MEMOIRS
OF THE
NEW YORK BOTANICAL GARDEN
VOL. IV

EFFECTS OF THE RAYS OF RADIUM ON PLANTS

BY

CHARLES STUART GAGER, PH.D.

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PREFACE

The investigations embodied in this Memoir were begun in the autumn of 1904, with the intention of making them a minor problem during a year's residence at the New York Botanical Garden. On account of the scarcity of radium the work progressed slowly, for it was seldom that more than one or two experiments could be conducted simultaneously. When it became certain that the facilities of the Garden laboratories would be available for an indefinite period, other work was made secondary to the radium problem, for the relatively large quantities of radium and radium preparations placed at my disposal created an opportunity too valuable to let pass unimproved.

Chapter I. contains, in concise, non-technical language, information that is widely scattered in publications, many of which are not easily accessible or familiar to botanists. This information, however, is absolutely essential in order to understand the discussions that follow, and it was deemed advisable to include it.

The problem was originally suggested by Dr. William J. Gies, Consulting Chemist of the New York Botanical Garden, and it is a pleasure to acknowledge his helpful suggestions during the earlier progress of the work.

The investigations would not have been possible had it not been for the munificent liberality of Mr. Hugo Lieber, of the firm of H. Lieber & Co., of New York City. Mr. Lieber has freely placed at my disposal some $3,000 to $4,000 worth of standard preparations of the purest radium bromide yet obtained, as well as weaker preparations of radium, polonium, and radio-tellurium. As stated in detail in the pages that follow, he has devised apparatus, and made other apparatus at my suggestion, without which many of the experiments could not have been performed. It is not possible adequately to express in words my sincere gratitude to Mr. Lieber, not only for his great liberality, but also for the kindly interest he has maintained in the research throughout its entire progress.

C. STUART GAGER.

NEW YORK BOTANICAL GARDEN.

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EFFECTS OF THE RAYS OF RADIUM ON PLANTS

CHAPTER I

THE DISCOVERY AND NATURE OF RADIOACTIVITY

THE DISCOVERY OF CATHODE RAYS: The discovery of radioactivity was dimly foreshadowed as far back as the year 1838, when Michael Faraday,[51] studying the discharge of electricity through rare gases, noted the fact that there was always a dark space between the glows surrounding the positive and negative poles in the vacuum tube. Fourteen years later Grove[59] observed and described the stratified appearance of the electric discharge through very rare gases. Gassiot[54] further studied the stratification, and found that there were two distinct forms of stratified discharge, and that they could be deflected and thus separated by a magnet. He[55] later described the negative discharge in a vacuum tube, and ascertained, not only that it could be deflected by a magnet, but that, wherever the charge impinged, "a brilliant blue phosphorescent spot is perceivable, which spot is in a short time sensibly heated." Gassiot concluded that there is the appearance of "a direction of a force emanating from the negative," and inferred from his experiments that an electric current cannot pass through a perfect vacuum; the intervention, he said, of a certain amount of matter is necessary. Four years later he[56] published the results of experiments confirming these conclusions, and stated that there was "an actual disruption of particles from the negative terminal," which indicates force there.

The idea of cathode rays was more fully and accurately expressed by Hittorf,[63] whose paper, "*Ueber die Electricitäts-Leitung der Gase*," bears the date of October 9, 1868, and was published in 1869, six years after Gassiot's announcement.

Lord Kelvin's[68] statement * that the "kathode torrent" was dis-

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* Made also by Rutherford,[115] p. 73.

covered by Varley seems, therefore, rather extreme. Varley[145] correctly interpreted the cathode rays as "attenuated particles of matter projected from the negative pole by electricity in all directions"; he also caused these rays to produce motion by deflecting a thin plate of talc suspended in the vacuum tube by a silk fiber. The rays were deflected so as to impinge on the talc by a magnet, and the spot where they struck the talc was observed to become luminous.

In the year 1879, Sir William Crookes[28] published the results of his wonderful experiments on the passage of electricity through very rare gases. These experiments confirmed beyond question * the observation of Varley that, when the gas in a glass tube is exhausted to about one ten-thousandth of an atmosphere, a stream of material particles passes from the negative pole in the tube. It was already well known that a moving conductor carrying a charge of electricity could be deflected by a magnetic force. The stream of particles observed by Crookes could be thus deflected, and, furthermore, they could impart motion to a movable object placed in their path,† and could cast a shadow, as Hittorf had previously shown. Crookes concluded that they revealed a fourth state of matter. "The phenomena in these exhausted tubes reveal to physical science a new world — a world where matter may exist in a fourth state, where the corpuscular theory of light may be true, where light does not always move in straight lines, but where we can never enter and with which we must be content to observe and experiment from the outside."[27] It was to this stream of negatively charged particles that the name cathode rays was given. "We have actually touched the borderland," said Crookes, "where Matter and Force seem to merge into one another. . . ."[28]

Perrin,[86, 87] some years after, showed that the particles composing these rays carry charges of negative electricity. Two years later Thomson[135] verified the work of Perrin, and determined a most important figure, viz., the value of the ratio of the charge to the mass (e/m) of an individual particle from the cathode, and ascertained the

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* The German school was slow to abandon its position that the phenomena in a Crookes tube were a kind of ether-wave. The work of Varley and Crookes proved that hypothesis erroneous, and it is nowhere held at present.

† Thomson[140] later stated that the rotation of mill-wheels by the bombardment of the cathode rays is to be considered, not as due to momentum imparted by their particles, but to a secondary effect, due to the rays making the vanes hotter on one side than on the other, thus producing a radiometer action.

velocity of the particles. He found the mass to be one one-thousandth that of a hydrogen atom.

LENARD RAYS: Hertz[61] discovered that cathode rays will pass through thin aluminium foil inside of a Crookes tube. This proved that aluminium is transparent to the particles constituting these rays. Acting on this suggestion, Lenard[71] succeeded in producing cathode rays in a tube containing a window of aluminium. Some of the rays passed through the window, and Lenard was thus able to experiment with cathode rays outside of the tube, and to demonstrate that they carry a charge of negative electricity. Thus the statement of Crooks, quoted above, that we must be content to observe and experiment with cathode rays "from the outside," no longer held true. Cathode rays that have passed outside of a Crookes tube have been called *Lenard rays*.

The researches above referred to became the foundation stones for the new science of radioactivity.

DIVISIBILITY OF THE ATOM: In 1899, Professor J. J. Thomson found that carriers of negative electricity are given off from a red hot wire in a vacuum, and it was for these carriers that he proposed the name *corpuscle*. In Thomson's paper, also, we find one of the first statements, based upon experimental evidence, that the atom is not the limit of physical divisibility. "I regard the atom," says Professor Thomson,[137] "as containing a larger number of smaller bodies which I will call corpuscles. . . . In the normal atom this assemblage of corpuscles forms a system which is electrically neutral."

The term *electron* * has largely supplanted that of corpuscle. The mass of an electron is always the same, no matter from what gas, or from what solid produced, or by what means. It has the smallest mass of any known body. It will be noted later that an electron can be split off from the atom of probably every known substance. This fact leads to the immensely important inference that unit charges of negative electricity are constituents of the atoms of all matter, or, in other words, that the nature of all matter, organic or inorganic, is electrical. In fact the work of Professor Thomson lends much probability to the conclusion that the entire mass of the corpuscle is electrical, that it is a disembodied charge of negative electricity, and that matter and electricity are one and the same thing.

IONIZATION: When an electron passes through a mass of a gas

* Coined by Dr. G. Johnstone Stoney. Kelvin wrote it "*electrion*."

it will collide with some of the molecules of the gas. As a result of this collision, a unit charge of negative electricity (electron) may be torn from the molecule. The remaining portion of the molecule, by virtue of the loss of the electron, is positively charged. Such a separation of molecules into negatively and positively charged units is called *ionization*. Each of the units is a gaseous *ion*, and the gas is said to be *ionized*.

At low pressures, such for example as obtain in a vacuum tube, the electron is all there is of the negative ion, and the positively charged remainder of the molecule constitutes the positive ion; * but at atmospheric pressure each of these charged bodies becomes the center of aggregation of several molecules, and then the central charged nucleus, *together with the surrounding molecules*, is regarded as an ion. Gaseous ions are positive or negative according to their charge.

It is essential not to confuse the free gaseous ions with the ions of electrolytes in solution, and the employment of the same term in two senses is, in some ways, unfortunate. Negative gaseous ions are fragments of atoms, while the ions of electrolytes in solution result from the splitting up of molecules. The mass of a free negative gaseous ion is about $\frac{1}{770}$ the mass of a H ion in solution (Jones), but they both carry the same kind of a charge. The free negative ions, or electrons, are the same as the "satellites" of Kelvin, and the "corpuscles" or "particles" of J. J. Thomson. Electrons do not behave as a gas. They cling to positively charged bodies, and, if left quiet, settle on the walls of the containing vessel.

Discovery of X Rays: Three years previous to Thomson's proposal of the term corpuscle, Röntgen[97,99] read before the Würzburg Physico-Medical Society his epoch-making communication on the X rays, and later in the same year Perrin[87] and Stokes[131] showed that X rays are probably electro-magnetic pulses in the ether, and develop at any place where a body arrests the motion of the electrons of the cathode rays. This conception was subsequently more fully expressed by Thomson.[136] Thus when the cathode rays are stopped by the walls of the Crookes tube, X rays result. Here was a new kind of ray that could pass through bodies opaque to ordinary light, and darken a photographic negative.†

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* The mass of positive ions varies with the substances from which they are produced.

† The effect of X rays on a photographic negative was in reality a later discovery.

Rays of Niewenglowski: The phenomenon of X rays was always associated with phosphorescence, and Henri Poincaré[89] had already suggested that the two phenomena might bear a causal relation to each other. Experiments with phosphorescent sulfide of zinc, by Henry,[60] led to the discovery that a coating of that substance on a body, otherwise opaque to the X rays, rendered the body transparent to them. Rays that could penetrate matter opaque to ordinary light were obtained by Niewenglowski[81] in 1896 with several phosphorescent bodies after exposing them to sunlight. He obtained the image of a piece of money on sensitive paper that was protected from light rays by rays from phosphorescent sulfide of calcium that had been exposed to sunlight.

Becquerel Rays and the Discovery of Radioactivity: In the same eventful year of 1896 Becquerel[2] confirmed Niewenglowski's results, and experimented with, among other substances, various salts of uranium. Finally he[5] demonstrated that exposure to sunlight was not necessary, but that salts of uranium that had never been exposed to light gave out invisible rays that could pass through opaque objects and darken a photographic plate. Furthermore, while uranic salts are phosphorescent, the uranous salts are not, though both possess the property of radioactivity. Thus it was shown that the phenomenon is not necessarily connected with phosphorescence.

It was recognized[6] that Becquerel rays were very similar to X rays, and since all the salts of uranium, whether they had ever been exposed to light or not, and whether crystallized or dissolved, gave rise to the rays, the latter were thought to be due to uranium. Experiment showed that metallic uranium was strongly active. "Uranium," said Becquerel,[7] "is the first example of a metal manifesting a phenomenon of the nature of an invisible phosphorescence." These invisible rays from uranium that can pass through matter opaque to ordinary light and darken a photographic plate, are known as Becquerel rays.

The Discovery of Radium: After Becquerel's discovery, Monsieur and Madame Curie, of Paris, began to examine different minerals containing uranium in order to see if they gave off Becquerel

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Röntgen's first hint of the rays was their effect on barium platino-cyanide paper (Nov. 8, 1895). In the following year Troost[144] announced that artificial hexagonal blend gave off X rays, and could be substituted for the Crookes tube in many experiments.

rays. Some thirteen minerals were found to possess this property, and among them pitchblende was the most active.* One of the first results of their work was the discovery of *polonium*, the first substance emitting Becquerel rays to be isolated from pitchblende. It was named by Madame Curie from Poland, her native country. In this paper Madame Curie[34] also proposed the term *radioactive* for all substances giving rise to rays of this nature. Later in the same year M. and Mme. Curie and Bémont[37] announced the discovery of radium,† and the fact that it was a new element was confirmed spectroscopically by Demarçay.[44] The atomic weight of radium, as determined by Mme. Curie, is 225, while Wilde's[148] determination gives 232. The atomic weight of uranium is 240. The results of Mme. Curie's researches up to the year 1904 are embodied in her *Thésé*.[43]

It remains now to trace very briefly the researches that have led to a clear understanding of the nature of radioactivity, and its general distribution in nature.

THE COMPLEXITY OF THE RAYS: In 1899 Rutherford[101] made the discovery that the rays of uranium are complex, consisting of at least two kinds, to which he gave the names α rays and β rays.

CATHODE RAYS FROM RADIUM; BETA RAYS: In the same year Giesel,[57] Meyer and von Schweidler,[79] and Becquerel[8] all discovered independently that rays from radium were deflected by a magnet, and in the following year (1900) Becquerel[9] showed that their behavior in the magnetic field was quite similar to that of cathode rays. Evidently, then, here was a type of cathode ray given off by certain bodies spontaneously, and at atmospheric pressure. Following up these experiments, M. Curie[33] demonstrated, as Rutherford had the year previously for uranium, that the rays given off by various radioactive bodies are complex, consisting of at least two kinds of rays, one deviable by the magnetic field, and the other not. The deviable rays, said M. and Mme. Curie,[35] are charges of negative electricity. In 1902 Rutherford[103] announced that the negatively charged particles emitted by both uranium and radium are similar in all respects to cathode rays. The β particles vary considerably in velocity between certain limits, thus introducing a complexity into the nature of the β ray.

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* Afanasjew (1900) has since examined 51 minerals, and found that all containing uranium and thorium can blacken the photographic plate.

† See also citation No. 40.

POSITIVE IONS FROM RADIUM; ALPHA RAYS: It was Strutt[132] who first suggested that the less deviable, or α rays were streams of positively charged particles, and the experimental confirmation of this hypothesis was reported by Crookes[31] in the following year. One year later Rutherford[106] demonstrated that the α rays could be deviated in a magnetic field, and in a direction opposite to that of the β rays, thus further confirming the fact that they carry a positive charge.* The α particles from radium were shown to have twice the mass of a hydrogen atom. Further communications by Thomson[141,142] and by Rutherford[112] established beyond reasonable doubt that the α rays of radium and other radioactive substances, consist of a stream of positively charged particles, shot off from radioactive bodies, but with a velocity much less than that of the electrons. Rutherford[120] has recently calculated that the velocities of expulsion of the α particles from various radioactive substances all lie within a range of 1.56×10^9 and 2.25×10^9 cm. per second.† Thomson[142] also showed that bodies struck by the α rays from polonium have a positive charge communicated to them. Because of their greater size and less velocity, the particles of the α rays are much less penetrating than are those of the β rays. Rutherford and Grier[124] have shown that a layer of aluminium .09 mm. thick completely absorbs α rays. Not all of the α particles have the same velocity, for, since their motion is retarded by passing through matter, those coming from different depths of a thick layer of radium will have unequal velocities, those from the surface moving fastest. For this reason α rays like β rays are not simple but complex.‡ The α particle, losing its positive charge, becomes a helium atom.[121a]

Thomson found, in the course of his experiments, that both polonium and radium emit slowly moving cathode rays which cannot penetrate aluminium foil easily penetrated by β rays. Swinton[134] has recently shown that anode rays that have passed through perforations in the cathode terminal ("canal rays") may cause rapid motion of mill wheels, just as Crookes demonstrated for cathode rays.

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*Experiments indicate that the α particle acquires its positive charge as the result of ionization through collision. See Bibliography, p. 14, Nos. 17, 18, 113, and 77.

Whether the α particle is a molecule of hydrogen, an atom of helium, or a helium molecule carrying twice the ionic charge, is not easily decided (Rutherford[118]), but the results obtained by Cameron and Ramsay[23] seem to indicate that the α particle and helium are not identical.

†By the photographic method Des Coudres[45] found the velocity (V) of the α particles from radium to be 1.65×10^9 cms. per second.

‡Cf. citations Nos. 17, 12, 21, 116.

X Rays from Radium; Gamma Rays: It was Villard [146, 147] who discovered that radium, besides emitting α and β rays, is the source also of a non-deviable ray, analogous to very penetrating or "hard" X rays. Becquerel,[10] Strutt,[133] and Eve [49] also identified the very penetrating, non-deviable rays. Now a type of X ray arises whenever a β particle is either started or stopped, and Strutt considered that the non-deviable rays from radium arise secondarily by the self-bombardment of the radium by the β particles. Ashworth,[1] on the other hand, supported the theory that these rays were not of a secondary nature, but resulted directly from the disintegration of the radium-atom. Finally Rutherford [109] showed that this "hard" type of X ray (a narrow electro-magnetic pulse) arises from radium by atomic disintegration, while a "soft" type of X ray (a broad electro-magnetic pulse) arise at the points where the β particles strike another body. The non-deviable rays were named by Rutherford γ rays.

The Emanation: In addition to the giving off of three types of rays as above indicated, it was found by Rutherford [102] that a radioactive gas diffuses from thorium, and in the same year Dorn [47] determined the same fact to be true of radium. To this heavy, radioactive gas Rutherford gave the term *emanation.** Its gaseous nature was confirmed by Rutherford and Brooks.[122, 123] The emanation was found to condense at – 150° C.,[127] and in 1904 its spectrum was mapped by Ramsay and Collie.[93] Ramsay,[91] also, gave further evidence that the emanation is a gas by showing that it obeys Boyle's law. About three fourths of the activity of radium, according to Rutherford,[107] is due to the emanation.

The Nature of Radioactivity: In brief, then, we know that certain elements of very high atomic weight are giving rise, spontaneously, to three types of invisible radiation, as follows:

1. A stream of positively charged particles, with slight penetrating power, with a mass twice that of an atom of hydrogen, and moving with about one tenth the velocity of light. Streams of these particles constitute the α rays. The α rays consist of "veritable atoms of matter projected at a speed, on an average, of 6,000 miles

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* Sir George Stokes proposed to Crookes [32] a systematic nomenclature in radiology as follows: "Ray—A disturbance propagated in the ether. Jet—A discharge of electrons. Emanation—To include both Rays and Jets." The distinction between the first two terms has obvious advantages, and is used by Crookes in the paper above cited. The term emanation, however, is now firmly established as referring to the radioactive gas.

an hour. They cause most of the ionization observed near an uncovered radioactive substance." The α particle travels 3.5 cm. through air before it is stopped, breaking up, or ionizing, in this path about 100,000 molecules.

2. A stream of negatively charged electrons, moving with a velocity of from $\frac{1}{5}$ to $\frac{9}{10}$ the velocity of light, having a mass $\frac{1}{1000}$ the mass of a hydrogen atom, very penetrating to substances opaque to ordinary light, and giving rise, when stopped, to a ray analogous to, if not identical with, the X ray. Streams of these particles constitute the β rays.

3. An electromagnetic pulse in the ether, exceedingly penetrating to opaque bodies, and similar in all respects to the Röntgen or X ray. These pulses are called γ rays. Not all radioactive substances give off all three kinds of rays. Only α rays, for example, are emitted by polonium.

In October, 1907, Bragg[19] put forward the hypothesis that, in addition to positive and negative particles, atomic disintegration may give rise to the emission of neutral particles, "such as, for example, a pair consisting of one α or positive particle and one β or negative particle." It is not impossible, he says, that the γ rays, instead of being ether pulses, may consist of streams of these neutral pairs, and all the known properties of the γ rays, as well as of X rays, are satisfied on this hypothesis. In a later paper he[20] states that ether pulses are a component of both X and γ rays, but do not compose the entire phenomenon of the ray. Cooksey,[25] from his experiments, was unable to accept Bragg's view, and Kleeman's[70] experiments led him to adhere to the older theory that γ and X rays are, in general, alike, both consisting of electro-magnetic pulses produced by the acceleration of electric charges.

4. In addition to the three types of rays, there is given off a very dense, chemically inert, radioactive gas, which slowly diffuses from radium (and also indirectly from thorium and actinium). The atom of the emanation gives off, in its diisntegration, only α particles.*

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*Experiments of Rutherford[117] indicate that the emanations of radium, thorium and actinium differ from the other inert gases of the argon family in the fact that, in the small amounts in which it is available, it is absorbed by charcoal.

By comparing the rate of diffusion of radium emanation with that of mercury vapor, Perkins[85] determined the molecular weight of the emanation to be 235. This excess over the atomic weight of radium is explained by Perkins as due to experimental errors. On the basis of the disintegration theory of Rutherford, and considering the emanation as a monatomic gas, its molecular weight should be nearly that of the atomic weight of radium.

Beta rays and alpha rays are both vehicles of energy. Owing to their high speed and relatively minute size, the particles of the β rays may penetrate great numbers of atoms, passing through the spaces between the electrons that compose them. Under such conditions they continue in their path without deflection; but when a β particle collides with an electron the latter may be torn from the atom and set free (ionization). The ion may not always be torn from the atom by the collision, for the impact may serve only to deflect it from its path. In either event the β particle will lose energy, will therefore travel more slowly, and hence be thereafter more easily deflected.

The particles of the α rays are more effective ionizers than the electrons, but, owing to their relatively large size, they are not easily deflected by collision. Hence the α particle loses energy chiefly by collision.

It is partly for the reasons just described that both the α and the β rays, as stated above, are complex. They are both composed of streams of particles possessing widely varying amounts of energy.

THE COMPLEXITY OF THE ATOM: A conception of the atom such as the discoveries in radioactivity compel us to adopt, has been expressed by Perrin in a very striking figure. He likens the atom to a miniature planetary system. If a suitable force acts on an atom strongly enough it disengages a negative planet, or electron, producing thereby ionization. If the atom is very unwieldly, that is, relatively very large, and the corpuscle far from the center — the Neptune of the system — it will be loosely held by the electrical attraction of the remainder of the atom, and so more easily separated from it. In a somewhat speculative calculation, based upon the energy liberated in radioactive processes, Campbell[24] has estimated that "the number of electrons in a radium atom must be greater, and probably very much greater than 1,200."

THEORY OF ATOMIC DISINTEGRATION: The researches of Rutherford and Soddy,[125] of Thomson,[139] and of Rutherford all indicate that radioactivity is a manifestation of sub-atomic change. "In its simplest form," says Rutherford,[121] "the theory (of atomic disintegration) supposes that every second a certain fraction (usually very small) of the atoms present become unstable and explode * with

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* Sir Oliver Lodge, in a recent discussion of Lord Kelvin's philosophy (Nature 76: 198. 2 Jl 1908), has called attention to the difference between the static and the kinetic conceptions of the atom. "The internal energy of Lord Kelvin's model atom

great violence, expelling in many cases a small portion of the disrupted atom at a high speed. The residue of the atom forms a new atomic system of less atomic weight, and possessing physical and chemical properties which markedly distinguish it from the parent atom. The atoms composing the new substance formed by the disintegration of the parent matter are also unstable, and break up in turn. The process of disintegration of the atom, once started, proceeds through a number of distinct stages. These new products formed by the successive disintegrations of the parent matter are in most cases present in such extremely minute quantity that they cannot be investigated by ordinary chemical methods. . . . For any simple substance, the average number of atoms breaking up per second is proportional at any time to the number present. In consequence the amount of radioactive matter decreases in a geometrical progression with time." Rutherford[121] illustrates these changes by the following diagram * (FIGURE I). The time periods given indicate how long is required for the given product to become half transformed. Thus, it requires 2,000 years † for a given quantity of radium to become half transformed into the next following product.

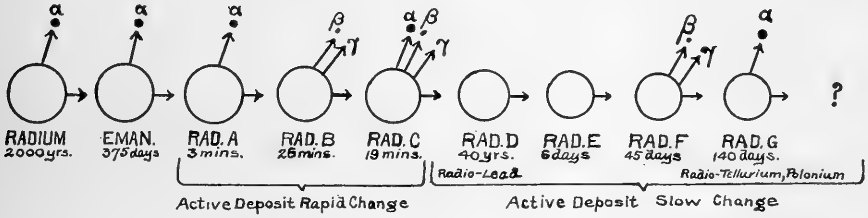

FIG. I. Theory of Atomic Disintegration. (After Rutherford.)

Uranium is now generally regarded as the ancestor of radium, but there are several intermediate disintegration products of uranium, and of these *ionium*, discovered by Boltwood,[15] may be the immediate

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is static or potential. The internal energy of the hypothetical atom at which others are working is kinetic.

"The disintegration of radium in the former case is comparable to the explosion of an unstable chemical compound, like gun-cotton. In the latter case it must be represented by something more akin to the flying to pieces of a single rapidly spinning unit, such as a fly wheel."

* Duane[48] states that radium *B*, formerly held by Rutherford[115] to be non-radioactive, emits as much negative electricity as does radium *C*.

† Results obtained by Boltwood[16] on the growth of radium in preparations of ionium separated from uranium minerals indicate that the half-value period for radium is 2,000 years. Rutherford[121a] gives 1,760 years.

parent of radium. All the α rays given off by the same product behave alike, but the particles from the different products are quantitatively unlike, though qualitatively similar.

PHOSPHORESCENCE: It was the association of phosphorescence with X rays that led to the discovery of radioactivity. As might be expected, radium is phosphorescent. The source of this light is not definitely known, but experiments of Lord and Lady Huggins[66] seem to indicate that the luminosity is not due to the β rays.

MEASUREMENT OF ACTIVITY: Eve[115] has shown that the activity of radium is a function of the amount present; therefore the purer the radium salt the greater the activity, weight for weight. The degree of activity is stated in terms of that of uranium as a standard. Thus, to indicate that a given preparation is of 10,000 activity (10,000 ×), means that it is ten thousand times as active as an equal weight of uranium. The purest radium bromide so far obtained has an activity of 1,800,000. The activity of a given radium preparation enclosed in a sealed glass tube shows no signs of decreasing with time. The intensity of activity, according to Rutherford,[108] does not vary with the concentration of the salt. In his experiments, "a distribution of the radiating matter over a thousand times its original volume has no appreciable influence on its original activity."

AMOUNT OF ENERGY EVOLVED: The results of Curie and Laborde,[39] confirmed by Runge and Precht,[100] indicate that one gram of radium emits energy at the rate of 100 gram-calories per hour, or 2,400 per day. Rutherford[115] has pointed out that this is nearly as much energy as is required to dissociate one gram of water (3,900 gram-calories). Rutherford[115] has calculated that the total energy of radiation during the disintegration of one gram of radium is 1.6×10^9 gram-calories, and the energy radiated may not represent all of the energy involved in the change. The energy of the transformation is at least twenty thousand times, and may be a million times as great as the energy of any molecular change, such, for example, as that involved in the union of hydrogen and oxygen to form water. Ramsay[92] states that the emanation given off by one gram of radium evolved 75 calories in one hour.

In replying to expresssions of doubt as to the validity of the foundations of the theory of atomic disintegration, Soddy[130] emphasized the fact that it is firmly grounded on experimental evidence, that no fact has been adduced that does not conform to it, and adds that it is impossible to form "from words or reading the least idea of the really startling character of some of the new discoveries."

EFFECT OF EXTERNAL CONDITIONS: As might be inferred from the fact that radioactivity is neither a molecular nor an atomic, but a sub-atomic change, external conditions have no effect upon it. It is not affected by temperatures ranging from 200° C. to that of liquid air, nor by variations in atmospheric pressure. The emanation, of course, being a gas, diffuses from the radioactive body more slowly under increased pressure and lower temperature, and the rate of escape is greater in moist air, and when the radioactive salt is in solution, but its radioactivity is not thereby affected in the least.*

EXCITED RADIOACTIVITY: It has been found that bodies under certain conditions of exposure to radioactive substances become themselves radioactive. Radioactivity thus produced is called *excited radioactivity.* Rutherford[105] demonstrated that the presence of the emanation is necessary in order to produce excited radioactivity, and later (1903) showed that the excited radioactivity is due to a deposit of radioactive matter (called the "active deposit") from the emanation of either thorium or radium.

ORIGIN OF RADIUM: The observation of Lord and Lady Huggins[64] of the gradual appearance of the spectrum lines of helium in the spectrum of radium is the first demonstration in the history of science of the origin of one chemical element from another. Ramsay and Soddy[95] in the same month (July, 1903) announced their observation that gases occluded by 20 mg. of radium bromide contain helium, and soon thereafter they[95] observed the spectrum lines of helium gradually appear in the spectrum of the radium emanation after it has stood for four days, thus confirming the results of Lord and Lady Huggins. Further confirmation of this transformation was published by Dewar and Curie,[46] and by Himstedt and Meyer.[62]

The observation of the actual transmutation of one element into another suggests the question as to the origin of radium. In 1903 Rutherford and Soddy[127] suggested that radium is a disintegration product of one of the other radioactive substances found in pitchblende. Soddy[128] in 1904 expressed his belief that uranium is the source of radium, and in 1905 obtained[129] experimental evidence that

* Makower[77] states that the activity of the radium emanation measured by the rays it gives off can be changed by high temperature, but this has not yet been confirmed.

the radium atom results from the disintegration of the atom of uranium. Two years later McCoy[75] stated that the results of his experiments confirm the conclusion that uranium is the parent, not only of radium, but also of all other active substances that accompany it in uranium compounds.*

Soddy[128] also finds experimental evidence that there is a steady production of radium from uranium, though the observed amount is of a lower order of magnitude than is indicated by the disintegration theory. This discrepancy between observation and theory is doubtless due to the fact, discovered by Boltwood[14] in 1906, that the transformation of uranium into radium is not direct. The intermediate transformation product was later found to be, not actinium as at first thought, but a new radioactive element, which "emits both α and β radiations, which produces no emanation and which resembles thorium in its chemical properties." For this new substance Boltwood[15] proposed the name *ionium*, from ion, because of its ionizing properties due to its emission of α radiations.

Conceptions on this subject obtaining at the time of this writing will doubtless be more or less modified by researches now in progress.

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* The investigations of Coppock,[26] Martin,[78] Winkler,[149] Boltwood,[13] Murray,[80] and of Cameron and Ramsay,[23] and Ramsay and Soddy,[94] which rendered highly probable, if not positively demonstrated, the evolution of the chemical elements by radioactive transformations, are not reviewed here, as they have only a remote bearing on the purpose of this résumé.

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CHAPTER II

RADIOACTIVITY A FACTOR OF PLANT ENVIRONMENT

Previous to the present decade it was possible to classify the known inorganic factors of the plant's environment as either molar, molecular, or undulatory. The discovery of radioactivity and other revelations closely related to it, as briefly outlined in the preceding chapter, together with the investigations referred to below, lead to the recognition, not only of another element of environment, but to an entirely new kind of environmental factor, viz., radioactivity.

It is a matter of considerable interest to ascertain the effects of this new kind of energy on the life-processes of plants and animals, but when we realize that it forms a part of the natural surroundings of all living things, and must be reckoned with as a possible factor in all their vital activities, this interest greatly deepens.

The life-processes of plants are regarded as reactions to stimuli. Permanently remove all stimuli and all processes (*i. e.*, life itself) cease. In last analysis these different stimuli are all different manifestations of energy. We are familiar with some of the effects of the more ordinary ones, such as water, air, heat, sunlight, and gravity, with the corresponding tropisms (hydrotropism, geotropism, etc.), and with the condition of adjustment to the normal for each factor, tonus (phototonus, thermotonus, etc.). The discoveries in radioactivity show us that the effects of these long-recognized factors cannot be completely interpreted unless we take into account the newly discovered facts.

Probably all plants are in a state of *radiotonus*, or adjustment to the radioactive forces of their normal environment, and evidence is at hand that we shall be able to add radiotropic response to the other and well-known tropisms. Whether we consider water, sunlight, air, or soil, radioactivity is a factor involved, and the following paragraphs will briefly outline the investigations which compel to this conclusion.

Radioactivity in Water: Thomson [126, 128, 129] was the first to announce that air bubbled through Cambridge (England) tap-water was decidedly radioactive. Since then a large number of investigations seem to establish the fact that various waters, widely distributed, are sources of radioactivity. This property was present in 15 out of 18 specimens of water examined later by Thomson,[130] and in water from Italian springs examined by Pocchettino and Sella.[92] The radioactivity is due to the presence in the water of some radioactive substance, usually radium or its emanation. Allen and Blythswood[7] obtained a radioactive emanation from the water of the hot springs of Bath and Buxton, and, as might have been expected, Dewar has found helium in the same waters. Himstedt[69] demonstrated the evolution of a radioactive emanation from water- and oil-springs, and, later in the year, Adams[2] announced that a radioactive gas is evolved from the water of deep wells. When the emanation was removed by boiling, the water recovered the power of giving it off. It is probable that there is a slight amount of radioactive material dissolved in the water. The emanation was very similar to that of radium, and probably identical with it.

Artesian water from several wells, and the town water of Ely, Birmingham, and Ipswich was found by Thomson[129] to give off a radioactive gas when boiled. Bumstead and Wheeler[20] obtained a radioactive gas from a well 1,500 feet deep near New Milford, Connecticut (U. S. A.), and found that the city water of New Haven, Connecticut, was radioactive, whether taken directly from the reservoir or from a faucet after passing through the city mains. The latter writers[21] established the identity of this emanation with that of radium.

Vichy from Chomel was found to contain a radioactive emanation,[32, 33] as was also the water from the hot springs of Baden-Baden,[69] from Aix-les-Bains,[11] and from Karlsbad.[67] Radioactivity in the water from many German springs was detected by Schenck,[104] from Buxton springs by Blythswood and Allen,[14] and Strutt.[117], and from Lavey-les-Bains by Sarazin, Guye, and Micheli.[103] Strutt found traces of the salts of radium in the mud deposited from the hot springs of Bath and of Buxton, as well as radium emanation in their waters, while Boltwood states that he found radium dissolved in the waters of Bath and Baden-Baden under high pressure and temperature. Mache[79] tested the water in 17 springs in different parts of Germany, Schmidt and Kurz[108] that of 117 springs, and Dienert and Bouquet[36] the waters of four springs in France. Radioactivity was found associated with them all, and one hot spring contained both radium and thorium. Schmidt and Kurz conclude that all spring waters contain an emanation, usually that of radium, but in some cases that of thorium. Skinner[110] states that Mr. H. Cottam did not succeed in finding "any marked quantity of active gas" from samples of clays from various districts near London (Eng.), but found a radioactive gas in water of a deep well which goes below the clay to the green sand.

The radioactivity of deep well waters, mineral water, and water from a cistern in Columbia, Missouri, is attributed by Schlundt and Moore[105] to the presence of radium emanation in the water. Waters from 123 springs in southwest Germany, Austria, and Italy, and from an old Roman spring, the "Queen Isabella," in the island of Ischia, bay of Naples, were, in nearly every instance, found to be decidedly radioactive by Engler and Sieveking.[55] The sediment deposited by the water of the springs was often active, and self-luminous radium preparations were prepared from one of the sediments. The radium emanation was detected by Sury[123] in spring water of Baden, Switzerland, Leuker Bad, Garasp, and Disentis. The water from St. Placidus spring contains a radioactive salt which produces the emanation. In 1908 Joly[70] measured the activity of a sample of ocean water from Valencia, County Kerry, and suggested that oceanic radioactivity is due to radioactive materials brought to the ocean by streams. Much uranium, he says, is carried in solution or in fine suspension, and deposited in the ooze.

RADIOACTIVITY IN MUD AND ROCKS: The following investigators have reported radioactivity in deposits from various springs, and in mud from other sources: Elster and Geitel[51, 53] in the "fango," or mud from the hot springs of Battaglia, North Italy, and also[52, 54] in deposits from Baden-Baden, Nauheim and Wiesen baths. The activity of fango they attribute to the presence of radium, and state that 1,180 tons of this mud would yield 1 gr. of radium chloride. Borgmann[17] in mud from Odessa, and from Arensburg, on the island of Oesel; Blanc[12] in sediment from 9 springs in the region of the Alps; Castorina[29] in lava from Mt. Aetna. Giesel[63] found radium and radioactive rare earths in fango mud, and in earth from the fields of Capri; Vincentini and Da Zara,[137, 138] radium emanation in the water and sediments from a number of hot springs in northern Italy. Sediments from 20 springs from different parts of Germany were found radioactive by Schmidt and Kurz,[108] and in 1906 Mogri[89] detected the same property in the deposits from the thermal springs at the Institute of Bogni di Lucca, in Tuscany.

Tomassina[133] detected radioactivity in the lava thrown out by Vesuvius in the eruption of 1904, and, in a study of 28 samples of igneous rocks, including granite, basalt, hornblende, and serpentine, Strutt[120] found radium present in all. More was found in granite than in any other igneous rock studied, while the basic rocks contained the least amount. Iron meteorites were found to contain very little, if any, while stone meteorites contained about as much as the terrestrial rocks which they resembled.

The same author[120] found radium present in sedimentary rocks generally, marble, chalk, flint, clay, roofing slate, oil-bearing sandstone, deposits from the hot springs of Bath, sea salt, boiler-rust (Cambridge, Eng.), and in the rock-forming minerals, zircon, apatite, hornblende, tourmaline, labradorite, white feldspar, white and brown mica, white quartz and others. The specimens came from various widely separated regions in continental Europe, Africa, India, Asia (Ural Mts.), England, and the United States. It was found that more than one half of the radium is contained in the heavy minerals, though these form only about one eighth of the whole mass of the rock.

The association of radioactivity with the "ashes" and lava of Vesuvius was reported by Becker,[10] and a slight radioactivity of soils, clays, basaltic tufas, basalts, soft calcium carbonate, etc., was detected by Accolla[1] in 1907. Sands and mud of the seashore, and mud from the sulfur spring of Brucoli also possess a weak activity.

Analyses by Eve and McIntosh[60] showed in sedimentary Ordovician rock from $.92 \times 10^{-12}$ to .91 gr. of radium per gram of rock; in igneous Devonian .26 to 4.3 gr. per gram of rock, and in sedimentary Quaternary .16 to .8 gr. per gram of rock; and the probability that the internal heat of the earth is due to radium is discussed *pro* and *contra* by H. A. Wilson,[146] Strutt,[122] and others.

RADIOACTIVITY IN AIR: Elster and Geitel[45, 46] were the first to show that the atmosphere contains a radioactive emanation. They suspended a negatively charged wire for some hours in the air, then coiled it up and tested it with the electroscope. The wire was found to be radioactive, but this result was not obtained if the wire was first given a positive charge. Now a negatively charged wire immersed in the emanation of thorium or radium can collect a deposit formed by the disintegration of the atoms of the emanation, hence the conclusion from the above experiments, that the air contains a radioactive emanation.

Evidence of the presence of negative ions in the air was known, however, long before radioactivity was discovered. Thus Giese, [62] in 1882, observed the electrical conductivity of gases from flames, and in 1897 Kelvin and MacLean,[73] investigating the flames of a bunsen burner, a candle, an alcohol lamp, and a paraffin lamp, found a small negative charge in the gases drawn from them. Charcoal and coal "both gave negative electrification when there was a flame; and both gave positive electrification when they were glowing without flame." These investigations were extended by MacLean and Goto,[80] who showed that air is electrified by the burning of matches, wood, paper, and many other substances. So also McClelland[81] in 1898. Waves of ultra-violet light, and point-discharges of electricity produce negative ions in the air.[134]

The formation of ions by a candle flame was demonstrated by Ayrton,[8] who observed that such a flame can discharge an electroscope in 40 seconds at a distance of 40 cm. "The flame of a match had no less power, and an electric arc no more power than an uninsulated candle flame placed at the same distance."

Traubenberg[135] found the atmosphere in the vicinity of the crater of Vesuvius strongly ionized, and in 1904 Allan[6] showed that the excited radioactivity from the atmosphere behaves like that from thorium and radium, and contains both α and β rays. In the same year Elster and Geitel[53] pointed out that the electrical conductivity of the atmosphere is due largely, if not wholly, to a radioactive emanation which issues from the earth's crust. These authors are of the opinion that the outer layers of the atmosphere doubtless become ionized by the sun's rays of short wave-length, and that perhaps β and γ rays also proceed from the sun and produce a like effect. In this connection, it is known, from the investigation of Bacon,[9] that an electroscope discharges nearly 18 times as fast in sunlight as in ordinary diffuse light in the middle hours of the day. McClelland[82] believed that the ionization of the atmosphere points to the presence in it of some radioactive substance, and Blanc[13] showed experimentally that transformation products of radio-thorium are, at least in the vicinity of Rome (Italy), a most important agent in atmospheric radioactivity.

C. T. R. Wilson[140] calculated that the number of ions being produced per second in each cubic centimeter of air is about twenty. The actual number of ions present per c.c. of air varies with the meteorological conditions, being greater in clear, sunny weather. The number usually fluctuates between 500 and several thousands, and the positive ions are more numerous than the negative.[39] Schuster[109] found the following numbers of ions per c.c. of air in Manchester (Eng.), under the conditions indicated: 2,370 (snowing at intervals, with spells of sunshine. East wind. In a field). 3,600 (cold, bright, gusty. East wind. On a roof). 3,660 (cold, dull. East wind, on a roof). In addition to swiftly moving ions, Langevin[75] found slowly moving ones also present. The latter are about forty times as numerous as the former. Langevin's experiments were made on top of the Eiffel tower.

A negatively charged conductor placed in the open air becomes temporarily radioactive,[47] and this radioactivity can be removed by solution in an acid. This experiment was repeated in Canada by Rutherford and Allan,[98] who found that the greatest amount of excited radioactivity on a negatively charged wire was obtained during a strong wind. Allan[3] thinks the activity of the air is "probably due to an emanation of positive electrons in the air, ever present though in varying quantities."* Elster and Geitel[50] found that the radioactivity of the air of any given place varies with the nature of the soil of the locality, and later in the same year[50] found that sea air is only about one third as active as air over the land at Wolfenbüttel. Soon after this Himstedt[68] showed that air through which water is blown, or air that has passed through or over surfaces moistened with water has its electrical conductivity increased (by ionization) more than 100 times. This, he thinks, is the origin of the conductivity of the air from cellars and soil, observed by Elster and Geitel, as well as of the conductivity constantly present in the free atmosphere. In the following year Himstedt[69] reported that air sent through the water from many springs, and fresh ground water has its conductivity increased, and he contends that there is widely (perhaps universally) distributed in the earth a radioactive material from which a gaseous emanation proceeds which is absorbed by water and oil, is carried by these to the surface, and thence distributed to the atmosphere.

In connection with Elster and Geitel's observation it is interesting

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* Cf. also Stewart, R. M.[111]

to compare the variations in the penetrating radiation from earth, studied by Wright[149] in 1908. Measurements of this radiation made in different localities on the north and south shores of lake Ontario, and at different points on the surface of the lake during the passage of a steamer, show that there is greater activity inland and at points along the shore than over the lake between Toronto harbor and the Niagara river bell-buoy. The activity was measured by the number of ions generated per c.c. per second within a lead cylinder.*

Studies on the effect of altitude enabled Saake[100] to state that there was a larger amount of emanation in the high altitudes of the valley of Arosa, Switzerland, than is normal at lower levels. Ebert[40] has found that a radioactive emanation can be removed from the atmosphere by condensation in liquid air, while Ramsay and Soddy[93] have demonstrated that the inert gases in the atmosphere (helium, neon, argon, krypton, and xenon) have no radioactivity of their own. That the atmosphere at New Haven, Connecticut, contains the emanation of both thorium and radium was announced by Bumstead[18, 19] in 1904.

Antedating these discoveries, was the paper of Elster and Geitel[44] on the existence of electrical ions in the atmosphere. The authors found that the free atmosphere contains positively electrical masses, and considered the existence of these free ions as the most suitable basis for a rational theory of atmospheric electricity. This theory was further elaborated by Geitel[64] in 1901.

Elster and Geitel did not explain the origin of these ions in the free atmosphere, but, in the year following their discovery, Lenard[77] found that the rays of ultra-violet light generate cathode rays, and two years subsequently he[78] published the results of further investigations along the same line, stating that the cathode rays thus generated show diffusiveness, and must be largely absorbed by gases.

In the light of Lenard's experiments C. T. A. Wilson[143] concluded that the sunlight ionizes the atmosphere through which it passes, especially in the upper layers, where the sunlight is still strong in ultraviolet rays.

Certain facts, says Wilson, render it not improbable that pene-

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* It has not been thought necessary, in reviewing the above literature, to explain that electrical conductivity, through wires or air, or whatever else, is accomplished by the passage of electrons, or other ions. Increase in the conductivity of air and water means an increase in the number of the ions they contain. The fundamental ideas here involved are clearly set forth by Fournier (Bibliography, p. 17, No. 53), and others.

trating cathode rays traverse our atmosphere without being absorbed until they encounter the solid mass of the earth which thus becomes negatively charged. An excess of negative electricity is also carried from the atmosphere to the earth by rain. The positive charge thus left behind in the air is carried by convection currents to other regions. Thus the negative charge of the earth is maintained. As Wilson states, the fact that the earth's surface is negatively charged, and that free positive ions exist in the atmosphere, must result in a continuous flow of positive electricity from the atmosphere into the ground.

The hypothesis has been proposed by Villard[136] that cathode rays giving rise to the aurora borealis have their origin, not in the sun, but in the earth itself. Even the spraying of liquids may produce ionization, the negative ions, as Eve[57] has shown, being greatly in excess of the positive. Strong[112, 113] thinks that the penetrating radiation that causes ionization in closed vessels is probably due to gamma rays from radioactive products in the air, rather than in the ground, but it is considered probable that these products originate in the ground, as the theory of Elster and Geitel indicates. The products vary much in quantity according to atmospheric conditions. Eve[58] states that the ionization of the air is due to radioactive changes in both the air and the soil. Of course the amount of emanation coming into the air from the soil would vary with the rise and fall of the water-table in the soil, the soil temperature, the entrance into the soil of rain water, and the decrease of barometric pressure, all of which would be accompanied by a flow of emanation out of the soil into the lower layers of the atmosphere.* According to McLennan's[83] calculation, "approximately 9 ions per c.c. per second are generated in free air by the penetrating radiation from the earth."

Eve[59] has called attention to the fact that the radium C in the air is carried to the earth not only by falling rain, snow, dust, or smoke, but also by the potential difference in the atmosphere. Thus the radioactive matter in the air is decreased, that in the soil increased. Thorium C has been found in the atmosphere of both hemispheres, with an activity about one half that of the radium C present. Since

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* Another paper by Strong[113a] has appeared since the above sentence was written. He finds a relatively enormous amount of "external radiation" during the forenoon from 8 A. M. until 2 or 3 P. M. "This may be due," he says, "to the expansion of the air in the soil and an increase of emanating power due to the heating by sunshine, or it may be due to a change of barometric pressure."

the emanation of thorium decays about 6,000 times as fast as that of radium, and "has a poor chance of escaping from the soil," the amount of thorium C in the ground exceeds the amount of radium C, and the thorium C in the ground will be more than 15 times that in the air. Eve says that, "in most localities the penetrating radiation due to active matter in the air is less than one fifteenth that due to active matter in the earth." Recent determinations by Dadourian[35] indicate that the amount of radium emanation present in the air at New Haven, Conn. (U. S. A.), is from 20,000 to 50,000 times as great as the amount of thorium emanation.

RADIOACTIVITY IN SNOW AND RAIN: In 1902 Professor J. J. Thomson observed that water drops falling through air that contains ions remove the ions, and in the same year C. T. R. Wilson[141, 144] tested freshly fallen rain and found it radioactive. By adding barium chloride to rain water and precipitating the barium with H_2SO_4 he found the precipitate to be radioactive. Subsequently Allan[4] obtained a radioactive residue by evaporating freshly fallen snow to dryness. From about one liter of snow that fell during a heavy storm, there could be obtained about the same effect as from 0.1 gr. of uranium. The amount of activity varied with the amount of snow falling per second, and was constant so long as the fall of snow was constant. Later in that year the same writer[5] found that the radioactive residue could be rubbed off onto a piece of cotton, and when this was burned the ashes were still radioactive. Allan supported the theory that some process is continually going on in the air producing radioactive carriers which are removed by the snow-flakes. C. T. R. Wilson[144] also detected the activity of freshly fallen snow. Kaufmann has determined that such snow is, under similar circumstances, as active as rain, but snow falling on roofs loses its activity sooner than snow that falls on the ground.* The loss of activity appears to vary with atmospheric pressure. Righi[94] found that, during a snowfall, the electrical conductivity of the air is more than doubled.

Soil air from Wolfenbüttel was found to contain a radioactive material by Elster and Geitel,[49] and by Ebert and Ewers[41] at Munich. Geitel[50] found that the activity of the air from the soil in his garden did not apparently diminish in eight months, but the ashes of plants which had grown on active earth gave off no appreciable rays.

* This result was confirmed by Constanzo and Negro,[31] in 1906.

Carbon-dioxide from great depths of old volcanic soil was markedly radioactive, and Sarasin [101, 102] demonstrated the same property in air from the so-called "breathing wells" in Europe. The radioactive gas in the air and soil of New Haven (Connecticut, U. S. A.) was determined by Bumstead and Wheeler[21] to be identical with the emanation of radium. McLennan[84] also found that natural gas from wells in the Welland district, near Niagara Falls, and near Brantford, was charged with a radioactive emanation, and in the experiments of Dadourian[34] the activity of underground air was found to be due to thorium. A radioactive emanation, evolved on heating raw petroleum, is considered by Burton[22] to be due solely to the presence of radium in the soil.

THE GENERAL DISTRIBUTION OF RADIOACTIVITY: Evidence from the general distribution of radioactive substances, that radioactivity is a factor in the normal environment of plants has been shown to be abundant. Numerous other researches give added emphasis to this fact. The discharge of positive electrification by metals at a temperature of 270° C., or over,[42, 43, 95, 114] the evidence that the photosphere of the sun is emitting electrons in large quantities,[125] which travel throughout the solar system, the probable existence of electrons in metals at all temperatures moving freely between the molecules,[125] and causing a "soft" X ray when they collide with the molecules, are all facts pointing to the same conclusion.

Drops of spray on striking wet rocks at the foot of water-falls communicate a positive charge to the water and a negative charge to the air.[76] A very penetrating radiation has been found to exist inside buildings,[98, 99] and zinc and tinfoil, lead, copper, glass coated with phosphoric acid, silver, copper, platinum, aluminium,[115] dried earth, polished tin, brick, iron,[30] and hydrogen-peroxide,[37] have all been found to be radioactive, most of these substances giving off rays comparable to the β rays of radium. Experiments of McLennan and Burton[88] indicate that all metals in varying degree are sources of marked, though feeble, radioactivity, and that "the ordinary air of rooms is traversed by an exceedingly penetrating radiation," such as comes from thorium, radium, and the excited radioactivity produced by them. Campbell,[23, 25] after testing nine different metals, all of which are included in the lists given above, states that "the emission of ionizing radiation is an inherent property of all the metals investigated," and adds, "I see no reason why it should not be extended to all substances." The larger portion of the rays, according to Campbell, are α rays.

During the course of experiments at the foot of Niagara Falls, McLennan[83] found that three stretches of No. 24, insulated copper wire, of about 30 meters each, exposed to the spray, immediately became negatively charged to a potential of about 7,500 volts. The charge was shown to be caused by the spray, and to vary with its density. The spray also excited radioactivity in the air, but when evaporated it left no radioactive residue.

