeger, extraction of the soil for 24 hours with 12 percent cold hydrochloric acid removes all of the inorganic phosphorus readily soluble in dilute acids. Would not such treatment also remove any inorganic phosphorus readily soluble in dilute alkali? It would certainly seem that the subsequent extraction with dilute ammonia of the acid extracted residue ought to dissolve only organic phosphorus. This idea was confirmed by experimental evidence and thus additional information regarding the nature of the ammonia-soluble phosphorus was obtained.

Two samples of 10 grams each of the soil under consideration were extracted for 24 hours with 100 c.c of 12 percent cold hydrochloric acid, then filtered and washed with hot water until the filtrate was free from chlorides. One of the samples was then extracted with 4 percent ammonia for 36 hours in the usual way and the amount of ammonia-soluble phosphorus determined: the second sample was again extracted with 12 percent cold hydrochloric acid for 36 hours and the amount of soluble phosphorus determined. Both experiments were duplicated. The dilute ammonia extracted 540 and 570 pounds of phosphorus or an average of 555 pounds of phosphorus per two million pounds of soil which had previously been extracted with cold 12 percent hydrochloric acid, while a second extraction with cold 12 percent hydrochloric acid yielded only 94 and 96 pounds or an average of 95 pounds of phosphorus per two million pounds of soil.

It would seem reasonable to suppose that both extractions of the soil with cold 12 percent hydrochloric acid removed some organic phosphorus since Berthelot and André (31) have demonstrated that organic matter of the soil is somewhat soluble in this reagent.

It would also seem very *unreasonable* to suppose that dilute ammonia possessed as great a solvent power for inorganic phosphorus as does 12 percent cold hydrochloric acid. But, assuming, for the sake of argument, that only inorganic phosphorus is extracted by the hydrochloric acid and that dilute ammonia has as great a solvent power for inorganic phosphorus as the hydrochloric acid, the above experiments seem to demonstrate that at least 460 pounds of phosphorus (555-95) of the ammonia-soluble phosphorus has been derived from organic sources and that at least 83 percent of the ammonia-soluble phosphorus has been derived from organic sources.

(c) ORGANIC PHOSPHORUS BY SCHMOEGER'S METHOD

The second method for determining the organic phosphorus is the one proposed by Schmoeger. Eggert, Nilson, Tache and others have shown that simple ignition increased the solubility of the phosphorus in cold hydrochloric acid. The increased solubility of the phosphorus was believed to be due to the destruction of the organic phosphorus compounds. Therefore, the amount of phosphorus in the original soil, soluble in cold hydrochloric acid, subtracted from the amount in the ignited soil soluble in cold hydrochloric acid of the same strength was regarded as having been derived from the organic phosphorus compounds. This assumption was confirmed by Schmoeger by hydrolyzing the soil under pressure at a temperature of 140°-160°C. This treatment of the soil decomposed the organic phosphorus compounds so that the organic phosphorus was rendered soluble in cold hydrochloric acid. The difference, therefore, between the amount of phosphorus extracted from the original soil by cold hydrochloric acid and the amount extracted from the soil which had been hydrolized gave the amount of organic phosphorus. Schmoeger found that, as a rule, concordant results were obtained by the two methods altho in certain cases slightly higher results were obtained by the latter method.

It was decided to determine the organic phosphorus by both of the above methods. Thus, 10 grams of the original soil was treated with 100 c.c. of 12 percent hydrochloric acid and digested in the cold with an occasional shaking for 24 hours. A second sample of 10 grams was ignited and then extracted with 12 percent cold hydrochloric acid in a similar manner. At the end of 24 hours the extract was diluted with water and separated by filtration. The residue was washed with cold water until the filtrate was free from chlorides: the filtrate was then made up to 500 c.c. and 100 c.c. used for the phosphorus determination. The results recorded in Table 24 show that there are 271 pounds of phosphorus in the original soil soluble in 12 percent cold hydrochloric acid while there are 814 pounds in the ignited soil soluble in the same reagent. These results show, therefore, that there are 543 pounds of organic phosphorus in two million pounds of the surface soil.