From the fact that air bubbled through distilled water in which lead acetate or lead nitrate has been dissolved is more radioactive than when bubbled through pure water, Thomson[127] infers that lead is radioactive. Brick,[97] metals generally,[88] the minerals samarskite, pitchblende, and monazite, from North Carolina, Cornwall, Norway, and Brazil,[117] and tin, zinc, graphite, platinum, lead, aluminum, and carbon,[23] all manifest radioactivity. Muñoz[90] has studied minerals, earth, water, and gases of the atmosphere in various parts of Spain, and finds radioactivity widely distributed throughout that country.

"From the kinetic theory of gases," says Lord Kelvin,[72] "it seems certain that every kind of matter has some radioactivity; that is to say, shoots off both vitreously and resinously electrified particles. Hence it is only in their extraordinarily great abundance and great velocities of shooting, that polonium and radium differ from ordinary matter." *

Campbell[27] concludes from his experiments that potassium and rubidium are radioactive substances. The rubidium is less active than potassium, while the activity of the latter is $\frac{1}{1000}$ that of uranium. The rays given off by potassium are β rays. Tests with sodium gave negative results.† The radioactivity of potassium has been confirmed by McLennan.[86] The fact that potassium is an essential constituent of the food of most plants renders its radio-

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* In the same paper Lord Kelvin describes a model of an atom to illustrate the mode by which, according to his idea, the α and β particles are given off.

† In 1907 Professor J. J. Thomson[131] succeeded in getting electrification from both heating and rubbing various salts (phosphates and oxides), and explains the result as due to the hypothetical fact that the salts are covered superficially with a double layer of electrification, one layer negative, the other positive. The heating or rubbing removes the outer layer and leaves the inner one unbalanced. He suggests that the electrifying of bodies by friction results from the removal of one or both layers of electrification by the rubbing.

activity a matter of especial interest and importance to the plant physiologist.*

Reasoning from various experiments, Thomson[130] states that, "a radioactive substance, apparently radium, is exceedingly widely distributed, occurring in the most unexpected places," *e. g.*, soil, bricks, glass, sea sand from Whitby beach, Yorks ("exceedingly rich"), one specimen of wheat flour (others none), all clays, sands, and gravels that were examined. Nearly all bodies, he states, emit radiations which can ionize a gas. Experiments of Wood[148] lead to the same conclusion, and Strutt[119, 120] has calculated that the amount of radioactivity necessary to compensate for the radiation of heat by the earth is much exceeded by the activity of ordinary materials. The spontaneous ionization of the air in closed glass vessels may possibly be partly accounted for by the fact that clay and other silicates are known to emit large numbers of ions.[66] †

In this connection may be recalled the paper by Eve,[56] describing the infection of the entire physics-building of McGill University with radium. The use of the radium had been confined to one portion of the building, but, later, objects from all parts were found to be strongly radioactive. "Sheets of mica, lead foil, iron, zinc, and tin were all active, even when taken from drawers and cupboards." About 90 per cent. of the activity could be removed by solution in strong hydrochloric acid, and the acid thereby became active. The influence has spread from room to room, and from floor to floor in the large building.

Lenard[78] has found that when light of short wave-length falls on metal surfaces slowly moving ions are shot into space. Their initial velocity varies, not with the intensity, but with the quality of the light, and they ionize gases through which they pass, thus making them conductors of electricity.

THE EFFECT OF RADIOACTIVITY ON SURROUNDINGS: In addition to causing excited radioactivity, as noted above, radioactivity may affect the plant's surroundings in other ways. Oxygen, for example, is changed to ozone by the rays from radiferous barium. Furthermore a slight rise in temperature may result. Curie and Labord[31a] were the first to announce the emission of heat from radium. One

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* Phillips[91] has recently (28 May, 1908) reported evidence of an emanation from sodium that will discharge an electroscope negatively (but not positively) charged. Whether this is a true radioactive gas, or not, has not yet been demonstrated.

† Cf. also Wood, A.[147]

gram of "pure" radium emits a quantity of heat equal to 100 gram calories per hour, or 2,400 gram calories in one day—nearly enough to dissociate one gram of water (3,900 gram calories).*

RELATIVE IMPORTANCE OF THE EMANATION: A large part of the radioactivity to which the sub-aërial portions of plants are exposed is due to the emanation diffused in the atmosphere. Rutherford states that about three fourths of the activity of radium is due to the emanation, and he has calculated that, if we could get 1 c.c. of radium emanation, it would raise to a red heat the glass tube containing it, and light up an X ray screen brilliantly through one foot of solid iron. This evolution of energy would continue for several days without much change, and would be appreciable after one month.

This chapter contains a portion of the evidence, accumulated during the past ten years or more, that radioactivity and free electrons are ecological factors, a part of the normal environment of probably every plant. Undoubtedly there will be considerable modification of detail as the science progresses, but the main fact of the wide distribution of radioactivity in nature seems now to be firmly established. In what way, and to what extent it influences physiological processes and morphological expression remains largely to be determined.

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* Though Paschen[90a] finds reasons for thinking that 50 per cent. of the heating effect is due to the gamma rays, the experiments of Rutherford and Barnes[98a] indicate that this effect is due mainly to the bombardment by the a rays.

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CHAPTER III

PREVIOUS INVESTIGATIONS UPON ANIMALS

The recent development of the science of the electron has so unified physical and chemical phenomena that only artificial boundaries now exist between the physics of heat waves, electro-magnetic waves, light waves, ultra-violet light, X rays, and the γ rays of radium. They differ only in degree, not in kind, and so their physiological rôles are unified, and effects caused by any one of them throw light on the results produced by all the others. Likewise, as is well known, it is becoming increasingly difficult to draw the line between physiological investigations in botany and those in zoölogy. They both center in the common field of cell-physiology, so that very often results obtained with plants can be adequately interpreted only in the light of investigations made with animals, and *vice versa*.

For these reasons it has seemed best to include, in the following historical review, the literature dealing with the effects, not only of radium rays, but of X rays and of ultra-violet rays as well on both animals and plants. The results of the studies of the physiological and therapeutic effects of X rays were brought together by Schiff[51] in 1901, and no attempt is here made to compass the voluminous literature bearing on X ray therapy and radium therapy.

In 1899 Schaudinn[50] announced that the various protozoa react differently to the stimulus of Röntgen rays; certain species do not react at all, some only slightly, and still others very strongly. The dissimilarity in reaction appears to be connected with differences in the structure of the protoplasm. Those which react quickly possess a looser plasm than those which react slowly or not at all.

The now familiar fact that radium rays will cause a painful "burning" of the skin was first recorded by Walkhoff,[60] in October, 1900. This effect was noted in the same year by Giesel,[24] who laid a double walled celluloid capsule containing 0.27 gr. of radium-barium bromide for 24 hrs. on the inner surface of his arm. The first effect noticed was a slight reddening of the exposed region, and, after two or three weeks, a strong irritation with pigmentation, and finally a peeling off of the skin, followed soon thereafter by healing. Rutherford[46] describes an inflammation that lasts from 10 to 20 days, after which the skin peels off, while the pain endures for two months. He states that these results are due mainly to the α and β rays. Becquerel and Curie[7] obtained the burns with a preparation of only 800,000 activity, and found that the effect varied with the intensity of the active rays and the duration of the exposure. Personally I have never experienced any inconvenience in handling sealed glass tubes of radium bromide of activity as high as 1,800,000, though I have never taken any special care to avoid injury. I have carried a wooden, velvet-lined case containing the preparation in my vest pocket for several hours, and in my ungloved hand for as long as half an hour, without ever having experienced any "burning" or other unpleasant effects.

Joseph and Prowazek,[32] exposing *Paramoecia* and *Daphnia* to X rays, found that those organisms show a negative tropism with reference to the rays. The plasm of *Paramoecia* and *Bryopsis* undergoes modifications which were interpreted to signify either injury or exhaustion.

In much of the literature, especially the earlier, the amount of radium used and its activity are not given. Bohn[9] found that embryos of *Bufo vulgaris* grew more slowly after exposure to the rays of radium, and exposed tadpoles of the frog developed into monstrosities. He also says that if the rays of radium act upon the body of an animal for several hours, the tissues acquire new properties, which may remain in the latent state for a long period, but manifest themselves as soon as the normal activity of the tissues is resumed. The activity of his preparations is not given.

In a later paper Bohn[10] states that if sea-urchins (*Strongylocentrotus lividus*) are exposed for from 20 minutes to 2 hours after gastrulation, the plutei are small and atrophied. The rays rapidly enfeeble or kill spermatozoids, but, on the contrary, eggs submitted to their influence seem to become more susceptible of being fecundated, and are increased in vitality. The rays affect the nuclear chromatin especially, increasing its activity or destroying it, according to the duration of exposure. Spermatozoids are more sensitive than ova, because they consist of almost naked chromatin, but the chromatin of eggs, protected by the cytoplasm, is so stimulated, he says, as to produce parthenogenesis. This last result has not been confirmed.

Danysz[14, 15] found the tissues in the peritoneal cavities of guinea pigs less sensitive than the skin, but the central nervous system infinitely more sensitive. One centigram of radium in a sealed glass tube, placed above the backbone and part of the cranium of a mouse one month old, produced phenomena of ataxia and paresis in about three hours. Tetanus convulsions followed in from seven to eight hours, and, if the tube was left in place, death ensued in twelve to eighteen hours. The older the mouse, the less sensitive he was to such exposure. At the age of three to four months death did not result for from three to four days, and at the age of one year not until from six to ten days. Danysz attributed the increased resistance to the transformation of the walls of the neural cavity from cartilage to bone. Caterpillars of *Ephertia* are paralyzed by the emanation, and anthrax germs cease developing after an exposure of 24 hours in an atmosphere charged with the emanation. The microbes which produce the proteolytic enzymes of autodigestion are specially sensitive. In his later paper he[16] states that the epithelial tissues of young animals are more sensitive than those of adults.

A. Exner[22] found that blood, hair, nails, and muscle fiber, and especially the crystalline lens, are made phosphorescent by the rays, and Hardy and Anderson[26] concluded that the sense organs of higher animals are not at all affected by them. The sensation of diffused light, caused by bringing a few milligrams of a radium salt near the head is purely of retinal origin, and not due to a response of the optic nerve or brain. The tissues of the eyeball give out this diffused light under the influence of the β and γ rays. Fresh lenses of sheep, ox, and rabbit, and also skin, fat, and muscle are made to glow when exposed to the rays. The eyelids are extremely opaque to radium rays, and this possesses added interest in view of the fact that a penetrating radiation exists in the air (p. 25). Exner's observations were confirmed in 1904 by Bouchard, Curie and Balthazard.[12] In the same year S. Exner,[23] exposing the tails of mice, determined that the β and γ rays are both physiologically active, the former less so than the latter.

According to Perthes,[42] an intensity of X rays not sufficient to kill the cells of man and the chick, greatly retard cell-division. The rays similarly affected the cells of *Ascaris*.

Radium rays were found by Schwartz[54] to affect the yolk of eggs more than the albumin. He considered that the rays decomposed the lecithin of the yolk, and that they affect all tissues in the same way, for cells rich in lecithin, he says, are the most sensitive to the rays. On the contrary Neuberg[41] concluded from his experiments that the rays can decompose neither pure lecithin nor proteins, and probably do not decompose them within the cell.

In 1904 von Baeyer[1] found that both the alpha and the more penetrating rays from radioactive lead, polonium, and induced silver and palladium cause the death of bacteria, but that the alpha rays do not affect the skin. He holds the opinion of Scholtz that the effect on the skin is to be ascribed to the penetrating rays.

The effect of Röntgen rays on regeneration in planarians was investigated by Bardeen and Baetjer.[5] Their experiments showed that cell-division may be retarded and entirely stopped by a sufficient exposure to the rays. They also noted that the effects did not appear for some days after the first exposure, and that the rays have an effect, not so much upon tissue differentiation, as upon cell reproduction. The rays were found to affect primarily cells possessing reproductive capacity, and the authors suggest that "death in exposed specimens may possibly be due to a necessity on the part of the organism for a certain amount of cell-reproduction."

The physiological effects of the radium emanation were studied by Bouchard, Curie, and Balthazard,[12] and it was found to kill guinea pigs and mice within one or two hours, according to the quantity of the emanation used. The effect was shown not to be due to ozone produced by the radioactivity of the emanation.

In 1904 Caspari[13] reviewed the physiological investigations with the rays of radioactive substances since Becquerel announced the discovery of Becquerel rays, and in the same year Danysz[16] observed that the epithelial tissues of young animals were more sensitive to these rays than those of adults.

According to Dorn and Wallstabe,[18] rabbits were not affected by drinking tap-water which had absorbed the emanation, but were poisoned by an exposure of one and one half weeks to air which contained it. Their lungs were found to be hyperemic.

Dunham[21] observed that *Chilomonas* and two species of *Paramoecium* were killed by six exposures of three minutes each to X rays on three successive days, while rotifers, *Arcella*, and *Cryptomonas* were not affected by that treatment.

The first recorded attempt I have found to ascertain the independent effect of the different kinds of radium rays was made by A. Exner[22] in 1904. He separated the β from the γ rays by means of the magnet, and found that they both produced the same kind of result on the tails of mice, but the β rays were less active than the γ rays. In the same year Gillman and Baetjer[25] found that the eggs of *Amblystoma*, exposed to X rays, developed faster than normally for a few days, though eventually their development was markedly altered and checked. These authors announce that similar results were obtained by Bardeen with the hen's egg.

When invertin, emulsin, and trypsin were exposed to the radiations from radium, Henri and Mayer[27] found that their activity gradually diminished and was finally entirely lost after several days exposure.

Perthes[43,44] found that eggs of *Ascaris megalocephala*, exposed in drop cultures to radium and to Röntgen rays, had their first division delayed, and their further development made irregular and slower than normally. Eggs in the resting or in the dividing condition served equally well for the experiments. Centrosomes and spindle fibers were unaffected, but in *Ascaris megalocephala univalens* the characteristic number of chromosomes was doubled. In the course of the chromatin loops there appeared knotty swellings instead of the normal, club-shaped enlargements. In a few eggs, instead of the usual two chromosomes on the equatorial plate, there were observed numerous, unequal pieces, though Perthes suggests that this may have resulted from the mode of sectioning, and says, "I cannot consider that a disintegration of the chromosomes by Röntgen rays has been demonstrated." The eggs exposed to X rays[44] gave rise to abnormally developed worms.

Phisalix[45] exposed the venom of a viper for periods of 6, 20, and 58 hours to radium rays. Its toxicity was decreased by the shorter exposures and finally destroyed by the longer. By means of radium rays Tizzoni and Bongiovanni[55-58] and Novi[41a] obtained an attenuated virus of rabies (*in vito*). The length of exposure necessary to render the virus inoffensive varied with the organ in which the injection was made. *Nassula* and *Trypanosoma Brucei*, studied by Salomonson and Dryer,[47] were killed in from two to three hours by rays from radium, and cyst-forming amoebae were injured. The contractile vacuoles of ciliates were distended, and their period of contraction prolonged. In 1905 Bongiovanni[11] also announced to the Bologna Academy that radium rays rapidly destroy the virus of rabies, both in tubes and when applied to animals, within an hour of their infection. "Animals already suffering could be cured with certain results." Danysz * was unable to reproduce the results of Tizzoni and Bongiovanni.

According to Schaper,[49] an exposure to radium rays retarded and modified the regeneration of the tail in *Triton* larvae, and regeneration of *Planaria lugubris* was similarly affected by an exposure of three and one half hours to the rays, and of 5 mm.-larvae of *Rana esculenta* by the emanation. Cell-division, embryonal differentiation, and growth were inhibited after a longer or shorter latent period. Eggs of *Rana esculenta*, exposed at various stages of segmentation or early differentiation of the embryo, were retarded in development, and the embryos were small and deformed.

The destruction of the activity of chymosin by radium rays was attributed by Schmidt-Nillsen [52, 53] to the ultra-violet rays caused by the phosphorus in his preparations. When Venenziani[55] placed specimens of *Opalina ranarum* (a ciliated parasite, living in the intestinal fluid of frogs) in a 5 per cent. sodium chloride solution, and then exposed them to rays from 0.1 gm. of radium salt of 10,000 activity, they lived longer than control specimens, similarly placed, but not exposed to the rays. In water the exposed organisms survived still longer.

Willcock[61] found that *Euglena viridis* manifested no tendency to avoid or to accumulate in the path of rays from barium, but Jennings[31] had already found it almost impossible to obtain any directive response to other stimuli from *Euglena*. The barium rays seemed to hasten spore formation in small encysted forms, and encysted forms of the larger variety were made active by 24 hours' exposure to the rays from 5 mg. of radium bromide. *Stentor viridis* contracts when the rays fall upon it. Repeated exposure results in a marked decrease in irritability and in the capacity for contraction and extension. *Hydra fusca*, which contains no chlorophyll, gave no tropostic response, even during exposures that resulted in death, but *Hydra viridis*, which contains a green algal symbiont, does manifest a negative radiotaxis which decreases with fatigue, as with *Stentor*. The response takes place both in full daylight and in absolute darkness. An exposure of two hours to α, β, and γ rays acting together caused *Hydra fusca* to disintegrate, and the result was attributed to the effect

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*Ann. Inst. Pasteur 20: 206. 1906.

on the nervous system of the oral disc, for this was the region first affected, and no response at all was obtained when the foot was exposed. *Opalina*, *Nyctotherus*, and *Balantidium*, organisms without chlorophyll, suffered no obvious harm, and manifested no response when exposed for 24 hours to rays from 50 mg. of radium bromide, though these organisms are very sensitive to ordinary stimuli. Negative results were also obtained with *Actinosphaerium* and rotifers. Thus, only forms containing chlorophyll appeared to be sufficiently sensitive to the rays to react, and the author suggests that the green algae, living symbiotically with *Hydra viridis* and *Stentor*, act as sense organs for the beta rays, possibly through a modification of the metabolism of the alga by the rays, and a consequent disturbance of the balance between the two organisms. This author's brief note on experiments with plants is referred to on page 62.

In harmony with an inference of Willcock with reference to *Hydra*, is the result of Beck,[6] who found that radium rays either deaden or destroy the sensibilities of the peripheral nerves. In the same year (1905) Salomonson and Dryer[48] stated that rays from 5 mg. of "pure" radium bromide, passing through a sheet of mica, had little effect on the protozoan *Nassula*, even after six days. Some specimens of amoebae were killed in twelve hours, while others survived for four days. *Trypanosoma Brucei* was killed in from two to three hours, and on red blood corpuscles the rays exerted a haemolytic power.

Hewlett[29] has suggested that the inhibition of cancerous growth by radium and X rays may be because the rays cause proliferation of the connective tissue elements of the growth, and thus interfere with its nutrition. "It is possible," he further says, "that the stimulus of these rays may also act like fertilization, and cause the gametoid once more to revert to the somatic cells." No experimental evidence, however, is adduced in support of either of these hypotheses.

A thorough study of the effects of radium rays on chlorophyllous and non-chlorophyllous organisms was made in 1905 by Dr. Margaret Zuelzer.[62] She exposed large numbers of *Spirostemum ambiguum* and the chlorophyll-containing *Paramoecium bursaria* to the rays, and found that, after 24 hours' exposure, from four to seven out of ten *Paramoecia* had divided, but not so in the case of the *Spirostema*. Cell-division of the chlorophyll-containing *Paramoecia* had ceased after 30 hours of exposure. The animals which had not been noticeably altered by a radiation of 36 hours then moved more slowly, and, after four to seven days of exposure, gradually swelled up and disintegrated. In preserved specimens the macro-nucleus appeared swollen, and deeply staining, and Dr. Zuelzer suggests that the greater resistance of the chlorophyll-containing organism is possibly due to an inhibition of the injurious effects of the rays by the oxygen given off by the chlorophyll in the presence of light.

The source of this suggestion of Dr. Zuelzer's was the work of Hertel,[28] who studied the effect of waves of ultra-violet light on organisms. He found that organisms that contain chlorophyll are more resistant to the injurious effects of the ultra-violet rays of a wave-length of 280 $\mu\mu$ than the chlorophylless organisms. The protoplasmic streaming in cells of *Elodea* (*Philotria*) was retarded more promptly in the dark than in light, though darkness in itself, at least within one hour, does not affect the process. Illuminated leaves were not affected as readily as those not illuminated, and *Paramoecium bursaria* Ehrenberg, which contains chlorophyll, was less sensitive *in light* than species without chlorophyll. Hertel thinks that the oxygen given off in the light by photosynthetic processes inhibits the injurious effects of the ultra-violet rays. He further found that the rays accelerate and favor the decomposition of hydrogen peroxide when that process has been started by a catalyzing body, and finally concludes that the effects of the rays on the living cell are due to this influence on the proportion and distribution of oxygen in the cell. Hertel's results with hydrogen peroxide possess added interest in light of the studies of Neilson and Brown[40] on the effects of ions on the decomposition of hydrogen peroxide by platinum black. The cation has an inhibiting or depressing effect, the anion an accelerating influence. In these experiments solutions of 22 different salts of sodium, and of nine different chlorides were used, with concentrations of from $n/8$ to $n/512$.

If it shall be determined, says Dr. Zuelzer, after referring to Hertel's work, that ultra-violet rays, as well as those of radium, can influence the metabolism of the cell by depriving it of oxygen, then the results of Venenziani[59] may find here their explanation, for the *Opalina ranarum* with which he experimented, lives normally in a medium (the intestine) poor in oxygen. Continuing, she says that the disturbance of spermatogenesis and the killing of the spermatozoa in healthy tissue, the quick destruction of the skin and of malign tumors with their rapidly dividing cells, may possibly depend upon the same property of the rays. If the exposure of the animals was not too prolonged they recovered. Thus, rapidly creeping specimens of *Amoeba limax* and *Pelomyxa palustris* came to rest and contracted after an exposure of three to four hours. In this condition they lay unchanged for 24 hours, but if the radium influence was then withdrawn they revived completely, and crawled about normally after two hours. Dr. Zuelzer's results were confirmed the following year by Hussakof,[30] who states that from one to two hours exposure produced no effect upon amoeba and paramecium.

The experiments of Berg and Welker[8] on the metabolism of young dogs led them to the conclusion that: "Radium preparations of low activity (240, 1,000, 10,000) containing barium bromide in preponderating proportions were without special influence on metabolism when administered per os or subcutaneously in relatively large quantities. Equal or larger doses of pure radium bromide also failed to show any decisive effects before fatal results were inaugurated."

The effect of prolonged exposure to radium of weak activity was studied by London.[35, 36] He does not give the activity of his preparation, but rabbits were exposed to 260 mg. of it for 14 months with injurious effects both to external and internal organs. The electric organ of the torpedo was exposed by Mendelssohn[37] to rays from 3 mg. of radium bromide of 1,800,000 activity in a glass tube. No effect was observed until the end of the first hour of exposure. Then, after a period of 20–30 minutes of stronger activity, there took place a gradual weakening of the discharge, which fell to a minimum in five to six hours. This enfeebled discharge continued for six to eight days, but no complete suppression of the function of the electric organ of the torpedo, due to the radium rays, has been observed.

In 1906 Meyer[38, 39] ascertained that, "By whatever channel radium is introduced (into the animal body) it seems to find its way into practically all the fluids of the body," and in 1907 Bardeen[3] demonstrated that, when the ova of toads were fertilized by spermatozoa that had been previously exposed to the X rays, the ova developed abnormally. Similar results were obtained later in the same year[4] with *Rana pipiens*, and the least abnormal and longest survivor out of 250 larvae, died one week after the eggs were fertilized.

The coagulation of albumins by the actions of ultra-violet light and of radium was studied by Dreyer and Hanssen.[20, 21] They found that both serous and egg albumin are coagulated under the action of an intense light; the serum of the horse only slightly. A solution of peptone remains clear, though becoming yellow, and the same effect was noted with casein. These results are all attributed to the ultra-violet portion of the light. Radium rays were found to coagulate vitellin, but not other albumins.

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CHAPTER IV

PREVIOUS INVESTIGATIONS UPON PLANTS

1. Effects of Röntgen Rays on Plants

In the year 1892 Leo Errera,[3, 4] by a series of painstaking experiments with *Phycomyces nitens* and other plants, demonstrated the fallacy of the notion of "physiological-action-at-a-distance," which had been put forward in 1890 by Elfving.[1, 2]* It is not surprising, then, to find him one of the first investigators to study the physiological action of the then new kind of rays, the X rays, on living plants, for Röntgen's discovery belonged to a series of brilliant investigations which have resulted in the almost, if not wholly, complete abandonment of the idea of action-at-a-distance of any kind. Errera,[22] however, was unable to determine the slightest response of *Phycomyces nitens* when it was exposed to these rays.

Soon after the discovery that Röntgen rays are a component of sunbeams, Müller[50] raised the question as to whether or not the rays thus occurring exerted any influence upon plants. He placed specimens of garden cress in a dark chamber, protected from sunlight, and found that their stems turned towards the rays as in phototropism. By this means, and also by using phosphorescent substances, Müller reached the conclusion that the X rays present in sunbeams do act upon plants.

Contrary to these results, Schober[55] failed to observe in oats any tropistic response with reference to X rays, but found that an exposure of one hour to the rays did not cause the seedlings to lose their phototropic sensitiveness.

Studies of the action of X rays on bacteria also began in the year 1896. The literature on this subject alone has become so voluminous that it would not be desirable nor profitable to review it here in detail, and no attempt is made even to cite all of the published papers. Practically all investigators have obtained one or the other of two kinds of results; either negative results, or an injurious effect on the bacteria. Among those who have obtained negative results are Mink[47]

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* Errera's interpretation of "physiological-action-at-a-distance" as merely a special case of hydrotropism, was confirmed by Stryer[5] in 1901.

with the typhus bacillus, Wittlin[63] with *Bacillus coli-communis*, *B. typhi*, diphtheria bacilli, *Staphylococcus aureus*, *Spirochaeta cholerae*, and *Tyrothryx tenuis* Duclaux; Beck and Schultz[11] with various bacilli; Atkinson[9] with bacteria; and Freund,[23] who exposed bacteria to both X rays and the rays from uranium.

Röntgen rays were found either to inhibit growth or to kill by Rieder[52, 53] with *B. prodigiosus*, the cholera bacillus, and other forms; by Tolomei[58] with *B. anthracis;* by Strebel[57] with bacteria exposed to Becquerel rays; and by Aschkinass and Caspari[8] with bacteria exposed to Becquerel rays and to cathode and X rays. Rieder's results disagree with those of Krüger and Friedenthal, who, he says, state that bacteria could be killed only when electricity is conducted through the culture, thus by electrolysis, forming anti-bacterial substances. He states that he carefully excluded from his cultures any possible influence from heat rays, rays from fluorescent light, and electric currents.

In 1897 Atkinson[9] reported that etiolated plants recovered their green color less rapidly than normally after exposure to the rays, and interpreted this as suggesting some injury to the chloroplasts. No other influence was observed, and studies on the absorption of the rays by species of *Mucor*, bacteria, and *Oscillatoria* gave negative results. On the question of the absorption of the rays by the parts of plants, however, Hinterberger[35] had already shown that fruits containing little sap, and large cavities, such as beans and pea pods are most easily penetrated by the rays, while thick buds and fleshy fruits, such as pears and cucumbers, are very impenetrable.

Lopriore[44] studied the action of X rays on the protoplasm of the living vegetable cell. With an exposure of not more than half an hour the protoplasmic streaming of *Vallisneria spiralis* was accelerated. After this time, if the influence of the rays is removed, the motion again becomes normal. An exposure for one hour is deleterious; the protoplasm continues to stream, but takes on a yellow tint, and becomes vacuolated and granular. After an exposure of two hours the streaming had not ceased, but the chloroplastids had begun to fade. Pollen-grains of *Genista* and of *Darlingtonia* did not germinate while exposed to the rays, but began to do so after the influence of the latter was removed.

In 1898 Atkinson[9] reported negative results from experiments with germinating seeds, seedlings, and mature seed plants, *Mucor*, and *Mimosa pudica.* He found that the various plant tissues absorbed the rays differently, so that X ray photographs may be taken which disclose certain details of internal structure, such as vascular bundles.

Maldiney and Thouvenin[45] succeeded in accelerating the germination of seeds of *Convolvulus arvensis*, *Lepidium sativum*, and *Panicum miliaceum* by exposing them to the action of X rays. From the fact that these seedlings as they came from the seeds were yellow as usual, the authors conclude that the rays are without influence on the formation of chlorophyll.

Tolomei[59] states that Röntgen rays act upon plants like light. Under their influence the leaves of *Philotria canadensis*, in water containing CO_2, give off bubbles of gas as in sunlight. Like light also, the rays retard the absorption of oxygen by *Mycoderma aceti*, and also the production of CO_2 by beer-yeast.

By exposing seeds for about an hour daily for several days to X rays, Wolfenden and Forbes[64] induced an acceleration of germination, and two years later (1902) Seckt[56] published the results of his studies on the influence of X rays on the plant organism. He found, as did Lopriore, that the rays have a decided accelerating influence upon the protoplasmic streaming in hairs of *Cucurbita Pepo*, *Tradescantia virginiana*, and *T. Selloi.* Seckt remarks that this effect may be similar to that called forth by poison or by wound stimulus, by which the organism is stimulated to an abnormally aggravated vital activity. *Spirogyra*-cells were plasmolyzed at a distance of 10–12 cm. from the X ray tube, but were indifferent to the rays at greater distances. Increased turgor could also be called forth by the rays. The guard cells of the stomata of *Tradescantia Selloi*, and the pulvini of *Mimosa* and of *Oxalis*, under the influence of the rays undergo an increase of cell-pressure which may have its cause in a peculiar influence upon the protoplasm of the cells.

The latest studies of the effects of Röntgen rays on plants are those of Koernicke[38, 40, 41]. In his first paper (1904) he announced that the immediate effect of exposure is an acceleration of growth, an effect similar to that which Townsend[60] found to occur in plants after a slight wounding. Finally, however, growth was retarded. The time intervening between exposure and the decrease in the rate of growth varies with the plant and its physiological condition at the time it was exposed. *Brassica napus*, for example, is especially resistant to the rays, not being at all affected by an intensity of ray that calls forth a strong reaction in *Vicia Faba*. If the rays are not sufficiently strong no retardation occurs at all. Roots whose growth is inhibited by the rays for a certain period will resume their growth. The centgener power of seeds was not affected by two exposures to rays of the intensity employed (20 Holzknecht units), and the author suggests that perhaps Röntgen rays of a certain intensity may act as a stimulus to germination, but such experimental effects were not recorded. Further results obtained by Körnicke by treatment with X rays are mentioned in connection with his experiments with radium.[39, 42]

2. Effects of Radium Rays on Plants.

The first recorded observations of the effects of radium rays on plant tissues were made by Giesel,[28] two years after Madam Curie's discovery. He announced that the rays produced a bleaching of the chlorophyll in leaves, causing them to assume an autumnal yellow throughout, with a brown coloring on the side exposed to the radium. Paper in which radium preparations have lain for a long time becomes brown and brittle, and celluloid loses its firmness. These effects of intense radium rays, said Giesel, hint at a molecular rearrangement, whereby their physiological effects upon plant and animal cells may be explained.

Aschkinass and Caspari[8] reported that Becquerel rays "of the second type" (β rays), as well as X rays, are injurious to bacteria, and a few weeks before this Becquerel[12] announced that an exposure of a week or more to radium rays destroyed the germinating power of seeds of cress and white mustard. Negative results followed an exposure of only 24 hours. These experiments were made in Becquerel's laboratory by Louis Matout.

Danysz[14] studied the pathogenic action of the rays and "emanations"* given off by radium on different tissues and organisms, and found that all species of bacteria are hindered in their development by radium rays, but that certain kinds, notably those which produce proteolytic enzymes, are more sensitive than others, and are killed under certain conditions of exposure.

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*As explained in Chapter I, there is but one "emanation" given off by radioactive substances. It is difficult to know to just what Danysz refers by "emanations."

In November, 1903, Dixon[18] reported that the growth of cress seedlings was retarded by the rays from 5 mg. of "pure" radium bromide in a sealed glass tube supported at a distance of 1 cm. over seeds sown uniformly on moist sand in the dark. The retardation was apparent only in those plants situated within a radius of about 2 cm. from the tube, and on these seedlings the root-hairs were fewer and shorter than on the others. No curvatures were evoked by the rays, and when the tube was placed in a vessel of water containing *Volvox globator* in the dark, the *Volvox* showed no signs of attraction or repulsion, or other response. Later in the same year Dixon and Wigham[19] announced that β rays from radium bromide exercise an inhibitory action upon the growth of *Bacillus pyocyaneus*, *B. typhosus*, *B. prodigiosus*, and *B. anthracis* in agar cultures. An exposure for four days at a distance of 4.5 mm. to rays from 5 mg. of radium bromide did not kill all of the bacilli, for a tube of broth, inoculated from a patch thus exposed, developed organisms.

Hoffmann[36] found *Staphylococcus pyogenes aureus*, and milk bacteria more resistant than *B. prodigiosus*. The latter on an agar plate was killed by an exposure of three hours to rays from 5 mg. of the bromide, passing through a mica plate at a distance of 3.5 mm. Pfeiffer and Friedberger[51] also found the spores more resistant than the bacteria of typhus and cholera, the latter being killed by an exposure of 48 hours to rays from radium bromide.

On the contrary, Van Beuren and Zinsser[61] obtained negative results in every experiment with *B. typhosus*, *B. pyocyaneus*, and *Staphylococcus pyogenes aureus*. They employed 12 mg. of radium of 300,000 activity, with exposures varying from 8–14 hours, and at distances of from 1 cm. to 0.5 cm. Negative results also followed the exposure of the fore arm for one and a half hours with one thickness of a linen handkerchief intervening.

Abbe[6] stated that the germination and growth of rape were retarded in proportion to the duration of the exposure to rays from "a grain or two of radium salt," of activity not given. The seeds were exposed before planting in soil, and exposure of the same kind of seeds to X rays was followed by similar results. Later in the same year (1904) Abbe[7] reported that the power of seed-germination was weakened and finally inhibited by exposure of from two to ten days, but he does not mention the kind of seed nor the amount nor tivity of the radium salt used.

Experiments on the physiological effect of α rays were made by Baeyer.[10] He employed radioactive lead and "induced" silver, palladium, and polonium. The α rays from these preparations killed bacteria, but did not affect the skin.

Dauphin[15, 16] was the first to investigate the effect of radium rays on the lower fungi. He found that growth was retarded in cultures of *Mortierella*, *Mucor*, *Piptocephalis*, and *Thamnidium*. Spores of *Mortierella* would not germinate in the neighborhood of the radium tube, but germinated as soon as the tube was removed. The growth of hyphae was arrested, and outgrowths were formed on the filaments. The plasma withdraws from the influence of the rays, and septation of the hyphae takes place, followed by encystment. On the removal of the radium, growth recommences.

The most extensive paper, up to the date of its publication, was that of Dixon and Wigham,[19, 20] which appeared in March, 1904. In all of their experiments these authors used 5 mg. of radium bromide in a sealed glass tube, but the activity (presumably 1,500,000) is not given. They found that the seedlings of *Lepidium sativum* were not radiotropic, but grew less rapidly, and had fewer and smaller root-hairs when exposed to the rays. *Volvox globator* ("positively photoscopic") gave no reaction to the rays. The failure of *Bacillus pyocyaneus*, *B. prodigiosus*, *B. typhosus*, and *B. anthracis* to develop under the influence of the rays is attributed, not to the direct effect of the rays on the organisms, but to a probable change in the agar. The effects are thought to be due more to the β than to the γ rays, for the latter are too penetrating to be absorbed by 30 mm. of air, and beyond this distance the radiations were apparently without effect. It is suggested that the electrons, emitted directly by the radium, or produced indirectly by it, were partly absorbed by the bacterial cultures. They possibly attach themselves to the positive ions of the cultures, among which are the hydrogen ions of the water. Thus OH ions would be set free, and the water in the protoplasm would become alkaline. This would check the action of the enzymes on which the metabolism of the cells depends, for most enzymes, except trypsin, are inhibited in an alkaline solution. In support of this hypothesis it was found that colorless phenolphthaline, diffused through an agar culture, acquired a feeble pink color when the preparation was exposed for a day or two to the radium rays. This the authors explain as being due to the liberation of negative OH ions through neutralization of the H ions by the electrons from the radium. It is recognized that the coloration may be due to direct ionization of the phenolphthaline as well as of the water. Further experimentation, however, showed that the detection of electrons by means of this indicator is very uncertain. The same agar preparation became pink when supported over a solution of caustic potash * in a closed chamber.

In a note on the action of radium on microörganisms, A. B. Green[29] states that the specific germ of vaccine is killed after 22 hrs. exposure to radium rays from 1 cg. of "practically pure radium bromide," contained in a vulcanite and brass capsule, fronted with thin talc. Only the β and γ rays were thus available. *Staphylococcus pyogenes aureus*, *S. pyogenes albus*, *S. cereus flavus*, and *S. cereus albus* were less resistant than the specific vaccine organism, and spore-forming bacteria were most resistant, requiring 72 hrs. to kill. It is further stated that, after an exposure of from 24–120 hrs., the microörganisms themselves may exhibit radioactivity. It is considered uncertain as to whether or not living organisms can acquire this power, but those killed by the rays can do so. It is impossible to conceive how the organisms, dead or alive, could become radioactive as a result of their exposure to a radium salt in a *sealed* container. If the container was not perfectly sealed, however, then traces of the emanation might diffuse out and thus cause induced activity of the organisms.

Willcock[62] stated that an attempt by Mr. Hardy to secure photosynthesis in liverworts by the energy of radium rays was unsuccessful, and that Dr. Anderson had shown that the tissues of leaves may be killed by the rays.

Koernicke's[39] paper on the influence of radium rays on germination and growth appeared twelve days after that of Dixon and Wigham. He found that the growth of the roots of *Vicia Faba* in sawdust was inhibited when a sealed glass tube of radium bromide was placed close by the elongating tip. The radium was removed at the end of four days, but growth was not resumed, though the roots remained alive for over a month after they had ceased to grow. When dry seeds of *Vicia Faba* were exposed for 24 hrs. the subsequent growth of the root was retarded, but the shoot did not appear

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* The radioactivity of potassium, discovered by Campbell (p. 32), was not known at the time of publication of Dixon and Wigham's experiments.

affected. Adventitious roots sprang from the epicotyl, and three days after this the main root died. The intervention of three days between exposure of seeds and planting seemed to make no difference in the result. The effect of the rays decreased with the distance, and apparently ceased at a distance of four centimeters. When roots of *Vicia Faba* seedlings 3–11 cm. long were exposed to rays from 5 mg. of radium bromide for five days, the roots near the radium tube grew longer at night, and the portions lying back of the tip grew a little to one side in the region of the radium influence. Growth ceased on the fourth day.

Seeds of *Brassica napus* seemed specially resistant both to radium and to X rays. Five days exposure with 10 mg. of the bromide did not interfere with germination and further development. Swollen seeds irradiated with the same mass of radium salt showed an acceleration of germination, but subsequently the rate of growth became normal. Removal of the seed-coats of *Brassica* did not make them any more sensitive, for development of these seeds was normal after they were exposed. Ten days' exposure to rays from 10 mg. retarded germination and growth, and exposure for a longer time completely destroyed the power to germinate.

Growth of both root and shoot of *Vicia* was retarded by exposing the vegetative points to the rays, and in this respect etiolated and non-etiolated specimens behaved alike. The growth of callus on wounds of *Populus alba* was retarded by the rays.

Germination of the spores of *Aspergillus niger* was inhibited by two days' exposure under the radium tube, and the mycelia that developed near the tube did not fruit. Körnicke considered these non-fruiting hyphae in a condition of "latent life," an interpretation which Dauphin had applied to spores of *Mortierella* which failed to germinate while exposed, but did so after the radium was removed. Dry conidia of *Aspergillus niger*, irradiated for 1–4 days with 10 mg. of the bromide, did not lose their power to germinate, though the germination was more or less delayed according to the length of exposure.

London's[43] experiments indicate that the vitality of bacterial cultures was destroyed after an exposure of two days to the "emanations." *

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* See footnote, p. 8. This word is doubtless not used here in the sense of *emanation*, as defined by Rutherford, but as a collective term, referring to all the influences from the radium, especially the rays.

Dorn, Baumann, and Valentiner,[21] experimented with the gaseous emanation, and found that *Bacillus typhosus*, cholera germs, and bacilli of diphtheria, exposed to its influence, were killed. The effects were attributed to the β rays * given off by the emanation, and not to the gas itself, apart from its radioactivity.

In August, 1905, were published Koernicke's[40] further researches concerning the effect of Röntgen and radium rays on plants. His earlier results were confirmed by these experiments, in which he used 0.75 gm. of about 4 per cent. radium-barium chloride in a thin aluminium and glass capsule. *Brassica* was found very resistant, and no difference was noticed in the effect of rays from the large quantity of radium preparation passing through thin aluminium, and of those from the smaller amount in the thicker glass capsule. The growth of roots of *Vicia Faba* was inhibited, and, in some instances, was resumed after the radium was removed, in other cases not. The shoot, once inhibited, never resumed growth, but luxuriant adventive sprouts developed in the axils of the cotyledons. Tissues were made brown by the exposures, and marked individual variations were found in the behavior of the seeds of *Vicia*. The growth of *Pisum* seedlings was retarded in proportion to the length of exposure of the seed before germination.

These experiments indicate that roots are more sensitive than shoots, and, in the light of Willcock's and of Hertel's work, this was explained by the presence of chlorophyll in the shoots. All attempts to affect the geotropic sensitiveness of roots and shoots were unsuccessful, except in those cases where growth had been completely inhibited by the rays. Starch was not found in roots or shoots of seedlings grown from the seeds exposed for two days before planting. Exposure to X rays gave similar results. Exposed seedlings also responded phototropically to light so long as they were growing, but, says Koernicke, " at the conclusion of growth they were in a condition which I may designate as 'radium-rigor and Röntgen-rigor.' " The light waves coming from the preparation induced phototropic curvatures in sporangiophores of *Phycomyces nitens* after an exposure of about 15 hours, at a distance of more than 3 cm. This response failed when the light rays were cut off by wrapping the radium tube in black paper, and Körnicke feels sure that the result

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* So the authors. The emanation, however, does not give off β rays, but only α rays. See p. 9.

was not caused by the moisture that gathered on the tube, as was shown by Errera and Steyer with reference to the experiments of Elfving. Positive phototropic response to the phosphorescent light of the radium is also claimed for *Vicia Faba* seedlings if the activity of the preparation is sufficiently strong.

In November of the same year (1905) appeared Koernicke's[41] paper on the effect of Röntgen and radium rays on plant tissues and cells. In plants developing from seeds exposed to either radium or X rays, the epidermis of the roots was wrinkled over the entire surface. Direct exposure of growing roots produced the same result. The undulatory curvatures of the vascular bundles, present in normally contractile roots, was not observed. Multiplication of nuclei was also observed in cells of the periblem and plerome of exposed roots.

Cell-studies were made of roots of *Vicia Faba* and of *Pisum sativum*, after exposure of 1, 2, and 3 days to the radium, and also of roots that developed from exposed seeds. No results followed exposure of the roots for one day, but after an irradiation of two days cell-divisions appeared normal, but less numerous, while treatment for three days sufficed to practically inhibit mitosis. Resting nuclei appeared unaffected. Not until after the roots had ceased growing did peculiar forms appear which could be attributed to the influence of the rays on the chromatin. Here spindles occurred in which the daughter chromosomes were separated from each other with difficulty, so that their progress toward the poles was delayed. Formation of the cell-wall occurred normally in exposed tissues, and for this and other reasons it is held that the polynucleate cells resulted from amitosis.

Flower buds of *Lilium martagon* of various ages, from the youngest to the oldest, were exposed to the rays for varying lengths of time, and fixed at different periods following the exposure. Mitosis was retarded and inhibited, and the reproductive cells were more sensitive than the vegetative cells to the influence of the rays. Numerous irregularities in mitosis were observed. For example, anthers fixed 20 hrs. after irradiation for 5 hrs., showed the nuclear thread of the pollen-mother-cell separated into smaller and more numerous double segments than is normal in the species. The small segments were later drawn together on a normally formed spindle, and, in one instance, on a multipolar spindle. In the subsequent longitudinal splitting and distribution to the daughter-nuclei these segments moved irregularly, some lagging behind on the equator, or appearing distributed along the spindle between the equator and the poles. Several cases were observed where two and three daughter-nuclei were formed on each side of the cell-plate. In some cases an increased number of tetrads resulted from the division of the pollen-mother-cell. Following the exposure to the rays, there was observed a tendency to a stronger formation of kinoplasm than formerly.

The first recorded experiments of the effect of radium rays on plant respiration are those of Micheels and de Heen,[46] who found that the respiratory energy of germinating pea seeds was diminished by exposure to the rays from 0.5 mg. of a radium preparation of 240 activity. The authors describe this result as being in good agreement with other physiological phenomena due to radium.

Molisch[48] was not successful in any attempts to produce a tropistic curvature by the direct influence of the rays of radium, but found that the phosphorescent light produced by mixing radium bromide of 3,000 activity with zinc blend in a sealed glass tube, caused positive phototropism of stems in *Vicia sativa*, and *Ervum Lens*, but called forth no curvatures in *Helianthus annuus*. In the case of *Phycomyces nitens*, sporangiophores were positively, and mycelium negatively phototropic. The phototropic sensibility of the seedlings above mentioned was greatly intensified by exposure to the rays. As Molish indicates in the title of his paper, these curvatures may be attributed only indirectly to the radium; they are responses only to the phosphorescent light caused in the zinc-blend by the rays.

The ability of *Bacillus pyocyaneus* to secrete its characteristic pigments was found by Bouchard and Balthazard[13] to be diminished by exposure to radium emanation, and the power of reproduction and division was diminished and finally destroyed. Rays from "the radioactive residues from the treatment of pitchblend from Joachimsthal" affected various species of *Aspergillus* in a similar manner (Dauwitz[17]). Sensitiveness to the rays varies according to the species, *A. niger* and *A. fumigalus* being most sensitive, and *A. ochraceus*, *A. clavatus*, and *A. varians* less so, in the order named. Spores formed while the plant was under the influence of the radioactive bodies germinated poorly, and produced a meager mycelium that bore no spores. Analogous effects followed the treatment of different species of *Penicillium*. Experiments with *B. prodigiosus* and *B. pyocyaneus* confirmed the results of Bouchard and Balthazard.

Dorn, Baumann, and Valentiner[21] caused the emanation from 30 mg. of radium bromide (activity not given) to be bubbled through sterilized bouillon for five minutes, and then added 1 oz. of a typhus-bouillon culture. Then daily, for 10–12 days, two or three times a day for ten minutes at a time, the emanation was bubbled through the mixture. On the tenth and thirteenth days the cultures were platted out, and in three days thereafter the exposed culture was only a tenth as much developed as the control. Beta and gamma rays from 5 mg. of "pure" radium bromide inhibited the growth of germs of typhoid, cholera, and diphtheria. The authors hold that the emanation itself, behaving as a heavy gas, has no physiological effect, and their results are, therefore, to be attributed to the radioactivity of the emanation.

Guilleminot's[33] study of the comparative effects of X rays and those of radium on the plant-cell were first published in November, 1907. This author had previously * indicated a process for determining the effectual strength of X rays, and defined a unit M, obtained by a comparison of the fluorescence of barium platynocyanide and that of a standard of radium. Using seeds, of Mahon's gilliflower, he obtained the following results: 1. The true characteristic action of the rays is a retardation of growth when the strengths are rather great. 2. The strength that slightly retards appears to be 3,000 M, radium, and 15,000 M, X rays. 3. The fatal strength is in the neighborhood of 10,000 M, radium, while 20,000 M, X rays permits of a feeble development. 4. The accelerating action, if such exists, is apparently reached at about 250–500 M, radium, and 5,000–7,500 M, X rays. The differences (in result), he says, are too feeble to warrant the unqualified assertion of an acceleration, and, in his longer paper, he[32] states that an "exciting" dosage probably does not exist.

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* Guilleminot[30, 31] adopts as a unit of intensity (M) of the field of irradiation the quadruple of the intensity producing the same luminescence as a standard of 0.02 gm. of radium bromide of 500,000 activity, spread over a circular surface of 1 cm. in diameter, and placed at a distance of 2 cm. from the phosphorescent surface. Then the unit of quantity of irradiation will be the quantity acting for one minute when the field has unit of intensity.

3. Effects of Radium Rays on Plant Fibers

In addition to the physiological results reviewed above, the action of radium rays on vegetable fibers is both interesting and pertinent in this connection. This effect was first noticed by Giesel,[28] and afterward accidentally by Lord Blythswood,[65] in 1904. "I happened," says the latter, "to replace the usual mica plates, used to keep in the small quantity of radium in its ebonite box, with a piece of cambric, so as to permit the whole of the emanations to pass out, mica stopping the α rays. In four days the cambric was rotted away. I have replaced it now several times with the same result."

In the following year Martin and Morton[67] experimented on the effect of the rays on unspun silk fibers and ordinary unbleached cotton thread. The threads were exposed to "bare radium," at a distance of about one half centimeter. After a certain period of exposure the average breaking strength of the threads was taken and plotted against time. The points obtained lay closely on a smoothly descending curve. In the case of the silk the loss of strength went on at a practically uniform rate from the beginning up to the end of the longest exposure given (seven days). The initial strength of 78 gm. decreased by about 4 gm. per day. The cotton threads gave a curve which fell more rapidly in the early than in the later stages. The strength began at 370 gm., and decreased at first by about 60 gm. per day. After ten days the rate of weakening was about half this. At the end of 17 days the strength was reduced to 17 gm. The difference in behavior of the two kinds of fibers is attributed to the greater thickness of the cotton threads. The effect is due entirely to the α rays, for only negative results were obtained when those rays were screened out.

Wet threads, with the same length of exposure, were less weakened, and this, the authors state, was plainly due to the decreased emission of rays on account of the solution of the radium salt, and the consequent removal of the emanation.*

In a subsequent paper, McKee and Morton[68] state that the constant removal of the emanation does not affect the result. It was further found that when threads or a piece of filter paper, after exposure to the rays are dyed with methylene blue, the exposed part takes a deeper color than the rest, thus indicating the presence of

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* "The rate of escape of emanation is much increased by solution of the compound." Rutherford, p. 255 (see Bibliography, p. 20, No. 115).

oxycellulose. The weakening effect became inappreciable at a distance of 18 mm. Under the microscope the broken ends of the fibers of exposed threads were straight at their ends, those not exposed curled, indicating a loss of elasticity through exposure to the rays.

In July, 1907, Hussakof[37] published a review of the more important papers bearing on the physiological effects of the rays of radium. Previous announcements of my own results have appeared, with one exception, as abstracts of papers presented before various scientific societies; citations to them will be found in the Bibliography appended to this chapter, and further reference to them here is unnecessary.