Another sample of 10 grams of the soil was treated with acidulated water and heated in an autoclave for 12 hours at a temperature of 140°-145°C. The sample was then digested for 24 hours with cold hydrochloric acid, filtered and the filtrate made up to 500 c.c. An average of two determinations show that 878 pounds of phosphorus were obtained. This would indicate that there were 607 pounds of organic phosphorus in two million pounds of the surface soil. Slightly higher results for organic phosphorus are thus obtained by the autoclave method but it is probably more nearly correct since it is difficult to conceive how the treatment in the autoclave would render any inorganic phosphorus soluble which would not be rendered soluble by ignition while the work of Leavitt and LeClerc (81, 82) would indicate that ignition might render some of the organic phosphorus insoluble in cold hydrochloric acid of any strength.

The calculation method shows that 423 pounds of phosphorus per two millions of the surface soil are in organic combination: the ammoniacal extraction method shows 504 pounds of organic phosphorus and the ignition method shows 543 pounds, while Schmoeger's method shows that there are 607 pounds of organic phosphorus. The calculation method is, therefore, very conservative in nature and it can be safely stated that at least that much phosphorus is in organic combination. Table 26 gives the summarized results of the organic phosphorus obtained by the several methods.

TABLE 24.—PHOSPHORUS SOLUBLE IN TWELVE PERCENT HYDROCHLORIC ACID: RESULTS EXPRESSED AS POUNDS IN TWO MILLION POUNDS OF SOIL

Number	Original soil	Ignited soil	Organic phosphorus
A	266	819	
B	276	809	
Average	271	814	543

TABLE 25.—PHOSPHORUS SOLUBLE IN TWELVE PERCENT HYDROCHLORIC ACID: RESULTS EXPRESSED AS POUNDS IN TWO MILLION POUNDS OF SOIL

Number	Original soil	Evaporated soil	Organic phosphorus
A	266	876	
B	276	880	
Average	271	878	607

TABLE 26.—TOTAL PHOSPHORUS AND ORGANIC PHOSPHORUS BY SEVERAL METHODS

	Organic phosphorus by calculation		Organic phosphorus by solution in dilute ammonia		Organic phosphorus by ignition		Organic phosphorus by evaporation (Schmoeger)	
Total phosphorus	Pounds per two million pounds of soil	Percent of total	Pounds per two million pounds of soil	Percent of total	Pounds per two million pounds of soil	Percent of total	Pounds per two million pounds of soil	Percent of total
919	423	46	504	55	543	60	607	66

CONCLUSIONS

1. The phosphorus-nitrogen ratio in the surface soil of the brown silt loam soils is 1:13.5 while the same ratio in the black clay loam soils is 1:11.4.

2. Under normal conditions the nitrogen-carbon ratio of the soil has a tendency to become narrower as the age of the organic material increases: the ratio, however, never becomes narrower or even equal to the ratio of the more common proteins contained in the humus producing materials.

3. The nitrogen-carbon ratios of the ordinary brown silt loam soils of Illinois are 1:12.1, 1:11.5 and 1:8.9 in the surface, subsurface, and subsoil respectively.

The ratios in the black clay loam soils are 1:11.7, 1:11.9 and 1:9 in the surface, subsurface, and subsoil respectively.

4. The phosphorus-carbon ratio in the surface soil of the brown silt loam is 1:165.2 while the ratio in the surface soil of the black clay loam soils is 1:163.6.

5. The calculation method for determining organic phosphorus is very conservative in character and can be relied upon in drawing broad general conclusions.

6. The evaporation on the water bath of the ammoniacal solution, in the preparation of the *matière noire* in quantity for analysis, causes a hydrolysis of the organic phosphorus compounds.

7. The determination of the phosphorus associated with the precipitated *matière noire* is not a quantitative method for the determination of the total organic phosphorus of the soil. It should be regarded only as a good qualitative evidence of the existence of organic phosphorus in the soil.

8. The contention of Fraps that, "There is no evidence that the phosphoric acid in the filtrate is in organic combination" and that, "It is probably derived from the iron and aluminium phosphates" is entirely untenable.

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