The details of the investigations reviewed above are numerous, but the results may be briefly summarized. There is very general agreement on the following points:

1. Radium rays have the power to modify the life-processes of both plants and animals.

2. Röntgen rays and radium rays produce similar physiological results.

3. Sensitiveness to these rays varies with the species of either plant or animal.

4. Younger, and especially embryonic tissues, are more sensitive than those more mature.

5. With only one or two exceptions, exposure to radium rays has been found to either retard or completely inhibit all cell-activities. The rays may cause irregularities in mitosis.

6. Experimental evidence for or against the existence of a radiotropic response is conflicting.

7. Whatever the immediate, internal change produced in the protoplast may be, the result, with animals as well as with plants, appears to be more or less profoundly modified by the presence of chlorophyll in the cell.

8. Radium rays appear to retard the activity of enzymes.

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CHAPTER V

BIO-RADIOACTIVITY, EOBES, RADIOBES

1. The Supposed Radioactivity of Plants and of Wood

Soon after the discoveries of "contact" electricity and "animal" electricity by Volta and Galvani, plant physiologists began to look for electric currents in plants, and to find therein the explanation of "vital" activity. In a similar manner the announcement of the discovery of radioactivity has been followed by numerous supposed observations of a natural or acquired radioactivity of plants and plant tissues.

Professor A. B. Green[2] was among the first to report that micro-organisms, especially species of *Staphylococcus*, after an exposure of from 24–120 hrs. to radium rays at a distance of 0.5 mm., themselves exhibit phenomena of radioactivity. He considers it uncertain as to whether living organisms can acquire this property, but states that those killed by the action of radium rays can do so. In his experiments the radium salt was enclosed in a vulcanite and brass capsule, and the radioactivity acquired by the organisms, lasted for three minutes after the termination of the exposure, and enabled them to photograph themselves on a sensitive plate. Their spores were found to be best for this purpose. I have already discussed these results on page 62.

Lambert[3] stated in 1904 that ferments that digest albuminous matter emit Blondlot rays, and that the emission of these rays is the cause of the action of the soluble ferments.

The experimental demonstration of the emission of the so-called N rays by plants of the garden cress was reported by Meyer. Their emission, he said, varies with the activity of the protoplasm, and is diminished when the plants are exposed to the vapor of chloroform, and is modified by mere compression of the tissues.

In 1904 Russel[5] described before the Royal Society the rather startling discovery of the action of wood on a photographic plate in the dark. This property, he said, belongs probably to all woods. Conifers are especially active, and the spring wood most of all, but the dark autumn wood produced no such effect. Oak, beech, acacia

(*Robinia*), Spanish chestnut, and sycamore possess this property, but ash, elm, the horse-chestnut, and the plane tree only to a slight degree. Most resins manifest it, but not so the true gums, such as gum senegal and gum tragacanth. Exposure to sunlight, especially to the blue rays of the spectrum, increases the activity. Cork, printer's ink, leather, pure India rubber, fur, feathers, and turpentine are reported to have their activity increased in the same way. Since bodies such as slate, porcelain, flour and sugar, in which there is no resinous or allied body, do not react in this way, nor affect the plate at all, the activity of the various kinds of wood is attributed to the resinous substances in them. Tommasina's[8] paper was also published in 1904. He reported that all freshly gathered plants, fruits, flowers, and leaves possess a radioactivity which is stronger in the young and in individuals in action than in those at rest, being apparently proportional to the vital energy. For this phenomenon he proposed the term *bio-radioactivity*. Buds of lilac, and leaves of *Thuja* and of laurel were found by him to be bio-radioactive.

In the following year Tarchanoff and Moldenhauer[7] published their preliminary note on the induced and natural radioactivity of plants, and on its probable rôle in their growth. When seeds of various grains, and of the pea were exposed to the radium emanation, the seedlings growing from such seeds showed induced radioactivity in their roots, but the stem and small leaves remained inactive. Also when a mature plant was exposed to the emanation the roots became strongly radioactive, the stem somewhat less so, the leaves only slightly, and the flowers not at all.*

This distribution of the radioactivity in the plant body is constant, and the authors consider that there is in the plant a special substance, sensible to the emanation, and capable of becoming radioactive under its influence. This substance occurs in the roots, but gradually diminishes up the stem. It is found also in seeds. According to this same paper plants possess a natural radioactivity, which is distributed throughout the plant similarly to the induced radioactivity. This natural radioctivity is strong enough to affect a photographic plate, and plays an important rôle in the development of the plant.

In a second paper Russel[6] gives a list of 33 native and 22 foreign woods that are active, and says that the activity of resins and gums is increased by exposure, not only to sunlight, but to the arc-light as

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* Results not confirmed by Acqua, Rend. Accad. Lincei V. 6: 357. 1907.

well. Photographic plates often contain a negative of the plate-holder. That this is not a case of radioactivity appears to be proved, says the author, for a glass or a mica screen of one thousandth of an inch in thickness entirely protects the plate from being acted on.

Finally Paul Becquerel[1] undertook a careful study of "plant radioactivity." He tested pea seeds, moss (*Hypnum*), and branches of boxwood for radioactivity, but found not a trace of it manifest when the electroscope was carefully guarded from water-vapor. This explains the condition found necessary by Tommasina, that the parts of plants must be freshly picked in order to manifest bio-radioactivity. According to Becquerel, the discharge of the electroscope in Tommasina's experiments was due to the water in the plants.

From all the investigations noted above, the general conclusion seems to be warranted that radioactivity is not a property of protoplasm nor of living tissues. A clear understanding of the nature of radioactivity would lead, *a priori*, to the same inference.

2. The Professed Artificial Creation of Life

Radioactivity and vital activity are in two respects very roughly, but only very superficially analogous. Both radioactive bodies and living organisms are undergoing a destructive process; atomic disintegration in the one, molecular transformation in the other; both, with exceptions, maintain themselves constantly at a higher temperature than their surroundings. These analogies have in two or three instances proven dangerously attractive.

A consideration of radioactivity led Dubois,[18] in 1904, to the view that the distinction between "matter of life" and "living matter" is superficial. He proposed the term *bioproteon*, meaning the particular state of the "proteon" in living beings, and suggested the desirability of determining the radioactivity proper of the bioproteon. In a subsequent paper[21] he says: "The unique principle of everything, of both force and matter, I have called 'proteon,' and when it pertains to a living being, 'bioproteon.'" Proteon and bioproteon are only two different states of the same thing. When the bioproteon is dead it has only ceased to be radioactive and becomes simply proteon. He claimed also to have discovered the emission, from the lamellibranch mollusc, *Phalade dactyle*, of rays that could penetrate paper and opaque substances, and darken a sensitive plate.

Early in the year 1905 appeared his paper[19] on "*La création de l'être vivant et les lois naturelles*" in which he announced the formation of living organisms in bouillon gelatine by placing on it crystals of the bromide of both barium and radium. Later in the same year[20] he claimed to have secured a kind of spontaneous generation by radium. By the contact of certain crystalloids with organic colloids, there are obtained, he says, granulations, or vacuolides, possessing the optical and morphological characters of simple life, more rudimentary than bioproteon, or living matter. These bodies arise, grow, divide, grow old, and die, returning to the crystalline state like all living things, and Dubois applied to them the generic term *eobe* (dawn of life). Eobes are held to form the transition between the organic and the inorganic world. In his essay[21] on "*La radioactivité et la vie,*" he elaborates the hypothesis that the energy irradiated by living beings has two distinct origins — one from the environment, and one ancestral or hereditary. By their "ancestral energy" living beings are similar to radioactive bodies. They both give off heat rays, light, chemical rays, electricity, and possess molecular motion, and atomic and other movements.

Leduc's[26, 27] profession to have created life was controverted by Bonnier,[10] Charrin and Gaupil,[17] and by Kunstler,[25] in 1907.

The most extravagant claims made in this direction are those of Burke [11-16], whose observations on the spontaneous action of radioactive bodies on gelatine media form the basis of a voluminous work entitled "The Origin of Life." While these experiments have little of the scientific importance they have been held to possess in the popular mind, it is desirable to state, in Burke's own words, what he did, and his own interpretation of the results.

"An extract of meat of 1 lb. of beef to 1 liter of water, together with 1 per cent. of Witter peptone, 1 per cent. of sodium chloride, and 10 per cent. of gold labelled gelatine was slowly heated in the usual way, sterilized, and then cooled. The gelatine culture medium thus prepared, and commonly known as bouillon, is acted upon by radium salts and some other slightly radioactive bodies in a most remarkable manner."[12]

When the mixture above described was placed in a test-tube and sterilized, and the surface sprinkled with 2.5 gr. of radium bromide (activity not given), after 24 hours (three to four days when radium chloride was used), "a peculiar culture-like growth appeared on the surface, and gradually made its way downwards, until after a fortnight, in some cases, it had grown nearly a centimeter beneath the surface." From this growth Burke was not able to make subcultures. He considers them not bacteria, and not contaminations, but "highly organized bodies." They have "nuclei," subdivide when a certain size is reached, and "the larger ones appear to have sprung from the smaller ones, and they have all probably arisen in some way from the invisible particles of radium." He regards them as colloidal, rather than crystalline, "of the nature of 'dynamical aggregates' rather than of 'static aggregates,'" and coins for them a new name, *radiobes.* This forms the experimental basis for a volume of 351 pages.

With reference to these discoveries, Dubois[22] claims priority over Burke, and rejects his term radiobe in favor of eobe, because these bodies may be obtained with non-radioactive substances.

A few months after Burke's announcement Rudge[28, 29] showed that the alleged growths were "nothing more than finely divided precipitates of insoluble barium salts." He was unable, in a preparation similar to the one described by Burke, to observe anything like cell-division, and believes that an occasional grouping of the particles in pairs must be purely fortuitous. The appearance of growth of the radiobes is explained as due to a diffusion of the precipitate through the gelatine from a point of concentration where the radium salt was in contact with the gelatine. Salts of barium, lead, and strontium produced effects exactly similar to those caused by radium preparations.

Again repeating Burke's experiments, Rudge[30] was unable to secure the radiobes when agar-agar was substituted for gelatine and distilled water was used. If tap-water was employed a slight growth resulted, while the addition of a soluble sulfate resulted in a very dense growth. An examination of 30–40 samples of gelatine showed that they all contained enough H_2SO_4 to give a distinct, sometimes a dense precipitate with barium chloride in the presence of HNO_3. This precipitate was found, on analysis, to be $BaSO_4$. Gelatine was then prepared free from sulfates and gave no growth. Negative results were obtained with salts of uranium, thorium, pitchblende, and metallic uranium, thus clearly indicating that there is not the slightest connection between the formation of the radiobes and radioactivity.

A sample of gelatine from which H_2SO_4 had been removed was sealed with a radium salt from June until September. At the end of that time no growth appeared, but when a soluble sulfate was added to a portion of this gelatine the growth began at once.

"The cellular form of these precipitates," said Rudge, "is probably due to the circumstance that the gelatine is liquefied by the action of the salt, and each particle of precipitate is formed about a core of gelatine, so that the layer of barium sulfate forms a kind of sac or cell which is surrounded by the solutions of the salt in the liquefied gelatine. This 'cell' may be permeable to the liquefied gelatine containing a salt in solution, which, passing through the cell-wall, causes an expansion to take place, the limit of growth being controlled by some surface tension effect." No trace of a nucleus or of mitosis was observed under the very highest magnification, and "cells" under a cover-glass sealed down with cement were observed to suffer no alteration during four months.

Reference to the extreme claims noted in the literature above cited may be fittingly concluded by the following quotation from Lord Kelvin:[24]

"But let not youthful minds be dazzled by the imaginings of the daily newspapers that because Berthelot and others have . . . made foodstuffs they can make living things, or that there is any prospect of a process being found in any laboratory for making a living thing, whether the minutest germ of bacteriology or anything smaller or greater."

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CHAPTER VI

RADIUM PREPARATIONS AND METHODS OF EXPOSURE

The radium preparations used in physical and physiological experiments are salts of that element. The one most commonly employed is radium bromide, though the more expensive chloride is sometimes used. The salt may be obtained in at least three different kinds of containers. (1) Thin glass tubes, usually about 30 mm. long, and, in the newer preparations, protected by being placed in metallic holders with openings extending nearly the length of the tube. (2) Aluminium tubes, devised by Mr. Hugo Lieber. (3) Hard rubber cells with brass caps having a mica window. By all of the above containers the α rays are practically cut off, as they cannot penetrate the walls.

In addition to these preparations, a radium coating has been invented by Lieber,[2-5] by the use of which, not only the α rays, but also the radium emanation becomes available for experimentation. The process of preparing this coating is thus described by Lieber:

"Radium coatings are made in the following manner: Radium is dissolved in a suitable solvent and into this solution a suitable material is dipped. This material is then withdrawn, with radium solution adhering to it. The solvent quickly evaporates, leaving the material covered with an exceedingly thin film of radium. The kind of solvent to be used is determined by the nature of the material to be coated. Such solvents are employed as have a tendency to soften the material which is to be coated. Thus, if celluloid rods, discs, or similar instruments are to receive a radium coating, in order to be used for therapeutic purposes, solvents such as alcohol, amyl acetate, etc., may be employed. These solvents have a tendency to soften the celluloid temporarily. When the solvent evaporates, the radium has been uniformly distributed over the celluloid, and has also been incorporated in its surface. In order to prevent accidental removal of the radium in such coatings, the celluloid instruments produced in this way are dipped for a short time in a collodion solution. By this process the whole radium coating is covered by a very thin film of collodion. In the course of a few days this film of collodion becomes so tough that it will strongly resist destruction, thus affording ample protection for the underlying radium. . . ."

"In the preparation of these coatings both the radium and the collodion solutions are colored with an analine dye. This is done to show the part that has been coated. Besides, if the radium happens to be removed by accident or otherwise, as by scraping, etc., the disappearance of the color makes such removal evident.

"The great difference between radium used in containers, composed even of exceedingly thin aluminium, and radium used in the form of the coatings here described, is shown by their relative influences on the electroscope; a delicate rod coated at its tip with radium bromide of 10,000 activity and holding, therefore, very little radium, compared very favorably in its effects with 1 grm. of radium bromide of 10,000 activity in a glass tube, or with 10 mgrms. of radium bromide of 1,000,000 activity in a very thin aluminium tube."

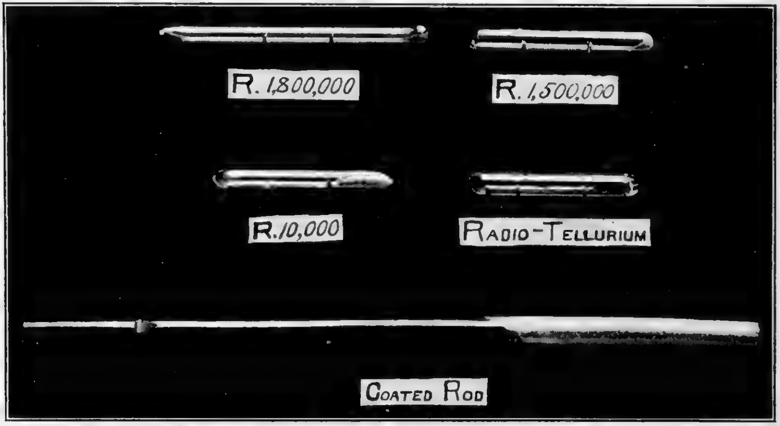

FIG. 2. Sealed Glass Tubes of Radium Bromide and a Rod Coated with Lieber's Radium Coating.

That these coatings permit of the escape of the gaseous emanation is shown by the fact that the presence of the emanation may be demonstrated in a current of air passed over a coating. That the α rays penetrate through the thin collodion film is proved by the fact that the scintillations produced on a zinc sulphide screen by alpha rays may be caused by the use of a coated rod or disc. The activity of the radium is not affected by this treatment, for Rutherford[6] has shown that, "a distribution of the radiating matter over a thousand times its original volume has no appreciable influence on its radioactivity."

An illustration of some of the radium preparations used in the following experiments is given in FIGURE 2. See also Experiment 40, p. 147, and Experiment 42, p. 149.

For the purpose of observing under the microscope the effect of radium rays on individual living protoplasts, the writer[1] has devised a radioactive microscopic slide, the preparation of which has been perfected by Mr. Lieber. This slide has been described as follows:

"A solution of any desired concentration of radium bromide of known activity is made in a suitable solvent, and applied to the surface of the slide near the center. When the solvent evaporates a film of the salt remains on the slide. The film is protected by a coating of a specially prepared substance. Living cells may now be mounted as on an ordinary slide, and their response, if any, to the stimulus of the rays observed. The coating has the advantage, not only of being sufficiently transparent to light, but easily transparent to the β and γ rays, and in less degree to the α rays also."[1]

The methods of applying to living plants the various preparations here described are explained in detail in connection with the experiments. It may be mentioned here, however, that in no case has the radium salt been permitted to come into contact with the plant, nor with any portion of it. The effects produced are due solely to the action of the rays coming from either the radium bromide direct, or from the radioactive emanation.

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CHAPTER VII

EFFECTS ON GROWTH OF EXPOSING SEEDS TO RADIUM RAYS

1. Effects on Growth of Exposure of Unsoaked Seeds

The object of the following experiments was to ascertain the effect on germination and growth of exposing unsoaked seeds to the rays of radium.

Experiment 1

January 28, 5:15 P. M.

Six seeds of *Lupinus albus*, unsoaked, were placed under the radium tube (10 mg., 1,500,000 ×), with the tube resting on the hilum edges of the seeds. Six control seeds were similarly placed under an empty glass tube.

February 2, 4 P. M.

After five days radiation the seeds were planted in soil in separate pots, and transferred to the propagating-house.

February 9, 9 A. M.

Radium	*Control*
The cotyledons of five seeds are just emerging; the sixth is just arching the soil.	The cotyledons of three seeds are raised about 5 mm. above the surface of the soil, the cotyledons of two seeds have just emerged, and the cotyledon of one seed is just breaking the soil.

February 11, 3:30 P. M.

Radium	*Control*
Only four seedlings have the cotyledons clear of the soil, one of which has the cotyledons spread showing the plumule.	Five seedlings have the cotyledons entirely clear of the soil, and well spread apart, showing the plumules. The cotyledons of the other seedling are just emerged. This culture is more vigorous, and further advanced in every way than the radiated one.

The lengths of the hypocotyls above the surface of the soil are as follows:

Radium	*Control*
10.00 mm.	10.00 mm.
7.00	21.00
8.50	12.00
9.00	18.00
6.00	24.00
5.00	00.00
45.50 mm. Total.	85.00 mm. Total.
Average height, 7.58 + mm.	Average height, 14.16 + mm.

February 16, 12 M.

Length of Hypocotyls

Radium	*Control*
12.50 mm.	26.00 mm.
4.50	29.00
7.50	24.50
12.50	23.50
12.50	20.50
7.00	20.50
56.50 mm. Total.	144.00 mm. Total.
Average height, 9.41 mm.	Average height, 24.00 mm.
The cotyledons are not reflexed, and the epicotyl has begun to elongate in only one seedling.	The cotyledons are well reflexed, and the epicotyls are elongating with leaves well expanded.

February 22.

The plants were carefully removed from the pots and the soil washed from the roots. The roots of the radiated plants are very short, and secondary roots are only slightly, and in two specimens not at all, developed.

The roots of the control plants are several times as long as those of the radiated specimens, and with secondary roots well developed.

The radiated plants did not lose the nyctitropic movements of the leaves, and measurements of the length of the stomata in the two showed no difference between the normal and the radiated plants.

Experiment 2

February 2, 4 P. M.

Four seeds of *Lupinus albus*, after having lain for four days in a cylinder lined with Lieber's " radium coating," were planted in soil in the propagating-house. Control culture same as in Experiment 1.

February 9, 9 A. M.

The cotyledons of all four seeds are emerging, but they are not yet raised above the surface of the soil.

February 11, 3:30 P. M.

The cotyledons are all clear of the ground, all spreading and showing the plumule. In height the plants are intermediate between the radiated and the control in Experiment 1. Length of hypocotyls above the surface of the soil as follows:

Radium	*Control*
7.00 mm.	10.00 mm.
13.00	21.00
15.00	12.00
15.00	18.00
50.00 mm. Total.	24.00
	00.00
	85.00 mm. Total.
Average height, 12.50 mm.	Average height, 14.16 + mm.

February 16, 12 M.

LENGTH OF HYPOCOTYLS ABOVE THE SURFACE OF THE SOIL

Radium	*Control*
24.50 mm.	26.00 mm.
19.00	29.00
23.50	24.50
14.00	23.50
81.00 mm. Total.	20.50
	20.50
	144.00 mm. Total.
Average length, 20.25 mm.	Average length, 24.00 mm.

EXPERIMENT 3

April 15, 6:20 P. M.

Twenty seeds of "Lincoln" oats, with the glumes removed, were placed in two parallel rows with the radicle ends touching and the embryo side upermost. Over them was laid the tube of $RaBr_2$ (1,500,000 ×), resting on the radicle ends of the seeds.

Control with empty tube, and both sets placed in the dark room.

April 22, 11:30 A. M.

After an exposure of 6 days and 15 hours, both sets of seeds were planted in separate jars and placed in the propagating-house to germinate.

April 25.

(Record taken by the gardener.)

All the seeds in the control culture have broken the soil, save one that decayed.

None of the radiated seeds have yet come up.

April 27.

Two of the radiated seeds have come up.

April 28.

Seventeen more of the radiated seeds are just breaking through, the surface of the soil. The average height of the control seedlings from the surface of the soil to the tip of the first leaf, is 60.40 mm.

On April 29 the cultures were photographed (FIGURE 3), and up to May 5 there had been no appreciable growth of the radiated seeds since they were photographed, and the experiment was closed.

FIG. 3. Experiment 3. Germination of Oats Retarded by Exposure, while Dry, to Radium Rays.

EXPERIMENT 4

May 6, 9:35 A. M.

Eight dry seeds of *Phaseolus* (Henderson's "Long Yellow Six Weeks") were placed with their hilum-edges touching the radium-coated rod (25,000 ×) four seeds on each side of the rod.

May 12, 5 P. M.

After an exposure of 145 hours the rod was removed from the seeds, and six hours after its removal the seeds were planted in soil (pot R), with the control seeds (pot C) in an adjacent pot in the propagating house.

May 19, 5 P. M.

The gardener reported that the seedlings in R broke through the soil before those in C, but he did not have the exact data.

The radiated culture has four seedlings with the arch of the hypocotyl not yet straighted, while the control culture has only one such seedling. The heights of the hypocotyls above the surface of the soil are as follows:

Radium		*Control*	
25.00 mm.	To top of arch.	114.00 mm.	
103.00		99.50	
56.00	“	112.50	
34.00	“	91.50	
62.00	“	112.00	
115.50		30.00	To top of arch.
80.00		89.00	
71.00		114.00	
546.50 mm.		762.50 mm.	
Average length, 68.31 + mm.		Average length, 95.31 + mm.	

FIG. 4. Experiment 5. Acceleration of Growth of Wheat Following Exposure to *a* Rays from Polonium.

Experiment 5

Object: To ascertain the effect of α rays only (from polonium), on the germination and growth of wheat.

Sixteen grains of wheat (*Triticum vulgare*, Henderson's "Wellman Fife") were exposed to α rays by being placed in contact with a metallic rod coated with polonium.

June 7, 9:30 A. M.

After 10 days' exposure the exposed grains and 16 unexposed grains were planted in soil.

The heights of the seedlings above the surface of the soil were measured as follows:

	June 11, 10 A. M.		June 18, 10 A. M.	
	Polonium	*Control*	*Polonium*	*Control*
1	5.50 mm.	29.50 mm.	125.00 mm.	76.50 mm.
2	31.00	27.50	116.00	86.00
3	25.00	31.00	91.50	107.00
4	9.00	20.50	132.00	83.50
5	10.00	4.00	60.00	66.50
6	19.00	18.50	143.00	59.00
7	16.50	10.00	118.00	50.00
8	24.00	7.00	134.00	27.00
9	4.00	30.00	121.00	91.00
10	23.00	30.00	124.50	100.00
11	19.50	33.50	145.00	88.00
12	28.00	24.00	118.50	83.00
13	32.50	24.50	134.00	68.00
14	24.00	21.50	144.00	71.00
15	19.00	30.00	123.00	96.50
16	31.50	15.00	144.00	56.00
	321.50 mm.	356.50 mm.	1,973.50 mm.	1,209.00 mm.
Av.	20.00 mm.	22.20 mm.	123.34 mm.	75.56 mm.

While at first there was no significant difference in the heights of the exposed and the control seedlings, the exposed plants later grew much more rapidly than the control. See FIGURES 4 and 5.

Summary

From the above experiments it is seen that both germination and growth of *Lupinus albus*, *Phaseolus*, and *Avena sativa* are retarded

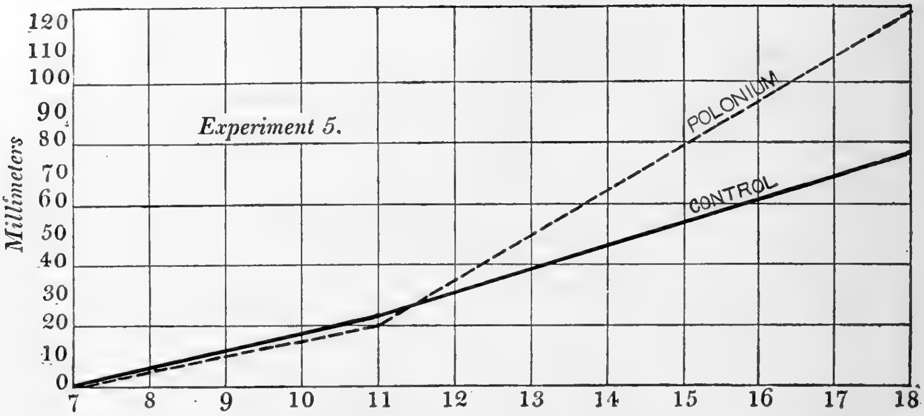

FIG. 5. Acceleration of Growth of Wheat Following Exposure to α Rays from Polonium.

by exposure to the rays. A five days' exposure of *Lupinus* to radium of 1,500,000 × retarded germination and growth, but did not appreciably affect irritability, as shown by the fact that the plants still performed their nyctitropic movements. Exposure in the radium-lined cylinder (of much weaker activity, but permitting the α rays to act) retarded germination and growth less than exposure to a stronger preparation. The effect of the radium-coated rod of 25,000 × was also less than that of the preparation of 1,500,000 ×. Oats exposed to this last preparation for a little over six and one half days were greatly retarded in germination, and had their subsequent growth completely inhibited. In the rod- and cylinder-exposures the α rays were in large part available, as well as the β and γ rays, but the added effect of the α rays was not sufficient to compensate for the weaker activity of the preparations.

An exposure of dry wheat grains (*Triticum vulgare*) for ten days to α rays by placing them in contact with a metallic rod coated with polonium was followed at first by a slightly less rapid growth, and then, eleven days after planting, by a more vigorous and rapid growth than that of seeds similarly grown but not exposed.

2. EFFECTS ON GROWTH OF EXPOSING SEEDS WHILE SOAKING

The object of the following experiments is to ascertain the effect on germination and growth of exposing seeds during imbibition of water to the rays of radium.

EXPERIMENT 6

February 2, 6 P. M.

Numerous seeds of timothy grass were scattered evenly over the bottom of an earthenware germinator, and the glass tube of $RaBr_2$ (10 mg., 1,500,000 ×) was supported over the center of the dish with the end containing the radium resting on the bottom among the seeds.

A control was similarly arranged with an empty tube, and both cultures were placed in the dark room.

February 9, 10 A. M.

Radium

No seeds directly under the tube have germinated. Germination has begun about 2 mm. from the tube, and plumules and radicles are shortest near the tube, and increase in length toward the edge of the dish. There is a slight (in some cases, marked) tendency for the plumules to bend in toward the center. This suggests a positive *radiotropism*, but is probably a hydrotropic response to the more humid conditions at the center of the germinator.

Control

The seeds have germinated evenly over the bottom of the dish, at the center as well as elsewhere. No tendency to bend toward the center or in any other direction was observed.

February 11, 9 A. M.

Radium

The seeds at the center are beginning to germinate, but slowly. The following measurements of the height of the shoots from the circumference to the center of the germinating dish is typical: 10 mm., 9 mm., 6 mm., 2 mm., 1 mm.

All the seedlings are decidedly pale and those within a radius of 7.50 mm. are entirely etiolated.

All the hypocotyls are excessively elongated beyond the radius of 7.50 mm. from the center, making the average height of the seedlings beyond this radius much taller than those not radiated.

Control

All of the seedlings are of about the same height and average much taller than the radiated set.

The seedlings are uniformly of a dark green color, in striking contrast to those of the radiated set.

The heights of the shoots average much less than those of the radiated plants outside of the 7.50 mm. radius.

February 12, 11 A. M.

The relative condition of the radiated and control sets remains as yesterday. Measurements of the heights of the seedlings gave the following averages at the distances indicated:

Distance	*Height*	
	Radium	*Control*
0 mm.	2.00 mm.	10.00 mm.
5	5.00	10.00
10	7.50	13.00
15	11.00	14.00
20	17.00	9.00
25	23.00	10.00

February 16, 3:30 P. M.

All the seedlings in both cultures have apparently grown, but the relative condition and size remains as on the 12th.

EXPERIMENT 7

February 10, 9:30 A. M.

Experiment arranged as in No. 6, using seeds of alfalfa.

February 15, 5:30 P. M.

Seeds have germinated in both cultures. In the center of the radiated set germination is greatly retarded, but in the control dish germination is generally uniform.

Both cultures were placed near a window in the laboratory.

February 18, 11:30 A. M.

The seedlings within a radius of 5 mm. of the radium tube have not developed further than the protrusion of the radicle. All the seedlings in the control culture have grown, and are taller than those exposed.

Rhizopus nigricans attacked both cultures, but while growing vigorous and rank in the control culture, it grew weakly and poorly in the radium culture.

February 22, 10 A. M.

The leaves of the radiated seedlings are much lighter green than those of the control.

EXPERIMENT 8

February 26, 12:30 P. M.

Unsoaked seeds of timothy grass were arranged in 8 rows, radiating from the center of a circular piece of blotter, which was placed on the bottom of the germinating dish. Over the center was placed the radium-tube (10 mg., 1,500,000 ×), with all the radium in the lower end, and the tube in contact with the underlying seeds.

Control similarly arranged with empty tube.

March 1, 5:30 P. M.

No seeds have germinated in either culture.

March 9, 9 A. M.

The control seedlings are all of about the same height and color, being normally green. Average height (estimated), 30 mm.

The radiated plants are entirely etiolated under the radium-tube, and for a radius of 4 mm. on all sides. The height of the seedlings gradually increases from 3 mm. at the center to about 30 mm. at the circumference.

FIG. 6. Experiment 8. Retardation of Germination and Growth of Timothy Grass by Radium Rays. Activity, 1,500,000.

March 10, 12 M.

The exposed culture was photographed (FIGURE 6).

March 11, 3:30 P. M.

Microscopic examination of the control plants shows in the cells of the leaves normal chloroplastids, with healthy, green color.

In the non-green radiated seedlings, the plastids in many cells were apparently of normal size and shape, but destitute of green, while in other cells the plastids, although green, had largely lost their shape and individuality, and were aggregated in a disorganized mass against the vertical cell-walls, and often massed in one end of the cell.

A similar result, though less marked, was obtained by exposure made through eight layers of sheet tin, a total thickness of 9 mm.

EXPERIMENT 9

March 19, 6 P. M.

Seeds of timothy grass were arranged along 8 radii of a circular piece of blotter in a Zurich germinator and a tube of $RaBr_2$ (7,000 ×) was placed vertically over the center with the end containing the radium resting on the seeds.

Control with empty tube, and both cultures placed to germinate in the dark-room.

March 23, 10:30 A. M.

The seeds have germinated quite uniformly in the control culture, averaging about 3–6 mm. high.

In the radium culture the germination is similar to that in the control, except under the radium-tube, where germination, though not entirely inhibited, has been greatly retarded, the seedlings not averaging more than 1–2 mm. high for a radius of about 5 mm. from the center of the tube in all directions.

The glass tubes were removed and both cultures were placed in the laboratory in front of a window. Cylinders of blotter were placed around the culture dishes to prevent phototropic curvature.

March 26, 11 A. M.

The plants in both cultures are of about the same height, except in the radium culture, where those in the center, directly under the radium, are noticeably shorter than the others, and the height increases rather abruptly from the center toward the circumference.

The tubes were removed and the cultures left to develop further.

March 30, 3 P. M.

The retarded seedlings in the center of the radiated culture have reached about the average height of the others, and the difference in the average height of the seedlings in the two cultures is very slight. The balance, if any, is in favor of the radiated set being slightly taller.

March 31, 5 P. M.

The radiated seedlings average decidedly taller than the control set. Thus, following exposure to the radium, there has been at first a retardation of germination and growth, then, after the removal of the radium, an apparent recovery and subsequent acceleration.

EXPERIMENT 10

March 23, 4:30 P. M.

The experiment was set up as in No. 9, only a sealed glass tube containing radio-tellurium was used instead of the radium tube. Timothy seed was employed.

March 30, 3:45 P. M.

The seedlings in both cultures have an average height of about 25 mm., except those immediately under the tube of radio-tellurium, which are only about one half as tall as the others, but the influence has not extended to any appreciable distance from the tube in any direction. The tube was in contact with the underlying seeds.

EXPERIMENT 11

May 12, 6 P. M.

Eight seeds of the bean (*Phaseolus*) were placed, four in a row, on moist sphagnum, with the hilum-edges adjacent. Over them, resting on the adjacent edges, was placed the coated rod (10,000 ×). Eight dry seeds were similarly exposed.

May 14, 12:30 P. M.

After 42 hrs. exposure, the seeds, together with eight unexposed seeds, were planted in soil.

May 26, 9 A. M.

The seeds in all three cultures germinated poorly, but of those exposed while imbibing water, only one has come up, of those exposed dry, five have come up with an average length of hypocotyl above the surface of the soil of 39.60 + mm. Four, only, of the control seeds came up, but the average length of their hypocotyls above the soil surface is 59.25 mm. The hypocotyls of those exposed dry, and of the control seeds were preserved for histological study. (See p. 226.)

EXPERIMENT 12

May 14, 4 P. M.

On damp cotton in a moist chamber were placed 8 seeds of the bean (*Phaseolus*), in two rows of 4 seeds each, with the hilum edges of opposite seeds adjacent. The radium-coated rod (25,000 ×) was placed in contact with the adjacent edges.

Control of 8 seeds in the same chamber, but with no radium.

May 19, 9 A. M.

All the seeds have germinated in both cultures. The lengths of the radicles are as follows:

Radium	*Control*
6.00 mm.	18.50 mm.
24.00	25.00
9.00	30.00
13.00	22.00
9.00	21.00
8.00	30.00
17.00	14.00 *
12.00	25.00
98.00 mm.	185.50 mm.
Average of 8, 12.25 mm.	Average of 8, 23.18 + mm.

The amount of growth following exposure to the radium rays during imbibition was only about one half the normal.

EXPERIMENT 13

May 14, 4 P. M.

Eight seeds of *Lupinus albus* were placed on damp cotton in a moist chamber in two rows of four seeds each, with the hilum-edges of opposite seeds adjacent. Over the seeds was placed a radium-coated rod (10,000 ×), in contact with the adjacent edges.

Control of eight seeds in the same chamber with no rod.

May 19, 9 A. M.

All the seeds have germinated in both cultures, but the radicles of the radiated seeds are shorter than those of the control seeds. The exposed radicals are also noticeably more yellowish than are those of the control.

The lengths of the radicles are as follows:

Radium	*Control*
41.00 mm.	41.00 mm.
19.00	44.00
12.00	22.00
16.00	41.00
17.00	41.50
22.00	37.00
30.00	26.00
16.00	35.00
173.00 mm.	287.50 mm.
Average, 21.62 + mm.	Average, 35.93 + mm.

---

* Caught in seed-coat.

Experiment 14

May 25, 12 M.

Eight dry seeds of the bean (*Phaseolus*. Henderson's "Long Yellow, Six Weeks") were placed in moist sphagnum with their hilum-edges touching a rod coated with $RaBr_2$ (10,000 ×). Control, similarly placed, but with no radium. Rod continued throughout the experiment.

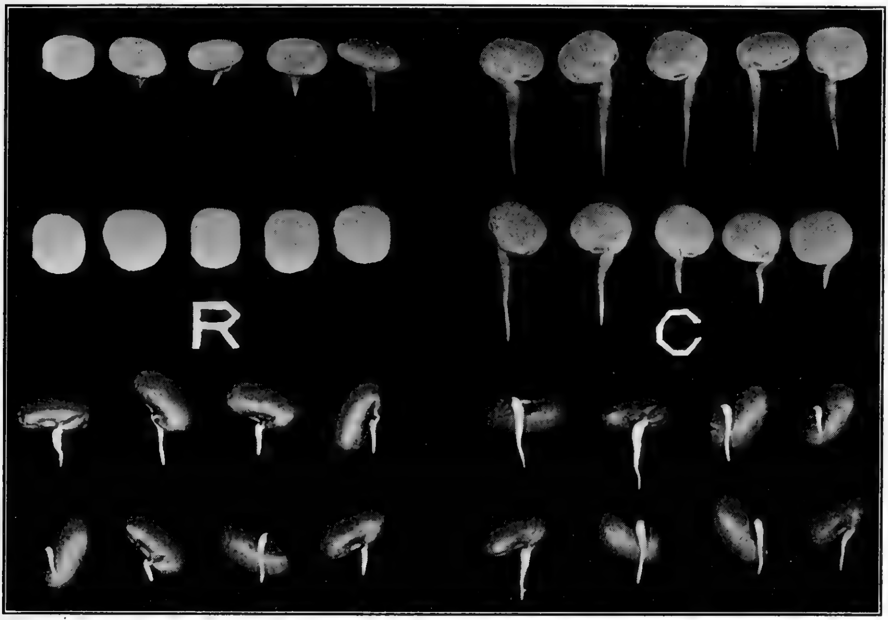

Fig. 7. Experiments 14 (Bean) and 17 (Lupine). Germination Retarded by Rays from Coated Rods of 10,000 × (Bean), and 25,000 × (Lupine).

May 27, 3 P. M.

The radicles have protruded in one radiated seed, and in seven of the control seeds.

May 28, 10 A. M.

The seeds were photographed with those of Experiment 17 (figure 7).

The lengths of the radicles are as follows:

Radium	*Control*
11.50 mm.	20.00 mm.
9.50	20.00
13.50	12.00
15.00	20.00
16.00	18.00
11.50	23.00
16.50	20.00
12.00	18.00
105.50 mm.	151.00 mm.
Average, 13.18 + mm.	Average, 18.87 + mm.

EXPERIMENT 15

May 28, 11 A. M.

Repetition of Experiment 14.

May 30, 11 A. M.

Lengths of the radicles as follows:

Radium	*Control*
5.00 mm.	8.00 mm.
0.00	17.00
0.00	6.00
11.00	13.00
3.00	12.00
7.00	11.00
5.00	15.00
8.00	10.50
39.00 mm.	92.50 mm
Average, 4.87 + mm.	Average, 11.56 + mm.

June 1, 9:30 A. M.

Radium	*Control*
38.50 mm.	43.00 mm.
39.00	54.00
38.00	49.00
49.00	42.00
34.00	56.00
47.50	47.00
39.50	59.00
51.00	45.00
336.50 mm.	395.00 mm.
Average, 42.06 + mm.	Average, 49.37 + mm.

June 3, 2:30 P. M.

Portions of the hypocotyls were placed in formalin for sectioning. See page 226.

Experiment 16

May 28, 11 A. M.

Repetition of Experiment 14, using *Lupinus albus*. Experiment started as there indicated. Rod, 25,000 ×.

May 30, 10 A. M.

The lengths of the radicles are as follows:

Radium	*Control*
0.00 mm.	15.00 mm.
0.00	12.00
0.00	12.00
0.00	11.00
0.00	0.00
0.00	0.00
0.00	7.50
0.00	9.00
7.00*	1.00
13.00*	0.00
20.00 mm.	67.50 mm.
Average of 10, 2.00 mm.	Average of 10, 6.75 mm.

June 1, 2:30 P. M.

Radium	*Control*
44.00 mm.	62.00 mm.
36.00	75.00
20.50	61.50
55.00	66.00
53.00	27.00
52.00	54.00
28.00	59.00
57.00	44.00
63.00	54.00
73.00	26.50
481.50 mm.	529.00 mm.
Average, 48.15 mm.	Average, 52.90 mm.

* Very poorly exposed.

June 3, 12 M.

Radium	*Control*
83.00 mm.	100.00 mm.
63.00	132.00
58.00	99.00
94.00	105.00
97.00	78.50
91.00	96.00
70.00	110.00
112.00	87.00
108.00	111.00
135.00	60.00
911.00 mm.	978.50 mm.
Average, 91.10 mm.	Average, 97.85 mm.

EXPERIMENT 17

May 25, 12 M.

Ten dry seeds of *Lupinus albus* were placed in moist sphagnum, with the hilum-edges in contact with a rod coated with $RaBr_2$ (25,000 ×). Control of ten seeds, similarly placed, but with no radium.

May 27, 3 P. M.

The radicles have protruded from none of the radiated seeds.

The radicles have protruded from 9 of the control seeds.

Two of the radiated seeds were poor, and have not absorbed water.

May 28, 10:30 A. M.

The lengths of the radicles are as follows:

Radium	*Control*
13.00 mm.	15.00 mm.
0.00	18.00
0.00	23.50
6.50	10.50
6.50	26.00
4.50	22.00
0.00	27.50
0.00 Injured.	26.50
30.50 mm.	26.50
Average, 3.81 + mm.	22.50
	218.00 mm.
	Average, 21.80 mm.

September 19.

The plants were photographed together with those of Experiment 14, and portions of the hypocotyls and roots were preserved for sectioning (FIGURE 7). The radium rod was continued throughout the experiment.

EXPERIMENT 18

Object: To ascertain the effect of the rays from a radium-coated rod on the germination and growth of *Lupinus albus*.

June 16, 3 P. M.

Eight seeds of *Lupinus albus* were placed in two parallel rows in moist sphagnum, with a rod coated with Lieber's "radium-coating" (about 10,000 ×, and one year old) placed in contact with the hilum-edges of the seeds.

Eight control seeds similarly placed, but with no radium.

June 20, 3 P. M.

The lengths of the hypocotyls from the cotyledons measured as follows:

	Radium	*Control*
1	50.00 mm.	54.00 mm.
2	61.50	46.00
3	63.00	69.50
4	70.00	50.00
5	83.00	44.00
6	59.50	63.00
7	33.00	55.00
8	81.50	49.00
	501.50 mm.	430.50 mm.
	Average, 62.68 mm.	Average, 53.81 + mm.

Acceleration of growth has followed exposure to the rays.

EXPERIMENT 19

Five seeds of Henderson's "Long Yellow Six Weeks" bean (*Phaseolus*) were exposed for five days (120 hours), during imbibition from the dry condition, to a radium-coated rod (10,000 ×).

At the end of the 120 hours the hypocotyls of the exposed seeds averaged 50.80 mm. in length, those of the control seeds 56.00 mm. Portions of the root and hypocotyl of the exposed seeds and of control specimens were preserved for sectioning (p. 226).

Summary

Seeds were exposed to radium preparations in sealed glass tubes, and of activities of 7,000, 10,000, and 1,500,000. Under the conditions imposed, exposure to rays from all of these preparations was followed by a retardation of growth, the amount of retardation varying directly with the activity of the preparation.

When seeds of timothy grass (*Poa pratense*) and alfalfa were exposed to β and γ rays during imbibition and subsequent stages of germination, both germination and growth were retarded and the plants were etiolated. Microscopic examination of the etiolated plants showed that in some cases the plastids had apparently failed to form chlorophyll, while in others the green chloroplastids seemed to have been disorganized. Bean seeds placed in contact with the coated rod (a, β and γ rays) during germination were also retarded. When timothy grass seeds were exposed to radium of weak activity (7,000 ×) an initial retardation was followed by apparent recovery after an interval of five days. At the end of this period the exposed seeds averaged even taller than those of the control culture, though other conditions of growth had been practically identical.

A similar recovery of growth vigor followed exposure of bean seeds to the coated rod of 10,000 ×, and of lupine seeds to the coated rod of 25,000 ×. These results suggest the establishment of a condition of *radium tonus*, in which the organism becomes adjusted to a certain intensity of radioactivity in its environment, and the rays of preparations of this strength cease to act as a stimulus.

Exposure of seeds to a preparation of radio-tellurium in a sealed glass tube was followed by results similar in kind to those following exposure to radium rays. The germination of fungus spores (Exp. 7) was also apparently less vigorous in radium cultures than normally.

In one case (Exp. 18) the germination of seeds of *Lupinus albus* and the subsequent growth of the radicle was apparently accelerated 16 per cent. by placing a radium-coated rod of about 10,000 × in contact with the seeds as they germinated in moist sphagnum.

3. Effect on Growth of Exposing Soaked Seeds

The aim of the following experiments was to ascertain the effect on germination and growth of exposing soaked seeds to the rays of radium.

EXPERIMENT 20

Six seeds of *Lupinus albus* were soaked in water for 17 hours, and the seed-coats were removed from over the hypocotyls. The seeds were then placed in a Zurich germinator in two rows, with the hypocotyls facing each other, and the glass tube of $RaBr_2$ (5 mg., 1,500,000 ×) was placed over the seeds in contact with the hypocotyls.

Control of six seeds similarly treated except that an empty glass tube was used instead of the radium-tube.

November 26, 8:30 A. M.

All seeds have begun to germinate, and an ink mark was placed on each radicle as a reference mark in measuring.

November 27, 4:30 P. M.

Increase in length of the radicles in the past 32 hours as follows:

Radium	*Control*
1.00 mm.	3.00 mm.
5.00	8.00
5.00	8.00
2.00	10.00
6.00	5.50
6.00	11.00
25.00 mm.	45.50 mm.
Average, 4.16 mm.	Average, 7.58 mm.

November 28, 3:30 P. M.

Radium	*Control*
7.00 mm.	15.00 mm.
10.50	broken
10.00	12.50
6.00	17.00
12.00	13.00
9.00	20.00
54.50 mm.	77.50 mm.
Average, 9.08 mm.	Average, 15.50 mm.

The radicles of the control seeds have elongated almost twice as rapidly as those exposed to the radium.

EXPERIMENT 21

November 28, 4:30 P. M.

Twenty-four seeds of buckwheat (*Fagopyrum esculentum*), soaked in water for 24 hours, were divided into two groups, *R* and *C*, of 12 seeds each, and placed to germinate in the Zurich germinators; *R* with the radium-tube (1,500,000 ×) lying over and in contact with the seeds, *C* with a short glass rod lying over the seeds.

December 1, 8:30 A. M.

In *R* three radicles have protruded, in *C* four. The seeds were placed to develop further in two pots of earth of equal size.

December 18, 12 M.

In the radiated culture four arched hypocotyls are above the surface of the soil; in the control culture, six.

January 2, 9:30 A. M.

Radium	*Control*
Only 5 seeds have germinated. Lengths of hypocotyls above surface of soil as follows:	Seven seeds have germinated. Lengths of hypocotyls above surface of soil as follows:
35.00 mm.	78.00 mm.
23.00	90.00
14.00	90.00
9.00	69.00
5.00	65.00
86.00 mm.	392.00 mm.
Average, 17.20 mm.	Average, 78.40 mm.

The petioles of the cotyledons in *C* were longer than those in *R*, and the seedlings were in every way more vigorous and healthy.

EXPERIMENT 22

January 5, 9 A. M.

Six flax seeds (*Linum*), soaked in water over night, were placed in Zurich germinators, and the glass tube of $RaBr_2$ (1,500,000 ×) was laid over them, with the radium evenly distributed along the bottom of the tube.

Control, similarly arranged with an empty glass tube, and both cultures placed in the dark room.

January 6, 2 P. M.

None of the seeds have germinated.

January 7, 9 A. M.

All of the seeds under the radium have germinated; all but one of the control seeds.

An exposure of 48 hrs. to the $RaBr_2$ of 1,500,000 activity has not been sufficient appreciably to affect germination.

EXPERIMENT 23

January 11, 9 A. M.

Eight seeds of white mustard (*Brassica alba*), soaked in water for 15 hrs., were placed in Zurich germinators in the dark-room with the glass tube of $RaBr_2$ (5 mg., 1,500,000 ×) resting on the seeds, the radium evenly distributed along the bottom of the tube.

Control of 8 seeds similarly treated and placed under an empty glass tube.

January 12, 9 A. M.

One radiated seed has begun to germinate; two control seeds. The seeds were planted in the same pot of soil, the exposed seeds on one side, the control seeds on the other side of the pot.

January 14, 9 A. M.

Of the radiated seeds, two hypocotyls have pierced the soil; of the control seeds, five.

January 16, 4:30 P. M.

Radium	*Control*
Lengths of hypocotyls of six seedlings above soil surface as follows:	Lengths of hypocotyls of six seedlings above soil surface as follows:
33.50 mm.	30.50 mm.
23.00	37.00
22.00	14.00
18.00	39.00
20.00	14.00
30.00	38.00
146.50 mm.	172.50 mm.
Average, 24.41 mm.	Average, 28.75 mm.

January 17, 3:45 P. M.

One seedling was injured in each culture and discarded.

Radium	*Control*
46.00 mm.	49.50 mm.
36.00	45.50
35.50	30.00
41.50	53.00
38.00	25.00
27.00	49.00
224.00 mm.	252.00 mm.
Average of 6, 37.33 mm.	Average of 6, 42.00 mm.

January 19, 9 A. M.

Radium	*Control*
67.00 mm.	69.50 mm.
60.50	76.50
36.00	30.50
70.00	64.50
57.00	73.50
54.50	45.00
345.00 mm.	71.50
	431.00 mm.
Average of 6, 57.50 mm.	Average of 7, 61.57 + mm.

January 22, 11 : 30 A. M.

Radium	*Control*
80.50 mm.	80.50 mm.
91.50	61.50
91.50	79.00
49.50	100.00
83.00	65.00
87.50	89.00
483.50 mm.	79.00
	554.00 mm.
Average of 6, 80.58 + mm.	Average of 7, 79.14 + mm.

Germination and growth were at first retarded, following exposure to the radium, but after ten days (January 21–22) the exposed and the control seedlings are of practically the same length.

Experiment 24

February 26, 1 P. M.

Six seeds of *Lupinus albus* were soaked in water for 18 hrs. and after the removal of the seed coats, were placed to germinate in a Zurich germinator in two rows, with the hilum-edges in contact, and a sealed glass tube of $RaBr_2$ (10,000 ×), with the radium salt evenly distributed over the bottom of the tube was placed lengthwise over the hypocotyls, and in contact with them.

Five control seeds were similarly placed with an empty tube.

March 1, 5 : 30 P. M.

The seeds have all germinated in both cultures, with hypocotyls from 15–35 mm. long. The relative lengths of the radiated and control seed hypocotyls were not recorded.

Both sets of seeds were placed to develop further in moist sphagnum.

March 17, 9:30 A. M.

The tap-roots of the radiated plants have failed to develop normally, being not more than one half to one sixth as long as those not exposed to the radium rays. Correlated with the failure of the tap-root to develop, the secondary roots are more fully developed in the radiated seedlings, but there was no difference in growth of secondary roots on the side of the hypocotyl that was next to the radium-tube during exposure and those on the opposite side.

EXPERIMENT 25

March 22, 4 P. M.

Ten soaked seeds of " Lincoln " oats (*Avena*), with the glumes removed, were placed in a Zurich germinator, in parallel rows of 5 each, in contact with the glass tube of $RaBr_2$ (10,000 ×) with the radium evenly distributed over the bottom of the tube. The radicle ends of the oats were all touching the glass tube.

Control of 5 seeds with empty tube, similarly placed, and both sets placed in the dark room to germinate.

March 23, 11 A. M.

All the grains have germinated in both cultures, but those under the radium are only about one half as far developed as the control grains.

March 26, 11 A. M.

All the grains have grown since the last observation, but the control grains have grown more than those radiated. Two secondary roots have developed on each control grain, no secondary roots on the radiated grains. The grains of both sets were planted in soil and placed in the propagating house to develop. The total length of exposure to the radium has been 67 hrs. 35 min.

March 31, 9:30 A. M.

The experiment was photographed. The control plants are several times taller than those from exposed seeds (FIGURE 8).

FIG. 8. Experiment 25. Retardation of Germination and Growth of Oats Exposed to Radium Rays During Early Stages of Germination. Cf. FIGURES 9 and 24.

April 1, 3 P. M.

After removing the plants and carefully washing the soil from the roots, the following observations were made:

Roots	*Radium*	*Control*
Number.	3.5 (av. for 10 plants).	5.7 (av. for 7 plants).
Length.	Average (estimated) about ½ as long as the control.	Estimated twice the length of the radiated.
Secondary roots.	None, except a few *very* short ones on the most vigorous plant.	Numerous on all plants save one, which had only 2–3 small ones.
Diameter.]	Decidedly club-shaped in five of the specimens.	Normal appearance: not enlarged near the root-tip.
Root-hairs.	Appear to be slightly more numerous, and about twice as long as in the control plants. (See *R*, FIGURE 9.)	Normal.
Root-tips.	Much darker colored than in the control plants.	Of normal appearance. Decidedly lighter than those of the radiated plants.

SUMMARY

The exposure to radium rays of seeds previously soaked was followed, in all instances by a retardation of growth. When seeds of *Lupinus albus* were exposed for 17 hrs. to rays from radium bromide of 1,500,000 × the seedlings produced by them grew only one half as rapidly as normally. Exposure of soaked seeds of buckwheat for 24 hrs. to rays of the same strength retarded growth, and impaired the general vigor of the plants. One experiment was tried with flax seed, but an exposure of 48 hrs. (1,500,000 ×) was followed by no appreciable effect on the germination of the seeds.

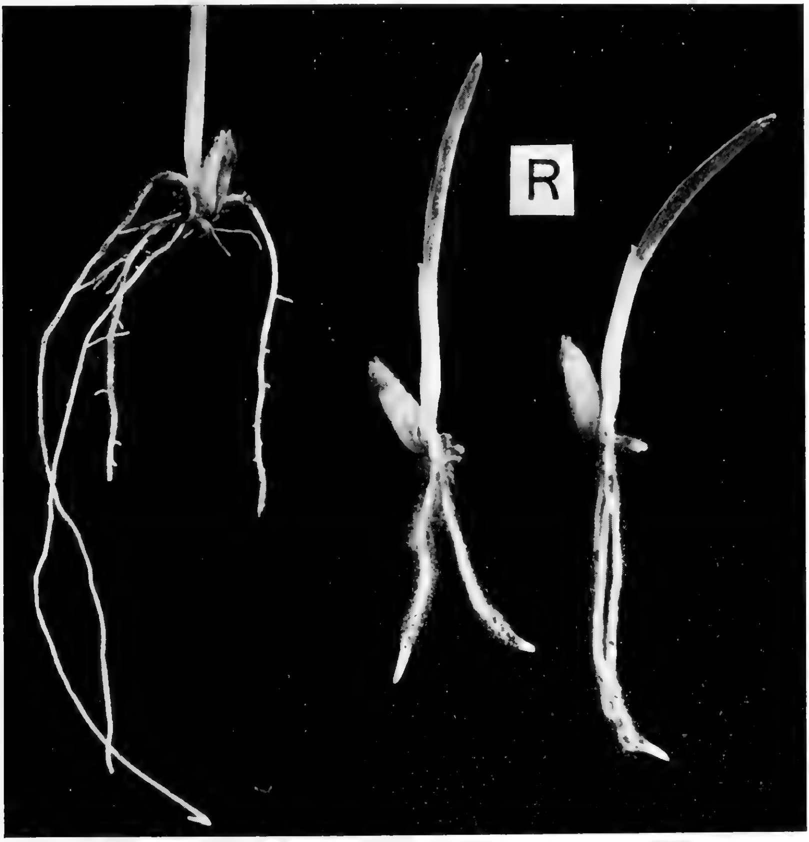

Fig. 9. Experiment 25. Roots and Root-Hairs of Seedlings from the Exposed Culture (*R*), and from the Unexposed. Slightly Enlarged. Cf. figure 8.

An initial retardation of the growth of white mustard after the soaked seeds were exposed for 15 hrs. (1,500,000 ×), was followed by a recovery and apparent acceleration of growth, for the plants from exposed seeds, within ten days after planting, had caught up with and even slightly surpassed the control plants in height. The effect of this exposure of the white mustard was slight, and in harmony with results obtained by Koernicke,* who found *Brassica napus* especially resistant. He reported that swollen seeds were accelerated in growth by exposure to 10 mg. of radium bromide, but does not state the activity of the preparation he used.

When seeds of *Lupinus albus* were first soaked in water, and then exposed while germinating for 18 hrs. (10,000 ×), and planted in soil, the tap-roots grew very abnormally. After 16 days' growth in the soil the tap-roots had evidently ceased growing, and, correlated with this fact, the secondary roots had developed more vigorously than normally.

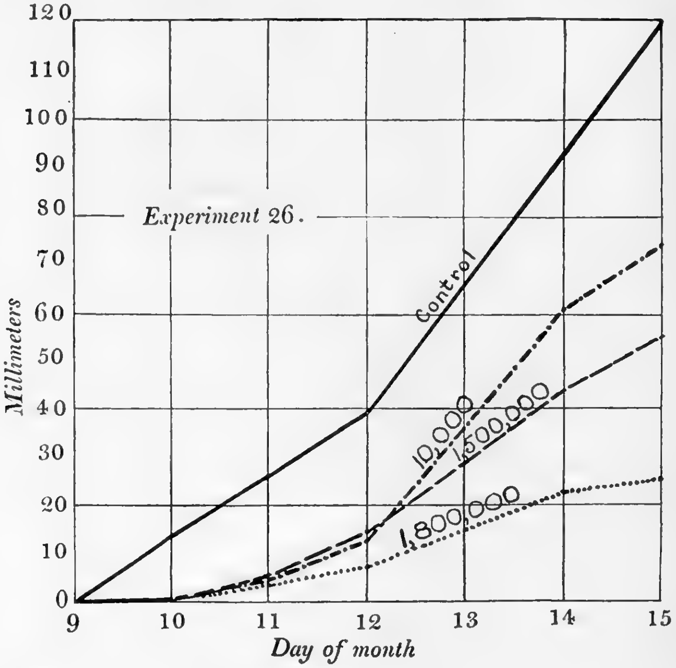

Fig. 10. Effect on the Growth of *Zea Mays* of Exposing the Grains, before Planting, for 16 Hours, to Rays from Radium of Different Degrees of Activity.

Oat grains were exposed for 67 hrs. after being soaked in water, and then planted in soil. The subsequent growth of the shoots was

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* Bibliography, p. 72, No. 39.

greatly retarded (FIGURE 9), and roots were fewer in number, shorter, thick, and club-shaped, with the slightly more numerous root-hairs nearly twice the normal length. The root-tips of the exposed plantlets were of a decidedly darker color than those of the control.

4. EFFECT OF DURATION OF EXPOSURE AND DEGREE OF ACTIVITY

In the preceding experiments it has become more and more clearly evident that the effect of exposure to radium rays varies, as Koernicke also found, with the duration of the exposure and the degree of activity of the radium. The purpose of the following six experiments was to confirm this fact, and get more accurate data on the relation between the duration, the degree of activity, and the effect produced.

EXPERIMENT 26

The object of the following experiment is to ascertain the effect on the germination and growth of corn (*Zea Mays*) of exposure for

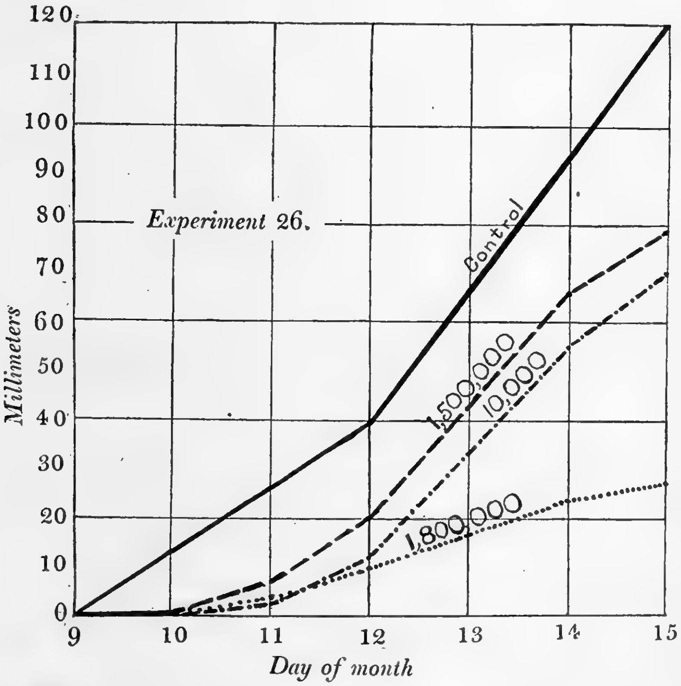

FIG. 11. Effect on the Growth of *Zea Mays* of Exposing the Grains, before Planting, for 24 Hours, to Rays from Radium of Different Degrees of Activity.

the same period of time to rays from radium of different strengths of activity.

May 4, 10 A. M.

Four sets of four seeds each of *Zea Mays* were exposed for 16 hours to radioactive preparations as follows:

Set A to RaBr$_2$, 1,800,000 ×, in sealed glass tube.
Set B to RaBr$_2$, 1,500,000 ×, in sealed glass tube.
Set C to RaBr$_2$, 10,000 ×, in sealed glass tube.
Set D to radio-tellurium in sealed glass tube.
Set E, Control — not exposed.

Similar exposures were made for 24 hours and for 33 hours, and the seeds were then planted in soil.

Exposure for 16 Hours

May 10, 10 A. M.

The heights of the seedlings above the surface of the soil are as follows:

	A	B	C	D	E
	1,800,000 ×	1,500,000 ×	10,000 ×	Ra. Tel.	Control
1	0.00 mm.	0.00 mm.	just up	0.00 mm.	16.50 mm.
2	0.00	just showing	0.00 mm.	0.00	18.00
3	0.00	arching soil	0.00	0.00	11.00
4	0.00	just showing	0.00	just up	12.00
	0.00 mm.	0.00 mm.	0.00 mm.	0.00 mm.	57.50 mm.
					14.38 mm.

May 11, 10 A. M.

	A	B	C	D	E
	1,800,000 ×	1,500,000 ×	10,000 ×	Ra. Tel.	Control
1	4.00 mm.	7.50 mm.	9.00 mm.	0.00 mm.	29.50 mm.
2	0.00	3.00	3.50	4.00	28.00
3	3.00	6.00	3.00	4.00	23.00
4	0.00	6.00	3.00	9.00	22.50
	7.00 mm.	22.50 mm.	18.50 mm.	17.00 mm.	103.00 mm.
	3.50 mm.	5.62 mm.	4.62 mm.	5.66 mm.	25.75 mm.

May 12, 10 A. M.

	A	B	C	D	E
	1,800,000 ×	1,500,000 ×	10,000 ×	Ra. Tel.	Control
1	7.00 mm.	18.00 mm.	18.00 mm.	0.00 mm.	44.00 mm.
2	0.00	7.00	13.00	11.50	37.00
3	9.00	16.00	10.00	15.00	37.00
4	5.00	13.00	13.50	20.00	36.00
	21.00 mm.	54.00 mm.	54.50 mm.	46.50 mm.	154.00 mm.
	7.00 mm.	13.50 mm.	13.62 mm.	15.50 mm.	38.50 mm.

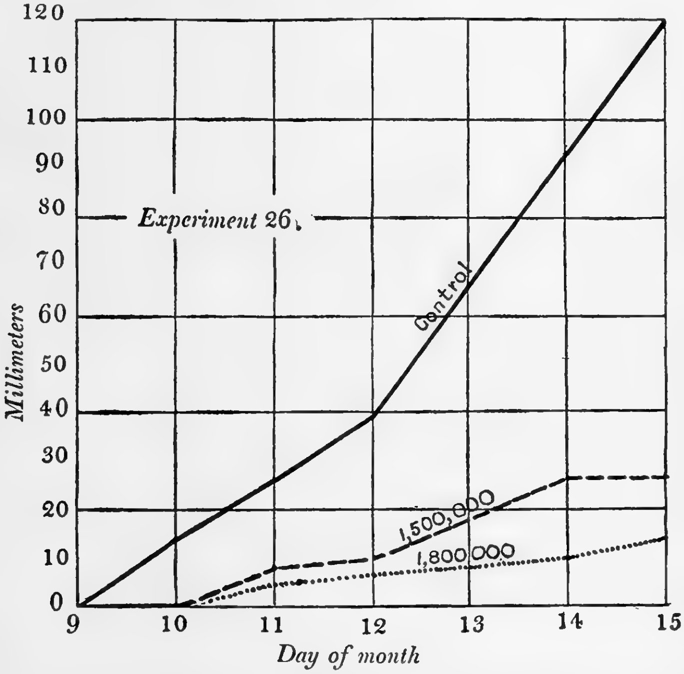

FIG. 12. Effect on the Growth of *Zea Mays* of Exposing the Grains, before Planting, for 33 Hours, to Rays from Radium of Different Degrees of Activity.

May 14, 10 A. M.

	A	B	C	D	E
	1,800,000 ×	1,500,000 ×	10,000 ×	Ra. Tel.	Control
1	27.00 mm.	57.00 mm.	68.00 mm.	0.00 mm.	108.00 mm.
2	10.00	29.00	62.00	50.00	80.00
3	30.00	52.00	62.00	65.00	95.00
4	20.00	38.00	51.00	68.00	90.00
	87.00 mm.	176.00 mm.	243.00 mm.	183.00 mm.	373.00 mm.
	21.75 mm.	44.00 mm.	60.75 mm.	61.00 mm.	93.25 mm.

May 15, 10 A. M.

	A	B	C	D	E
	1,800,000 ×	1,500,000 ×	10,000 ×	Ra. Tel.	Control
1	34.00 mm.	69.00 mm.	86.00 mm.	0.00 mm.	135.00 mm.
2	10.00	36.00	73.00	57.00	111.00
3	35.00	71.00	75.00	86.00	121.00
4	20.00	45.00	62.00	93.00	115.00
	99.00 mm.	221.00 mm.	296.00 mm.	236.00 mm.	482.00 mm.
	24.75 mm.	55.25 mm.	74.00 mm.	78.67 mm.	120.50 mm.

None of these plants exposed to the radium of 1,800,000 activity are erect, but grow parallel with and close to the surface of the soil.

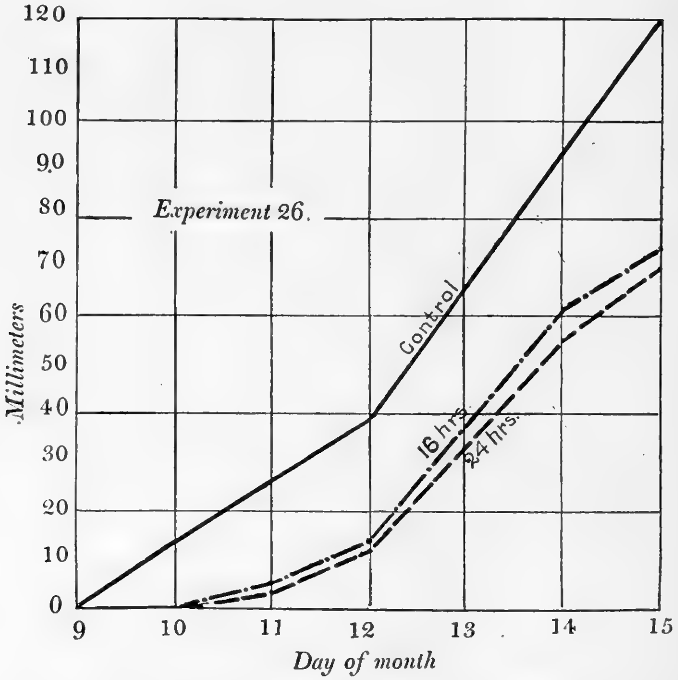

FIG. 13. Effect on the Growth of *Zea Mays* of Various Durations of Exposure of the Grains, before Planting, to Radium Rays. Activity, 10,000.

EXPOSURE FOR 24 HOURS

The heights of the seedlings above the surface of the soil are as follows:

May 10, 10 A. M.

	A	B	C	D	E
	1,800,000 ×	1,500,000 ×	10,000 ×	Ra. Tel.	Control
1	just showing	just showing	not up	not up	16.50 mm.
2	0.00 mm.	“	“	“	18.00
3	0.00	“	“	“	11.00
4	0.00	“	“	“	12.00
	0.00 mm.				57.50 mm.
	0.00 mm.				14.38 mm.

May 11, 11 A. M.

	A	B	C	D	E
	1,800,000 ×	1,500,000 ×	10,000 ×	Ra. Tel.	Control
1	4.00 mm.	7.00 mm.	1.00 mm.	3.00 mm.	29.50 mm.
2	2.00	7.50	4.50	0.00	28.00
3	6.00	7.00	0.00	0.00	23.00
4	0.00	11.50	3.00	5.00	22.50
	12.00 mm.	33.00 mm.	8.50 mm.	8.00 mm.	103.00 mm.
	4.00 mm.	8.25 mm.	2.83 mm.	4.00 mm.	25.75 mm.

May 12, 10 A. M.

	A	B	C	D	E
	1,800,000 ×	1,500,000 ×	10,000 ×	Ra. Tel.	Control
1	10.00 mm.	14.00 mm.	9.00 mm.	13.00 mm.	44.00 mm.
2	6.50	21.00	14.00	0.00	37.00
3	14.00	17.00	0.00	0.00	37.00
4	0.00	26.50	12.00	15.00	36.00
	30.50 mm.	78.50 mm.	35.00 mm.	28.00 mm.	154.00 mm.
	10.16 mm.	19.62 mm.	11.66 mm.	14.00 mm.	38.50 mm.

May 14, 10 A. M.

	A	B	C	D	E
	1,800,000 ×	1,500,000 ×	10,000 ×	Ra. Tel.	Control
1	31.00 mm.	52.00 mm.	42.00 mm.	53.00 mm.	108.00 mm.
2	19.00	74.00	61.00	0.00	80.00
3	20.00	67.00	0.00	29.00	95.00
4	21.00	70.00	61.00	70.00	90.00
	91.00 mm.	263.00 mm.	164.00 mm.	152.00 mm.	373.00 mm.
	22.75 mm.	65.75 mm.	54.67 mm.	50.67 mm.	93.25 mm.

May 15, 10 A. M.

	A	B	C	D	E
	1,800,000 ×	1,500,000 ×	10,000 ×	Ra. Tel.	Control
1	45.00 mm.	62.00 mm.	53.00 mm.	71.00 mm.	135.00 mm.
2	20.00	83.00	79.00	0.00	111.00
3	19.00	85.00	0.00	41.00	121.00
4	22.00	83.00	77.00	88.00	115.00
	106.00 mm.	313.00 mm.	209.00 mm.	200.00 mm.	482.00 mm.
	26.50 mm.	78.25 mm.	69.66 mm.	66.66 mm.	120.50 mm.

All the plants exposed to the radium of 1,500,000 × have failed to become erect, and grow horizontally over the surface of the ground (FIGURE 14).

FIG. 14. Experiment 26. Absence of Geotropic Response in Shoots of *Zea Mays*, following a 24 Hours' Exposure of the Grains, before Planting, to Radium of 1,500,000 ×. From left to right, $RaBr_2$ 1,800,000 × ; $RaBr_2$ 1,500,000 × ; Ra. Tel.; Control. Cf. FIGURE 62.

On June 4, the culture exposed to the radio-tellurium was photographed with the control set (FIGURE 15).

EXPOSURE FOR 33 HOURS

The heights of the seedlings above the surface of the soil are as follows:

May 10, 10 A. M.

	A	B	D	E
	1,800,000 ×	1,500,000 ×	Ra. Tel.	Control
1	0.00 mm.	8.50 mm.	12.50 mm.	16.50 mm.
2	0.00	just up	5.00	18.00
3	0.00	"	0.00	11.00
4	0.00	"	10.00	12.00
	0.00 mm.		27.50 mm.	57.50 mm.
	0.00 mm.		9.16 mm.	14.37 mm.

FIG. 15. Experiment 26. Retardation of Growth of *Zea Mays* by Exposing Grains, before Planting, to a Sealed Glass Tube of Radio-Tellurium. Duration of Exposure, 24 Hours.

May 11, 10 A. M.

	A	B	D	E
	1,800,000 ×	1,500,000 ×	Ra. Tel.	Control
1	0.00 mm.	13.50 mm	22.50 mm.	29.50 mm.
2	6.00	7.00	17.00	28.00
3	0.00	7.00	0.00	23.00
4	4.00	3.00	20.00	22.50
	10.00 mm.	30.50 mm.	59.50 mm.	103.00 mm.
	5.00 mm.	7.62 mm.	19.83 mm.	25.75 mm.

May 12, 10 A. M.

	A	B	D	E
	1,800,000 ×	1,500,000 ×	Ra. Tel.	Control
1	0.00 mm.	19.00 mm.	34.00 mm.	44.00 mm.
2	8.00	14.00	30.00	37.00
3	5.00	12.00	0.00	37.00
4	7.00	6.00	32.00	36.00
	20.00 mm.	51.00 mm.	96.00 mm.	154.00 mm.
	6.66 mm.	12.75 mm.	32.00 mm.	38.50 mm.

May 14, 10 A. M.

	A	B	D	E
	1,800,000 ×	1,500,000 ×	Ra. Tel.	Control
1	0.00 mm.	29.00 mm.	76.00 mm.	108.00 mm
2	11.00	33.00	85.00	80.00
3	18.00	27.00	0.00	95.00
4	12.00	13.00	80.00	90.00
	41.00 mm.	102.00 mm.	241.00 mm.	373.00 mm.
	13.66 mm.	25.50 mm	80.33 mm.	93.25 mm.

May 15, 10 A. M.

	A	B	D	E
	1,800,000 ×	1,500,000 ×	Ra. Tel.	Control
1	0.00 mm.	29.00 mm.	93.00 mm.	135.00 mm.
2	11.00	33.00	107.00	111.00
3	19.00	27.00	0.00	121.00
4	12.00	13.00	90.00	115.00
	42.00 mm.	102.00 mm.	290.00 mm.	482.00 mm
	14.00 mm.	25.50 mm.	96.66 mm.	120.50 mm.

The results of this experiment are rather difficult to interpret, and do not harmonize in some places with the hypothesis that the effect of radium rays on growth varies directly with the activity of the radium, and the duration of the exposure. In the first place, the results of exposure to the various preparations are not in all points strictly comparable. For example, there was 100 times (.52 gm.) as much of the radium bromide of 10,000 × and of radio-tellurium as of the stronger radium preparations. Again, in laying the tubes over the seeds, it is more than probable that the salt was not distributed along the tube precisely similarly in any two cases, and doubtless not all of the four seeds of any given set were equally exposed, though great care was taken in this regard. The necessity of using such a small number of seeds was doubtless here (as in other experiments) a source of error, by emphasizing individual variations, but this could not well be eliminated on account of the scarcity of the radium, and the comparatively short period of time that any given preparation was at my disposal.

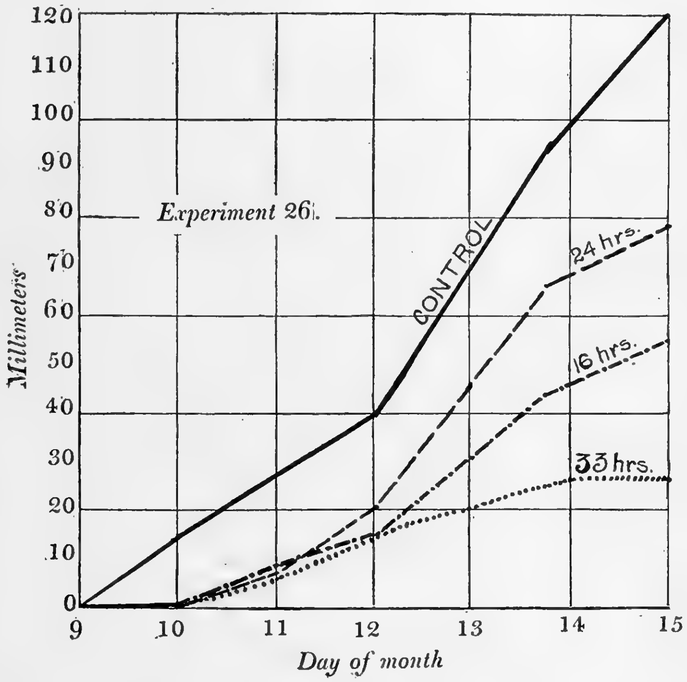

FIG. 16. Effect on the Growth of *Zea Mays* of Various Durations of Exposure of the Grains, before Planting, to Radium Rays. Activity, 1,500,000.

Interpretation of the results is greatly facilitated by inspection of the curves (FIGURES 10–12). In FIGURE 10 it is clearly shown that, for an exposure of 16 hrs., the effect of the rays from the preparations of 1,500,000 × and 1,800,000 × varied directly as the activity of the radium. The same fact is clearly indicated in FIGURES 11 and 12, where the exposures were respectively for 24 and 33 hours.

The immediate effect of exposure for 16 hrs. to the radium of 10,000 × (FIGURE 10) was a greater retardation of growth than that produced by the next more active preparation, but from the third day the plants of this culture grew faster, so that, from this time on, the rate of growth varied inversely as the strength of the radium. In FIGURE 11, also, growth following exposure to the larger amount of the weaker radium is shown to be less at first than after exposure to the radium of 1,800,000 ×, but soon becomes greater, though the indicated effect of the radium of 1,500,000 × is less than that of either the 10,000 × or the 1,800,000 × radium.

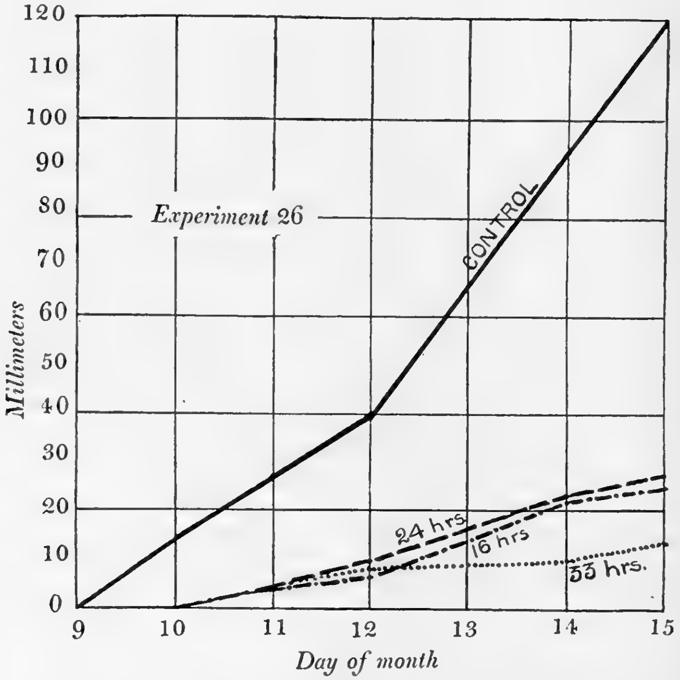

FIG. 17. Effect on the Growth of *Zea Mays* of Various Durations of Exposure of the Grains, before Planting, to Radium Rays. Activity, 1,800,000.

If we consider the effect of rays of the same strength allowed to act for varying lengths of time (FIGURES 13, 16, 17), we shall not find, in this experiment, that the hypothesis is very well verified. With the radium of 10,000 × (FIGURE 13) the results are what the hypothesis calls for.*

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* The 10,000 × culture of 33 hrs. exposure went bad, and was discarded, but this is only partially true in the other three cases (FIGURES 16, 17). These results being at variance with the expectation warranted by previous experience, it was impossible to avoid the conclusion that some unknown source of error crept into the experiment. I think it best, however, to give the results as they were obtained.

The experiment suggests that the degree of activity of the radium is a more important factor than the duration of the exposure. That is, that a slight increase in activity is more effective than a relatively slight increase in the duration of the exposure. If this is so, the facts concerning the effect of varying the length of exposure will be more readily ascertained by exposing for longer periods of time. With this in view the following experiment was made, and the greatest care was exercised in choosing seeds apparently as nearly alike as possible in size and vigor, in distributing the radium salt evenly over the bottom of the horizontal tube, in having the tube in contact with all the seeds, and at the same region of the seed, and in having the pots in which the exposed seeds were planted, and all other conditions of growth as nearly uniform as possible for all four cultures.

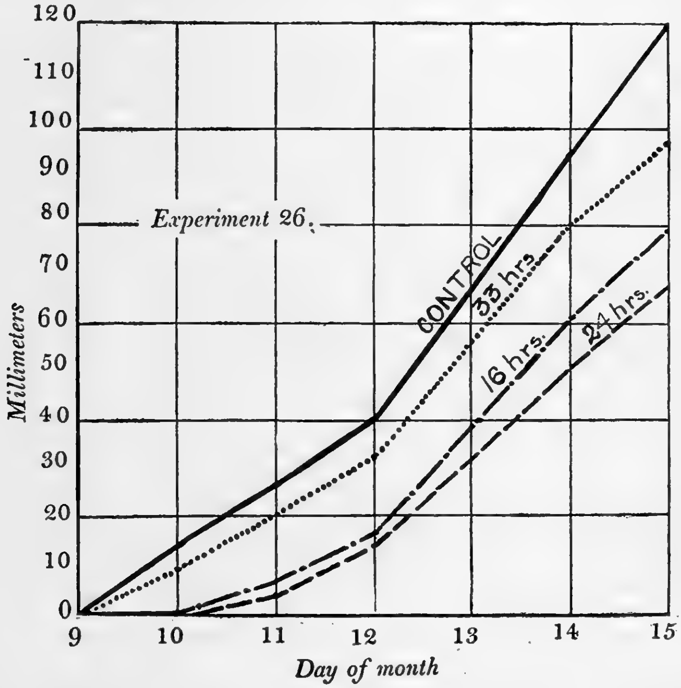

FIG. 18. Effect on the Growth of *Zea Mays* of Various Durations of Exposure of the Grains, before Planting, to Radio-Tellurium in a Sealed Glass Tube.

EXPERIMENT 27

Object: To ascertain the effect of varying lengths of exposure to the rays of radium of the same activity on the germination and growth of *Lupinus albus*.

October 22, 10 A. M.

Three sets of six dry seeds each, A, B, and C, were exposed as follows, to the rays from 10 mg. of $RaBr_2$, 1,800,000 activity, in a sealed glass tube:

A. 72 hrs. Planted 4 days after the end of the exposure.
B. 50 hrs. " 2 " " " " " "
C. 26 hrs. " 21 hours " " " " "
D. Control, not exposed.

The seeds were then planted without soaking, in separate pots in soil.

Fig. 19. Experiment 27. Effect of Duration of Exposure to Radium Rays on Germination and Growth of *Lupinus albus*. Length of Exposures, from Left to Right, 72 Hours, 50 Hours, 26 Hours, Control. Cf. FIGURE 20.

The lengths of the hypocotyls above the surface of the soil were measured as follows:

October 29, 10 A. M.

	A	B	C	D
	72 hrs.	50 hrs.	26 hrs.	Control
1	6.00 mm.	10.00 mm.	10.00 mm.	12.00 mm.
2	8.00	9.00	11.00	17.00
3	5.00	10.00	9.00	11.00
4	4.50	10.00	10.00	16.00
5	10.00	just up *	11.00	12.00
6	12.00	11.00	12.00	13.00
	45.50 mm.	50.00 mm.	63.00 mm.	81.00 mm.
	7.58 mm.	10.00 mm.	10.50 mm.	13.50 mm.

* Poor seed. Discarded.

October 30, 10 A. M.

	A	B	C	D
	72 hrs.	50 hrs.	26 hrs.	Control
1	10.00 mm.	16.00 mm.	16.00 mm.	18.00 mm.
2	10.00	11.00	17.00	22.00
3	9.00	15.00	16.00	16.00
4	5.00	14.00	16.00	20.00
5	12.00	0.00	15.00	19.00
6	16.00	15.00	16.00	19.00
	62.00 mm.	71.00 mm.	96.00 mm.	114.00 mm.
	10.33 mm.	14.20 mm.	16.00 mm.	19.00 mm.

October 31, 9:30 A. M.

	A	B	C	D
	72 hrs.	50 hrs.	26 hrs.	Control
1	13.00 mm.	19.50 mm.	20.00 mm.	19.50 mm.
2	10.00	12.00	20.00	25.00
3	9.50	19.50	21.00	20.00
4	6.00	19.00	20.00	24.50
5	12.00	0.00	19.50	25.00
6	19.50	20.00	21.00	19.50
	70.00 mm.	90.00 mm.	121.50 mm.	133.50 mm.
	11.66 mm.	18.00 mm.	20.25 mm.	22.25 mm.

November 1, 9:30 A. M.

	A	B	C	D
	72 hrs.	50 hrs.	26 hrs.	Control
1	14.00 mm.	19.50 mm.	20.00 mm.	21.00 mm.
2	12.00	12.00	20.00	27.00
3	10.50	20.00	21.00	21.50
4	8.50	19.00	22.00	24.50
5	14.00	0.00	20.50	25.00
6	20.50	21.00	24.00	21.00
	79.50 mm.	91.50 mm.	127.50 mm.	140.00 mm.
	13.25 mm.	18.30 mm.	21.25 mm.	23.33 mm.

November 2, 9:30 A. M.

	A 72 hrs.	B 50 hrs.	C 26 hrs.	D Control
1	15.00 mm.	19.00 mm.	20.00 mm.	21.00 mm.
2	12.00	12.50	21.00	29.00
3	10.50	20.00	21.00	21.50
4	10.00	19.00	22.00	25.00
5	15.00	0.00	21.00	26.00
6	23.00	21.00	24.00	21.00
	85.50 mm.	91.50 mm.	129.00 mm.	143.50 mm.
	14.25 mm.	18.30 mm.	21.50 mm.	23.92 mm.

November 3, 9:30 A. M.

	A 72 hrs.	B 50 hrs.	C 26 hrs.	D Control
1	15.00 mm.	19.00 mm.	21.00 mm.	21.00 mm.
2	12.00	12.50	21.00	29.00
3	12.00	21.00	24.00	22.00
4	10.00	21.00	22.00	25.00
5	16.00	0.00	21.50	27.00
6	23.00	21.50	25.00	21.00
	88.00 mm.	95.00 mm.	134.50 mm.	145.00 mm.
	14.66 mm.	19.00 mm.	22.41 mm.	24.16 mm.

In the 72-hrs. culture the plumule is above the partly opened cotyledons in only two seedlings. In the other three cultures the plumule is well above the spreading cotyledons. The effect on the root-system is shown in FIGURE 20.

November 5.

No apparent growth of the hypocotyl has taken place in any of the cultures since November 3, but the epicotyl has grown in all the plants of all four cultures, except four plants of the 72-hrs. exposure.

November 14.

The experiment was photographed (see FIGURES 19 and 20), and portions of the hypocotyls were preserved for sectioning (see p. 224).

The experiment clearly shows that, when the activity of the radium is the same, retardation of germination and growth varies directly with the duration of the exposure (see FIGURE 21).

The results are in perfect accord with the hypothesis, and with the results of other experiments. The rate of growth is seen to vary inversely with the duration of exposure (FIGURE 21), and to decrease most rapidly as the activity of the radium increases. The experiment was continued until all the hypocotyls ceased growing in all four cultures.

FIG. 20. Experiment 27. Root Systems of Seedlings of *Lupinus albus* from 72-Hour and from Control Cultures Shown in FIGURE 19.

EXPERIMENT 28

To ascertain the effect of exposing dry seeds for short periods of time to radium of strong activity, the following experiment was made.

Five sets of eight dry seeds of *Lupinus albus* each were exposed respectively to the rays of radium of 1,500,000 activity by having the sealed glass tube of the radium bromide (10 mg.) placed in contact with their hilum-edges. The duration of exposures was 2, 3, 4, 6, and 14 hrs., respectively.

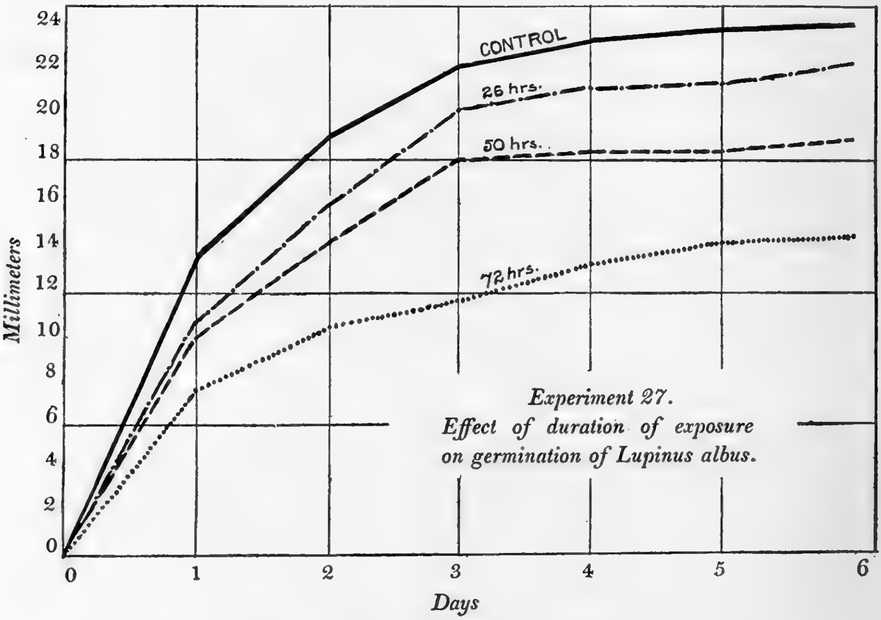

FIG. 21. Activity of Radium, 1,800,000.

April 17, 11 A. M.

The exposed seeds, together with eight control seeds, were planted in soil, in similar pots.

April 21, 2 P. M.

No marked difference could be observed in the six cultures. No measurements were taken.

April 23, 10 : 30 A. M.

Average lengths of the hypocotyls above the surface of the soil:

14 hrs.	6 hrs.	4 hrs.	3 hrs.	2 hrs.	Control
14.50 mm.	15.87 mm.	14.70 mm.	18.28 mm.	17.70 mm.	15.12 mm.

April 26, 11 A. M.

14 hrs.	6 hrs.	4 hrs.	3 hrs.	2 hrs.	Control
23.60 mm.	25.75 mm.	24.00 mm.	25.62 mm.	26.85 mm.	23.50 mm.

At the end of five weeks (May 24) there was no appreciable difference in the six cultures that could be attributed to the radium rays. An exposure of dry lupine seeds for 14 hrs. to radium of 1,500,000 × was not sufficient appreciably to affect germination or growth.

EXPERIMENT 29

The relation between the degree of activity of the radium and the retardation of growth is clearly shown by the following experiment.

Object: To ascertain the effect on the germination and growth of *Lupinus albus* of exposure for the same period of time to rays of radium of different degrees of activity.

Oct. 30, 1 : 30 P. M.

Three sets, A, B, and C, of six dry seeds each of *L. albus* were exposed to the rays of $RaBr_2$ in sealed glass tubes by laying the tubes against the hilum-edges of the seeds. The time of exposure was 91.50 hrs., and the strengths of the radium as follows:

FIG. 22. Experiment 29. Effect on the Germination and Growth of *Lupinus albus* of Exposure for the Same Period of Time to Rays from Radium of Different Activities, as Follows (from Left to Right): 1,800,000; 1,500,000: 10,000; Control.

A. 1,800,000 ×.
B. 1,500,000 ×.
C. 10,000 ×.
D. Control — not exposed.

Nov. 3, 9:30 A. M.

The seeds were planted in soil, each set in a separate six-inch pot.

Nov. 8, 8:30 A. M.

Seedlings are breaking through the soil in all cultures, but with no significant difference in the four sets.

Nov. 9, 8:30 A. M.

Relative growth the same as yesterday. Only 5 or 6 plants with hypocotyls long enough to measure. No measurements were taken.

Nov. 10, 10 A. M.

Measurements of the lengths of the hypocotyls were taken as follows:

	A	B	C	D
	1,800,000 ×	1,500,000 ×	10,000 ×	Control
1	8.00 mm.	9.00 mm.	5.00 mm.	5.50 mm.
2	0.00	4.50	4.50	13.00
3	7.00	0.00	14.00	5.00
4	3.00	5.00	5.50	8.00
5	7.00	5.00	9.00	11.00
6	12.00	6.00	5.50	11.00
	37.00 mm.	29.50 mm.	43.50 mm.	53.50 mm.
	6.16 mm.	4.91 mm.	7.25 mm.	8.91 mm.

Nov. 12, 9:30 A. M.

	A	B	C	D
	1,800,000 ×	1,500,000 ×	10,000 ×	Control
1	13.00 mm.	20.00 mm.	15.00 mm.	18.00 mm.
2	5.00	14.00	18.00	27.00
3	12.00	0.00	27.00	16.00
4	9.00	17.00	17.00	16.00
5	10.00	15.00	17.00	25.00
6	18.00	19.00	16.00	23.00
	67.00 mm.	85.00 mm.	110.00 mm.	125.00 mm.
	11.16 mm.	14.16 mm.	18.33 mm.	20.83 mm.

November 13, 9:30 A. M.

	A	B	C	D
	1,800,000 ×	1,500,000 ×	10,000 ×	Control
1	14.00 mm.	22.00 mm.	15.00 mm.	20.00 mm.
2	10.00	17.00	19.00	31.00
3	10.50	6.00	28.50	16.50
4	10.00	18.00	17.50	18.00
5	13.50	16.00	17.00	26.00
6	18.00	20.00	17.50	24.00
	76.00 mm.	99.00 mm.	114.50 mm.	135.50 mm.
	12.66 mm.	16.50 mm.	19.08 mm.	22.58 mm.

November 14, 9:30 A. M.

	A	B	C	D
	1,800,000 ×	1,500,000 ×	10,000 ×	Control
1	14.00 mm.	22.50 mm.	16.00 mm.	21.00 mm.
2	10.00	17.00	21.00	33.50
3	11.50	10.00	30.00	19.00
4	10.50	20.00	18.00	20.50
5	14.50	17.00	17.50	27.50
6	18.50	21.00	19.00	27.00
	79.00 mm.	107.50 mm.	121.50 mm.	148.50 mm.
	13.16 mm.	17.91 mm.	20.25 mm.	24.75 mm.

November 15, 9:30 A. M.

	A	B	C	D
	1,800,000 ×	1,500,000 ×	10,000 ×	Control
1	14.00 mm.	26.00 mm.	19.00 mm.	23.00 mm.
2	10.00	17.00	24.00	38.50
3	11.50	12.00	34.00	21.00
4	10.50	21.00	20.00	20.00
5	14.50	18.50	19.00	30.00
6	19.00	23.00	20.00	30.00
	79.50 mm.	117.50 mm.	136.00 mm.	162.50 mm.
	13.25 mm.	19.58 mm	22.66 mm.	27.08 mm.

November 16, 9:30 A. M.

	A	B	C	D
	1,800,000 ×	1,500,000 ×	10,000 ×	Control
1	14.00 mm.	28.00 mm.	20.00 mm.	26.00 mm.
2	10.00	18.00	27.00	42.00
3	11.50	15.00	34.50	23.50
4	10.50	24.00	21.00	23.00
5	14.50	20.00	19.50	31.00
6	19.00	24.00	22.00	34.00
	79.50 mm.	129.00 mm.	144.00 mm.	179.50 mm.
	13.25 mm.	21.50 mm.	24.00 mm.	29.91 mm.

November 17, 9:30 A. M.

	A	B	C	D
	1,800,000 ×	1,500,000 ×	10,000 ×	Control
1	14.00 mm.	30.00 mm.	22.00 mm.	28.00 mm.
2	10.00	18.00	28.50	45.00
3	11.50	17.00	35.50	26.50
4	10.50	25.00	23.50	25.50
5	14.50	20.50	20.00	31.50
6	19.00	25.50	24.00	37.50
	79.50 mm.	136.00 mm.	153.50 mm.	194.00 mm.
	13.25mm.	22.66 mm.	25.58 mm.	32.33 mm.

When the duration of the exposure is the same, then the retardation of growth varies directly with the strength of the radium employed. (See FIGURE 23.)

The 1,800,000 × curve is made to cross the 1,500,000 × curve during the first time interval by the excessive growth of one seedling (No. 6) of the 1,800,000 × culture. It is very probable that this seed was not as effectively exposed as the others. Likewise the initial difference between the average growth of the 10,000 × culture and the control on November 10 would have been greater but for the exceptional growth of seed No. 3, which was either not well exposed to the rays, or else was unusually resistant to their influence.

In experiments 26, 27, and 28, it was necessary that a longer time should intervene between exposure and planting in some cultures than in others. The following experiment was made in order to see if this variation in time-interval had any effect on the result of exposure for the same length of time to radium of the same activity, at least within the limits imposed in the preceding experiments.

EXPERIMENT 30

Object: To see if a variation in the time-interval between the exposure of seeds to radium-rays and their germination, alters the efficacy of the exposure to affect germination.

November 19, 10 A. M.

Three sets of six seeds each of *Lupinus albus*, treated as follows, were planted without soaking, in soil in flower pots.

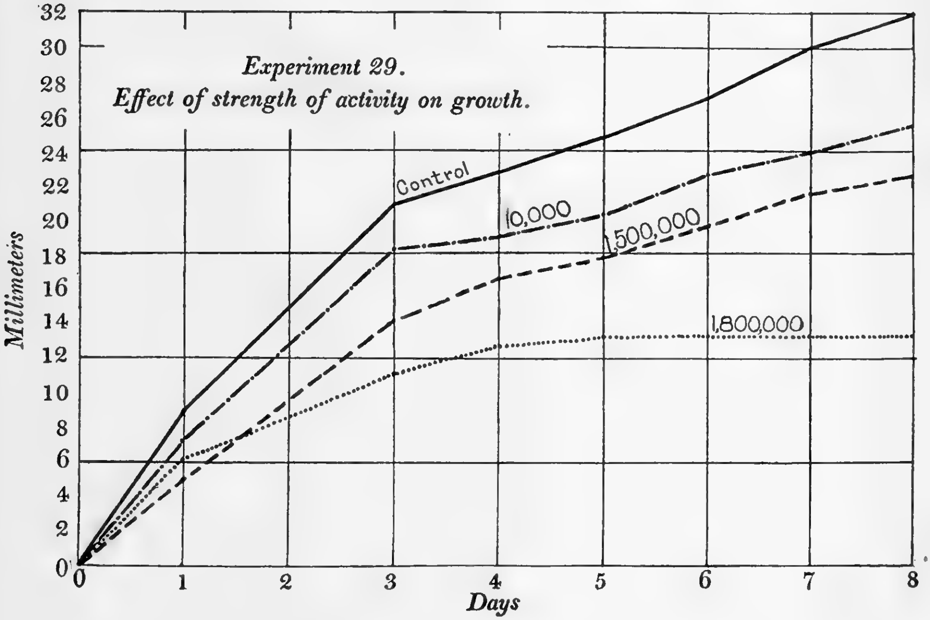

FIG. 23. *Lupinus albus.* Duration of Exposure, 91.50 Hours.

A. Exposed for 72 hrs. to $RaBr_2$ 1,500,000 ×. Interval between exposure and planting, 6 days.

B. Exposed for 72 hrs. to $RaBr_2$ 1,500,000 ×. Interval between exposure and planting, 3 days.

C. Exposed for 72 hrs. to $RaBr_2$ 1,500,000 ×. Interval between exposure and planting, 0 days.

D. Control, not exposed. Planted at same time as A, B, and C.

Heights of the hypocotyls above the soil-surface as follows:

November 26, 10 A. M.

	A	B	C	D
	6 days	3 days	0 days	Control
1	7.00 mm.	6.50 mm.	4.50 mm.	9.50 mm.
2	6.50	9.50	5.50	11.50
3	8.00	not up	6.50	8.50
4	8.50	9.50	7.50	10.00
5	7.00	9.00	5.50	10.00
6	8.00	5.00	6.50	11.50
	45.00 mm.	39.50 mm.	36.00 mm.	61.00 mm.
	7.50 mm.	7.90 mm.	6.00 mm.	10.16 mm.

November 27, 10 A. M.

	A	B	C	D
	6 days	3 days	0 days	Control
1	7.50 mm.	7.50 mm.	6.50 mm.	13.50 mm.
2	8.50	13.00	6.00	15.50
3	8.50	not up	11.00	13.50
4	11.50	11.00	11.50	16.50
5	8.50	10.00	9.00	14.00
6	8.50	8.50	13.00	19.00
	53.00 mm.	50.00 mm.	57.00 mm.	92.00 mm.
	8.83 mm.	10.00 mm.	9.50 mm.	15.33 mm.

November 28, 10 A. M.

	A	B	C	D
	6 days	3 days	0 days	Control
1	8.00 mm.	10.00 mm.	8.50 mm.	16.00 mm.
2	9.50	15.00	9.50	21.50
3	12.50	not up	13.00	16.50
4	14.00	13.00	15.00	19.50
5	9.50	10.00	9.00	18.00
6	9.50	10.00	15.00	23.50
	63.00 mm.	58.00 mm.	70.00 mm.	115.00 mm.
	10.50 mm.	11.60 mm.	11.66 mm.	19.16 mm.

November 29, 10 A. M.

	A	B	C	D
	6 days	3 days	0 days	Control
1	10.00 mm.	12.00 mm.	13.00 mm.	21.00 mm.
2	15.00	21.00	12.00	29.00
3	16.00	just up	13.00	21.50
4	20.00	13.50	18.00	25.00
5	13.00	12.00	10.00	24.50
6	10.00	11.00	19.00	30.00
	84.00 mm.	69.50 mm.	85.00 mm.	151.00 mm.
	14.00 mm.	13.90 mm.	14.16 mm.	25.16 mm.

November 30, 10 A. M.

	A	B	C	D
	6 days	3 days	0 days	Control
1	10.00 mm.	13.00 mm.	14.00 mm.	21.00 mm.
2	15.00	21.00	12.00	29.00
3	16.00	—	13.00	21.50
4	21.00	13.50	18.00	25.00
5	13.50	12.00	10.00	24.50
6	10.00	11.00	19.00	30.00
	85.50 mm.	70.50 mm.	86.00 mm.	151.00 mm.
	14.25 mm.	14.10 mm.	14.33 mm.	25.16 mm.

A time-interval of from three to six days between exposure and planting makes only a very slight, if any difference, at first, and any early difference, if one really exists, does not persist.

EXPERIMENT 31

Object: To ascertain the result of varying the distance between the radium-tube and the seeds, on the effect of radium rays on the germination and growth of *Lupinus albus*.

Four sets of six seeds each of *L. albus* were exposed for 72 hours to the rays from the same amount of radium bromide of 1,500,000 activity, contained in a sealed glass tube. The distances from the bottom of the radium-tube to the upper surfaces of the seeds were respectively 80 mm., 40 mm., 20 mm., and 0 mm.

December 7, 10 A. M.

The seeds, exposed as above, together with a control (unexposed) set of six, were all planted without soaking, in soil in pots.

The lengths of the hypocotyls above the soil-surface are as follows:

December 17, 10 A. M.

	80 mm.	40 mm.	20 mm.	0 mm.	Control
1	25.00 mm.	20.00 mm.	21.00 mm.	16.00 mm.	not up
2	not up	20.00	20.00	8.00	24.00 mm.
3	17.00	18.00	21.00	12.00	17.00
4	27.00	20.00	20.00	10.00	19.00
5	20.00	17.00	28.00	12.00	23.00
6	30.00	23.00	25.00	16.00	30.00
	119.00 mm.	118.00 mm.	135.00 mm.	74.00 mm.	113.00 mm.
	23.80 mm.	19.66 mm.	22.50 mm.	12.33 mm.	22.60 mm.

December 18, 10 A. M.

	80 mm.	40 mm.	20 mm.	0 mm.	Control
1	30.00 mm.	25.00 mm.	25.00 mm.	20.00 mm.	not up
2	——	28.00	25.50	12.00	26.00 mm.
3	23.00	22.00	27.00	18.00	22.00
4	30.00	25.00	24.00	13.00	23.50
5	24.00	23.00	30.00	15.00	28.00
6	35.00	29.50	29.00	20.00	35.00
	142.00 mm.	152.50 mm.	160.50 mm.	98.00 mm.	134.50 mm.
	28.40 mm.	25.41 mm.	26.75 mm.	16.33 mm.	26.90 mm.

It is seen that the effect produced when the radium-tube is in immediate contact with the seeds is marked, and in agreement with the results of preceding experiments. No effect, however, can be detected when the exposure is made at distances of 20 mm., 40 mm., and 80 mm., and this indicates that the β rays are physiologically more effective than the γ rays, for, while the former are eliminated by these distances in air, the latter are not, or not completely.

CHAPTER VIII

EFFECTS OF RADIUM RAYS IN THE SOIL ON GERMINATION AND GROWTH

It has been ascertained, as previously pointed out, that soil-air is radioactive. Therefore it becomes a matter of considerable interest to ascertain the effect on germination and growth of passing radium rays through soil in which seeds are planted. The object of the following experiments is to answer this question.

Experiment 32

March 4, 4 P. M.

Unsoaked seeds of "Lincoln" oats (*Avena*) were sown in soil in three concentric circles, at distances of 7 mm., 22 mm., and 45 mm. from the center of the pot. The sealed glass tube of $RaBr_2$ (1,500,000 ×) was inserted in the soil at the center of the pot, with the end containing the radium at a depth of about 15 mm. below the surface.

FIG. 24. Experiment 32. Acceleration of Germination and Growth of Oats by Placing a Sealed Glass Tube of Radium (1,500,000 ×) in the Soil. The Glass Tube in *C* is Empty. Cf. FIGURES 8, 25, and 26.

Control culture similarly arranged, but with empty tube.

March 10, 2 P. M.

After an exposure of 106 hours, the seedlings in the pot containing the radium are all up, and are most decidedly taller than those in the control culture, three of which were not yet up, and all of which were less developed in every way than those exposed to the radium.

The plants in the outer circle of the exposed culture average 50 mm., those in the middle circle 46 mm., and those in the inner circle 42 mm. taller than those in the corresponding circle of the control.

FIG. 25. Experiment 32. The Former Control (*C*) is now Exposed to the Radium (*CR*), and the Culture Formerly Exposed (*R*) becomes the Control. The Tube in *CR* now Contains the Radium, while the Tube in *R* is now the Empty Tube. Cf. FIGURE 24.

After the plants were photographed (FIGURE 24), one seedling was carefully removed from each culture, and the soil washed from its roots. The plant from the exposed culture has root-hairs from 2 to 3 times as long as those on the control, and they are also more numerous.

March 10, 5 P. M.

I now changed places with the radium-tube and the empty tube, placing one end of the radium-tube in the center of the soil of the culture that had served as the control. This pot was labeled CR, and the other pot R. Five days later the control plants (CR), under the influence of the radium, averaged nearly as tall as those first radiated. Thus, by changing the radium-tube back and forth from time to time, one can accelerate either culture at will (FIGURE 25).

EXPERIMENT 33

Repetition of Experiment 32, using seeds of *Lupinus albus*.

March 19, 11:30 A. M.

Six unsoaked seeds of *L. albus* were planted about 5 mm. deep around the margin of a 5-inch pot. All the seeds were placed with the hypocotyl facing the center of the pot and pointing downward. In the center of the soil was inserted one end of the sealed glass tube containing $RaBr_2$ of 1,500,000 activity, to a depth of about 10 mm.

Control pot with six seeds and empty glass tube.

March 24, 11:30 A. M.

In the radium-culture no seeds are up. Two of them are just beginning to lift the soil.

In the control culture four seeds are up, from 10 to 15 mm. high. Two other seeds are just coming through the soil.

On March 26 only one exposed seed was up, with the hypocotyl 10 mm. high, while 5 of the control seeds were up with an average height for the five of 19.60 mm. The radium and empty tubes were removed from the soil.

Six days later (March 30) only two of the exposed plants had come up. Their average height was 30 mm. Five of the control seeds were well up, with an average height of 45.60 mm.

The plants were all removed from the soil. On the two radiated plants the primary and secondary roots were very slightly developed, but on the control plants the secondary roots were long and numerous, and the primary root was from three to five times as long as in the plants exposed to the radium.

EXPERIMENT 34

Repetition of Experiment 33, using white mustard (*Brassica alba*) seeds instead of lupine.

March 19, 11:30 A. M.

Exposed and control cultures were arranged as described in Experiment 33, except that white mustard seeds were used. Twenty-one unsoaked seeds were planted in each pot, at distances from the center as follows:

11 seeds 40 mm. from the center.
6 seeds 25 mm. from the center.
4 seeds 10 mm. from the center.

March 23, 10:30 A. M.

Eleven of the exposed seeds have germinated, and only six of the control seeds. The seedlings from the exposed seeds average at least one third taller than the control seedlings, but measurements were not taken. The exposed plants seem in every way more vigorous.

EXPERIMENT 35

April 3, 9 A. M.

Into each of three pots of soil were planted 12 unsoaked seeds of Henderson's "First-of-All" peas (*Pisum sativum*), in two circles, the outer one of 8 seeds 60 mm. from the center of the pan, the inner of 4 seeds 30 mm. from the center.

Into the center of the soil in the first pan was inserted the end of a tube of radium bromide (10,000 ×), in the center of the second the sealed glass tube of radio-tellurium, into the third an empty glass tube. The lower ends of all the tubes were depressed 25 mm. below the surface of the soil.

April 6, 10:30 A. M.

8 plants exposed to radium rays show the arch of the hypocotyl.
6 plants exposed to radio-tellurium show the arch of the hypocoytl.
3 plants of the control culture show the arch of the hypocotyl.

April 18, 10 A. M.

The plants grown in the soil with the radium-tube are taller, and in every way more vigorous looking than those of the other two pots. There is very slight, if any, difference between the height of the plants grown with the radio-tellurium in the soil and the control plants.

A count of the total number of root tubercles on the roots of the plants gave a total of 18 tubercles where the radium-tube was, 19 with the radio-tellurium, and 22 in the control. These differences are not considered significant.

EXPERIMENT 36

April 15, 11 : 15 A. M.

Twenty unsoaked seeds of "Lincoln" oats were sown in soil in a 6-inch pot, and a rod coated with a "Lieber's radium coating" (10,000 ×) was thrust vertically into the soil in the center of the pot. All the seeds were placed with the radicle vertical, and the embryo-side of the seed facing the radium-coated rod.

Control with no rod, and both cultures placed in a glass frame in the propagating house.

April 28, 5 P. M.

Heights of the seedlings from soil-surface to tips of second leaf:

Radium		*Control*	
77.00 mm.		113.00 mm.	
91.50		86.50	
94.50		107.50	
83.50		90.00	
64.00		0.00	injured
21.50		39.00	
61.00		20.00	
107.00		32.00	
44.50		109.00	
116.50		63.00	
0.00	injured	94.00	
91.00		37.00	
98.50		67.00	
44.00		0.00	
71.50		93.00	
69.00		170.00	
24.00		160.00	
78.00		148.00	
79.00		67.00	
0.00	injured	108.00	
1,316.00 mm.		1,604.00 mm.	
Average of 18, 73.11 + mm.		Average of 19, 84.42 + mm.	

April 29.

Cultures photographed (FIGURE 26), and experiment closed.

EXPERIMENT 37

May 12, 5 P. M.

Into each of three 6-inch pots were planted eight unsoaked seeds of the bean (*Phaseolus*). Into the center of the first pot (R)

FIG. 26. Experiment 36. Retardation of Germination and Growth of Oats by Placing in the Soil a Celluloid Rod Coated with Lieber's Radium Coating. Cf. FIGURE 24.

was placed the sealed glass tube containing the $RaBr_2$ (1,500,000 ×), into the second (R′) four rods coated with "Lieber's radium-coating" 25,000 ×), the third served as a control, with no radium.

May 19, 5 P. M.

The lengths of the hypocotyls above the surface of the soil were as follows:

R Tube, 1,500,000 ×	R′ Coated Rods	C Control
95.00 mm.	98.00 mm.	114.00 mm.
53.00 *	84.00	99.50
45.00 *	89.00	112.50
125.00	96.00	91.50
106.00	113.00	112.00
67.00	98.00	30.00 †
100.00	60.00 †	89.00
104.00	0.00	114.00
695.00 mm.	638.00 mm.	762.50 mm.
Average of 8: 86.875 mm.	Average of 7: 91.14 + mm.	Average of 8: 95.31 + mm.

* To top of arch. † To top of arch. Went bad.

EXPERIMENT 38

Object: To ascertain the effect on the germination and growth of wheat of the rays from $RaBr_2$ and radio-tellurium contained in sealed glass tubes placed in the soil.

June 4, 11 A. M.

Twelve grains of wheat (*Triticum vulgare*, Henderson's "Wellman Fife") were planted without soaking in each of four pots, A, B, C, and D. Each grain was placed in the soil vertically with the embryo-end down, and the embryo-side facing the center of the pot. In the center of pots A, B, and C, were placed vertically the tubes of $RaBr_2$ and radio-tellurium, as follows:

A. $RaBr_2$ 10 mg. 1,800,000 ×. End about 10 mm. below the surface.
B. " " 1,500,000 ×. " " " "
C. Radio-tellurium. " " " "
D. Control. No tube.

June 6.

The tubes have been removed for 18 hours since planting. The average heights of the seedlings that have come up are as follows:

A	B	C	D
1,800,000 ×	1.500,000 ×	Radio-tellurium	Control
7 up. Av. ht.	4 up. Av. ht.	3 up. Av. ht.	7 up. Av. ht.
3.28 mm.	4.87 mm.	6.33 mm.	2.58 mm.

The tubes were replaced.

June 7, 10:30 A. M.

A	B	C	D
1,800,000 ×	1,500,000 ×	Radio-tellurium	Control
12 up. Av. ht.	12 up. Av. ht.	10 up. Av. ht.	10 up. Av. ht.
23.60 mm.	22.90 mm.	24.35 mm.	15.80 mm.

The tubes were again removed at 11 A. M. for another experiment.

June 8.

Heights of seedlings above the surface of the soil as follows:

	A	B	C	D
	1,800,000 ×	1,500,000 ×	Radio-tellurium	Control
1	41.50 mm.	51.00 mm.	53.00 mm.	13.00 mm.
2	35.00	40.00	32.00	33.50
3	44.50	41.00	50.00	31.00
4	45.00	40.50	45.50	injured
5	47.00	36.00	47.00	39.00
6	47.00	48.00	48.00	injured
7	38.00	54.50	65.50	23.00
8	53.50	48.00	injured	injured
9	53.50	52.50	27.00	39.50
10	45.00	30.00	50.00	33.00
11	57.00	40.00	injured	41.50
12	37.00	38.00	45.50	39.50
	544.00 mm.	519.50 mm.	463.50 mm.	293.00 mm.
Av.	45.30 mm.	43.20 mm.	46.35 mm.	32.55 mm.

Acceleration of growth has followed exposure to all the radioactive substances. (See FIGURE 27.)

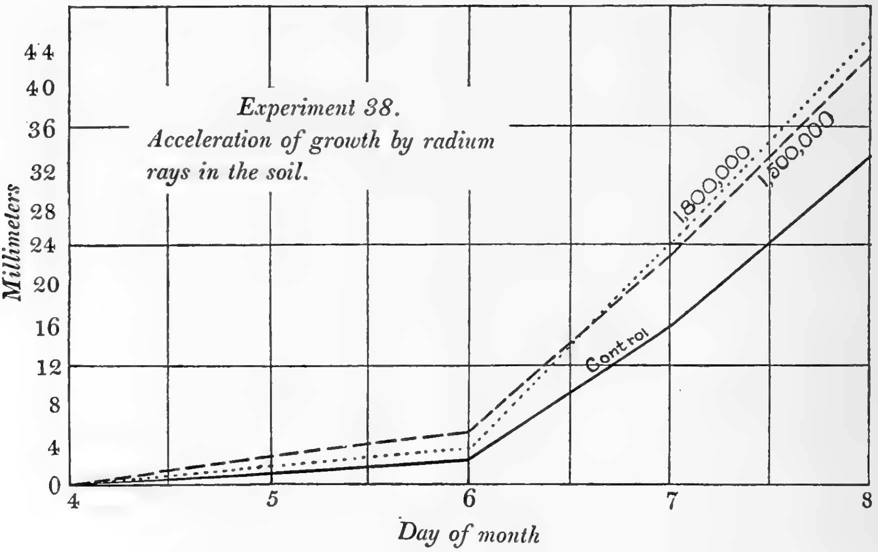

FIG. 27. *Triticum vulgare.*

Experiment 39

April 1, 4 P. M.

Unsoaked timothy grass (*Phleum pratense*) seed was sown on the surface of the soil in two flower pots. In the center of the soil of one pot was inserted the end of the glass tube containing $RaBr_2$ of 7,000 activity, to a depth of 15 mm.; in the control pan the empty glass tube.

Both pots were watered by being set in a pan of water until the top of the soil appeared entirely moistened. They were then covered with bell-jars.

April 10, 11:30 A. M.

The seedlings in the radiated culture are slightly taller than those of the control set, but the difference is not marked.

April 11, 5 P. M.

There is no appreciable difference in the height of the plants in the two pots.

Summary

When unsoaked oat grains were planted at distances of 7, 22, and 45 mm. from a sealed glass tube containing 10 mg. of radium bromide of 1,500,000 × inserted into the soil, germination and subsequent growth were accelerated. The seeds farthest from the radium were accelerated most, those nearest least. The root hairs on the exposed seedlings appeared to be more numerous, and were 2–3 times longer than normally. When seeds of *Lupinus albus* were exposed in a similar way to the same radium preparation the growth of the shoot was retarded, but the roots were from three to five times longer than normally. This is in agreement with the results of Willcock and of Zuelzer which indicate that tissues containing chlorophyll are more sensitive to these rays than other tissues. Under similar conditions the germination of seeds of *Brassica alba* was accelerated. When seeds of *Pisum sativum* were similarly exposed, using radium of 10,000 × and the sealed glass tube of radio-tellurium, acceleration of growth was produced by the radium rays, but none by the radio-tellurium. Careful counting disclosed no significant difference in the number of root-tubercles on the exposed plants and those of the control culture. More careful observation has not confirmed my earlier * statement to the contrary.

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* Bibliography, p. 71. No. 24.

The growth of oats exposed in the same way (10,000 ×) was accelerated, but bean seeds (*Phaseolus vulgaris*) exposed to radium (1,500,000 ×) in the sealed tube, and also to four coated rods (25,000 ×) had their germination and the growth of the seedlings retarded. Exposure to the rods (low activity) produced less retardation than did exposure to the preparation in the glass tube (high activity), though in the former case the α rays escaped for very short distances into the soil.

Exposure of wheat (*Triticum vulgare*) to radium of 1,800,000 × and 1,500,000 ×, and to radio-tellurium, each in a sealed glass tube inserted into the soil, was followed by acceleration of germination and growth. The amount of acceleration was about equal in each of the three exposed cultures, though slightly in excess in the culture exposed to the radio-tellurium, where the amount of the salt was larger than in the radium cultures. I am unable to explain how physiological effects can be obtained with radio-tellurium in a sealed glass tube, for this substance gives off only α rays, and these are not thought to be able to pass through the glass walls of the tube. The results, however, were constant and decided, leaving not the slightest doubt as to the physiological efficacy of the preparation.

When seeds of timothy grass were sown on the surface of soil into which a sealed glass tube of radium bromide of 7,000 × was inserted to a depth of about 10 mm. below the surface, germination and growth were very slightly accelerated.

Whether the acceleration of growth produced by inserting sealed glass tubes of radioactive preparations into the soil is due to the direct action of the rays, or to ions which they may possibly form in the soil-solution, remains to be demonstrated. If to the former, then the result must be attributed largely to the gamma rays, for the alpha rays do not leave the glass tube, and the beta rays would be stopped by at least one centimeter of moist soil. The gamma rays, however, on account of their high penetrability, might be effective through as much as one foot of moist soil.

Fischer[1] has shown that hydrogen ions and hydroxyl ions, whether of acids or of strong alkalis, stimulate germination. In his experiments the ions acted explosively, as he described it, a marked effect being produced by them by an exposure of one half a minute, a maximum stimulation on two minutes, while killing began with only four minutes' exposure. If the radium rays produce ionization in the mineral solutions in the soil then these ions would act as a stimulus to plants growing there, and, under suitable conditions, cause an acceleration of growth. It is not improbable that the results recorded above are due to a combination of both causes, that is, to the direct action of the gamma rays combined with that of ions produced by the rays in the soil-solution.

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1. **Fischer, Alfred.** Wasserstoff- und Hydroxylionen als Keimungsreize, Ber. Deut. Bot. Ges. **25**: 108. 1907.

CHAPTER IX

EFFECTS OF A RADIOACTIVE ATMOSPHERE ON PLANT GROWTH

The fact, pointed out in Chapter II, that the earth's atmosphere normally contains the emanation of radium and possibly of other radioactive substances, makes it desirable to ascertain the effect of this factor of environment on plants. Such an experiment was rendered comparatively easy by the invention by Mr. Hugo Lieber of a cylinder lined with a coating of a radium salt, and covered by a protecting layer of such a nature as to permit of the diffusion of the emanation into the surrounding space, as described more fully on page 81

FIG. 28. Experiment 40. Apparatus for growing Plants in an Atmosphere containing the Emanation of Radium. The inner Surface of the Tube (*T*) is coated with Lieber's Radium Coating. From the lower Tubulures of the Bell-Jars Rubber Tubing leads to a Suction Pump. Control Apparatus at the right. Cf. FIGURE 30.

of this Memoir. The details of the method are given in connection with the experiments that follow.

EXPERIMENT 40

Object: To ascertain the effect of germinating and growing timothy grass (*Phleum pratense*) seeds in an atmosphere containing the radium emanation.

March 12, 11 A. M.

Unsoaked timothy seeds were sowed in a pot of earth (thinly covered with the soil), and placed to germinate and develop under a bell-jar. From a hollow cylinder lined with "Lieber's radium-coating" there passes a glass tube through the upper tubulure of the bell-jar and down into the latter, ending, at the side of the pot of soil, in a dovetail gas tip, at a height of about 20 mm. above the surface of the soil. From the opposite side of the pot leads another glass tube through the bottom tubulure of the bell-jar to a suction pump. By this means a current of air containing the emanation from the radium-lined cylinder spreads out over the surface of the soil, and converges to the outlet tube, passing thence out of the jar. A control apparatus was similarly arranged, connected with the same suction pump, but with no radium tube attached (FIGURE 28).

FIG. 29. Experiment 41. Retardation of Growth of Timothy Grass by exposure to Air which has passed over Lieber's Radium Coating, and thus contains Radium Emanation.

March 15, 4:30 P. M.

There are no signs of germination in either pot.

March 16, 9:30 A. M.

The seeds in both pots have begun to germinate, but those exposed to the emanation appear to be slightly further advanced than the control.

March 17, 9:30 A. M.

The seedlings of the exposed culture are decidedly taller than those in the control.

This difference in height of the seedlings of the two cultures continued until March 24, when the experiment was discontinued.

EXPERIMENT 41

The object of this experiment is to ascertain the effect of growing seeds of timothy grass (*Phleum pratense*) in the apparatus described for Experiment 40, *i. e.*, in an atmosphere containing the radium emanation, but exposed more directly to the rays resulting from the decay of the emanation.

April 1, 2:30 P. M.

The experiment was set up as described in Experiment 40, except that the air and the emanation were delivered at a distance of only about 5 mm. above the seeds. Special care was taken to have the illumination equal on all sides, and both the light and the moisture conditions as nearly as practicable the same for both the exposed and the control cultures.

April 6, 8:30 A. M.

Germination has begun in both cultures, but is more advanced in the control pan.

April 10, 10 A. M.

In the exposed culture the plants are shorter and lighter colored on the side nearest the delivery of the emanation, and increase in size toward the opposite side of the pan. They manifest a slight phototropic curvature.

The plants of the control culture are of uniform height, and also have a slight phototropic curvature like those exposed.

The growth of the seedlings nearest the point of delivery of the emanation has been retarded. The cultures were photographed on April 12 (FIGURE 29).

This experiment was repeated with entirely confirmatory results.

EXPERIMENT 42

In order to test the effect on the germination and growth of the timothy seed, when grown in an atmosphere containing the radium emanation, but with the emanation delivered at a greater distance from the seeds than was the case in the preceding experiments, the following experiment was made.

May 27, 5 : 30 P. M.

Dry seeds of timothy grass were sown on the surface of the soil

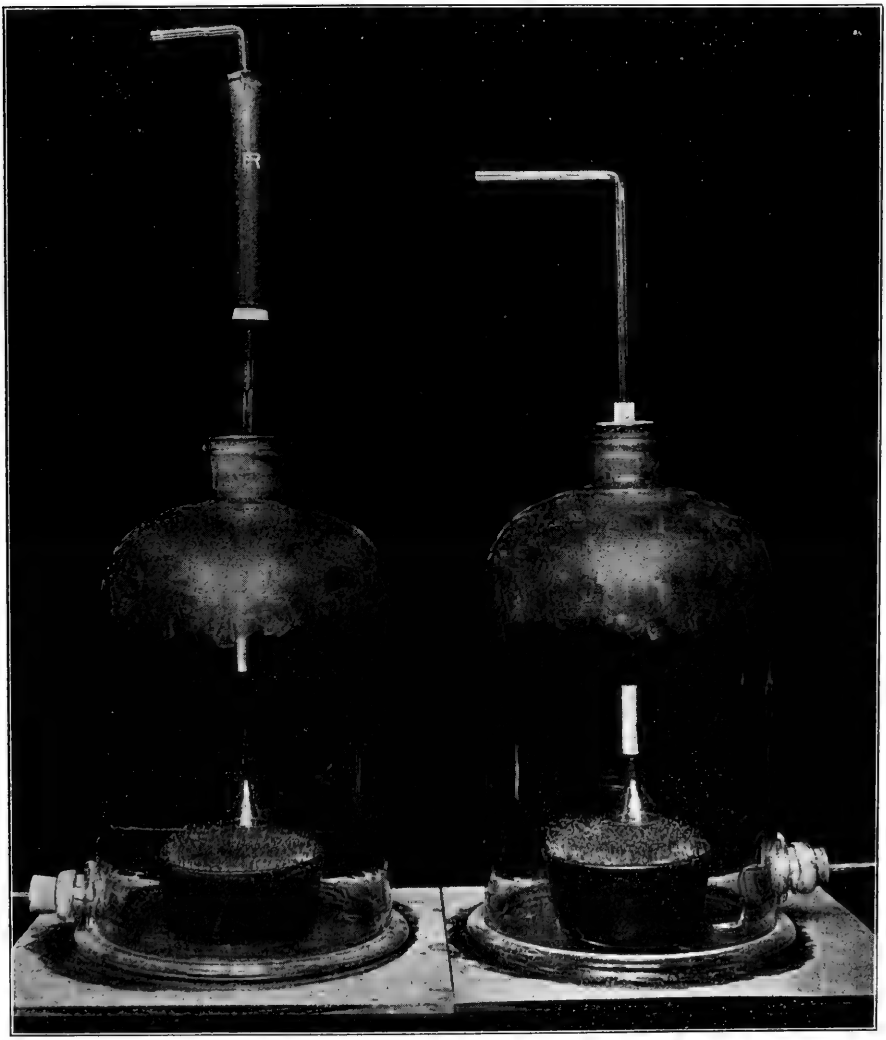

FIG. 30. Experiment 42. Apparatus for growing Plants in an Atmosphere containing Radium Emanation. The Air, after passing over the Radium Coating on the inner Surface of the hollow Cylinder (*R*), is delivered over the Cultures through a glass Funnel. Control Apparatus at the right. Cf. FIGURE 28.

in two pots, and the pots placed, one under each of the bell-jars, as described in Experiment 41, except that the emanation was delivered over the seeds from a small glass funnel, suspended over the center of the pot at a distance of 50 mm. The ordinary air in the control was similarly delivered over the control seeds (FIGURE 30).

May 30, 9:30 A. M.

The exposed seeds have germinated except directly under the funnel, where germination has not yet begun.

The control seeds have germinated uniformly, under the funnel as well as elsewhere.

June 1, 9 A. M.

The exposed seeds have practically all failed to germinate under the funnel, but are quite evenly germinated elsewhere. Numerous decidedly undersized seedlings are scattered throughout the culture.

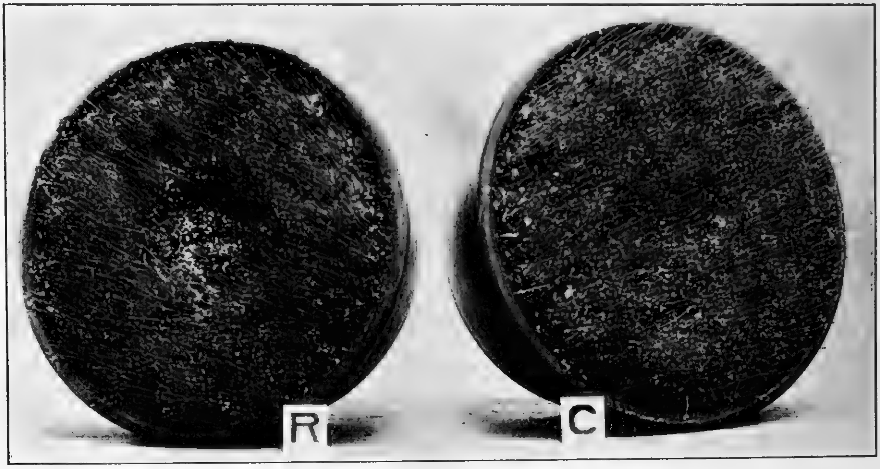

FIG. 31. Experiment 42. Inhibition of Germination of Timothy Grass Seed by direct exposure to Air that has passed over a surface coated with Lieber's Radium Coating, and thus contains Radium Emanation. Cf. FIGURE 30.

The control seeds are all evenly germinated, and there is no appreciable difference in their height under the funnel and elsewhere. There are also very few undersized seedlings — much fewer than in the exposed culture.

Complete inhibition of germination has followed exposure under the conditions described. On June 1 the cultures were photographed (FIGURE 31), and the experiment closed.

EXPERIMENT 43

Repetition of Experiment 42, only with the air and emanation delivered at a distance of 190 mm. above the seeds, planted on the surface of the soil.

June 1, 4:30 P. M.

Dry seeds of timothy grass were sown evenly over a blotter, kept moist by being placed on damp cotton, and placed under the bell-jar into which the radium emanation is to be drawn. The opening of the funnel through which the emanation is to be delivered is about 190 mm. from the surface of the blotter where the seeds lie.

Control with no radium.

June 3, 6 P. M.

No seeds have germinated in either culture.

June 4, 12 M.

In the exposed culture only one or two seeds have begun to germinate.

In the control culture the seeds have quite generally germinated, with plumules estimated at from 1 to 2 mm. long.

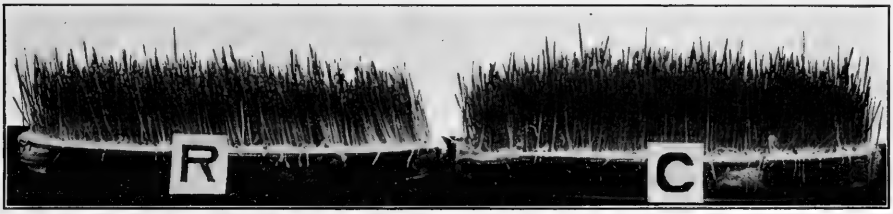

FIG. 32. Experiment 43. Retardation of Germination and Growth of Timothy Grass by exposure to Air containing the Emanation of Radium.

June 5, 12 M.

The seedlings in the control culture are about 5 mm. high, with the green color well developed. Growth is uniform throughout the culture.

The exposed seedlings are not more than one half as tall as the control, and only very slightly green. Growth seems uniform throughout the culture.

June 8, 3:30 P. M.

After seven days of exposure the radiated seedlings are still shorter than the control plants, but the difference is not so marked as it was 3 days ago, *i. e.*, 4 days after the exposure.

On June 10 the control seedlings averaged about one fourth taller than those exposed. On June 15 the cultures were photographed (FIGURE 32).

Retardation of germination and growth has followed exposure as described.

This experiment was repeated and similar results were obtained.

EXPERIMENT 44

Object: To ascertain the effect of germinating and growing timothy grass seed in an atmosphere containing the radium emanation.

Seeds of timothy grass (*Phleum pratense*) were planted on the surface of the soil in each of two pots, and each pot was placed under a bell-jar with a tubulure at the top and bottom. By means of a blast bulb the emanation from $RaBr_2$ was forced into the bell-jar at irregular intervals of from 2 to 24 hours from a cylinder lined with a "Lieber's radium-coating." Blasts of ordinary air were similarly forced through the control jar.

The emanation was delivered at a height of about 90 mm. above the seeds, and the periodic blasts were continued for ten days.

On the tenth day the exposed plants were slightly but definitely taller, on the average, than the unexposed plants, and on the eleventh day they were still taller than the control. On the twentieth day after planting the same difference in height was maintained and the experiment was discontinued.

EXPERIMENT 45

Object: To ascertain the effect of growing germinated seeds of *Lupinus albus* in an atmosphere containing the emanation of radium.

April 29, 12 M.

On the radicles of five germinated seeds of *Lupinus albus*, germinated in moist sphagnum until the radicles were over 10 mm. long, were placed reference marks in India ink, 10 mm. from the root-tip. The seedlings were suspended on glass spits under a bell-jar into which the radium emanation was drawn, as described in Experiment 44.

Five germinated seedlings were similarly arranged in a control jar.

Measurements of the growth in length of radicles were made as follows:

April 30, 11:30 A. M.

	Radium	*Control*
1	18.50 mm.	13.00 mm.
2	17.00	10.00
3	19.50	14.50
4	21.50	10.50
5	18.50	12.50
	95.00 mm.	60.50 mm.
	19.00 mm.	12.10 mm.

May 1, 9 A. M.

	Radium	*Control*
1	22.00 mm.	13.00 mm.
2	21.50	10.00
3	24.00	17.00
4	27.00	11.00
5	22.00	12.50
	116.50 mm.	63.50 mm.
	23.30 mm.	12.70 mm.

Exposure to the emanation has been followed by a decided acceleration in growth.

May 1, 12 M.

A repetition of the above experiment was started, using six seeds of *Lupinus albus* in each culture.

May 2, 12 M.

Measurements of growth were recorded as follows:

	Radium	*Control*
1	13.50 mm.	10.00 mm.
2	14.50	10.00
3	11.50	10.00
4	17.50	10.00
5	19.00	10.00
6	19.00	10.00
	95.00 mm.	60.00 mm.
	15.83 mm.	10.00 mm.

EXPERIMENT 46

Repetition of Experiment 45.

The experiment was arranged as described in Experiment 45. Measurements of growth in length were made as follows:

May 6, 10 A. M. (After 12 hours' exposure.)

	Radium	*Control*
1	19.00 mm.	13.00 mm.
2	16.00	13.00
3	16.00	16.50
4	20.00	17.50
5	22.50	17.00
6	17.00	15.00
	110.50 mm.	92.00 mm.
	18.41 mm.	15.33 mm.

May 7, 11 A. M.

	Radium	*Control*
1	24.50 mm.	14.50 mm.
2	16.00	14.50
3	23.00	16.00
4	25.50	32.00
5	28.00	injured
6	17.00	injured
	134.00 mm.	77.00 mm.
	22.33 mm.	19.25 mm.

Growth in the atmosphere containing the emanation is more rapid than the normal growth.

In a fourth repetition of this experiment the 5 exposed seedlings averaged 9.60 mm. growth in 20 hours, while the average growth of the control seedlings was only 7.30 mm. Again the average amount of growth in 24 hours was, for those exposed, 18.41 mm.; for the control, 15.80 mm.

In a fourth repetition of Experiment 45, the following measurements were made:

May 11, 5:30 P. M. (After 23 hours' exposure.)

	Radium	*Control*
1	20.00 mm.	14.00 mm.
2	25.00	17.00
3	25.00	14.00
4	21.00	12.00
5	25.00	17.00
6	20.00	15.00
	136.00 mm.	89.00 mm.
	22.67 mm.	14.83 mm.

May 12, 5:30 P. M.

	Radium	*Control*
1	26.00 mm.	14.00 mm.
2	32.00	20.00
3	32.00	15.00
4	25.00	13.00
5	31.00	19.00
6	25.00	15.50
	171.00 mm.	96.50 mm.
	28.50 mm.	16.08 mm.

The curves of growth given in FIGURE 33 are typical of the results obtained in all five experiments, which clearly indicate that the

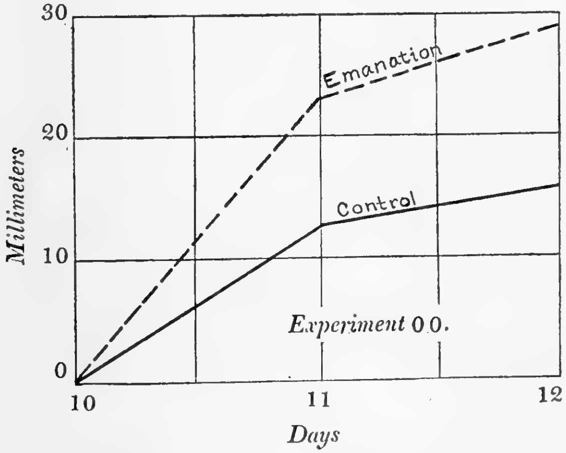

FIG. 33. Acceleration of Growth of Roots of *Lupinus albus* in a Radioactive Atmosphere.

growth of roots in an atmosphere containing the decaying radium emanation is more rapid than normally.

Experiment 47

In order to see if the emanation given off from about 10 mg. of $RaBr_2$ of 7,000 activity, contained in a glass tube open at one end, would affect germination and growth, timothy seed was sown on the surface of the soil in two pots, and each pot was placed under a bell-jar. One bell-jar contained the open tube of radium bromide throughout the experiment, the other served as a control. The radium tube had been open under the first bell-jar for 48 hrs. before the seeds were introduced.

Observations were continued for nine days, but with negative results. The seeds germinated and grew in both pots, but no significant difference was detected between the two cultures.

Summary

In harmony with results previously obtained under other conditions, it is seen that the effect of exposure to the radioactive emanation varies with the conditions of exposure. As in Experiment 32, when a layer of moist soil intervened between the radium and the seeds, the rays produced an acceleration of growth, so here, when unsoaked (and hence less sensitive) timothy seeds were planted beneath the soil surface, and the emanation delivered at a distance of 20 mm. above the soil, growth was accelerated. Under similar conditions of planting, but with the emanation delivered at a distance of only 5 mm. above the soil, growth was retarded. Again when the distance was 50 mm., but the timothy sown on the surface of the soil, retardation resulted. The same effect followed when the distance was increased to 190 mm. over timothy seeds sown on moist blotter.

If, now, the seeds are sown on the surface of the soil, and the emanation delivered, not continuously, as before, but by means of blasts at irregular intervals of from two to twenty-four hours, germination and growth are increased. The growth in length of radicles of *Lupinus albus* was uniformly accelerated in an atmosphere containing the emanation (FIGURE 33). This result was repeatedly verified and is additional evidence of the greater sensitiveness of tissues containing chlorophyll. An exposure which proved to be an over stimulus for the chlorophyll-bearing shoot system, causing a retardation of growth, approximated the optimum stimulation for the chlorophylless root, causing acceleration of growth.

No appreciable effect resulted from exposing germinating timothy seeds in a closed bell-jar to the emanation that diffused from an open glass tube of about 10 mg. of radium bromide of 7,000 activity.

These experiments with the radioactive atmosphere all point to the same general truth, namely, that *the rays of radium act as a stimulus to protoplasm. Retardation of growth following exposure to the rays is an expression of over-stimulation; acceleration of growth indicates stimulation between a minimum and an optimum point.*

CHAPTER X

EFFECTS ON PLANT GROWTH OF EXPOSED WATER AND FRESHLY FALLEN RAIN

1. Effects of Tap-Water Exposed to Radium Rays

Because of the fact, now so well known, that penetrating, or gamma-like radiations are present in probably all soil, the following experiments were made for the purpose of ascertaining the effect on germination and growth of water exposed to the penetrating rays of radium, for it is evident that the water in the soil is naturally thus exposed.

Experiment 48

Object: To ascertain the effect on the germination and growth of seeds of imbibition of water in which sealed glass tubes of radium bromide have been immersed.

April 9, 2 P. M.

Three beakers, *a*, *b*, and *c*, were arranged two thirds full of water, and containing (*a*) the tube of radium of 1,500,000 activity; (*b*) the tube of 10,000 activity; (*c*) no tube, and serving as the control.

April 10, 5:30 P. M.

After the water had been exposed to the radium rays for 26.5 hrs., seeds were put to soak in each of the beakers as follows:

12 seeds of lupine (*Lupinus albus*).
15 grains of oats (*Avena:* Henderson's "Lincoln").
8 grains of corn (*Zea Mays:* "Hickory King").

April 11, 5 P. M.

After soaking for 23.5 hrs. the seeds were all planted in soil in pots.

The records of observations of the different seeds are given separately, as follows:

Corn (*Zea Mays*)

On April 14 none of the corn grains had germinated, but on April 16 four seedlings were up in the 1,500,000 culture, and five in each of the other two. On April 17 measurements of the heights of the seedlings were recorded as follows:

No.	$RaBr_2$ 1,500,000 ×	$RaBr_2$ 10,000 ×	Control
1	29.50 mm.	18.00 mm.	16.50 mm.
2	23.00	12.50	25.00
3	25.00	18.50	4.50
4	not up	23.00	15.50
5	25.00	not up	10.50
6	16.00	13.00	7.50
7	not up	13.00	5.00
8	12.00	20.00	15.50
	130.50 mm.	118.00 mm.	100.00 mm.
	21.75 mm.	16.86 mm.	12.50 mm.

The relative heights of the plants in the three cultures remained as above until April 24, when observations were discontinued.

Lupine (*Lupinus albus*)

On April 14, none of the control plants were up, but in the other two cultures some of the seedlings were just breaking the surface of the soil. On April 16 seedlings were up in all of the pots, but there was no appreciable difference in their height or in any other character.

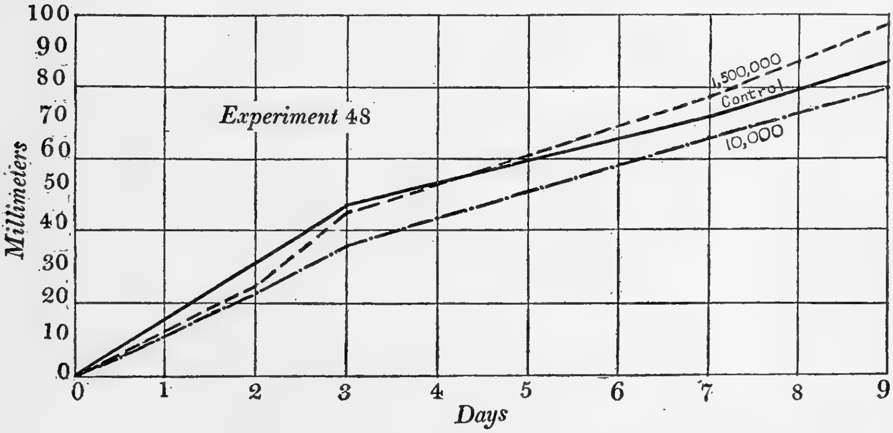

FIG. 34. Effect on the Germination and Growth of Oat of soaking the Grains in Water exposed for 26.5 Hours. to Rays from Radium of different degrees of activity.

On April 23 the average height of the seedlings of the 1,500,000 culture was 26.10 mm., of the 10,000 culture 25.38 mm., and of the control culture 25.00 mm. Thus, in the case of the lupines, no appreciable effect resulted from the treatment.

Oat (*Avena*)

On April 14 five seedlings of the 1,500,000 culture were just up, six seedlings of the 10,000 culture, and eight seedlings of the control.

April 16.

The following measurements of height were recorded:

	$RaBr_2$	$RaBr_2$	
No.	1,500,000 ×	10,000 ×	Control
1	30.00 mm.	35.50 mm.	16.00 mm.
2	15.00	35.50	27.50
3	23.50	32.50	not up
4	23.50	5.50	37.00
5	10.00	33.00	27.50
6	31.00	27.00	not up
7	38.00	15.00	23.50
8	34.50	23.00	34.50
9	27.00	22.50	40.00
10	35.00	17.50	36.00
11	28.50	6.50	35.50
12	27.00	32.00	25.00
13	not up	17.00	37.00
14	28.00	not up	not up
	351.00 mm.	302.50 mm.	339.50 mm.
	27.00 mm.	23.26 mm.	30.87 mm.

On April 17 the averages were, for the 1,500,000 culture, 45.34 mm.; for the 10,000 culture 35.80 mm.; and for the control 47.41 mm. On April 21, the average heights were, respectively, 77.10 mm.; 66.20 mm.; and 71.37 mm., on April 23, 97.60 mm.; 80.30 mm.; and 87.80 mm., and on June 4, 311.70 mm.; 290.70 mm.; and 283.88 mm. Thus the initial slight retardation is replaced by an acceleration. Experiment closed.

In the bean culture, from a comparison of the measurements, it is seen that soaking the seeds in the water exposed to the radium rays was followed by a much less rapid growth than the normal, but the difference between the 10,000 × and the 1,500,000 × cultures is negligible. At the end of six weeks there was no apparent difference in the height and vigor of the plants in the three cultures.

In the case of the lupines no appreciable effect was produced on germination by soaking in the treated water, but subsequent growth was slightly accelerated.

With the corn, acceleration of growth followed the soaking in the exposed water, but the rate of germination was apparently, in the early stages at least, not affected.

In the case of the oats, soaking in the treated water was followed, for two days after the seeds came up, by a retardation of germination and of growth. Then the rate of growth of the exposed plants increased. Acceleration was most rapid in the case of the plants soaked in the water exposed to the stronger radium, and these plants were taller than the control at the end of the fifth day after they came up. At the end of 51 days this relatively greater height was still marked, and the plants soaked in the water exposed to the radium of 10,000 activity were also taller than the control.

Part of the results of this experiment are shown graphically in FIGURE 34.

Experiment 49

The following experiment shows the effect on the germination and growth of corn grains (*Zea Mays*), bean seeds (*Phaseolus*), and oats (*Avena*), of soaking, before planting, in water in which sealed glass tubes of radium bromide had been immersed.

April 23, 4 : 30 P. M.

Into each of three beakers, *a*, *b*, and *c*, was placed 100 c.c. of water, and into the water of each beaker was suspended a sealed glass tube of $RaBr_2$, as follows:

Into *a* the $RaBr_2$ of 1,800,000 ×.
Into *b* the $RaBr_2$ of 1,500,000 ×.
Into *c* the $RaBr_2$ of 10,000 ×.

April 24, 4 : 30 P. M.

After 24 hrs. exposure to the rays of the radium, the radium-tubes were removed from the water, and 8 seeds each of corn, bean, and oat were placed to soak in each beaker. Also in a control beaker a like number of each kind.

April 26, 8 : 30 A. M.

After soaking for 40 hrs., the seeds were removed from the water and planted in soil in flower-pots. Observations of germination and growth follow.

Bean (*Phaseolus*)

For several reasons the 1,800,000 and 10,000 cultures were discarded. On May 2 in the 1,500,000 culture 4 seeds were partly up, while in the control only 3 seeds were just appearing.

	May 5, 10 A. M.		May 11, 9:30 A. M.	
	RaBr$_2$		RaBr$_2$	
No.	1,500,000 ×	Control	1,500,000 ×	Control
1	50.00 mm.	35.00 mm.	114.00 mm.	85.00 mm.
2	41.00	just up	106.00	82.00
3	75.00	25.00	124.00	100.00
4	7.50	17.00	100.00	128.00
5	21.00	18.50	85.00	107.00
6	20.00	36.00	95.00	108.00
7	45.00	35.00	injured	75.00
8	45.00	8.50	99.00	114.00
	304.50 mm.	175.00 mm.	723.00 mm.	799.00 mm.
	38.06 mm.	25.00 mm.	103.28 mm.	99.88 mm.

Germination and growth have been slightly more rapid in the seeds soaked in the water exposed to the rays of radium.

Corn (*Zea Mays*)

April 30, 9:30 A. M.

In the 1,800,000 culture one seed is just up, in the 1,500,000, six seeds, in the 10,000, eight seeds, and in the control culture four seeds.

May 1, 9:30 A. M.

	RaBr$_2$	RaBr$_2$	RaBr$_2$	
No.	1,800,000 ×	1,500,000 ×	10,000 ×	Control
1	7.50 mm.	13.00 mm.	10.00 mm.	18.00 mm.
2	not up	25.00	15.00	not up
3	15.00	15.00	16.00	23.50
4	11.00	7.00	14.00	not up
5	not up	18.00	15.00	5.00
6	17.50	11.50	10.00	20.00
7	not up	12.00	12.00	18.00
8	not up	15.00	18.00	11.00
	51.00 mm.	116.50 mm.	110.00 mm.	95.50 mm.
	12.75 mm.	14.56 mm.	13.75 mm.	15.91 mm.

May 2, 9:30 A. M.

	$RaBr_2$	$RaBr_2$	$RaBr_2$	
No.	1,800,000 ×	1,500,000 ×	10,000 ×	Control
1	27.00 mm.	35.00 mm.	27.00 mm.	40.00 mm.
2	5.00 *	44.00	36.00	5.00 *
3	39.00	33.00	40.00	47.00
4	31.00	24.00	32.00	5.00 *
5	5.00 *	42.00	37.00	18.00
6	39.00	32.00	33.00	36.50
7	9.00 *	35.00	34.00	38.50
8	not up *	39.00	40.00	32.00
	136.00 mm.	284.00 mm.	279.00 mm.	212.00 mm.
	34.00 mm.	35.50 mm.	34.88 mm.	35.33 mm.

May 3, 9:30 A. M.

	$RaBr_2$	$RaBr_2$	$RaBr_2$	
No.	1,800,000 ×	1,500,000 ×	10,000 ×	Control
1	47.00 mm.	54.50 mm.	44.00 mm.	61.50 mm.
2	20.00 *	64.00	55.50	18.00 *
3	61.00	50.00	60.00	67.00
4	54.00	45.50	53.00	7.00 *
5	17.00 *	62.00	56.00	35.00
6	61.00	52.50	55.50	54.50
7	25.50 *	58.00	53.50	53.00
8	not up *	60.00	57.00	injured *
	223.00 mm.	446.50 mm	434.50 mm.	271.00 mm.
	55.75 mm.	55.81 mm.	54.31 mm.	54.20 mm.

On May 4, the average height of the seedlings in the 1,800,000 culture was 77.00 mm.; of the 1,500,000 culture, 75.68 mm.; of the 10,000 culture 74.81 mm.; and of the control, 69.50 mm. On May 5 the average heights were as follows:

92.75 mm.; 93.81 mm.; 90.87 mm.; 85.28 mm.

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* Discarded and not added in.

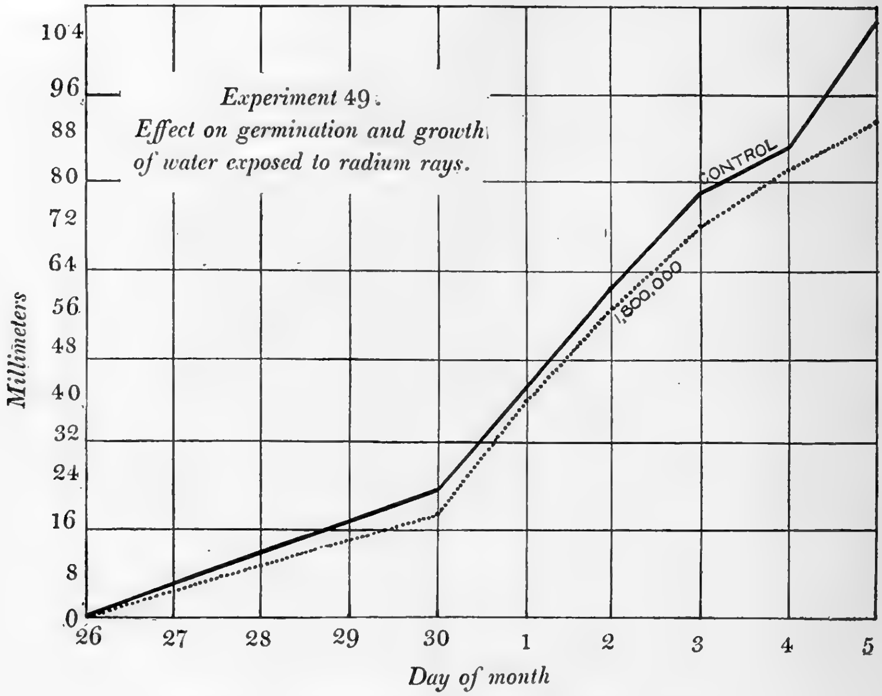

FIG. 35. *Avena sativa.* The Water was exposed to Radium Rays for 24 Hours.

Oat (*Avena*)

The heights of the seedlings were recorded as follows:
April 30, 9:30 A. M.

No.	RaBr$_2$ 1,800,000 ×	RaBr$_2$ 1,500,000 ×	RaBr$_2$ 10,000 ×	Control
1	23.00 mm.	20.00 mm.	26.00 mm.	22.00 mm.
2	17.00	4.00 *	14.00	23.00
3	4.00 *	8.00 *	23.00	25.00
4	19.00	21.00	not up *	24.00
5	20.00	not up *	17.00	18.00
6	13.00	not up *	11.00	25.00
7	18.00	21.00	not up *	22.00
8	20.00	21.00	16.00	not up *
	130.00 mm.	83.00 mm.	107.00 mm.	159.00 mm.
	18.57 mm.	20.75 mm.	17.83 mm.	22.71 mm.

* Discarded.

May 1, 9:30 A. M.

No.	$RaBr_2$ 1,800,000 ×	$RaBr_2$ 1,500,000 ×	$RaBr_2$ 10,000 ×	Control
1	45.50 mm.	35.00 mm.	46.00 mm.	43.50 mm.
2	39.50	47.50	19.50	44.50
3	29.00*	42.50	32.00	46.50
4	38.00	35.00	not up	47.50
5	40.00	not up	44.00	35.00
6	35.00	11.50	49.00	45.00
7	40.50	35.00	not up	42.00
8	41.00	41.50	48.00	not up
	279.50 mm.	248.00 mm.	238.50 mm.	304.00 mm.
	39.93 mm.	35.42 mm.	39.75 mm.	43.43 mm.

The average heights of the seedlings were as follows on the dates indicated:

	$RaBr_2$ 1,800,000 ×	$RaBr_2$ 1,500,000 ×	$RaBr_2$ 10,000 ×	Control
May 2.	56.93 mm.	53.50 mm	59.75 mm.	61.21 mm
May 3.	72.14	70.21	76.66	78.27
May 4.	81.71	80.85	90.25	85.92
May 5.	90.50	98.57	104.50	108.64

See FIGURE 35.

EXPERIMENT 50

Object: To ascertain the effect on the growth in length of the radicles of *Lupinus albus* of soaking in water in which sealed glass tubes of radium bromide have been immersed.

May 5, 12:30 P. M.

Into each of four beakers containing 100 c.c. of tap-water was placed a sealed glass tube of radium bromide, as follows:

A. Activity 1,800,000.
B. Activity 1,500,000.
C. Activity 10,000.
D. Control.

After the water had been exposed for 24 hours to the radium rays, the hypocotyls of germinated seeds of lupine were suspended

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* Discarded.

in it up to an ink mark, placed 10 mm. from the root-tip. Observations of the length of the hypocotyls were recorded as follows:

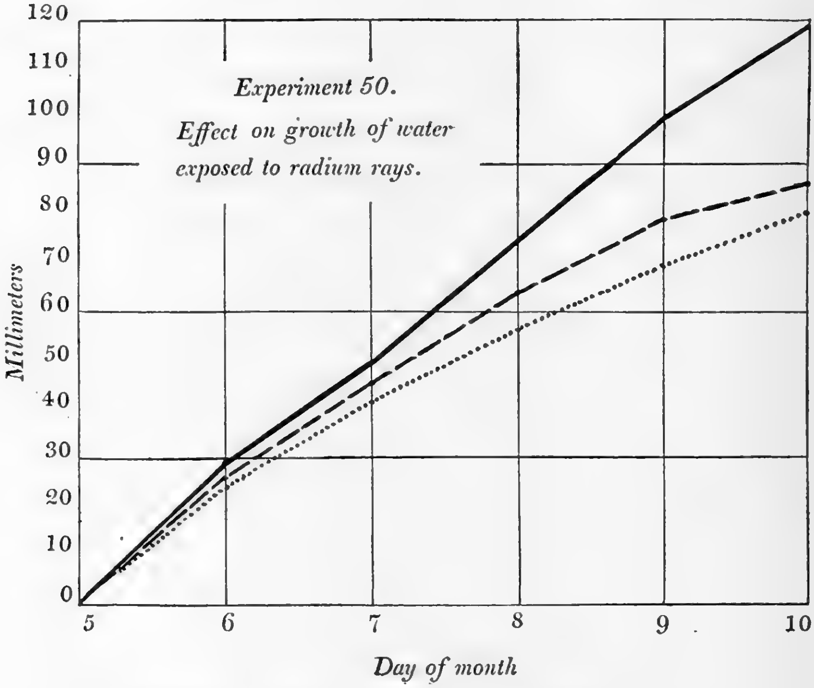

FIG. 36. *Lupinus albus.* Duration of exposure of the Water, 24 Hours. ——, Control; — — —, 1,500,000 ×; ......, 1,800,000 ×.

May 6, 10 A. M.

A. RaBr$_2$ 1,800,000 ×	B. RaBr$_2$ 1,500,000 ×	C. RaBr$_2$ 10,000 ×	D Control
24.00 mm.	27.00 mm.	29.00 mm.	28.00 mm.
24.00	26.00	27.00	29.50
24.50	27.00	28.00	27.00
26.00	24.00	24.00	27.50
98.50 mm.	104.00 mm.	108.00 mm.	112.00 mm.
24.62 mm.	26.00 mm.	27.00 mm.	28.00 mm.

May 7, 11 A. M.

A. RaBr$_2$	B. RaBr$_2$	C. RaBr$_2$	D
1,800,000 ×	1,500,000 ×	10,000 ×	Control
40.00 mm.	47.50 mm.	49.00 mm.	44.00 mm.
42.00	44.00	46.00	52.00
43.50	46.00	48.00	51.50
39.00	42.00	36.00	47.50
164.50 mm.	179.50 mm.	179.00 mm.	195.00 mm.
41.12 mm.	44.87 mm.	44.75 mm.	48.75 mm.

May 8, 11:30 A. M.

A. RaBr$_2$	B. RaBr$_2$	C. RaBr$_2$	D
1,800,000 ×	1,500,000 ×	10,000 ×	Control
54.00 mm.	68.00 mm.	69.00 mm.	63.50 mm.
55.50	64.00	64.50	82.00
59.00	62.00	71.00	76.00
54.00	57.00	41.00	76.00
222.50 mm.	251.00 mm.	245.50 mm.	297.50 mm.
55.62 mm.	62.75 mm.	61.37 mm.	74.37 mm.

May 9, 3 P. M.

A. RaBr$_2$	B. RaBr$_2$	C. RaBr$_2$	D
1,800,000 ×	1,500,000 ×	10,000 ×	Control
69.50 mm.	87.00 mm.	88.50 mm.	83.00 mm.
69.50	78.00	81.00	109.00
71.50	74.50	91.00	101.00
66.00	71.00	53.50	101.50
276.50 mm.	310.50 mm.	314.00 mm.	394.50 mm.
69.12 mm.	77.62 mm.	78.50 mm.	98.62 mm.

May 10, 11 A. M.

A. RaBr$_2$	B. RaBr$_2$	C. RaBr$_2$	D
1,800,000 ×	1,500,000 ×	10,000 ×	Control
82.00 mm.	99.00 mm.	102.50 mm.	99.00 mm.
79.50	81.50	95.00	129.00
81.00	81.50	106.00	120.00
76.00	80.00	56.00	123.00
318.50 mm.	342.00 mm.	359.50 mm.	471.00 mm.
79.62 mm.	85.50 mm.	89.87 mm.	117.75 mm.

The less rapid growth in the water in which the radium-tubes were immersed is shown graphically in another figure. The curve for the results of exposure to the radium of 10,000 activity is omitted for the sake of clearness.

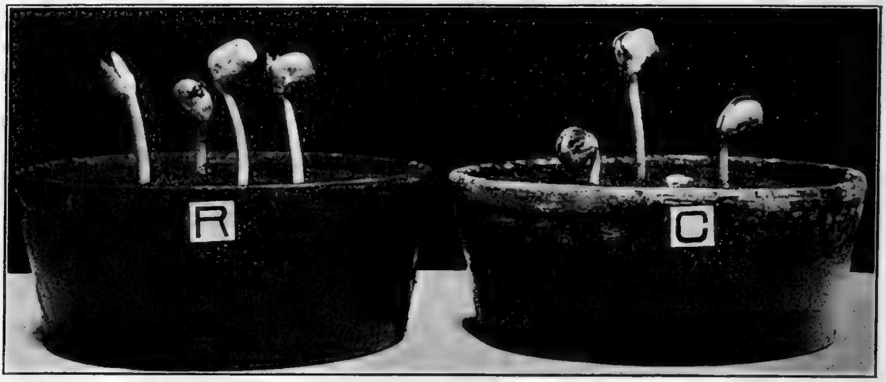

FIG. 37. Experiment 51. Acceleration of Growth of *Lupinus albus* by watering, after planting in Soil, with Water exposed to Radium Rays. Cf. FIGURE 38.

EXPERIMENT 51

Object: To ascertain the effect on germination and growth of watering with water in which a sealed glass tube of radium bromide has been immersed.

June 29, 9:30 A. M.

Into each of two pots of soil, thoroughly moist with tap-water, were planted four seeds of *Lupinus albus*, and into each of two other similar pots four seeds of *Zea Mays*.

June 30, 9:30 A. M.

One pot of each set was watered with 100 c.c. of tap-water, in which a glass tube containing 10 mg. of $RaBr_2$ of 1,800,000 activity had been immersed for 8 days. The other pot in each set was watered with a like amount of ordinary, unexposed tap-water.

This method of watering was continued daily until July 25, the radium tube being in the water from 1 to 9 days before each watering.

Observations of germination and growth were recorded as follows:

FIG. 38. Experiment 51. Retardation of Germination and Growth of *Zea Mays* by watering, after planting in Soil, with Water exposed to Radium Rays. Cf. FIGURE 37.

July 2, 10:30 A. M.

Zea Mays		*Lupinus*	
Radium	Control	Radium	Control
not up	15.00 mm.	just up	just up
not up	28.00	" "	" "
10 mm.	23.00	" "	" "
not up	22.00	" "	" "
	88.00 mm.		
	22.00 mm.		

July 3, 10:30 A. M.

Zea Mays		*Lupinus*	
Radium	Control	Radium	Control
3.00 mm.	45.00 mm.	16.00 mm.	10.00 mm.
0.00	66.00	10.00	2.00
32.00	73.00	7.00	7.00
0.00	60.00	15.00	17.00
35.00 mm.	244.00 mm.	48.00 mm.	36.00 mm.
17.50 mm.	61.00 mm.	12.00 mm.	9.00 mm.

July 4, 10 A. M.

Zea Mays		*Lupinus*	
Radium	Control	Radium	Control
18.00 mm.	102.00 mm.	35.00 mm.	15.00 mm.
20.00	107.00	33.00	5.00
60.00	116.00	20.00	18.00
just appearing	102.00	32.00	37.00
98.00 mm.	427.00 mm.	120.00 mm.	75.00 mm.
32.66 mm.	106.75 mm.	30.00 mm.	18.75 mm.

July 5, 11 A. M.

Zea Mays		*Lupinus*	
Radium	Control	Radium	Control
50.00 mm.	148.00 mm.	55.00 mm.	20.00 mm.
55.00	152.00	53.00	18.00
65.00	175.00	33.00	35.00
10.00	157.00	50.00	56.00
180.00 mm.	632.00 mm.	191.00 mm.	129.00 mm.
45.00 mm.	158.00 mm	47.75 mm.	32.25 mm.

July 6, 10:30 A. M.

Zea Mays		*Lupinus*	
Radium	Control	Radium	Control
72.00 mm.	179.00 mm.	63.00 mm.	27.00 mm.
82.00	172.00	60.00	22.00
65.00	199.00	45.00	46.00
30.00	185.00	60.00	65.00
249.00 mm.	735.00 mm.	228.00 mm.	160.00 mm.
62.25 mm.	183.75 mm.	57.00 mm.	40.00 mm.

The plants were kept growing and watered as described above until July 25, when the relative differences in height were the same as on July 6, and the experiment was closed. On July 4 the cultures were photographed (FIGURES 37 and 38; also FIGURES 39 and 40).

On July 8 one plant was removed from each culture, dried, and tested with the electroscope to see if the dry substance of the tissues was radioactive. Neither plant was radioactive. This was the result to be expected under the conditions of the experiment.

EXPERIMENT 51*a*

In order to ascertain the effect on the growth of roots in water of placing a sealed glass tube of radium bromide in the water, two glass beakers were partly filled with tap-water. Into the water of each

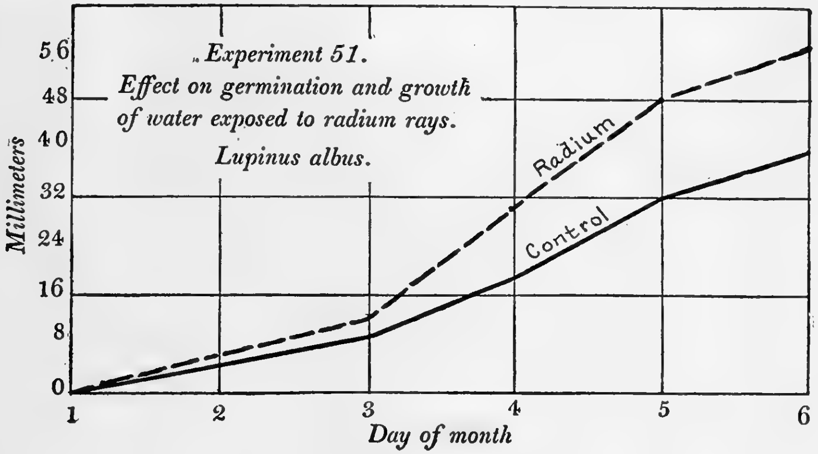

FIG. 39.

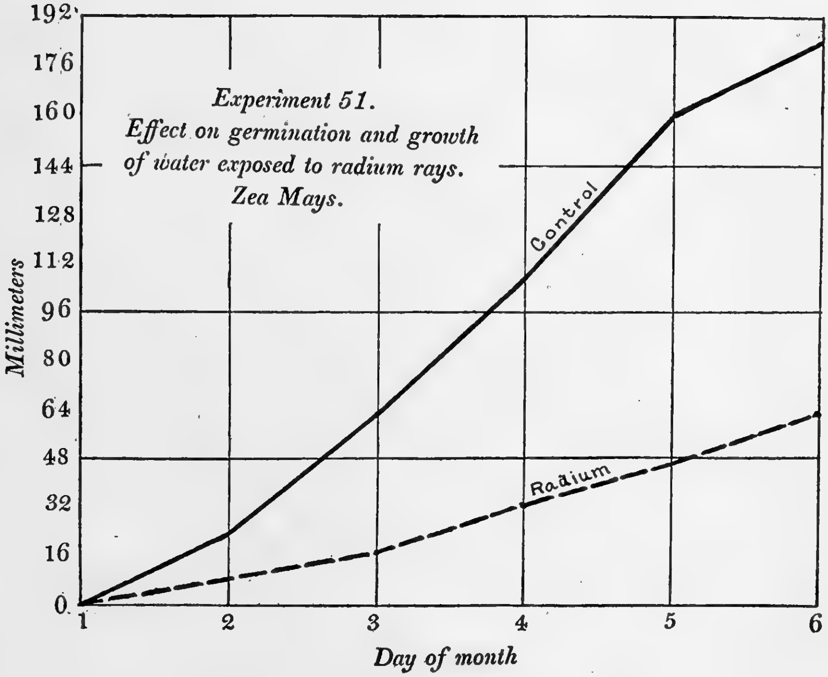

FIG. 40.

beaker were inserted the radicles of four germinated seeds of *Lupinus albus* to a uniform distance of 10 mm. from the root tip. The radicles were arranged in a circle, in the center of which was suspended the sealed tube, containing 52 mg. of radium bromide of 10,000 activity. The distance from the tube to the radicles was about 10 mm., and the radium salt was opposite the zone of maximum growth. The experiment was set up at 8:35 A. M., June 19. Measurements of the amount of growth in length of the radicles were recorded as follows:

June 19, 5:30 P. M.

	Radium		Control
1	18 mm.	1	16 mm.
2	16	2	20
3	15	3	18
4	17	4	18
Total,	66 mm.	Total,	72 mm.
Average,	16.50 mm.	Average,	18.00 mm.

June 20, 8 A. M.

	Radium		Control
1	38 mm.	1	35 mm.
2	34	2	37
3	32	3	37
4	32	4	39
Total,	136 mm.	Total,	148 mm.
Average,	34.00 mm.	Average,	37.00 mm.

June 20, 6 P. M.

	Radium		Control
1	48 mm.	1	50 mm.
2	49	2	57
3	46	3	51
4	45	4	51
Total,	188 mm.	Total,	209 mm.
Average,	47.00 mm.	Average,	52.25 mm.

The results are shown plotted in FIGURE 41.

2. The Radioactive Influence of Freshly Fallen Rain-Water

The fact that freshly fallen rain-water is radioactive suggested the following experiments to ascertain its effect on growth.

Experiment 52

May 28, 6 P. M.

In a glass beaker, washed chemically clean, was caught rain-water. The beaker was set in an open place to avoid drippings from buildings and trees. It had been raining almost constantly during the preceding day (2.26 in. precipitation), and slightly all the morning of the twenty-eighth. Thus the atmosphere was thoroughly washed, and the probability of traces of ammonia and CO_2 and any atmospheric dust in the rain-water was slight. No electrical disturbance had accompanied the rain.

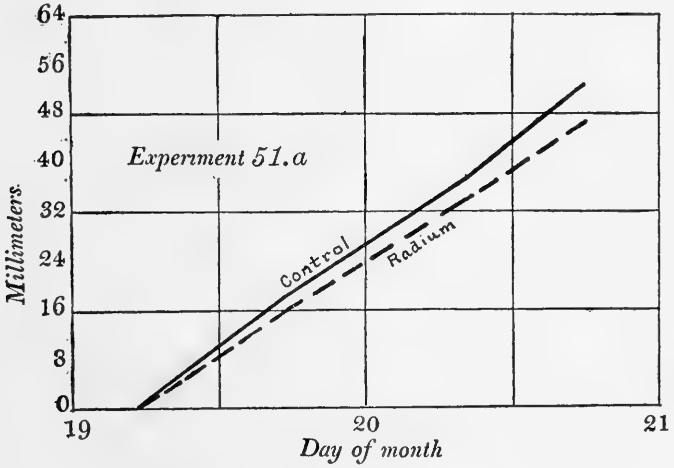

FIG. 41. Effect on Growth of placing a sealed glass Tube of Radium of 10,000 activity into Water in which Roots of *Lupinus albus* are growing.

Into the water thus collected were immersed, to a measured length of 15 mm., the tap roots of four germinated seeds of *Lupinus albus*. Since rain-water is practically distilled, a control was similarly arranged with distilled water, also in a chemically clean beaker.

The following observations were recorded of the amount of elongation of the roots:

May 29, 12 M. (Eighteen hours' growth.)

Rain	*Distilled*
10.00 mm.	15.00 mm.
11.00	10.50
(7.00) *	14.00
11.00	12.00
32.00 mm.	51.50 mm.
10.67 mm.	12.88 mm.

The difference of 2.22 mm. in favor of the roots grown in the distilled water may have been lessened by the possible toxicity of the latter, for it was not prepared in a glass still. Error from this cause was eliminated in the next Experiment (No. 53). The figures for the second day are omitted. (See FIGURE 42.)

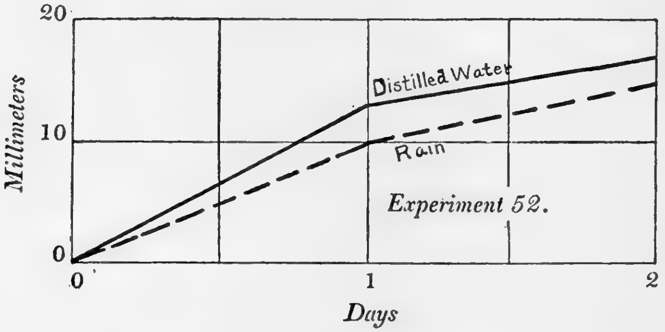

FIG. 42. Effect of freshly fallen Rain on the growth of Roots of *Lupinus albus*.

EXPERIMENT 53

In this experiment, rain-water was caught in chemically clean glass dishes in the open, on April 8, after about four hours of rain, and again on May 7, after over three hours' precipitation. The experiment was set up immediately after the last collection, using radicles of *Lupinus albus*, immersed in both the fresh and the stale rain-water to a depth of 5 mm. Two parallel cultures, A and B, in both the fresh and the stale rain-water were observed. The measured lengths of the radicles, in millimeters, are given in the following tables:

* Discarded.

May 7, 2:30 P. M.

A		B	
Fresh	*Stale*	*Fresh*	*Stale*
5.00 mm.	5.00 mm.	5.00 mm.	5.00 mm.

May 8, 9:30 A. M.

Fresh	*Stale*	*Fresh*	*Stale*
13.00 mm.	14.00 mm.	12.00 mm.	11.50 mm.
12.00	14.00	11.50	13.00
12.00	13.00	13.00	13.00
12.50	13.00	8.00	14.00
49.50 mm.	54.00 mm.	44.50 mm.	51.50 mm.
Av. 12.38 mm.	13.50 mm.	Av. 11.13 mm.	12.88 mm.

May 8, 5 P. M.

Fresh	*Stale*	*Fresh*	*Stale*
15.00 mm.	18.00 mm.	15.50 mm.	17.00 mm.
14.00	17.50	14.50	16.00
15.00	16.50	17.00	15.00
15.00	17.00	10.00	18.00
59.00 mm.	69.00 mm.	57.00 mm.	66.00 mm.
Av. 14.75 mm.	17.25 mm.	Av. 14.25 mm.	16.50 mm.

May 9, 9:30 A. M.

Fresh	*Stale*	*Fresh*	*Stale*
24.00 mm.	29.00 mm.	24.00 mm.	27.00 mm.
23.50	27.00	24.00	27.00
injured	25.00	24.50	25.00
23.00	29.00	17.00	29.00
70.50 mm.	110.00 mm.	89.50 mm.	108.00 mm.
Av. 23.50 mm.	27.50 mm.	Av. 22.38 mm.	27.00 mm.

Both sets of cultures, A and B, give the same kind of result, viz., a slower growth in the fresh rain-water than in that one month old.

The results are plotted in FIGURES 43 and 44, and are in substantial harmony with those obtained in the two preceding experiments. Care was taken in this experiment to have the temperature of the stale and of the freshly fallen rain-water alike by placing the carefully covered dish containing the former out of doors by the side of the latter while the fresh water was being collected. The fresh and the stale water were then kept side by side throughout the entire experiment.

The only known difference between the water in the cultures A and B is a difference of radioactivity, that of the water collected one month previous to the experiment probably being nearly or quite zero. It therefore seems a conclusion warranted by the conditions of the experiment that freshly fallen rain-water tends to retard the growth of roots of *Lupinus albus*, and that this effect is due to the radioactivity of the water.

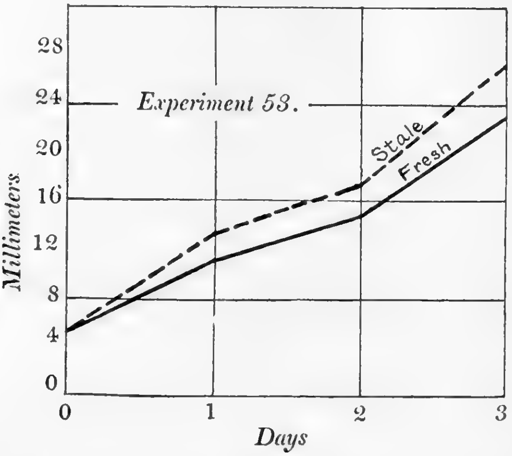

FIG. 43. Relative growth of Roots of *Lupinus albus* in fresh Rain-Water and in Rain-Water one month old. Culture B, Exp. 53. Cf. FIGURE. 44.

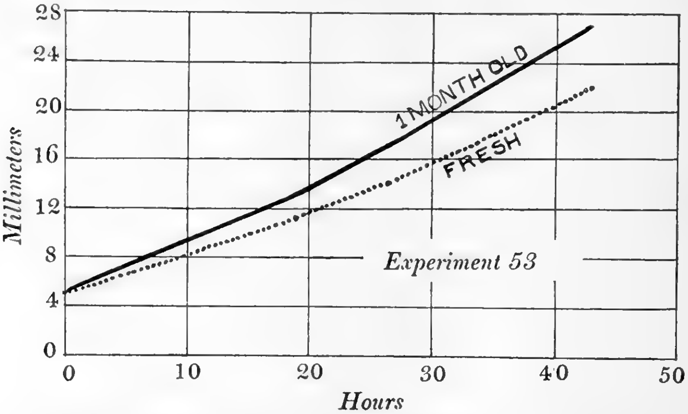

FIG. 44. Relative growth of Roots of *Lupinus albus* in fresh Rain-Water and in Rain-Water one month old. Culture A, Exp. 53. Cf. FIGURE 43.

In view of the experimental results previously obtained, and indicating that chlorophyll-bearing tissues respond to radium rays differently from tissues without chlorophyll, it is obvious that no conclusions may be drawn from this experiment as to the effect of the radioactivity of freshly fallen rain-water on green leaves and stems. Quite possibly these parts may be thus stimulated.

Experiment 54

The object of this experiment is to ascertain the effect on germination and growth of soaking corn grains (*Zea Mays*) in freshly fallen rain-water.

May 26, 6 P. M.

Five corn grains were placed in an empty, chemically clean beaker, left out in the open all night while it was constantly raining. There had been an almost continuous precipitation during the preceding 36 hrs. As a control, the same number of grains were placed in a covered, chemically clean beaker, in distilled water, placed near the other beaker to secure similar temperature conditions.

May 30, 11 : 30 A. M.

After the grains had soaked for 41 hrs., they were placed in soil moistened with ordinary tap-water. The average height of the seedlings was recorded as follows: (Two of the control seeds proved to be poor, so the growth recorded for the control is the average of only three seeds.)

	Rain	*Distilled*
June 4.	25.20 mm.	23.30 mm.
June 6.	79.60 mm.	86.60 mm.
June 7.	136.60 mm.	142.30 mm.
June 8.	161.60 mm.	179.00 mm.
June 9.	190.60 mm.	207.60 mm.

Up to 10 days after planting the exposed seeds grew less rapidly than the control, and on the tenth day they averaged 17 mm. shorter than those not exposed, but from this time on, though the conditions in the two cultures were maintained as nearly identical as possible as regards light, temperature, and moisture, and though the seeds of the two sets were of uniform size, the plants from seeds soaked in the rain-water grew much more rapidly, and, on June 18, the relative heights of the plants in the two cultures were as represented in the photograph (FIGURE 45).

FIG. 45. Experiment 54. Increased Growth of *Zea Mays,* following a ten-day Retardation, after the Seeds (before planting) were soaked in freshly fallen Rain Water.

SUMMARY

The experiments show that when corn grains were soaked for 24 hrs. in water exposed for 26.5 hrs. to radium rays growth was accelerated. The water exposed to the stronger radium caused the greater acceleration. With seeds of *Lupinus albus* similarly treated, the effects were very slight, but the same in kind as with corn. Oats seemed to be slightly retarded at first, but four or five days after the planting the oats soaked in the exposed water were much taller than the control plants, and tallest after soaking in the water exposed to the strongest radium.

In a similar experiment, after 40 hrs. of soaking in water previously exposed for 24 hrs., oats were accelerated from the beginning, and corn slightly. In this experiment the soaking of bean seeds in irradiated water was followed by decided acceleration at first, but the effect was not lasting, for the control plants eventually attained the same height as those soaked in the exposed water.

After seeds of *Lupinus albus* had germinated, further growth in length of the radicles was retarded in water previously exposed for 24 hrs. to radium rays of activities of from 10,000 to 1,800,000.

When dry seeds of corn and lupine were planted in soil, and watered with water exposed for from one to nine days to rays from radium of 1,800,000 ×, the growth of the corn was less and that of the lupines greater than of plants watered with fresh tap-water, but otherwise similarly treated.

The rate of growth of roots of *Lupinus albus* is less in freshly fallen rain-water than in rain-water one month old or in artificially distilled water. This effect is similar to that produced by placing a sealed glass tube of radium into water in which roots are growing (FIGURE 41), and the retardation is attributed, either directly or indirectly, to the radioactivity of the fresh rain.

CHAPTER XI

EFFECTS ON PLANT GROWTH: MISCELLANEOUS EXPERIMENTS

It is clearly recognized that in the preceding experiments a rather narrow range of plant material was employed. In the following miscellaneous experiments a greater variety of material was used.

Experiment 55

In order to test the effect of the rays of radium on the germination of pollen grains, ten pollinia of *Asclepias curassavica* were placed to germinate in a moist chamber on slices of beet, at varying distances from a sealed glass tube of $RaBr_2$ of 7,000 activity. Control of ten pollinia, similarly arranged, but with no radium.

Within three hours the three pollinia nearest the radium tube had begun to germinate, but there were no signs of germination in the control set, and the experiment was closed.

Experiment 56

March 10, 10:30 A. M.

In order to test the effect of radium rays on the form and rate of growth of yeast (*Saccharomyces*), a small portion of a Fleischmann's compressed yeast cake was mixed in a 5 per cent. solution of sucrose. The liquid was then divided, and one half placed in a small glass dish, the other half in a similar dish. In the former was placed the radium tube of 10,000 activity, supported vertically by a cork covering the dish, and with the end holding the radium extending to the bottom. In the other dish was similarly placed an empty glass tube.

March 11, 3:30 P. M.

There is no apparent difference in the microscopic appearance of the yeast contained in the drops that adhered to the tubes as they were withdrawn from the liquid in the two dishes.

March 12, 11:15 A. M.

The yeast cells seem to be rather more numerous in the radium culture, both to naked eye view, and as seen under the microscope, but the difference is slight. There is no significant difference in the form or size of the cells.

March 16, 11 A. M.

The odor of the radium culture is decidedly stronger than that of the control, and microscopic examination shows the cells to be more numerous in the former.

Exposure of the radium rays was followed by a slightly more rapid cell-division, and possibly by more rapid fermentation. On this last point see Chapter XIV.

EXPERIMENT 57

March 26, 12 M.

In order to ascertain the effect of radium rays on the growth of mushroom spawn (*Agaricus*), a half-pound cake of commercial mushroom spawn was divided into halves. Into the first half was placed two tubes of radium bromide of 1,500,000 activity, 15 mg. in all. The second half was used as a control.

April 3, 9 : 30 A. M.

After an exposure of 190 hrs., both portions of the spawn were planted in separate mushroom beds, in boxes, in the propagating house.

June 27.

The spawn in the control bed is fruiting, but there are no signs of growth in the radiated culture. The experiment was continued for several weeks, but the spawn exposed to the rays of radium never fruited.

EXPERIMENT 58

April 1, 12 M.

In order to see what effect, if any, the rays of radium would have on *Spirogyra*, a bit of the alga, freshly gathered from a stream, was placed in two small vials with some of the stream water. In one vial was suspended the glass tube containing 10 mg. of $RaBr_2$, 1,500,000 ×, in contact with part of the algal threads. In the control vial an empty glass tube was similarly placed.

April 1, 3 : 30 P. M.

No difference is discernible in the naked eye appearance or in the microscopic appearance of the threads or cells.

April 3, 8 A. M.

There is no significant difference in the microscopic appearance of the cells in the two cultures.

The experiment was repeated with similar results.

EXPERIMENT 59

Object: To ascertain the effect of the rays from radium on the growth of the gemmae of *Lunularia*.

December 5, 2 P. M.

A plant of *Lunularia* was placed under a bell-jar in the dark-room. Over the cupule, full of gemmae, was placed the sealed glass tube containing 10 mg. of $RaBr_2$ of 1,500,000 activity, vertically, with the end containing the radium at the height of the rim of the cupule.

A control plant was similarly arranged, but with an empty glass tube.

December 8, 9 A. M.

After an exposure of 67 hours, gemmae from the radiated cupule were sown on the surface of moist sterilized soil, and a control sowing was made from a plant not exposed. The cultures were placed in the propagating house to develop.

January 11.

The radiated gemmae have never developed, while those not exposed developed thalli of from 4 to 5 sq. mm. in area, and the experiment was then discontinued, owing to an accident to the young plants.

EXPERIMENT 60

Repetition of Experiment 59.

January 17, 11 A. M.

Gemmae of *Lunularia* exposed for 48 hours to the rays from $RaBr_2$ of 1,500,000 activity, were planted on the surface of soil in a pot, and non-exposed gemmae were similarly sown in another pot.

April 2.

Both the radiated and control gemmae have developed thalli, but those of the exposed culture are only about one half as large as those of the control. The plants from the radiated gemmae began to develop cupules about a month ago. These are now mature, with abundant gemmae, while as yet there are no signs of cupules on the control plants.

EXPERIMENT 61

A sealed glass tube of $RaBr_2$ (10 mg.; 1,500,000 ×) was placed in contact with the gemmae in a cupule of *Lunularia* for six days. The gemmae were then sown on one half the surface of soil in a pot. The other half of the surface was sown with unexposed gemmae.

Fourteen days later the unexposed gemmae are developing thalli, but those exposed have not grown at all, and are evidently dead.

This experiment was repeated, the exposure to the radium-rays being for 19 hours. At the close of the experiment, 42 days after the exposed and the control gemmae were sown on soil, none of the radiated gemmae have grown, but all of the control set have developed vigorous thalli.

EXPERIMENT 62

April 3, 9:45 A. M.

In order to see if the growth of gemmae of *Lunularia* would be affected by the presence of a tube of radium in the soil, 36 gemmae were sown on the surface of soil in a flower pot, and a tube containing 10 mg. of $RaBr_2$ of 1,500,000 activity was inserted vertically into the soil at the center of the pot, the end containing the radium being 25 mm. below the surface.

Control culture with 36 gemmae, but empty glass tube.

May 5, 2:30 P. M.

The gemmae are growing well in both cultures, with no appreciable difference in the two sets.

May 12.

There is no significant difference in the size of the developed thalli in the two cultures. The radium tube was removed. Duration of exposure 5½ weeks.

June 2, 10 A. M.

The thalli in the control culture are much larger and healthier in appearance than those exposed, and have well developed cupules with gemmae, while cupules have scarcely begun to develop at all on the radiated plants.

Experiment 63

Object: To ascertain the effect of radium rays on the "sprouting" of a potato tuber (*Solanum tuberosum*).

May 21, 12:30 P. M.

A glass tube containing $RaBr_2$ (1,500,000 ×) was inserted in a hole made in a tuber just beneath one of the "eyes," near the proximal end of the tuber. A portion of the other end was cut off, and the tuber was placed with the cut end in a tumbler of water in front of a window under a bell jar.

May 25, 10 A. M.

Sprouts are growing from all of the eyes, but there is no significant difference in their appearance or size. They all appear equally green and healthy.

Experiment 64

Object: To ascertain the effect of the rays from polonium on the germination and growth of timothy grass seed.

March 19, 6 P. M.

Several timothy grass seeds were placed in a narrow glass tube, and one end of a metallic rod coated with polonium was placed in among the seeds.

March 26, 11 A. M.

After 161 hrs. exposure, the seeds were sowed in soil in a flower pot, together with a sowing of unexposed seeds in another pot as a control.

March 30, 10:30 A. M.

The seeds are all growing vigorously, and there is no appreciable difference between the exposed and the control seeds.

Experiment 65

Object: To ascertain the effect of the thorium rays from a "Welsbach" gas mantle on the germination and growth of timothy grass seed.

June 9, 10 A. M.

Unsoaked seeds of timothy grass were sown in a row across the surface of the moist soil in a "Zurich" germinator. Lying horizontally over the seeds at right angles to the row, and about 3 or 4 mm. above them, was the Welsbach mantle, style "Yusia," No. 189, purchased in the market (FIGURE 46).

Control, with no mantle, and both cultures placed in the dark room, in a moist chamber.

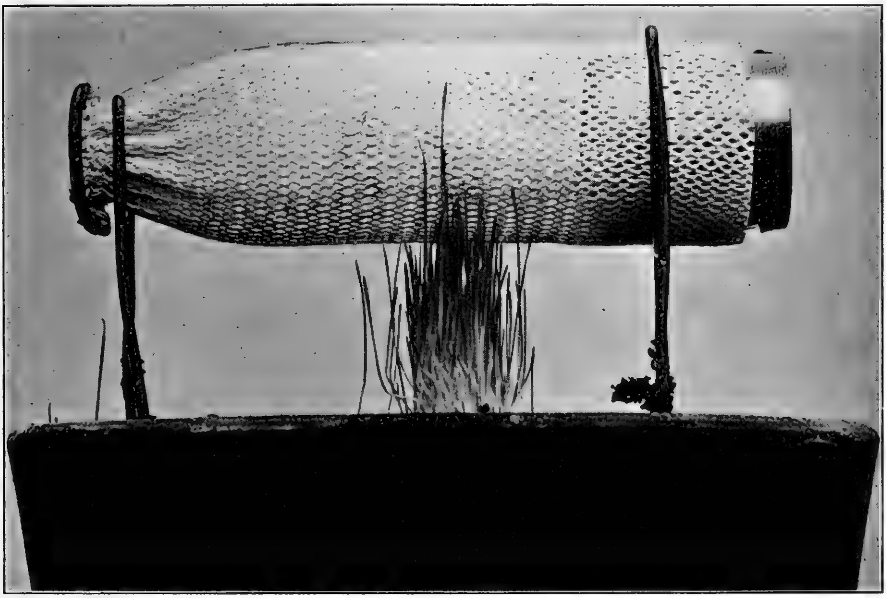

FIG. 46. Experiment 65. Method of exposing germinating Seeds to the Rays of Thorium from a Welsbach Gas Mantle.

June 13, 10 A. M.

The control seeds are evenly germinated.

The exposed seeds have germinated about the same amount as those of the control, except directly under the mantle, where there is only a very slight germination.

June 14, 10 A. M.

The seedlings in both cultures have grown since yesterday, but the relative conditions remain as then. Both cultures were placed in the light and illuminated only from above.

June 16, 10 A. M.

Retardation following the exposure is still evident, and the cultures were photographed (FIGURE 47).

This experiment was thrice repeated, with results the same in kind as those above.

EXPERIMENT 66

Object: To ascertain the effect of the rays from a Welsbach gas mantle buried in the soil on the germination and growth of timothy grass seeds planted in the same soil.

In a flower pot of earth was buried vertically the Welsbach mantle, style "Yusia," No. 189, purchased in the market, and on the surface of the soil were sown seeds of timothy grass. Control culture with no mantle, and both cultures placed in the dark room, and watered from below until the water had soaked up to the surface of the soil.

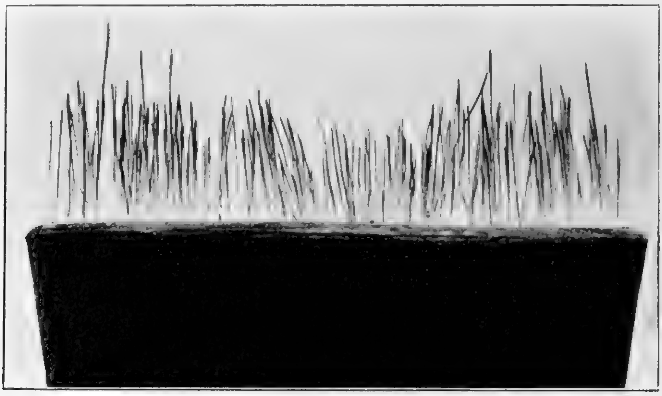

FIG. 47. Experiment 65. Effect of Thorium Rays from a Welsbach Gas Mantle on the growth of Timothy Grass. Cf. Fig. 46.

The seeds germinated in both cultures and produced good healthy plants, but throughout the duration of the experiment no difference could be detected in the height or other characters of the plants in the two pots.

The experiment was repeated, but with similar, *i. e.*, negative, results.

SUMMARY

The results of the preceding experiments, covering a wide systematic range of plants, are in conformity with those previously described. Rays from radium bromide of 7,000 × apparently accelerate the germination of pollen grains of *Asclepias curassavica* under the conditions imposed, while cell-division of commercial yeast was accelerated by rays from radium bromide of 10,000 activity. These rays produced no apparent morphological change in the yeast under the conditions of the exposure.

Mushroom spawn (*Agaricus campestris* L.) was killed by rays from 15 mg. of radium bromide of 1,500,000 ×, while rays of the same activity passing through water produced no apparent morphological change in *Spirogyra*.

When the end of the glass tube containing 10 mg. of radium bromide of 1,500,000 × was placed a few millimeters above the gemmae of *Lunularia* an exposure of 48 hours retarded the growth of the gemmae, and one of 67 hours completely inhibited it. The reaction of chloroplastids to radium rays is similar to their reaction to too intense sunlight (FIGURE 47*a*). This indicates that the gemmae are rather resistant to the rays. Cupules with brood-buds formed sooner than normally on thalli grown from the gemmae exposed for 48 hours. When the same radium tube was placed in contact with these gemmae they were killed within at least 19 hours, or possibly sooner. Growth of the gemmae was retarded when they were sown on the surface of soil into which the tube was inserted for five and one half weeks.

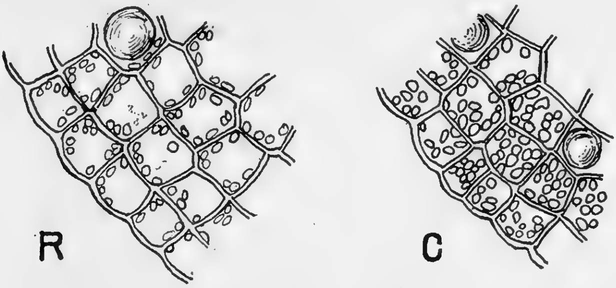

FIG. 47*a*. Experiment 60. Response of Chloroplasts to Radium Rays.

The sprouting of a potato was evidently not affected by having the radium tube (1,500,000 ×) inserted into the tissue of the tuber about 10 mm. from the "eye."

No effect on germination followed an exposure of dry timothy grass seed for 161 hours to the α rays from a rod coated with polonium.

The thorium rays from a Welsbach gas mantle retarded the growth of timothy grass when the mantle was placed over the seeds germinating on the surface of soil, but germination and growth did not appear to be affected in the least by burying the mantle in the soil.

CHAPTER XII

EFFECTS OF RADIUM RAYS ON THE SYNTHESIS OF CARBOHYDRATES

1. Effect on Photosynthesis

A few experiments were made to test the effects of radium rays on the photosynthetic activity of the green cell. These experiments are qualitative, and in many respects extremely crude. It is hoped, however, that they may serve to blaze the way for more accurate quantitative work.

Experiment 67

Object: To ascertain the effect of the rays from rods coated with Lieber's "radium coating" on photosynthesis in the bean leaf (*Phaseolus*).

June 9, 10:30 A. M.

A coated rod (25,000 ×) was placed horizontally about three mm. above a mature green leaflet of a bean plant, growing in a north window.

A rod of 10,000 activity was similarly placed over a leaflet of an adjacent plant.

Other leaves on the same plants served as controls, and a portion of the exposed leaf from each plant was tested and found to contain starch at the beginning of the experiment.

June 9, 5 P. M.

Nyctitropic movement has lowered both leaves away from the rods, and beyond their influence.

June 10, 10 A. M.

The rods were readjusted to their former positions.

June 10, 2:30 P. M.

The leaves were excised, dechlorophyllized, and tested with iodine for starch.

The control leaflet stained blue evenly throughout. The leaflet exposed to the rod of 10,000 activity showed no effect from the exposure, staining evenly throughout, but the leaflet exposed to the rod, of 25,000 activity, showed only slight starch reaction in the portion that was under the rod, but stained deeply in all other parts. Even though the rod of 10,000 activity showed no effect, it is possible that the effect produced by the more active rod was due either wholly or in part to the shading of the leaf by the rod. This source of error was eliminated by placing the coated rod under the leaf, as in the following experiment.

EXPERIMENT 68

May 29, 10 A. M.

A nasturtium (*Tropaeolum*) plant was removed to the light after being for 18 hours in the dark-room. Under one of the leaves was placed a Lieber's radium-coated rod of undetermined activity (probably 25,000 ×).

May 30, 10 A. M.

After 24 hours' exposure the leaf was tested for starch. Abundant starch was found in the part of the leaf farthest from the rod, but very slight traces only in the remainder of the leaf, particularly in the part that was directly over the coated rod.

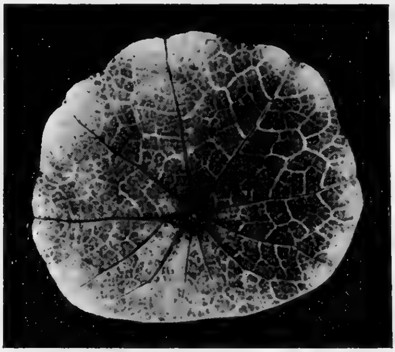

FIG. 48. Experiment 68. Retardation of Photosynthesis by Radium Rays. The print was made by placing the leaf itself in contact with velox paper in sunlight. Thus the darker portions of the print indicate the more translucent portions of the leaf-blade, due to absence of starch. The darker region was directly over the radium-coated rod.

EXPERIMENT 69

May 28, 5 P. M.

Under a healthy green leaf on a nasturtium (*Tropaeolum*) plant that had been in the dark-room for 7 hours, was placed 10 mg. of $RaBr_2$ (1,800,000 ×), in a sealed box, portected from the leaf by only a thin mica window.

After 19 hours' exposure in the dark-room to the radium rays, the radium was removed and the plant placed in the sunlight.

After an exposure of 30 minutes to sunlight, the leaf exposed to the radium rays, and a control leaf were tested for starch, with iodine. Starch was found in all parts of both leaves, but the iodine gave a much darker stain, and more evenly distributed in the control leaf, while in the radiated leaf the color was decidedly lighter, and lightest in the region that was directly over the radium, growing gradually darker toward the margin of the blade.

Apparently a slight retardation of starch-synthesis has followed the exposure to the radium rays.

Experiment 70

A nasturtium (*Tropaeolum*) plant was removed to the light after being in the dark-room for 18 hrs., and under one of the healthy leaves was placed the sealed glass tube of $RaBr_2$ (1,800,000 ×), the tube lightly touching the leaf in places.

After an exposure to the radium of 24 hours the leaf was tested for starch (10 A. M.), but the differences in the staining were not as marked as when the coated rod was used.

Experiment 70*a*.

In order to see if an exposure to radium rays for shorter periods of time would be followed by an acceleration of photosynthesis, the following experiment was tried:

From a healthy nasturtium plant (*Tropaeolum majus*) that had been in total darkness for 12 hours three vigorous leaves were detached and exposed to the direct rays of the sun as follows:

No. 1 lying on a glass tube of $RaBr_2$ of 1,800,000 activity.

No. 2 lying on a glass tube of $RaBr_2$ of 1,500,000 activity.

No. 3, Control. Not exposed to radium rays.

The leaves were all destitute of chlorophyll at the beginning of the experiment. Four exposures were made as described, one each of 2, 5, 10 and 15 minutes.

On decolorizing the leaves and staining with iodine no difference could be detected in the reaction for starch. No starch was indicated in any of the leaves. On examination with the microscope, also, no starch was found, except, of course, in the guard cells of the stomata, as would be expected under normal conditions.

The results were negative.

A repetition of this experiment with leaves that had been in the dark room for 48 hrs. also gave negative results after exposures of

5, 10, 15 and 30 minutes to the rays from radium of activity 1,800,000 and 1,500,000, and from a radium-coated rod (activity undetermined).

In order to give a more thorough test for photosynthetic activity, the experiments just described were repeated with the glass tubes containing $RaBr_2$ of activities 1,800,000 and 1,500,000 respectively. Exposures were made of 2, 5, 10 and 15 minutes, and the leaves were then tested for both starch and sugar (Fehling's test). Neither starch nor sugar was found in any of the exposed or control leaves.

2. Effect on the Conversion of Cane-Sugar to Starch in the Dark

Experiment 71

In order to test the effect of the rays of radium on the formation of starch by amyloplastids in the dark, two leaves of pumpkin (*Peppo*), taken from a healthy plant kept in the dark for 36 hours, were floated in separate glass dishes on the surface of a 10 per cent. solution of cane sugar, the petioles extending into the solution. Over one leaf (*R*) was placed a glass tube containing $RaBr_2$ (10,000 ×). The other leaf served as the control.

Tests for starch in both leaves at the end of three days showed no effects that could be attributed to the radium. Starch was formed irregularly in both leaves, and similar results were obtained in two repetitions of this experiment, using pumpkin leaves in each case.

The experiment was repeated with leaves of nasturtium as follows:

Leaves from a nasturtium plant that had been in the dark-room for 12 hours and found devoid of starch when tested, were floated on the surface of a 10 per cent. solution of cane sugar, in separate glass dishes. Over one of the leaves was suspended the sealed glass tube containing 10 mg. of $RaBr_2$ of 1,800,000 activity, about 3 or 4 mm. from the surface of the leaf.

After an exposure to the radium rays, as described, for 7½ hours, no starch was found in either of the leaves, exposed or control. After an exposure of 29 hours the control leaves were found to have made starch, but no trace of starch could be detected in the leaf exposed to the rays of radium.

In another repetition of the same experiment, after 20 hours' exposure (as above described) to the rays from $RaBr_2$ (1,800,000 ×), the result obtained in that experiment was confirmed, *i. e.*, starch had been elaborated in the control leaf, but not in the one exposed to the rays of radium.

The exposed leaf was very slightly etiolated.

The experiment was twice again repeated, once with an exposure to rays from $RaBr_2$ of 1,800,000 activity for 30 hours, and again for 20 hours. After the 30-hour exposure an abundance of starch was found in the control leaf, but absolutely none in the one exposed to the radium. The exposed leaf was greatly bleached.

After the 20-hour exposure the exposed leaf was only slightly bleached, and no trace of starch was found in it, while there was an abundance in the control.

EXPERIMENT 72

To test the effect of exposing the leaf to the radium before feeding it with the sugar solution, the following experiment was tried:

Over a nasturtium leaf still attached to a plant that had been in total darkness for 30 hours, was placed the sealed glass tube of $RaBr_2$ (10 mg., 1,500,000 ×), just touching the leaf. The plant was left in the dark, and after 29 hours' exposure to the radium, the leaf was cut off and floated on a 10 per cent. solution of cane sugar, in the dark, with two control leaves.

After 51 hours the three leaves were dechlorophyllized and treated with iodine. No trace of starch was indicated in the leaf that had been exposed to the radium, but an abundance of it was indicated in the control leaves.

The above experiments show that the rays from $RaBr_2$ of 1,800,000 and 1,500,000 activity inhibit the activity of plastids in converting cane sugar to starch.

EXPERIMENT 73

Two green nasturtium leaves, devoid of starch, were floated on a 10 per cent. solution of cane sugar in which the sealed glass tube of 10 mg. of $RaBr_2$, of 1,800,000 activity has been immersed for 15 hours, and then the tube of 10 mg. $RaBr_2$, of 1,500,000 activity for 12 hours. This solution had also stood for two days subsequent to its exposure to the radium rays, before being employed in this experiment.

Control with two leaves floated on a solution not exposed to the rays.

After an exposure for 8 hours, the leaves were tested for starch. Starch was found in all the leaves, but there was decidedly more starch apparent in those floated on the control solution, than in those floated on the solution exposed to the rays of radium.

Whatever effect the rays may have had on the solution persisted for at least two days, and resulted in a retardation of starch-making by the plastids.

3. Effect on Chlorophyll Solution and Chlorophyll Paste

Experiment 74

Object: To ascertain the effect of the rays of radium on an alcoholic solution of chlorophyll.

An alcoholic solution of chlorophyll was prepared by soaking filaments of *Spirogyra* in cold 95 per cent. alcohol. The solution was then filtered and divided into two halves. Into one was suspended a sealed glass tube containing 10 mg. of $RaBr_2$ of 1,500,000 activity, with the end containing the radium in about the middle of the solution. In the other was placed nothing. Both vials were set in the dark-room, at the same temperature.

Frequent observations during a period of 89 hours disclosed no appreciable difference in the appearance of the two solutions. The radium-tube was then removed. and the solutions left to stand in the dark-room for eight weeks. At the end of that time there was no difference in their appearance, and when both vials were then placed in the sunlight the solutions behaved precisely alike, so far as could be detected, while they were bleaching.

Repetitions of this experiment were followed by similar (negative) results. In the second and third experiments the solutions were made respectively from leaves of nasturtium (*Tropaeolum*) and bean (*Phaseolus*).

Experiment 75

A small box with a mica window, and containing 10 mg. of $RaBr_2$ of 1,800,000 activity, was placed over a small bit of chlorophyll paste ("Chlorophyll Puriss.," of supply houses), with only the mica between the radium and the paste. The exposure was made in the dark-room for 18 hours, but no visible effect from the radium resulted to the chlorophyll.

Experiment 76

Into a weak aqueous solution of the chlorophyll paste was placed the glass tube of $RaBr_2$ (10 mg., 1,500,000 ×), but after an exposure of four days the chlorophyll was not bleached, nor could any other effect be observed resulting from the influence of the rays of radium. The solution bleached in sunlight as normally.

Summary

Using the starch-iodine reaction as a test for photosynthesis, it was found that when a radium-coated rod of 25,000 × was placed over a healthy leaf of *Phaseolus vulgaris* for 10 hours in direct sunlight, and at a distance of only 2 or 3 mm., no starch was formed in the narrow region immediately under the rod, though abundantly in the remainder of the leaf. A rod of 10,000 × similarly placed at the same time over another leaf of the same plant had no apparent effect on starch formation. However, to be more sure that the first effect was not due merely to the shading of the leaf by the rod, the experiment was repeated, using a leaf of nasturtium (*Tropaeolum majus*), and placing the active rod under the leaf, and in contact with the under surface. By this method the effect, after an exposure of 24 hours, was not so marked, but there was a decided retardation of starch making. A leaf of a nasturtium plant that had been in the dark for 26 hours, and exposed to radium rays (1,800,000 ×) that had passed through only a thin sheet of mica for 19 hours out of the 26, was then brought into direct sunlight for 30 minutes. The starch-iodine reaction showed less starch in the exposed leaf than an unexposed one from the same plant, and the effect was greatest directly under the mica window through which the radium rays passed.

An exposure of 24 hours to the rays from radium bromide of 1,800,000 × in a sealed glass tube placed under a leaf was followed by no appreciable effect.

To see if photosynthesis could be stimulated by short periods of treatment, starch-free leaves of nasturtium were exposed in direct sunlight over sealed glass tubes of 1,500,000 × and 1,800,000 ×, and to a radium-coated rod for periods of 5, 10, 15, and 30 minutes, but no effect, of either acceleration or retardation, could be detected by the starch-iodine reaction. In a repetition of this experiment, tests were made for sugar as well as for starch, but with negative results.

It was Boehm,[1] who, in 1883, first demonstrated the power of chloroplasts to form starch in absolute darkness when supplied with cane sugar from a 10 to 20 per cent. solution. This process, in leaves of nasturtium, was completely inhibited, and the leaves bleached by exposures of from 20 to 30 hours to rays from the glass tube of 1,800,000 activity. When the leaf was exposed for 29 hours (1,500,000 ×) before being floated on the starch solution, it made no starch in darkness.

I then reversed the manner of exposure; that is, I exposed the sugar solution instead of the leaf to the rays. The sealed glass tube containing 5 mg. of radium bromide of 1,800,000 × was suspended in the solution for 15 hours, and then the tube of 1,500,000 × for 12 hours longer, giving a total exposure of 27 hours. It was then necessary to wait for two days before placing the leaves in the irradiated solution, but, whatever effect the rays had on the solution, it endured for at least two days, for, while some starch was found after eight hours in leaves floated on this preparation, the amount was very much less than that found in similar leaves floated for the same period of time on a portion of the same solution that had not been exposed to the radium. Undoubtedly the effect would have been much more marked had it been possible to use the solution immediately after it had been exposed, but I have not been able to repeat this experiment.

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CHAPTER XIII

EFFECTS OF RADIUM RAYS ON PLANT RESPIRATION

1. Effect on Aërobic Respiration

In order to test the effect of the rays of radium on plant respiration, the following experiments were tried:

Experiment 77

August 15.

Into each of three tumblers, 1, 2, and C, was placed a saturated solution of KOH, and above this, on a moist blotter, supported by a piece of wire gauze, were placed grains of wheat (*Triticum*, Henderson's "Wellman Fife") weighing 2 gr. (dry weight). Over the wheat and in contact with the grains were placed the radium-tubes as follows:

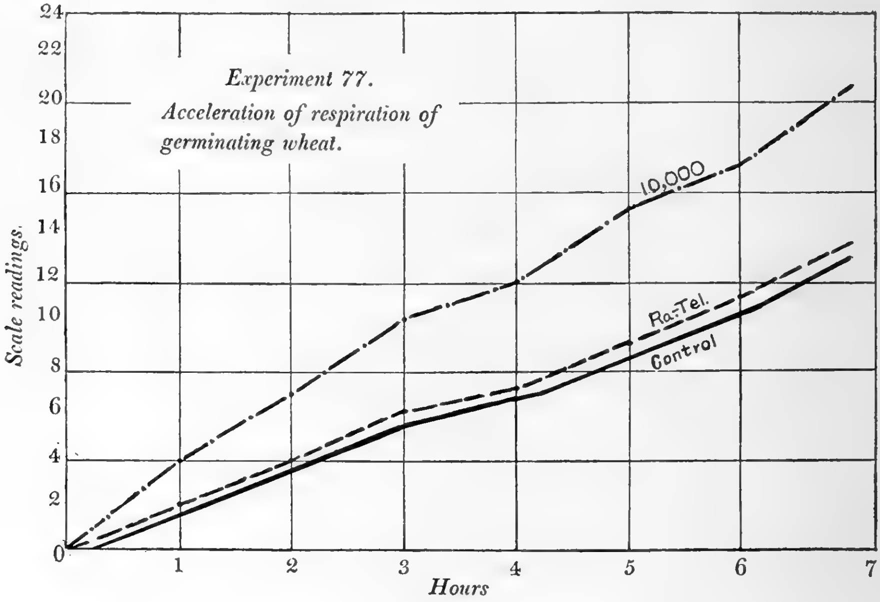

Fig. 49.

1. The sealed glass tube of $RaBr_2$ of 10,000 activity.
2. The sealed glass tube of radio-tellurium.
C. Control. No radium preparation.

Over all was placed, in each tumbler, a piece of moist blotter. Leading from each tumbler, through an air-tight rubber stopper into a mercury bath, was a glass tube, graduated to 1/100 of a mm. The entire tumbler in each case was immersed in water to insure similar temperature conditions, and also to make the respiratory chamber more surely air-tight.

The rise of mercury, following the absorption, by the KOH solution, of the CO_2 given off, was taken as the index of both the rate and the amount of respiration. The record of readings is given in the following table, and in FIGURE 49.

Readings of the Scale, Multiplied by 100

Time (Aug. 15)	$RaBr_2$, 10,000 ×		Radio-tellurium		Control	
9:15	74.25 =	0.00	81.25 =	0.00	71.00 =	0.00
		4.00		2.00		2.00
10:15	70.25		79.25		69.00	
11:15	67.25	3.00	77.25	2.00	67.00	2.00
12:30	64.00	3.25	75.00	2.25	65.00	2.00
1:15	62.25	1.75	74.00	1.00	64.00	1.00
2:15	59.00	3.25	72.00	2.00	62.00	2.00
3:15	57.00	2.00	70.00	2.00	60.00	2.00
4:15	53.50	3.50	67.50	2.50	57.25	2.75
	Total,	20.75	Total,	13.75	Total,	13.75

The radio-tellurium produced no effect that could be detected, but a decided acceleration in respiration followed the exposure to the rays from $RaBr_2$ of 10,000 activity.

EXPERIMENT 78

This experiment was arranged as in No. 77, using sealed glass tubes of $RaBr_2$, of 1,500,000 and of 10,000 activity. In each respirometer 20 grains of the wheat were used, with a total dry weight in each case of .50 gm.

Readings of the Scale, multiplied by 10

Time (Sept. 19)	$RaBr_2$ 1,500,000	Amt. of Change	$RaBr_2$ 10,000	Amt. of Change	Control	Amt. of Change	Temp. °C.
9:30	9.45	0.40	9.35	0.40	9.30	0.35	25
10:30	9.05	0.45	8.95	0.40	8.95	0.35	25
11:30	8.60	0.28	8.55	0.33	8.60	0.27	25
12:30	8.32	0.22	8.22	0.22	8.33	0.18	25
1:30	8.10	0.20	8.00	0.30	8.15	0.20	25.75
2:30	7.90	0.20	7.70	0.25	7.95	0.25	25.75
3:30	7.70	0.20	7.45	0.28	7.70	0.20	26
4:30	7.50	0.20	7.17	0.32	7.50	0.20	26
5:30	7.30		6.85		7.30		26
(Sept. 20)	Total,	3.05		4.25		2.90	
8:30	4.25	5.15	2.60	6.75	4.40	4.90	26
Total in 23 hrs. =		8.20		11.00		7.80	

The total amount recorded of CO_2 evolved in 8 hours is 3.05 units, following exposure to the rays from the salt of activity 1,500,000; 4.25 units following exposure to the rays from the salt

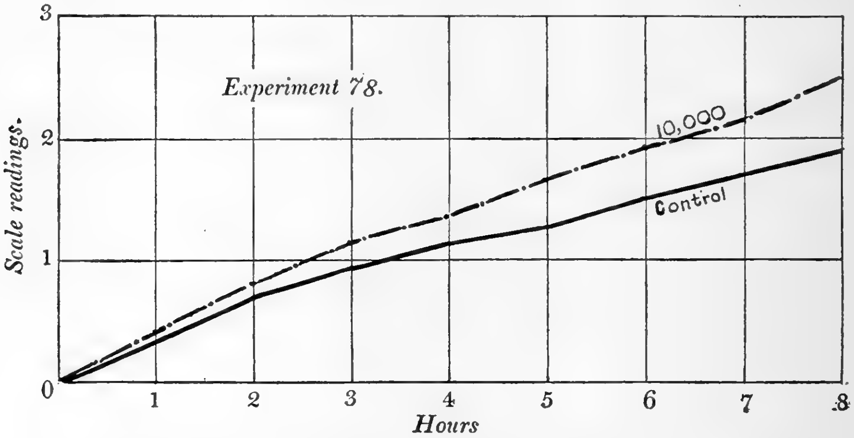

FIG. 50. Acceleration of Respiration of germinating Wheat by Radium Rays.

of 10,000 activity, and 2.90 units in the control. During the last 15 hours of the experiment, 5.15 units were recorded following exposure to the radium of stronger activity; 6.75 units following exposure to the radium of weaker activity, and 4.90 units in the control. The total amounts evolved during 23 hours were recorded respectively as follows: 8.20 units; 11.00 units; 7.80 units.

The greater acceleration following exposure to the radium of weaker activity may possibly be attributed to the fact that there was much more of the latter salt in the glass tube than of the former, the amounts being for the 10,000 × .52 gm., for the 1,500,000 × 10 mg. (FIGURE 50).

EXPERIMENT 79

The following experiment was arranged as in No. 77, using only the radium of 1,500,000 activity. Readings of the height of the mercury column in the graduated glass tube were taken as follows. The scale divisions are multiplied by 10.

Time	RaBr$_2$ 1,500,000	Amt. of Change	Control	Amt. of Change	Temp. °C.
9:30	9.80		9.70		26
10:30	9.50	0.30	9.45	0.25	26
11:30	9.20	0.30	9.25	0.20	26
1:30	8.65	0.55	8.85	0.40	26
3:00	8.20	0.45	8.46	0.39	26
3:30	8.05	0.15	8.35	0.11	26
4:30	7.73	0.32	8.10	0.25	26
5:30	7.50	0.23	7.85	0.25	25
		Total, 2.30		Total, 1.85	
		3.55		3.15	
		Total, 5.85		Total, 5.00	
9:00	3.95		4.70		25.5
10:30	3.55	0.40	4.40	0.30	25.5
11:30	3.32	0.23	4.25	0.25	25.5
1:30	3.03	0.29	3.99	0.26	25.5
2:30	2.88	0.15	3.80	0.19	26
3:30	2.70	0.18	3.56	0.24	26
4:30	2.53	0.17	3.50	0.06	26
5:15	2.40	0.13	3.40	0.10	26
8:30	0.20	2.20	1.33	2.07	25
		Total, 9.62		Total, 8.47	

Here also acceleration of respiration has followed exposure to the rays from radium of 1,500,000 activity. (See FIGURES 51 and 52.)

EXPERIMENT 80

In order to ascertain the effect, on the respiration of germinating wheat grains, of a previous exposure, in the dry condition, to the rays of radium, twenty wheat grains, weighing (dry weight) .50 gm., were placed before soaking, in contact with the walls of a sealed glass tube containing 10 mg. of $RaBr_2$, of 1,500,000 activity. The duration of exposure was 54 hours.

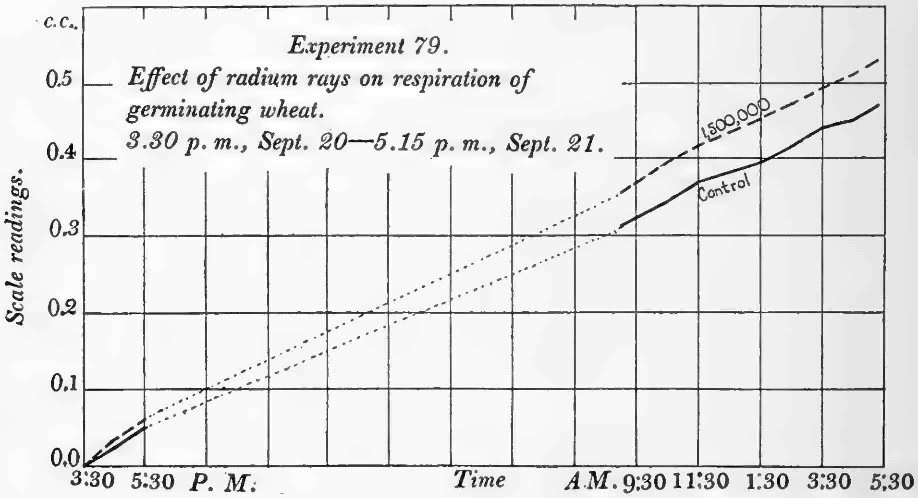

FIG. 51.

These seeds, together with 20 others, also weighing .50 gm., and serving as a control, were soaked in water over night, and arranged in the respirometer, as described in Experiment 77.

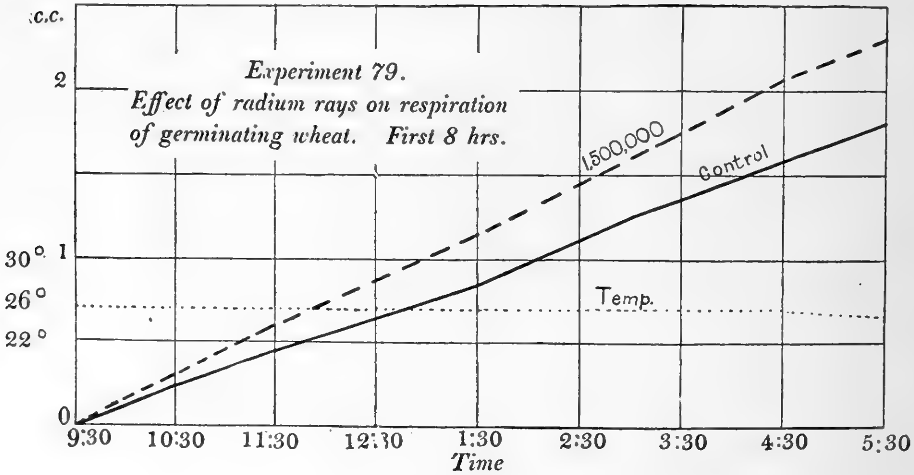

FIG. 52.

Observations of the amounts of CO_2 given off were recorded as follows:

Time	Radium		Control		
(Oct. 3)	Scale Reading	Amt. of Change	Scale Reading	Amt. of Change	Temp. °C.
9:00	9.53	0.13	9.10	0.17	19
10:00	9.40	0.03	8.93	0.01	19
11:00	9.37	0.07	8.92	0.02	19
12:00	9.30	0.10	8.90	0.05	19
2:00	9.20	0.10	8.85	0.05	19
3:00	9.10	0.05	8.80	0.00	19
4:00	9.05	0.10	8.80	0.10	19
5:45	8.95	1.25	8.70	0.80	19
		1.83		1.20	
(Oct. 4)					
9:00	7.70	0.10	7.90	0.05	18
11:00	7.60	0.03	7.85	0.02	18
12:00	7.57		7.83		18
(Oct. 5)		1.42		0.98	
9:30	6.15	0.15	6.85	0.15	19
3:30	6.00		6.70		18.5
(Oct. 6)		0.60		0.50	
9:00	5.40		6.20		19
(Oct. 7)		1.25		1.30	
10:00	4.15		4.90		18
(Oct. 8)		1.65		1.37	
9:00	2.50		3.33		16

The total amounts of gas evolved during the five days of the experiment were, for the exposed seeds, 7.03 units; for the control, 5.57 units. The results for the first nine hours are shown in FIGURE 53. There was an apparent initial retardation of respiration during the first hour of exposure, but thereafter the rate was more rapid for the seeds exposed. At the end of the first nine-hour period the radiated seeds had evolved a total of 1.83 units of gas, the control seeds a total of 1.20 units.

The effect on respiration of exposing dry seeds before soaking, and therefore before germination had begun, differs from that of making the exposure only after the seeds were soaked, in that at first there is apparently a retardation of respiration, followed by recovery and acceleration, while in the latter case respiration is accelerated from the start.

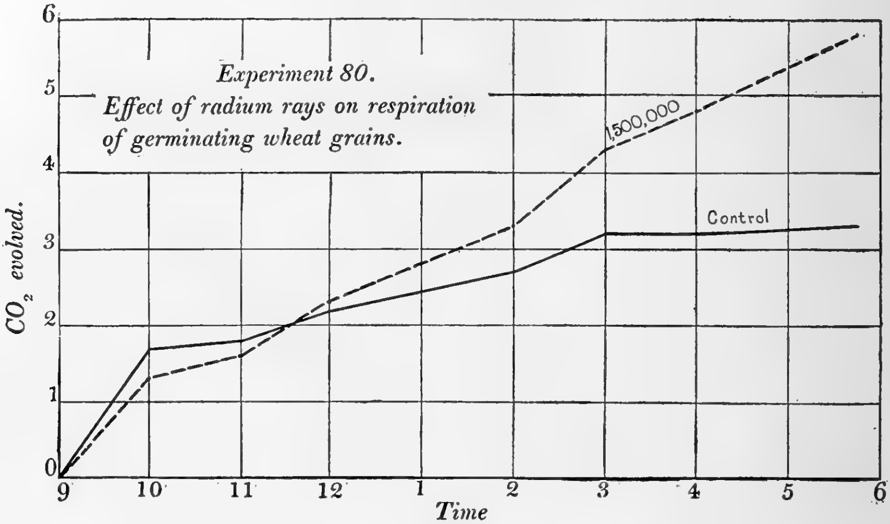

FIG. 53.

2. EFFECT ON ANAËROBIC RESPIRATION

EXPERIMENT 81

Three seeds of the pea (*Pisum sativum*), soaked in tap-water over night, were freed from their seed-coats and then found to weigh 1.41 gm. They were placed in a test-tube of mercury inverted over a mercury bath. In the tube with the peas, and in contact with them, was placed a celluloid rod coated with "Lieber's radium coating" of 10,000 activity. Thus the seeds came under the influence of the α rays and the emanation, as well as of the β and γ rays.

The control test-tube contained three seeds, also weighing, without their seed-coats, 1.41 gm.

Observations were recorded as follows:

Volume of Gas Evolved

Time of Observation	Radium	Control
April 15, 5:00 P. M.	0.00 c.c.	0.00 c.c.
" 16, 10:18 A. M.	.30	.30
" 17, 4:30 P. M.	1.70	2.10
" 18, 9:20 A. M.	.80	1.00
" 18, 6:40 P. M.	.40	.90
" 19, 9:30 A. M.	.50	.40
	Total, 3.70 c.c.	4.70 c.c.

EXPERIMENT 82

Five seeds of the pea (*Pisum sativum*), soaked over night and then freed of their seed-coats, were placed in a test-tube of mercury, inverted in a bath of mercury. The weight of the seeds, minus the seed-coats, was 2.29 gm. In the test-tube was first placed a sealed glass tube of $RaBr_2$ (1,500,000 ×), with a cork over its upper end to hold it at any desired level in the test-tube. Thus the seeds, when placed into the test-tube, were aggregated about the lower end of the radium-tube where the radium was, and so were brought closer to the radium salt, and more directly under the influence of the rays.

Control, with empty tube also supported by a cork, and with seeds having a total weight of 2.28 gm.

Observations of the amounts of gas given off by the seeds were recorded as follows (see also FIGURE 54):

Time of Observation	Radium	Control
April 22, 10:50 A. M.	0.00 c.c.	0.00 c.c.
" 23, 4:00 P. M.	1.50	1.60
" 24, 9:00 A. M.	1.60	1.30
" 25, 9:10 A. M.	1.80	1.75
" 28, 9:45 A. M.	7.30	8.05
	12.20 c.c.	12.70 c.c.

EXPERIMENT 83

Four test-tubes were arranged as described in Experiment 82, the total weight of the seeds (minus the seed-coats) in each case being 2.22 gm. The first three test-tubes contained radium preparations as indicated in the following table. Observations of the volumes of gas evolved in the test-tubes were recorded as follows:

Time of Observation	Radium 1,500,000 ×	Radium 10,000 ×	Radio-tellurium	Control
June 1, 6:00 P. M.	0.00 c.c.	0.00 c.c.	0.00 c.c.	0.00 c.c.
" 2, 10:00 A. M.	.40	.60	.60	.60
" 3, 11:00 A. M.	.80	1.60	1.30	1.10
" 4, 12:00 M.	1.35	1.30	1.25	1.00
" 5, 12:15 P. M.	.97	1.20	1.25	1.20
" 6, 11:00 A. M.	1.63	1.65	1.50	1.35
	5.15 c.c.	6.35 c.c.	5.90 c.c.	5.25 c.c.

These readings indicate, as in the preceding experiments, a retardation of respiration by the radium of 1,500,000 activity, but the volumes of gas evolved by the seeds exposed to radium of 10,000 activity and to radio-tellurium exceed the volume given off by the control seeds.

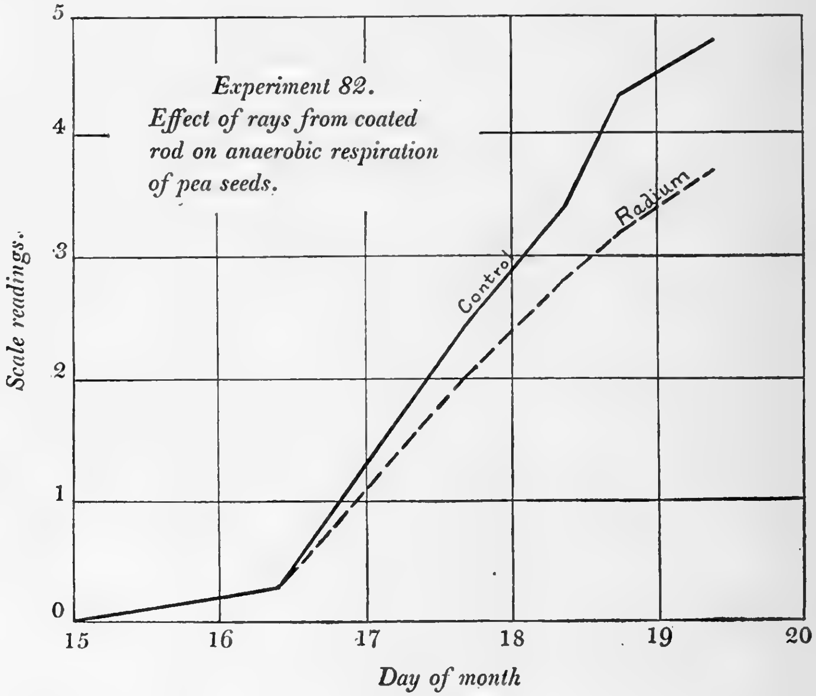

FIG. 54.

SUMMARY

When a sealed glass tube containing .52 gm. of radium bromide of 10,000 × is laid over, and in contact with soaked wheat grains placed to germinate, an acceleration of respiration results, but no effect could be detected when the grains were similarly exposed to radio-tellurium in a sealed glass tube. A like exposure to rays from 5 mg. of radium bromide of 1,500,000 × also resulted in an acceleration of respiration, but not to so great a degree as was caused by the preparation of 10,000 activity.

If the grains were exposed dry, before soaking, for 54 hrs. to rays from the radium of 1,500,000 ×, their respiration was at first retarded, but this was followed by recovery and marked acceleration.

Results obtained by the crude device employed for testing the effects of the rays on anaërobic respiration seem to indicate that an exposure to a radium-coated rod (α, β and γ rays, and the emanation) caused a retardation of the process in germinating pea seeds, and the same kind of result followed exposure to 5 mg. of radium bromide of 1,500,000 × in the sealed glass tube. In one experiment, however, a decided increase in the evolution of gas followed exposure to radium of 10,000 × in a sealed tube, and also exposure to radio-tellurium.

A comparison of the results seems to indicate that anaërobic and normal aërobic respiration of germinating pea seeds are affected in the opposite manner by radium rays, the former being retarded, the latter accelerated, but the data are quite too meager to permit of a more definite statement. The question needs further experimental investigation.

CHAPTER XIV

EFFECTS OF RADIUM RAYS ON ALCOHOLIC FERMENTATION

EXPERIMENT 84

To ascertain the effect of radium rays on alcoholic fermentation, a small piece of Fleishmann's compressed yeast cake was well mixed with 25 c.c. of a 5 per cent. solution of cane sugar. Two saccharimeters were then filled with 10 c.c. of the mixture. Into one saccharimeter was placed a sealed glass tube containing 10 mg. of $RaBr_2$, of activity 1,500,000, with all the radium in the submerged end of the tube.

A control saccharimeter contained an empty sealed glass tube, and both cultures were placed side by side at the same temperatures.

Readings of the saccharimeter-scale during fermentation were recorded as follows:

Time	Radium	Control
12 M.	0.00 c.c.	0.00 c.c.
2 P. M.	2.80	2.40
3 P. M.	3.50	2.80
4 P. M.	3.40	3.20

In a repetition of this experiment, using the sealed glass tube containing $RaBr_2$ of 7,000 activity, the following scale-readings were recorded:

Time	Radium	Control
2:50 P. M.	0.00 c.c.	0.00 c.c.
3:50 P. M.	.60	.30
4:50 P. M.	1.00	.50
5:50 P. M.	1.60	.80

The results are shown in FIGURE 55.

Again, using the radium of 1,500,000 activity, the following results were recorded:

Time	Radium	Control
12:30 P. M.	0.00 c.c.	0.00 c.c.
1:30 P. M.	.40	.40
2:40 P. M.	.60	.40
3:34 P. M.	.70	.45
4:28 P. M.	.70	.50

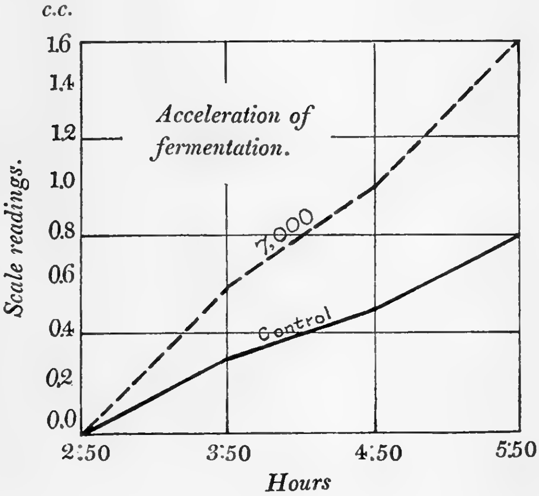

FIG. 55. Experiment 84.

The relative effect of the rays from radium and from radio-tellurium was ascertained from the following experiment.

EXPERIMENT 85

The experiment was arranged as described for Experiment 84.

Observations of the volumes of gas evolved in fermentation are recorded in the following table:

Time	$RaBr_2$ 10,000 ×	Radio-tellurium	Control	Temp. ° C.
9:20	0.00 c.c.	0.00 c.c.	0.00 c.c.	31
10:20	.95	.60	.45	35
10:53	2.00	1.30	1.10	35.5
11:20	2.95	2.10	1.60	33
11:50	4.00	3.10	2.10	33
12:20	4.75	3.85	2.55	33
12:50	5.30 *	4.60	3.00	33
1:20	5.50 *	5.05 *	3.30	33
1:50			3.60	33.5

* The last two readings for the radium, and the last reading for the radio-tellurium were estimated, as the surface of yeast mixture passed off the scale of the saccharimeter.

The above results are shown in FIGURE 56.

After fermentation had ceased in all three tubes they were allowed to stand for ten days (April 2–11), and as the gaseous products given off during fermentation were dissolved by the yeast mixture in the saccharimeters, readings were taken every 24 hours of the height of the liquid. The figures are omitted in tabular form, but the plotted curves are given in FIGURE 57. It is seen, consulting these curves, that the rate of absorption of the fermentation-product varied, being most rapid in the radium preparation, least rapid in the control, and intermediate following exposure to the radio-tellurium.

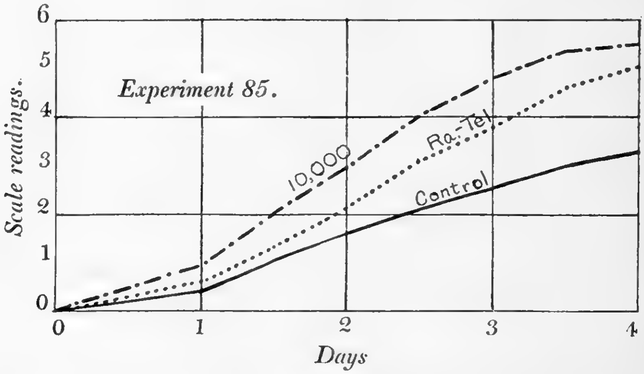

FIG. 56. Effect of the Rays from Radium and Radio-Tellurium on the Rate of Alcoholic Fermentation.

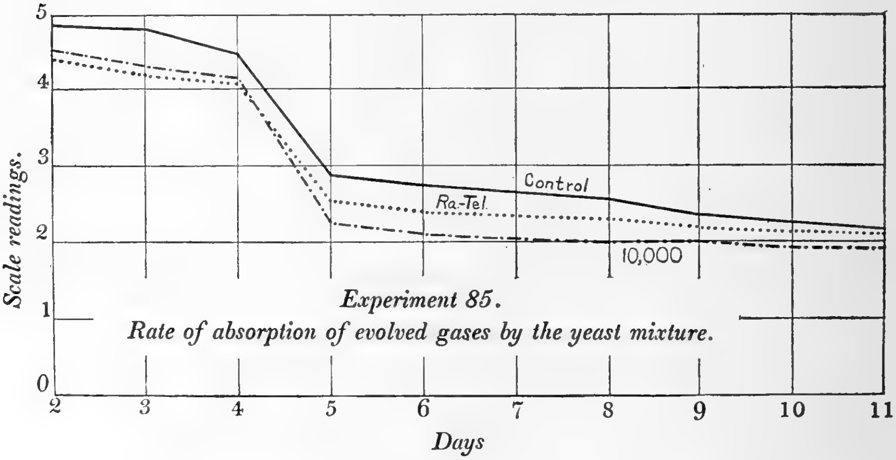

FIG. 57.

This result suggests that either the products of fermentation varied in the three cases, or else the yeast mixture was modified differently by the two radioactive preparations. Possibly the differential result is due to a combination of both conditions. Its real meaning must await further experimentation.

In a repetition of Experiment 85 the following readings were recorded of the volumes of gas given off in fermentation:

Time	$RaBr_2$ 10,000 ×	Radio-tellurium	Control	Temp. ° C.
10:30	0.00 c.c.	0.00 c.c.	0.00 c.c.	
11:30	1.00	.80	.60	
12:00	2.30	1.80	1.35	
12:30	3.85	2.85	2.40	
1:00	5.30 *	4.30	3.45	
1:30	5.60 *	5.00	4.15	
2:00	——	5.60 *	4.85	

Experiment 86

In the following experiment, into each of five saccharimeters, filled with 10 c.c. of a 10 per cent. solution of cane sugar, was placed .20 gm. of a Fleischmann's compressed yeast cake. Radium preparations were inserted into the saccharimeters as indicated in the following table, and the following readings of the saccharimeter scale were recorded:

Time	$RaBr_2$ 1,500,000 ×	$RaBr_2$ 10,000 ×	Radio-tellurium	Coated Rod	Control	Temp. ° C.
11:18	0.00 c.c.	0.00 c.c.	0.00 c.c.	0.00 c.c.	0.00 c.c.	25 †
11:48	.60	.60	.50	.30	.30	30
12:18	2.10	2.20	1.40	.80	.80	30
12:48	3.80	4.20	2.50	1.80	1.70	30
1:18	5.+ *	5.+ *	4.00	3.30	2.70	30
1:48	——*	——	5.20	4.60	3.50	29.5

Bubbles began to rise in the 1,500,000 and 10,000 preparations within two minutes after the preparations were placed in the thermostat oven, but no bubbles were rising in the control tubes up to four minutes after. The exact time in the latter was not observed, but vigorous fermentation did not begin in the control tubes as soon as in the other two tubes just mentioned.

---

* Estimated. The surface of the yeast mixture has passed off the scale.

† At 11:25 A. M. the thermometer registered 35°.

In a repetition of Experiment 86, readings of the saccharimeter scales were recorded as follows:

Time	$RaBr_2$ 1,500,000×	$RaBr_2$ 10,000×	$RaBr_2$ 7,000×	Radio-tellurium	Coated Rod	Control	Temp. °C.
2:30	0.00	0.00	0.00	0.00	0.00	0.00	29
3:00	.60	.60	.50	.40	.20	.10	22
3:30	2.10	1.80	1.60	1.50	.60	.50	31
4:00	4.00	4.00	2.60	3.00	1.20	1.20	31
4:30	5.50*	5.50*	3.85	5.10*	2.50	2.50	30.5
5:00	——*	——*	4.70	——*	3.40	3.30	30

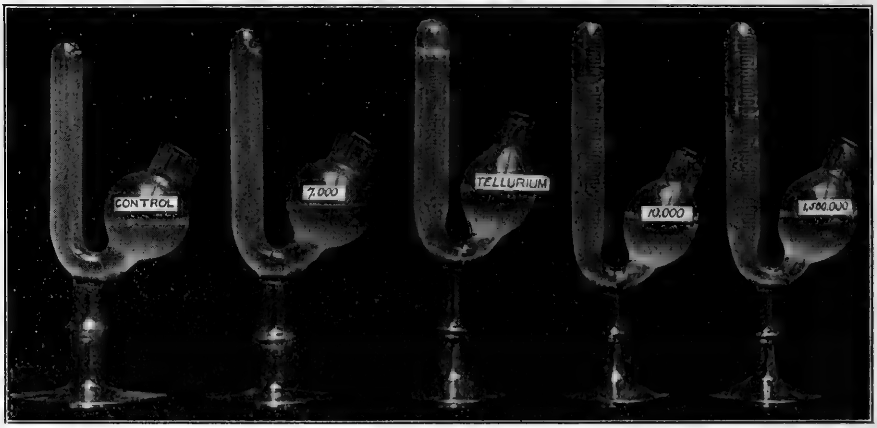

FIG. 58. Experiment 87. Acceleration of Alcoholic Fermentation by Rays from Radium and Radio-Tellurium.

EXPERIMENT 87

Into 250 c.c. of a 5 per cent. solution of cane-sugar was placed a piece of a Fleischmann's compressed yeast cake, weighing 2 gm. The mixture was thoroughly shaken, so as to distribute the yeast cells thoroughly throughout the liquid, and equal quantities were then placed in each of five fermentation tubes. Radium preparations were inserted into the fermentation tubes in sealed glass tubes as indicated in the following table, and the following readings of the scale were recorded (see also FIGURE 59):

---

* Estimated. The surface of the yeast mixture has passed off the scale.

Time	$RaBr_2$ 1,500,000 ×	$RaBr_2$ 10,000 ×	$RaBr_2$ 7,000 ×	Controls a	Controls b	Temp. °C.
10:07	0.00	0.00	0.00	0.00	0.00	*
11:30	.60	.58	.40	.20	.20	
12:00	1.20	.90	.50	.40	.40	
12:30	1.55	1.05	.50	.40	.32	
1:00	2.00	1.25	.60	.40	.40	
2:00	3.20	2.00	.70	.40	.40	

Between 1:20 and 1:45 P. M., the fermentation tubes were removed from the thermostat oven and photographed (FIGURE 58). A saccharimeter containing the sealed glass tube of radio-tellurium is also shown in the photograph. The readings of this preparation are not recorded in the above table.

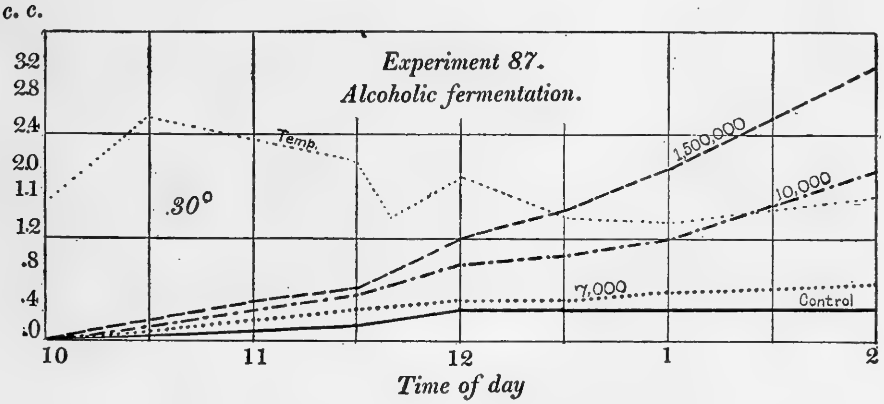

FIG. 59. Effects of the Rays from Radium of various Activities on the Rate of Alcoholic Fermentation.

EXPERIMENT 88

A yeast mixture was prepared by mixing thoroughly a piece of compressed yeast weighing 1 gm. in 100 c.c. of water, and filling the fermentation tubes with equal amounts of the mixture. Into one of the tubes was placed the radium bromide (1,800,000 ×) in the sealed glass tube, the other served as the control.

In the following table are recorded the observations of the volumes of gas evolved in each tube.

---

* The temperature is indicated in FIGURE 59.

Time	$RaBr_2$ 1,800,000 ×	Control	Temp. ° C.
2 : 00	0.00	0.00	30
2 : 30	2.00	1.75	30
3 : 00	10.00	7.00	31
3 : 30	15.50	12.00	29
4 : 00	20.50	16.00	31
5 : 00	34.00	23.00	33

The acceleration, following exposure to the rays, is shown in FIGURE 60.

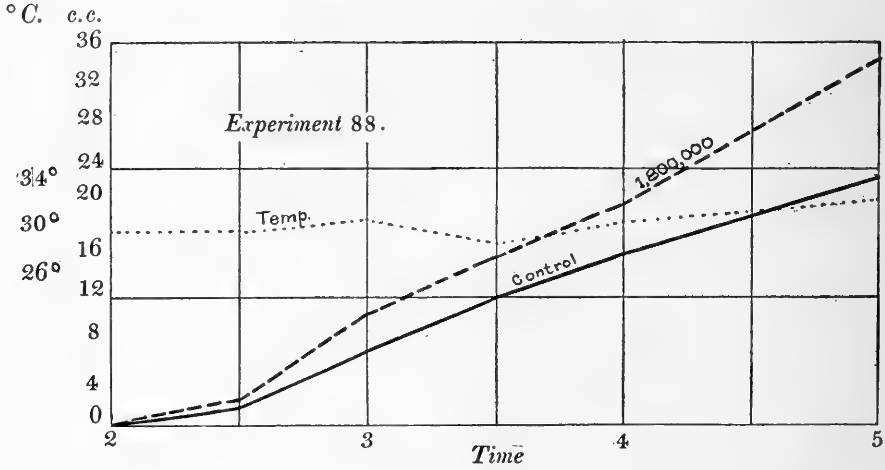

FIG. 60. Acceleration of Alcoholic Fermentation by Rays from Radium of 1,800,000 Activity.

EXPERIMENT 89

To ascertain the effect of exposing yeast-cells (*Saccharomyces*) to the rays of radium before fermentation began, 1 gm. of a Fleischmann's compressed yeast-cake was pressed closely around the end of three sealed glass tubes containing, respectively, $RaBr_2$ of 1,500,000 activity; $RaBr_2$ of 10,000 activity; and an empty glass tube.

After an exposure of 20 hours to the rays of radium, the yeast was removed from the tubes, and each piece was placed in a beaker containing 100 c.c. of a 10 per cent. solution of cane-sugar. After thoroughly mixing the yeast in the solution, three fermentation tubes were filled with the mixture from each beaker respectively, and placed in the thermostat oven. Readings of the scale showing the evolution of gas due to fermentation were recorded as follows:

Time	$RaBr_2$ 1,500,000 ×	$RaBr_2$ 10,000 ×	Control	Temp. ° C.
11:55	0.00	0.00	0.00	30
2:00	.89	.55	.68	27.5
2:30	1.30	1.00	1.00	32.8
4:00	2.30	1.80	1.15	30
4:45	2.65	2.20	1.20	30
5:00	2.90	2.50	1.20	30
5:45	3.20	2.90	1.30	30
6:15	3.40	3.20	1.30	30

A microscopical examination of portions of the mixture exposed to the radium of 1,500,000 activity, and from the control mixture, gave the following data concerning reproduction, as indicated by the number of yeast-cells having buds in the field of the microscope (B & L, $\frac{1}{6}$ obj. and No. 1 ocular). Five counts were made as follows:

$RaBr_2$ 1,500,000	Control
30	12
25	12
25	25
19	11
32	14
131	74
Av., 26.2	Av., 14.8

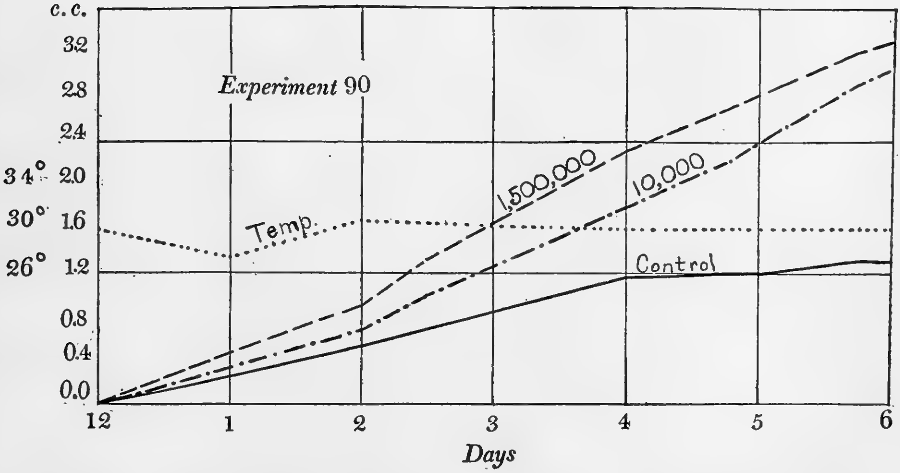

FIG. 61. Relative Rates of Alcoholic Fermentation in Saccharimeters that have been Several Times Exposed, in Previous Experiments, to Radium of 1,500,000 and 10,000 Activities.

Experiment 90

In order to see if the fermentation tubes had been affected in the preceding experiments by the rays of radium, they were filled with similar amounts of a yeast mixture, made as already described, and placed in the thermostat oven without any radium preparations. Observations were recorded as follows of the volumes of gas given off in fermentation:

Time	1,800,000 × Tube	1,500,000 × Tube	10,000 × Tube	Ra.-Tel. Tube	Control Tube	Temp. ° C.
8:00	0.00	0.00	0.00	0.00	0.00	30
9:30	1.25	1.30	1.60	1.10	1.40	31
10:00	1.95	2.30	2.60	1.90	2.10	30
10:30	2.22	3.10	3.50	2.76	2.60	28
11:00	2.80	3.72	4.16	3.60	3.00	27.3
11:40	3.48	4.63	5.10	4.66	3.55	26.7
12:00	3.90	5.10 *	5.50 *	5.10	3.90	27
	15.60	20.15	22.46	19.12	16.55	

Fermentation in the tubes that have been used a number of times with the radium preparations is more vigorous than the normal (FIGURE 61). The slower acceleration in the tube used with the strongest radium preparation (1,800,000 ×) is possibly due to the fact that this fermentation tube has only been used twice with the radium, while the others have been used 10 or more times.

Summary

When sealed glass tubes containing radium bromide of various activities, from 7,000 to 1,800,000, and radio-tellurium were inserted into fermenting mixtures of commercial yeast, the uniform result, as measured by the evolution of gas, was a stimulation of alcoholic fermentation. The same kind of result was produced by the celluloid rod coated with a Lieber's radium coating (10,000 ×).

Observation of the rate of absorption of the evolved gas by the yeast mixture showed different results following exposure to the radium preparations of different activities. This is taken to mean, either that the products of fermentation varied, or that the yeast mixtures were differently affected by the unlike treatment, one or both. No definite explanation is attempted at this time.

* Estimated. The surface of the mixture had moved off the scale.

When portions of a Fleischmann's yeast cake of like weight were exposed to radium rays before being placed in a sugar solution to ferment, the exposure to the preparation of 1,500,000 activity was followed by acceleration of fermentation from the start and throughout, but, following exposure to the salt of 10,000 activity, there was an initial retardation in the rate of fermentation, followed by recovery and acceleration. The quantity of the less active salt in this experiment was about five times that of the more active.

Microscopic examination showed that the budding of the yeast cells was considerably increased by exposure to the rays.

The fermentation tubes were found to be affected by the rays after being used a number of times so as to cause acceleration of fermentation, even though no radium was present. Reliable results, therefore, may be obtained only by using fresh tubes for each experiment.

CHAPTER XV

EFFECTS OF RADIUM RAYS ON TROPISTIC RESPONSE

It has already been shown in Chapter II that radioactivity in air and soil is a factor in the normal environment of all plants, and, therefore, that both roots and shoots are doubtless in a condition of radiotonus. Hence it is probable, *a priori*, that plants possess a radiotropic sensibility, enabling them to detect differences in either the direction or the intensity of the rays, and to respond to a unilateral stimulus of this kind. Moreover, since radioactivity is an environmental factor, such responses would be likely to be of an adaptive character. For example, the radioactivity of the soil is more intense than that in the air; therefore, within limits of intensity such that the rays do not become injurious to the tissues and produce a traumatropic response, we should expect the root to be positively radiotropic and the shoot negatively so.

In December, 1907, I very briefly discussed the probability of the existence of a true radiotropism, stating[4] that tropistic curvatures, being reactions to a stimulus felt unilaterally, could be called forth by radium rays only with difficulty, since the rays would ordinarily pass entirely through the tissues, and thus fail to be felt as a unilateral stimulus. In this communication the fact was overlooked that the organs of a plant can detect differences in direction as well as intensity of stimulus. Thus, as Pfeffer[10] has pointed out, gravity is felt with equal intensity throughout the diameter of a horizontally placed root, but its direction is perceived, and responded to. In like manner the root or shoot might be able to detect the direction of the radium rays, however slight might be the difference in their intensity on opposite sides of the organ.

In this chapter there will be briefly treated, first, the effects of the rays upon normal tropisms; second, a brief experimental investigation of the existence of a true radiotropism.

1. EFFECTS OF THE RAYS ON NORMAL TROPISMS

EXPERIMENT 91

To ascertain the effect of the exposure of dry seeds to radium rays on the geotropic response of the shoot, 5 grains of "Hickory

King" corn (*Zea Mays*) were exposed to the rays from $RaBr_2$ of 1,500,000 activity for 45 hours, and to the rays from $RaBr_2$ of 1,800,000 activity for periods of 24½ hours and 27 hours, by being placed in contact with the sealed glass tube containing the radium salt.

The grains thus exposed, together with a control set of five grains, were planted without soaking in similar pots of soil, and kept under similar conditions. The average heights of the seedlings were recorded as follows:

	45 Hours	*27 Hours*	*24.5 Hours*	*Control*
May 29 (4 days after planting).	3.75 mm.	2.00 mm.	3.00 mm.	8.00 mm.
May 30.	10.30 mm.	10.00 mm.	9.50 mm.	15.25 mm.
June 2.	47.60 mm.	33.00 mm.	18.20 mm.	72.80 mm.

On this date three of the seedlings from the seeds exposed for 45 hours (1,500,000 ×) had partly ceased to respond normally to the stimulus of gravity, and four of the seedlings in each of the cultures exposed to the radium of 1,800,000 activity had wholly or partly ceased to grow erect. On June 7 observations were recorded as follows:

45 Hours	*27 Hours*	*24.5 Hours*	*Control*
165.00 mm.	133.00 mm.	All have ceased growth since about 2 days ago, except one; height of that one equals 162.00 mm., with partial loss of power to make chlorophyll.	273.00 mm.
243.00	12.00 (ceased growth)		244.00
139.00	147.00 (etiolating)		205.00
20.90	25.00 (ceased growth)		217.00
188.00	60.00*		163.00
755.00 mm.	377.00 mm.		1102.00 mm.
151.00 mm.	75.40 mm.		220.40 mm.

The radium of 1,500,000 activity, though applied for nearly twice as long as that of 1,800,000 activity, has retarded germination and growth much less than the stronger. In the case of the radium of 1,800,000 activity, a difference in the length of exposure of 2.5 hours was at first followed by no marked difference in results, though subsequently a decided difference was noticeable.

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* Has ceased to grow erect.

EXPERIMENT 92

In order to ascertain the effect on geotropic response of growth in an atmosphere containing the radium emanation, 7 germinated pea seeds (*Pisum sativum*), were placed horizontally in conditions suitable for further growth, under a glass bell-jar containing the radium emanation as described in Experiment 45 (p. 152). In the control jar (no radium emanation) were similarly placed 8 seeds. After 24 hours 5 of the 7 exposed seeds were found to have curved geotropically, and 7 of the 8 control seedlings.

FIG. 62. Experiment 91. Absence of Geotropic Response (Pot 27) in Shoots of *Zea Mays* following Exposure of the Grains, before Planting, to Radium Rays: 45 exposed to Radium of 1,500,000 ×; 27 and 24 to 1,800,000 ×. Cf. FIGURE 14.

The experiment was repeated, using 6 germinated pea seeds under each bell-jar. After 24 hours all of the seeds showed positively geotropic curvatures in both cultures (except one seedling in the control jar which became injured), but the radicles exposed to the emanation have grown slightly more than those of the control.

Exposure to radioactivity as described appears to have no influence on geotropic sensibility, so far as can be detected by observing the growth curvature. The response, however, being a function of growth, may be varied according as the rate of growth is modified by the rays.

To further test the effect of the rays on geotropic response, 5 unsoaked grains of "Hickory King" corn (*Zea Mays*) were exposed in contact with the sealed glass tube of 10 mg. of radium bromide of 1,800,000 activity for 12 hours. Eight days after the exposed seeds had been planted in soil, they showed less than one half the growth in length of control seedlings, and two of them had failed to grow erect, and lay horizontally over the surface of the soil. They were perfectly turgid and hence failure to keep erect could not be attributed to loss of turgor.* In two other cases failure of corn seedlings to grow erect followed an exposure of the dry grain for 27 hours to the same radium-tube.†

These observations merely show that, under the conditions of exposure, the irradiated seedlings failed to grow upright as normally under the stimulus of gravity, and an examination of the tissues showed that, even if the stimulus has been perceived, response would have been practically impossible on account of the slight development of mechanical tissue, and other histological abnormalities.‡

Czapek[2] has proposed a chemical test for the perception of gravitational or other tropistic stimulus. If the stimulus is perceived, an anti-ferment is developed in seedlings of *Lupinus albus* and *Zea Mays*, which inhibits the complete action of tyrosinase. This anti-ferment is produced whether the organ responds by a curvature or not, as when a seedling is rotated horizontally on the clinostat. Even such a test would make it difficult to determine whether radium rays caused a loss of geotropic sensibility, for the rays might directly affect the ability of the protoplast to elaborate the anti-ferment when subjected to gravitational stimulus.

No experiments were performed with the express purpose of studying the effect of the rays on phototropism, but observations of all exposed plants in other experiments failed to indicate any loss of sensitiveness to the unilateral stimulus of light rays. Shoots of seedlings from seeds exposed to radium rays of varying degrees of activity and for various durations of exposure, manifested phototropic response. This is in full agreement with observations reported by Koernicke,[5] who found that both roots and shoots of *Vicia Faba*, *Lupinus albus*, and *Pisum sativum*, grown from seeds exposed to radium rays, were both geotropically and phototropically sensitive so long as growth continued, but not afterward. Radium stimulus, therefore, does not seem to interfere with sensitiveness to any other stimulus except in so far as it lowers or raises the general vitality of the protoplast as a whole.

2. Can Radium Rays cause Tropistic Response?

Experiments made for the purpose of detecting the existence of a radiotropism, or power to respond tropistically to a unilateral stimu-

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* One of the seedlings, and also one exposed for only 7 hours and a third exposed for 27 hours showed a partial loss of chlorophyll from the leaves.

† See p. 218.

‡ For details see Chapter XVI.

lation by the radium rays, are referred to in Chapter II. Koernicke's[5] experiments in this direction led to somewhat contradictory results, and he concludes that there is no response to the beta and gamma rays, but that, if the activity of the preparation is sufficiently strong, plants may bend toward the phosphorescent light of the preparation. He used *Phycomyces nitens* and *Vicia sativa*.

I have tried very many experiments with a wide variety of species, but always with negative or indifferent results, so far as concerns the direct influence of the beta and gamma rays. Neither roots nor shoots growing in air have ever shown the slightest tendency to curve toward or from sealed glass tubes containing radium of various activities.

The tropistic behavior of plants with reference to the electric current is a problem closely related to that of a possible radiotropism. It was Elfving[3] who, in 1882, first experimentally established the fact that roots grown in water or in sawdust may bend toward the positive electrode, and he introduced the term "galvanotropic" to designate this property. Twenty years later, Plowman[8] published a paper dealing with the same question, and confirming the correctness of Elfving's results. In 1904 he[9] proposed the term "electrotropism" as being more appropriate than galvanotropism. He found that negative charges of electricity "stimulate" and positive charges "paralyze" the embryonic protoplasm of the plants used, and therefore explained the electrotropic curvatures as due to the retarding effect of the positive anions on one side of the root, and the accelerating effect of the negative cations on the opposite side. This interpretation is in harmony with results obtained by Matthews[7] with the sciatic nerve of the frog. Matthews inferred that, "It is not the charge, but its motion and sign, which ultimately determines its action," and reached the conclusion that chemical stimulation, light stimulation, and electrical stimulation are identical in nature.

While nothing but failure followed all of my attempts to secure a direct tropistic response to radium rays, it was thought that, if these rays could be employed as ionizers of salts in solution, like the electric current in the experiments of Elfving and Matthews, tropistic curvatures could be thus indirectly induced.* In essentials most of

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* The entire question of the effect of radium rays on salts in solution needs further experimental investigation. In 1902 M. Curie[1] stated that the rays act on liquid dielectrics as on air, rendering them conductors to a slight degree. Kohlrausch,[6] how-

the experiments were arranged on the same plan, except for the nature of the liquid, and the distance of the radium from the roots.

EXPERIMENT 93

Roots of *Lupinus albus* were immersed in a glass dish of tap-water to a measured length of 10 mm. On account of the size of the seeds and the method of suspending them it was not practicable to employ more than two or three seeds at a time. A sealed glass tube containing 52 mg. of radium bromide of 10,000 activity was then suspended by threads so as to lie horizontally in the water with the salt distributed evenly over the bottom of the tube. The latter was placed in numerous trials at distances of from 2–25 mm. from the tips of the roots. At a distance of 25 mm. no effects were observed, but at distances of from 10 to 2 mm. curvatures took place toward the tube. Photographs of two of these trials are shown in FIGURES 63 and 64. The results in FIGURE 63 are quite similar to the electrotropic curvatures figured by Plowman. In this instance the tap-water was replaced by a nutrient solution made according to directions given on page 85 of MacDougal's Elementary Plant Physiology. No constant differences were observed between the curvatures in tap-water and culture solution.

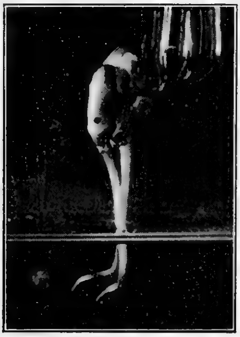

FIG. 63.

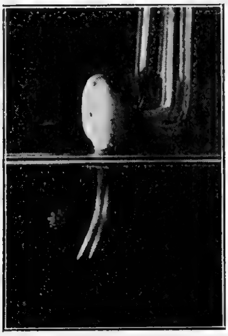

FIG. 64.

Experiment 93. Curvatures of Root-Tips of *Lupinus albus* toward a sealed Glass Tube of Radium Bromide in Water (FIGURE 64), and in a nutrient Solution (FIGURE 63).

ever, in the following year, stated that the electrical conductivity of water was not altered by exposure for a short period, but was slightly increased by exposure for as long as two days. The question is referred to further in Chapter XIX.

These curvatures are explained, not as responses to the direct influence of the radium rays, but as due to chemical ions produced by the rays in the liquid. It is probable that the exciting negative cations, travelling faster than the depressing, positive anions from the neighborhood of the tube toward the roots produce the curvature toward the tube by stimulating growth on one side. By this hypothesis the results are brought into harmony with those of Elfving and Plowman. It also follows that we have not here evidence of a true radiotropism. If such a tropism, either positive or negative, is possible, it has yet to be demonstrated. Theoretically, as pointed out above, we are led to expect its discovery.

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CHAPTER XVI

HISTOLOGICAL EFFECTS OF THE RAYS OF RADIUM*

The discovery of the fact that, following certain conditions of exposure to the rays of radium, growth is retarded, raises the question as to how the effect is produced. Several alternatives are suggested, involving the various factors concerned in the normal growth of an organism. If we disregard the numerous attempts at a rigid definition of growth, at least four such factors are to be considered: (1) increase in the size of the cells; (2) increase in mass; (3) cell-division;† (4) cell-differentiation.

Therefore when the growth of a plant or of a plant organ is diminished by any agency, the result may theoretically be accomplished by a retardation of any or of all of these four factors.

As to the effect of radium rays on constructive metabolism and resulting increase in mass we have no definite information as yet. My own experiments on the subject have so far yielded negative results. The same must be said in regard to the effect of these rays on osmosis, turgidity, and consequent cell-enlargement.

With reference to cell-division, however, we have more to say. It is known, for example, that karyokinesis may not only be interfered with, but completely inhibited by exposure to radium rays. The experiments demonstrating this are described in Chapters II and XVII. The presumption, then, is in favor of the theory that retardation or cessation of growth may be due in part to either partial or complete inhibition of cell-division. A histological examination of the tissues of plants that have suffered retardation of growth

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*The substance of this chapter was given before the bi-weekly Botanical Convention of the New York Botanical Garden, April 1, 1908.

†Of course cell-division and cell-differentiation are not a part of growth proper, but the discussion of this question need not be taken up here. Growth I would define as *increase in size, or increase in mass, one or both, with or without an accompanying change of form.* Cell-division is a factor because the number of the cells present modifies the amount of growth possible for the given tissue or organ. Cell-differentiation is a factor because it may modify the rate of growth. If a tissue rapidly matures (cell-differentiation), growth will rapidly cease. The longer the growth period of any given tissue or organ the slower the process of cell-differentiation in that tissue.

by the rays confirms the correctness of this presumption, and, at the same time, discloses the important part played by cell-differentiation.

In PLATE I are given reproductions of photomicrographs of cross-sections of the hypocotyl and roots of seedling lupines (*Lupinus albus*). FIGURE *D* shows the normal appearance of a fibro-vascular bundle and the adjacent tissues in the hypocotyl; *A* and *B* the corresponding region in the hypocotyl of plants grown from seeds that were exposed for 72 hours, while dry, to rays from radium bromide of 1,800,000 activity. After this exposure the seeds were planted in soil. The seedlings were those of Experiment 27 (p. 121). On the same plate, *C* illustrates the appearance of the corresponding region of the hypocotyl of one of the exposed plants of Experiment 29 (p. 127). This plant was grown from seeds exposed for 91.5 hours, while dry, to rays from radium bromide of 1,800,000 activity. The section was taken fourteen days after planting in the soil.

In *D* the fascicular and interfascicular cambium is well developed, as are also the phloem and xylem regions of the bundle. Comparison of *A* with *D* discloses profound modifications of these tissues. In *A* the cambium is entirely absent, all of its cells apparently having been differentiated into either phloem or xylem. The complete disappearance of the cambium clearly indicates a total inhibition of cell-division in that tissue. No new cells have been formed to replace those that have been transformed.

This effect was to be expected, being in harmony with the results of other investigators which show that the tissue most susceptible to radium rays is the embryonic. Thus Danysz * found that if 1 cg. of radium (activity not given), in a glass tube, was placed above the backbone and part of the cranium of a mouse one month old, phenomena of paresis and ataxia were produced in about three hours. Under a similar exposure a mouse three to four months old died with the same symptoms in three to four days, while a one year old mouse survived for from six to ten days.† The less mature tissue was the most quickly affected. The same author † found the epithelial tissues of young animals more sensitive than those of adults. The

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* Danysz attributed this result to the fact that, in the young mouse, the rays had to pass through cartilaginous tissue before reaching the cerebellum, whereas, in the older specimen, this tissue had become transformed into bone. It is possible also that the greater resistance of the older mice was due, in part at least, to the greater maturity of the nerve cells.

† See Chapter II. and the bibliography there given.

experiments of Bohn* on various animals, of Schaper* on the frog, of Zuelzer* on malign tumors, and of Hewlet* on cancer all indicate that embryonic tissues are more readily affected by radium rays than are mature tissues.

Now tissue-differentiation is distinguished from cell-division (tissue-formation) on the one hand, and from growth on the other. It is a function of maturity. The greater the degree of tissue-differentiation in any given species, the greater the maturity of the organ. The condition of the tissues in *A*, therefore, suggests that, as it were, the organ had become aged more rapidly following exposure to the radium rays. The youthful power of reproduction (cell-division) has been lost earlier than normally. It is also evident that, following exposure to the radium rays, the cells in the regions of the xylem and the phloem are smaller, and in every way less perfectly developed than normally.

In *B* is shown a cross-section of the hypocotyl of another plant of the same Experiment (No. 27, p. 121), exposed precisely as was *A*, and also of the same age as *D*. Here the cambium is still present, though within the bundle it is being differentiated. Xylem and phloem are less perfectly developed than normally.

Still a third variation in result is seen in *C*. Here again the cambium has disappeared, being entirely transformed into xylem and phloem. In the phloem-region of the bundle there has been more differentiation of tissue than in either *A* or *B*.

FIGURES *E* and *F* illustrate sections of the roots from plants of Experiment 29. *F* is from a control (normal) plant. In *E* the exposure of the seed was for 72 hours to rays from radium bromide of 1,800,000 activity. As in the case of the hypocotyl, the cambium has disappeared, the cells are smaller, and the tissues appear in every way abnormal.†

In PLATE 2, FIGURE *A* represents a cross-section of the hypocotyl of a *Lupinus albus* seedling grown from a seed exposed (Experiment 16, p. 99) to rays from a Lieber's radium-coated rod of 25,000 activity during imbibition of water in moist sphagnum. The material was collected six days after the seeds were placed to germinate in the sphagnum, and the exposure to the rays was continuous during this period. The hypocotyls of the control plants (*B*) were nearly 7 mm. longer than those of the exposed specimens.

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* See Chapter II. and the bibliography there given.

† The distortion of the cells of the cortex is an artifact.

Here, also, it is seen to be the cambium that is most profoundly affected. It has nearly, though not entirely, disappeared in the exposed plant, though some development of xylem and phloem has taken place. The cells of the cortex and pith are also smaller in the exposed plant than in the control.

FIGURES *C* and *D*, PLATE 2, represent, respectively, cross-sections of the roots of the same exposed and control plants as those from which *A* and *B* were taken. They show the same kind of result following exposure to the radium. The cambium is lacking, and the cells of the cortex and pith are much smaller than normally.

FIGURES *E* and *F* of the same PLATE (2) show cross-sections of the hypocotyl of the bean (*Phaseolus vulgaris*), *F* of a control plant, *E* of a plant exposed (Exp. 15, p. 98) during imbibition and germination in moist sphagnum, to a Lieber's coated rod of 10,000 activity. The section was made after growth under continuous exposure for five days.

The most noticeable difference here is in the size of the various tissue elements, especially evident on comparing the cells of the cortex and pith. The cambium has practically all disappeared from both the exposed and the control plant, though traces of it are seen in the control, indicating a more tardy tissue-differentiation. The same kind of differences appear in FIGURES *E* and *F* of PLATE 3, which show cross-sections of the stem of two seedlings of *Phaseolus vulgaris* exposed (Exp. 19, p. 101) as were those of PLATE 2. In FIGURES *E* and *F* (PLATE 2), growth took place entirely in darkness and in moist sphagnum, no foliage being developed, and no nourishment being supplied except tap-water.

The contrast between FIGURES *C* and *D* (PLATE 3) is most striking. These figures represent cross-sections through the hypoctyls of seedlings of *Phaseolus vulgaris* of Experiment 11 (p. 95). The seed from which the seedling of *C* was developed was exposed for 24 hours while dry, to the rays from a radium-coated rod, the rod being in contact with the seed. The activity of the rod was 10,000. At the close of the exposure the seeds were planted in the soil, and the seedlings developed under the most favorable conditions of heat, light, moisture, and nutrition from the soil. The section is from the hypocotyl twelve days after planting. The average length of the hypocotyl above the surface of the soil was, for the plants from exposed seeds 39.60 mm., for the control seedlings 59.25 mm.

Two facts stand out clearly in comparing *C* with *D*. First the smaller size of the medullary and cortical cells in *C*, and second the greater thickness of the cortex in *D*. Cambium is present in both, but the xylem cells are much smaller in *C*, and here, also, histological differentiation is slightly more advanced, the cell-walls of the xylem being thicker in proportion to the diamater of the cells. These differences in details combine to give *C* the appearance of a more mature tissue-complex than *D*.

If we compare the histology of the epicotyls, *A* and *B* (PLATE 3), of these same two plants, the same kind of differences are apparent. Though of precisely the same age (from time of planting to time of fixing), the tissue-differentiation of *A* is that of a more mature organ than that of *B*; ducts and woody cells are more numerous, and the bast fibers are larger. The cells of the cortex are of about the same size in the two epicotyls.

The FIGURES *A*, *B*, *D* and *E*, of PLATE 4, show the structure of the leaves of seedlings of *Zea Mays* grown from grains exposed dry to rays from radium of 1,800,000 activity (10 mg.) for 47 hours. The grains had their embryo-side in contact with the sealed glass tube containing the radium salt. These seedlings had almost wholly lost their power of responding to gravity by growing upright. Whether the capacity of perceiving the gravitational stimulus had been lost or not, we have no means of knowing, but a comparison of these sections with that of a normal leaf, shown at *C*, shows a profound structural modification. In *D*, for example, there is an almost entire absence of any mechanical tissue.

Here, doubtless, lies the explanation of the failure to grow upright. Even if the gravitational stimulus had been perceived, response would have been difficult or impossible.

In *E* the wider part of the section is through the midrib, the part of the blade to the left being wanting. Both *D* and *E* are from the same leaf, *D* being between the midrib and the leaf-margin. In *B* the epidermal layers appear to be hypertrophied, while in *A* there is both hypertrophy of the epidermis and atrophy of the mesophyll cells. In *E* the spongy parenchyma is greatly hypertrophied.

SUMMARY

In the above examples, where exposure to the rays of radium was followed by retardation of growth, histological examination discloses a cessation of cell-division, an acceleration of tissue-differentiation, a decrease in the size of the cells, a lack of coördination in histogenesis, either one or all in any given case.

Decrease in the size of the cells may be due, either to diminished turgor or to relatively* less vigorous constructive metabolism. It is not possible at present definitely to say which, though, since no partial loss of tissue-tension, nor any other evidence of a loss of turgidity, has been detected, following exposure to the rays, a less vigorous constructive metabolism appears to be the more probable cause.

The first two effects mentioned, viz., cessation of cell-division and accelerated tissue-differentiation, have both the same significance, that is, early senescence. In such instances as those shown in FIGURE *B*, PLATE 1, and FIGURE *D*, PLATE 2, where the cambium cells have persisted, and retained their characteristic appearance, they have evidently ceased to multiply.

It is true that, in each of these instances, the tissue-differentiation has not been vigorous, nor normal in any other respect, but the important point to emphasize is that such differentiation has taken place. Embryonic tissue has either entirely disappeared or its units have lost the peculiar function of such cells, the power of reproduction. Every protoplast passes normally from a period of youth through maturity and old age to ultimate death. At each stage it manifests certain morphological and physiological features peculiar to that stage. In general, the period of youth is marked by both structural and functional plasticity both of which features gradually diminish and finally disappear as old age approaches and advances.

Whatever picture we may try to form as to just what occurs in the protoplast when it is exposed to the rays of radium, the foregoing histological effects seem clearly to indicate that one of the ultimate results is an acceleration of the period of senescence. If this acceleration proceeds gradually enough, the cells and cell-complexes may assume during growth, the various morphological configurations characteristic of the successive ages; but if the acceleration is too rapid, physiological senescence is reached quickly, without the usually accompanying structural changes, while a sufficiently intense over-stimulus by the rays may be quickly followed by complete loss of vitality and death.

In this connection it is interesting to recall the fact that old age

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* Relatively, that is, to either the destructive metabolism, or to normal constructive metabolism.

finds histological expression in a diminished vitality and relative size of the nucleus. Careful measurements with an eye-piece micrometer of the size of cells and of their nuclei near root-tips of *Zea Mays* showed, as I have pointed out elsewhere,[1] that in roots not exposed to radium rays, the cells at a given region near the tips averaged 8.25 scale divisions, and their nuclei 2.75 divisions in diameter. This is the average of a number of cells taken from several different roots. Similar measurements of the same number of cells of roots exposed to radium rays, and at corresponding regions of the root, gave, for the average diameter of the cells, 6.10 divisions of the scale, and of the nuclei 2.17 divisions. In other words, the average diameter of the nucleus of the normal cells measured was 35.5 per cent. of that of the cell, while for roots of the same age, and similarly grown except for exposure to radium rays, the diameter of the nucleus is 33.33 per cent. of the diameter of the cell. This is a cyto-morphological expression of the fact that in tissues of the same "age," the period of senescence is reached sooner than normally after exposure to radium rays. Metchnikoff's[2] view, expressed by his statement, "*on résume la senilité par un seul mot: atrophie*," though probably too narrow for a generalization, is quite in harmony with the observations just described.

In discussing the problem of age, growth, and death, Minot[3] has recently said that, "the growth and differentiation of the protoplasm are the cause of the loss of the power of growth," and that "The older we are the longer it takes us to grow a definite proportional amount." These statements, originally made with reference to the human organism, apply with equal force in the realm of plant physiology, and make it more readily seen how a retardation or even a complete cessation of certain processes may really be an expression of what is fundamentally a stimulation. The facts here reviewed substantiate the conclusion, drawn from other results,* that *radium rays act as a stimulus to living protoplasm.*

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* See p. 157.

CHAPTER XVII

EFFECTS OF RADIUM RAYS ON NUCLEI AND NUCLEAR DIVISION *

Some of the reactions of cells to radium rays have already been indicated.† Koernicke[4] was the first to investigate the influence of the rays on nuclear division, and his results, noted on page 65, need not be restated here.

My own experiments were made with root-tips of *Zea Mays* (aerial roots) and *Allium cepa*. Negative results, so far as mitosis is concerned, were obtained with the corn, as the nuclei, in the control as well as in the exposed plants, proved to be not dividing at the time the material was collected and fixed. The effect of the rays on the relative size of the nuclei in the corn was observed, and has been described on page 229.

Root-tips of *Allium cepa*, grown from bulbs in a moist chamber in the dark, were exposed by placing the sealed glass tubes of radium bromide close to, but not touching them. Thus, as previously explained, only the beta and gamma rays were effective, the alpha rays being screened out by the walls of the tube. Roots for control were grown under precisely similar conditions, except for the absence of radium, and were collected and fixed at the same time and in the same manner as those irradiated.

The material studied was exposed, in part, as follows:

A. For 7 hours 20 minutes to $RaBr_2$ of 10,000 ×. Collected at 7 : 20 P. M.

B. For 52 hours 30 minutes to $RaBr_2$ of 10,000 ×. Collected at 3 : 20 P. M.

C. For 8 hours to $RaBr_2$ of 10,000 ×. Collected at 4 P. M. Other conditions of exposure are mentioned later.

Our incomplete knowledge of the periodicity of cell-division in the onion leaves some doubt as to the best hour for collecting material in order to secure the largest number of divisions. The subject was

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* The substance of most of this Chapter was given before the Botanical Society of America at the Chicago meeting, December, 1907.

† Pp. 181, 187, and 229.

first investigated by Lewis,[5] who found that, in normal light, the greatest percentage of dividing nuclei occurred at midnight, while in roots grown in darkness the lowest percentage was at midnight and the highest at 4 P. M.

Kellicott[3] reported the occurrence of two maxima and two minima in the rate of cell-division during 24 hours. A "primary maximum" was detected shortly before midnight (11 P. M), and a "primary minimum" about 7 A. M. "Secondary maxima" occurred at about 1 P. M., and "secondary minima" at about 3 P. M. No correspondence was observed between the rate of nuclear division and slight variations in temperature. On the basis of this fact the slightly higher temperature in the vicinity of the radium-tube has been disregarded as a factor in the following experiments.

In Kellicott's experiments the roots were grown in moist sand or pine sawdust, while in those of Lewis they were grown in moist air. Since the latter method was adopted in the radium experiments, it is probable that the roots exposed as in *B* and *C* above, if not in those of *A*, were collected at suitable hours for favorable results. At any rate division figures in all phases were very numerous in roots exposed in all three ways.

It has not seemed necessary to give here normal division figures for comparison, as this process in the onion departs little from the typical karyokinesis of the higher plants, and its individual peculiarities are well known through the work of Schaffner,[8]* Němec,[7] Merriman,[6] and Grégoire.[2] Miss Merriman's observations indicate that the number of chromosomes in *Allium cepa*, commonly reported as 16, is not constant, and may vary from 10 to 30 or more. In one instance she figures as many as 38 in one nucleus. In the material used for the radium experiments the number, as shown in the figures (PLATES 5 and 6), was clearly more than 16, and as it appeared to vary in the normal, unexposed roots, any attempt to detect a variation in number as a result of exposure to radium rays was impractical.

All the tissues exposed to the rays blackened more rapidly than those unexposed when placed in the Flemming solution for killing and fixing.

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* Subsequent cell-studies, as is well known, have not confirmed the occurrence of centrosomes in *Allium cepa* (or in any other higher plant) as reported by Schaffner. It is hardly necessary to add that no traces of such bodies were found in my material.

An exposure of not longer than 6 hours and 45 minutes to radium bromide of 1,500,000 activity was sufficient to completely inhibit nuclear division, and a marked tendency to double nucleoli was shown in cells of roots thus exposed. In material exposed for 24 hours to radium of the same strength similar effects were noted, and, in addition, the cytoplasm appeared disintegrated.

In roots exposed for 8 hours to rays from radium bromide of 10,000 activity, and collected at 4 P. M., the nuclei possessed an amoeba-like lobing that was not observed in the unexposed tissues (PLATE 5, FIGURES 1–10). The nuclei in roots exposed for 7 hours and 20 minutes to the same preparation and collected at 7 : 20 P. M. possessed this same lobate appearance and contained from two to three nucleoli. In some instances the nucleoli appeared to be dividing (PLATE 5, FIGURES 9–12).

Practically all of the mitotic figures, in whatever phase, appeared distorted, or abnormal in some other way.* In almost every instance the chromosomes advanced at unequal rates toward the poles of the spindle. Sometimes one or more chromosomes would appear to have been carried beyond the pole, and would then frequently fail to become incorporated in the daughter-nuclei (PLATE 5, FIGURE 17; PLATE 6, FIGURES 2, 5). Again there would be a lagging behind of some chromosomes near the equator of the spindle or at various points between the daughter-nuclei (PLATE 5, FIGURES 16–18; PLATE 6, FIGURES 5, 7–9). In some cells the chromosomes were displaced to one side of the spindle (PLATE 5, FIGURE 17; PLATE 6, FIGURES 4, 7–10), while in others they were distributed with the greatest irregularity all along the spindle fibers from pole to pole (PLATE 5, FIGURES 14, 15). Instances were numerous where one or more abnormally elongate chromatin masses would extend entirely across the spindle, connecting the two daughter-nuclei (PLATE 6, FIGURE 5), or would possess the appearance of having been stretched and drawn out into a fiber at one end or in the middle (PLATE 6, FIGURE 7). All combinations of these irregularities were found in individual nuclei.

In FIGURE 5 (PLATE 6), nine or ten chromatin masses (probably not all individual chromosomes) have failed to take part in the organization of one of the daughter nuclei. FIGURE 2 (PLATE 6) illustrates

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* For an explanation of the conditions of exposure for the figures of PLATES 5 and 6, see page 230.

a condition frequently observed, where some of the chromosomes appear to have been hindered in their advance to the poles, and project out from the mass of the daughter-nuclei along the spindle, extending almost to the equator. In FIGURE 13 (PLATE 5) is shown a tendency of the spindle to separate into two independent and parallel portions, suggesting that, if the process had continued, two separate nuclei would have formed in each daughter-cell. This figure should be compared with FIGURE 1, PLATE 6.

Frequently one or more of the chromosomes that failed to become incorporated into the daughter-nuclei would organize smaller, secondary nuclei, thus giving the cell the appearance of being multinucleate. As many as six of these secondary nuclei were observed in some cells, in addition to the main nucleus (PLATE 6, FIGURES 1, 3, and 6).

One of the most interesting variations observed is illustrated in PLATE 6, FIGURE 1. One nucleus had formed in one daughter-cell, and two in the other, all three appearing abnormal; but, in addition to these, a group of chromosomes that failed to participate in the major mitosis, has organized a secondary nucleus near one wall of the mother-cell, and this nucleus has undergone an independent and tardy karyokinesis, the late telophase of which is shown in the figure. Possibly an early stage of this process is illustrated in FIGURE 17, PLATE 5, and FIGURES 7 and 9, PLATE 6.

Such an instance as this is some evidence that a multinucleate cell in material exposed to radium rays, may not always be correctly explained as due to amitosis. This variation also suggests interesting possibilities in connection with sectorial variation and bud-sporting, and this will be referred to later in Chapter XVIII.

A number of exposures were made of ovaries of *Hemerocallis fulva* and *H. lutea*, to rays from radium of various activities, but, for some unexplained reason, none of these ovaries, nor indeed of those on neighboring plants, set seed, and, as no other suitable plants were at hand in flower when the radium preparations were available, only negative results can be reported on these experiments. The effect of the rays on vegetable cells, however, as just described, confirms Koernicke's[4] results with the pollen-mother-cells and embryo-sac-mother-cells of *Lilium martagon*, and indicates the ability of the rays profoundly to modify indirect nuclear division.

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CHAPTER XVIII

EFFECTS OF EXPOSING GERM-CELLS TO THE RAYS OF RADIUM

At the New York meeting of the American Association for the Advancement of Science (1906–7), I announced[7] before Section G that certain results had been obtained by exposing egg- and sperm-cells of *Onagra biennis* to radium rays, and that these effects were character changes that gave promise, if inherited, of being of specific value. That is, the results visible in ten-weeks old seedlings warranted the expectation that the mature plants would differ from their parent so profundly and fundamentally as to exclude their inclusion within the species of the latter. Individual variation of certain characters would fluctuate about a new mean. If these modifications should prove to be transmitted in sexual reproduction, then the new form would be entitled to at least the rank of the "elementary species" of de Vries.*

The species question, however, is here regarded as secondary to that of variation. By whatever method or combination of methods species are produced in nature, our immediate and fundamental concern should be with the causes and behavior of variations. Variations are the materials out of which species are manufactured, and it is essential in experimental work to center attention on the underlying question of variation before attempting to solve the problem of how nature handles these variations in the making of a new species. All this, in a sense, is a truism, but I state it here for the purpose of making it clear that I do not believe that I have experimentally produced a new species. Nor indeed do I believe it probable that we shall ever do so in the laboratory, at least with the higher green plants. However much we may differ as to what a species is, the term, as now used in taxonomy, always refers to *a group* of organisms. The characters that distinguish the group *as a group* are the truly specific characters, and they develop under the influence of forces that are not only physiological and ecological (in the strictest sense), but also geographical and cosmical. With these facts clearly

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* Or, following Britton's[4] terminology, to the rank of a "race."

in mind it seems almost self-evident that such a natural group of higher plants may not be artificially produced in the laboratory, nor, indeed, within the narrow confines of an experimental garden.

But whether we may artificially produce a parent or ancestor of a species is quite another question. A species has distinguishing marks because the individuals that compose it have those marks, and the group as a whole is separated from other groups of the same systematic rank for at least two reasons: (1) Because its individual members differ from the individual members of the other groups, and (2) because these distinguishing characters, within the range of

FIG. 65. *Onagra biennis.* Permanently Arrested Development. The Ovary, after Pollination, was exposed for 53 Hours to the Rays from Radium Bromide (10,000 ×) Contained in a sealed Glass Tube. Cf. FIGURE 66.

fluctuating variability, are transmitted from parent to offspring in sexual reproduction.

The modification of a specific group, therefore, is an expression of the variation of one or more of its component individuals, and it is clearly conceivable that the variation of only one individual, provided it is of the nature known as discontinuous, and transmissible by heredity, would be quite sufficient material out of which to form a new specific group.

FIG. 66. *Onagra biennis*. Permanently Arrested Development. The same Plant as is shown in FIGURE 65, six Months later. Cf. FIGURE 65.

It was from some such point of view as this that I undertook to see what would result on exposure of the germ-cells, male and female, of *Onagra biennis* to the rays of radium. I chose *Onagra biennis* because it was the only species available when the radium could be had that had a pedigree, and of whose purity I was therefore certain. I chose the germ-cells from a belief that, if they are uninfluenced by the environmental change, the resulting variation is not likely to be transmitted.*

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*Blaringhem,[1, 2, 3] however, has reported the inheritance of certain monstrosities produced in *Zea Mays*, oat, barley, and other herbaceous plants by traumatisms, such as compression, torsion, and cutting.

In the experiments here recorded the radium was used in the form of radium bromide of strengths of 10,000 and 1,500,000 activity, inclosed in sealed glass tubes, and also in the form of Lieber's radium-coated rods of 25,000 activity. Thus, as previously described,* by the use of the sealed glass tubes only the gamma rays and the more penetrating of the beta rays were available, while in the case of the coated rods the α rays could act. The method of treatment was to tie the tube or rod so that it touched the ovary or anthers or both.

FIG. 67. *Onagra biennis.* Functionally asymmetrical Rosette. The Pollen was exposed for 21 Hours to Rays from Radium of 1,500,000 Activity contained in a sealed Glass Tube. Ovary not exposed. Cf. FIGURES 68, 69, and 70.

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* Cf. p. 81.

In some of the experiments the ovary (egg-cell) only was exposed, in others the anthers (sperm-cell) only; in a third set both egg- and sperm-cells were treated. In each of these three cases the exposure preceded pollination and was discontinued after pollination. In still other cases exposure of the ovary was made after pollination with unexposed pollen, and again exposure of the ovary was made while it was maturing and the exposure continued after pollination and until fertilization was presumed to have taken place, or in some

FIG. 68. *Onagra biennis.* Functionally Asymmetrical Rosette. Close Pollination followed an exposure of both Ovary and Pollen to Rays from a Radium-coated Rod of 10,000 Activity for four Days. Cf. FIGURES 67, 69, and 70.

cases until the seed was mature. Throughout the experiments the usual precautions of pedigree-culture methods were followed, such as guarding with bags during pollination, and sowing in soil previously heated sufficiently to kill all seeds it might contain.†

Seeds gathered in September, 1906, after the various treatments described, were planted in soil in the propagating house, some in late September or early October, others in late January or early February, 1907. In no instance was germination completely inhibited by the radium treatment; that is, there was no case observed of failure of any considerable number of seeds to germinate. The percentage of germination seemed unaffected in any instance, though

Fig. 69. *Onagra biennis.* Asymmetrical Rosette. The Ovary was exposed before Pollination for four Days to the Rays from a Radium-coated Rod of 10,000 Activity; Pollen not exposed. Cf. FIGURES 67, 68, and 70.

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† See Shull,[13] p. 256.

rather more than usual variation was shown in the rate of germination of seeds from a given capsule. In all the cultures there were seeds that did not germinate until two months after the appearance of the first seedling in the same culture. Nor did the plants from the seeds that germinated late vary more in structure than any of the others.

One plant that followed exposure of the ovary after pollination with radium of 10,000 activity for 53 hours, never passed beyond the rosette stage. The stem elongated, lifting the rosette about 15 cm. above the surface of the soil, and one very short branch developed on the lower part of the stem (FIGURE 65). The rosette persisted all winter after the plant was removed from the experimental garden to the propagating house, the old leaves dying and new ones forming. But the plant finally died soon after it was photographed in the following spring (FIGURE 66). There was here a complete loss of reproductive capacity, and a generally diminished vitality. Careful examination during transplanting failed to disclose any fungus disease or other unfavorable circumstance that might account for the arrested development.

Among other effects that followed exposure to the rays, the following are worthy of mention:

1. FUNCTIONAL ASYMMETRY: FIGURES 67 and 68 illustrate this. These rosettes show a variation in leaf-character which is doubtless not to be attributed to the influence of the rays, falling as it does within the range of fluctuating variability. The asymmetry of the rosette followed exposure of either egg- or sperm-cell, and was estimated to occur in about one per cent. of the plants. It may result from either a retardation of growth on one side or an acceleration of it on the other, presumably the former.

In FIGURE 69, however, is shown an asymmetrical rosette from a seed whose ovary had been exposed for four days to a radium-coated rod of 10,000 activity. At the close of this exposure the stigma was pollinated with unexposed pollen. In this rosette the leaves on the more vigorous side are crisped, resembling the leaves of *O. Lamarckiana*, and some other species, but not typical in *O. biennis*. On the other side of the rosette the leaves are of the normal *biennis* type. If we look upon the crisping as an expression of growth-vigor, then the asymmetry of this rosette is logically to be attributed to an acceleration of growth on one side. Evidence warranting this interpretation is wanting in the case of the two rosettes first described.

Asymmetry of the rosettes is not unknown in other species of the evening-primrose without special antecedent treatment,* but its frequency in the radium-cultures, and more especially the subsequent behavior of the asymmetrical plants, and which is lacking in unexposed specimens, seems to justify the inference that, in these instances the asymmetry is a result of the exposure to the radium rays.

FIG. 70. *Onagra biennis.* Functionally Asymmetrical Rosette from a Seed whose Ovary was exposed for 53 Hours after Pollination to Rays from Radium of 10,000 Activity in a sealed Glass Tube. The Cauline Stem is beginning to grow horizontally, leaning toward the narrower Portion of the Rosette.

This subsequent behavior is illustrated in FIGURE 70, which is a

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* MacDougal,[8] FIGURE 13. A hybrid between *O. Lamarckiana* and *O. biennis.*

photograph showing a later stage of development of a plant whose rosette was asymmetrical. The main stem is developing, but instead of growing vertically up, as normally, it is bent over, growing nearly horizontally. In each instance of this kind the stem bends over toward the smaller side of the rosette, showing that growth in length is more rapid on the other side of the plant. The photograph also shows that on this main stem the leaves on the more rapidly growing side are larger than those on the opposite side. The tip in such plants is usually turned up, apogeotropically, as shown in the figure, and the stem eventually begins to grow vertically.

As to how the radium rays acted in order to bring about the effects just described, of course we do not know. That the asymmetry was not due to differences of illumination or to crowding is certain from the known conditions under which the plants were grown. Recalling the nutation of a growing stem, caused by the fact that the region of most rapid growth travels around the stem in a direction opposite to that of the nutation, it is possible that the asymmetry of the rosette and the bending of the stem is due to an inhibition of the migration of the region of maximum growth. Thus the effect is analogous to the modification of nutation by a unilateral stimulus in any tropism.

2. Morphological Asymmetry: Superficially resembling functional asymmetry, are results of which FIGURE 71 shows an example. Here the rosette is one-sided, but, on more careful observation, it is seen that the one-sided appearance is due to something more than mere difference in rate of growth. The leaves on one side of the plant are not only smaller than those on the other, but they are of different shape, being narrower in proportion to length, and with the margin of the basal portion not notched as normally. Furthermore the transition from the character of one side to that of the other is not absolutely abrupt. The individual leaves between the unlike sides are themselves asymmetrical, the side of the leaf next to the broader leaves of the rosette being wider from midrib to margin than the other side. The tips of these bilaterally asymmetrical leaves turn toward the narrower side of the leaf.

Here, of course, is functional asymmetry, but because the form also is modified and the asymmetry thus accentuated, I have called this result *morphological asymmetry*. There is a qualitative as well as a quantitative difference.

The morphological asymmetry was not confined to the rosette stage, but persisted throughout the entire life of the plant, giving narrow leaves on one side of the narrow upright stem, and on the secondary branches growing in their axils, and broad leaves on the opposite side and branches (PLATE 7, a different plant than FIGURE 71).

FIG. 71. *Onagra biennis.* Morphological Asymmetry. From a Seed developed in an unexposed Ovary whose Stigma was pollinated with Pollen exposed for 24 Hours to Rays from Radium of 1,500,000 Activity in a sealed Glass Tube. Cf. PLATE 7.

The difficulty of explanation is greater here than in the former case. The rosette (FIGURE 71), bearing leaves of both kinds suggests that a fertilized egg may have been unequally influenced by the radium rays, but such a condition is excluded by the fact that only the pollen was exposed. An untreated ovary was pollinated with pollen that had been exposed for 24 hours to the rays from radium bromide of 1,500,000 activity. There seem to exist in the germ-cells of this species two factors expressed in the mature organism by a different ratio between the length and breadth of the leaves. Most frequently the broad-leaved type appears, while at times, under some unrecognized environmental stimulus, the narrow-leaved form results. The radium rays may affect this unit-character in either germ-cell, and, when we recall that, after fertilization, the male and female chromatins do not fuse until synapsis immediately preceding the reducing mitosis in the sporogenesis of the mature zygote, it is not difficult to imagine how a plant with two unlike sides might result from altering the nature of either chromatin-mass.

In the normal production of an elementary form by mutation, the mutation is believed by de Vries,[15] to be " decided within the seeds." There is no experimental evidence, however, for not considering that, in sexual reproduction, the change may occur at any point in the life-cycle of the germ-plasm, at least from gametophyte mother-cell on, in either the maternal or the paternal line, or in both.

We may conceive of the morphologically asymmetrical plant as the result of some such sequence of events as follows:

1. A destruction (or change from a dominant to a recessive condition) by the radium rays, of the factor in the exposed pollen essential for the production of the *biennis* type of leaf.

2. By the fertilization of a presumably normal *biennis* egg by a sperm-cell from this pollen, an oösperm may result containing factors representing different peculiarities of leaf-form from each parent.

3. A unilateral expression of these peculiarities in the resulting plant. This step would result from a division of the fertitized egg in its first and subsequent mitoses in such a way as to confine the maternal chromatin to one side of the organism and the paternal chromatin to the opposite side. This would offer a reasonable explanation, also, of the transitional leaves between the opposite halves, for the primordia of these leaves are probably composed of adjacent cells from each side of the plant, thus giving rise to the observed bilateral asymmetry of the leaves. The plant, then, so far as this one feature is concerned, is really a hybrid between two elementary forms. The leaf-character of one of the parents has formerly existed only potentially in the male germ-cell, and finds morphological expression for the first time in the offspring of the first generation.

In " The Variation of Animals and Plants under Domestication," Darwin[5] records the testimony of Salter who, he says, " informs me that at first a branch often produces variegated leaves on one side alone, and that the leaves are marked only with an irregular edging or with a few lines of white or yellow. To improve and fix such varieties he finds it necessary to encourage the buds at the bases of the most distinctly marked leaves, and to propagate from them alone."

In his paper on infertile hybrids, Wilson[16] describes a zonal pelargonium-hybrid of interest in this connection. In making a cross between two variegated zonal pelargoniums, the variegation of the seed-parent, he says, was of the usual kind, "the peripheral zone of white enclosing a green center and sending into it projections of more or less intense variegation. In the pollen parent the variegation, also white, occupied the center and margin of the leaves. . . . The seedlings resulting from the cross were in the majority of cases non-variegated and coarse. A few were variegated from the first, but only one has been made special note of. Its cotyledons were blotched with white, etc. . . . Very soon three distinct vegetative regions were differentiated in the seedling under discussion; one including leaves with normal chlorophyll development, the next with variegated leaves, and the third with leaves quite destitute of chlorophyll. If a leaf arose in a plane between any two regions it embodied in itself features of both. . . . Ultimately a branch lying wholly in each region was produced. Variegation was only once seen in the green branch, a small patch of white occurring in one leaf. The variegation of the variegated branch was identical with that of the seed-parent. The albino portion showed marked persistence. . . . No trace of green was seen in the branches."*

Here we have recorded the case of a known hybrid in which the characters entering into combination expressed themselves in such a way that only one set of characters appeared on one side of the plant, the other set on another side, while the organs in an intermediate position partook, in bilateral fashion, of both sets of characters.

It is in view of the above facts and considerations that I believe that the morphologically asymmetrical evening-primrose is fundamentally a hybrid, and that its asymmetry may be due to a segregation, in opposite sides of the plant, of the characters brought together in the cryptomeric crossing.

One other possible explanation of this plant must not be overlooked, and that is an interpretation of it as a bud-sport in which only one half of the bud was affected. In this particular instance, and on this theory, the sectorial variation may be an expression of a change from dominancy to latency of some of the specific *biennis*-

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* The white portion was regarded as a parasite on the green portion.

characters, the effects being distributed unilaterally throughout the plant as a whole because of the fact that the sporting occurred in the terminal bud of the entire shoot-system. Even so, the initiation of the sporting may be logically attributed to the radium rays, their effects being brought into the offspring through the sperm-cell. But thus we are brought back again to the idea of the plant as fundamentally and essentially a hybrid.

Possible Induction of Mutation: The appearance in the radium-cultures of elementary forms already recognized in normal pedigreed cultures was rather to be expected, and the occurrence of such a form is to be attributed to the influence of the rays only with great caution. A description and discussion of a few aberrant forms that appeared after the radium treatment follow.

Among the progeny from an unexposed pistil whose stigma was pollinated with pollen that had been exposed to rays from radium bromide of 1,500,000 activity for 24 hours, there was found a seedling with unusually narrow rosette leaves. Some of the characters displayed by this plant at maturity are shown in the photograph, PLATE 8. The narrowness of the rosette leaves is seen to have persisted throughout the life of the plant. Furthermore its habit of growth differs considerably from that of a mature *biennis*. In numerous characters pertaining to the buds, petals, and mature capsules, it differs from the *biennis* type. The specimen closely resembles an elementary form observed by MacDougal[9] in a normal pedigreed culture, and described by him as a mutant. Other plants like this one followed the treatment with the radium.

The plant shown in PLATE 9 (5*c* of my cultures) is from seed produced in an unexposed capsule with the stigma pollinated with unexposed pollen, and then exposed to rays from radium bromide of 1,500,000 activity for 48 hours after pollination. It differs, not only from the typical *biennis*, but also from the other variant forms obtained. Following is a systematic description of the plant,* together with the description of *O. biennis* as given in Britton and Brown's "Illustrated Flora."

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*I wish to express here my best thanks to Dr. John K. Small, of the New York Botanical Garden, for writing all of the systematic descriptions in this chapter. They were written without any knowledge on Dr. Small's part of the antecedent history of the specimens.

Onagra biennis	*Onagra 5c*
Erect, generally stout, annual or biennial, simple and wand-like or branched, 1°–9° high, more or less hirsute-pubescent, rarely glabrate. Leaves lanceolate, acute or acuminate, narrowed and sessile at the base or the lowest petioled, repand-denticulate, 1′–6′ long; flowers spicate, terminal, leafy-bracted, bright yellow, open in the evening, 1′–2′ broad; calyx-tube slender, much longer than the ovary, the lobes linear, contiguous at the base, reflexed; capsules oblong, narrow above, erect, pubescent, ¾′–1′ long, 2½″–3″ thick, nearly terete, seeds angled.	Rosette leaves finely and sparingly pubescent, the larger ones 12–14 cm. long; blades narrowly linear-lanceolate, acuminate at both ends, undulate, much longer than the petioles; stems 7 dm. tall, with several rather large ascending branches below the middle; stem-leaves drooping; blades almost linear, often narrowly so, tapering to both ends, entire; bracts similar to the stem-leaves, but slightly smaller; hypanthium about 40 mm. long; sepals about 20 mm. long, fully one half as long as the free portion of the hypanthium, the free tips in the bud stout, 2.5–3 mm. long; petals about 15 mm. long, retuse at the apex.

Why, in the two aberrant plants described above, the entire specimen showed the changed characters, instead of one half only, as in the case of the morphologically asymmetrical specimen, cannot of course be said. Possibly the pollen-grains were differently affected in the different exposures to the rays, possibly, and quite probably, the mitoses that followed fertilization were different, resulting in the one instance in a segregation of the parental chromatins, but not so in the other case, or possibly the eggs that were fertilized by the irradiated pollen were unlike, or there may have been a combination of any two or of all three of these possibilities. Attention should also be called to the fact that these aberrant forms may not be results produced by the rays of radium, but only spontaneous mutations, whose cycle happened to coincide with the time of the experiment.

Of far greater interest were the two plants illustrated respectively in PLATE 10 (10*a* of my cultures), and PLATE 11 (11*b* of my cultures). These plants are as unlike each other as they are different from the parent *biennis*, or any of its hitherto observed mutants. The seed that produced 10*a* came from a capsule exposed after pollination with unexposed pollen for 53 hours to rays from radium bromide of 10,000 activity. Both sperm- and egg-cells, therefore, were exposed to the rays. A description of the plant follows:

Rosette leaves finely and rather copiously pubescent, the longer ones 9–10 cm. long; blades oblong to oblong-spatulate, undulate, longer than the petioles; stems 3 dm. tall, with very long spreading branches on the lower part; stem-leaves spreading; blades oblong to oblong-linear, acute at the apex, shallowly undulate-sinuate; bracts similar to the stem-leaves but much smaller; hypanthium 35–45 mm. long; sepals about 15 mm. long or shorter, one half as long as the free portion of the hypanthium, the free tips in the bud stout, 1–1.5 mm. long; petals 15 mm. long, or smaller, nearly truncate at the apex; capsules columnar, slightly tapering to the apex, about 20 mm. long, rather broader than the bracts. (See PLATE 12, FIGURES *f–k*.)

The second specimen (PLATES 11 and 12) is equally as distinctive. The spreading tips of the calyx in the bud (PLATE 12, FIGURES *c* and *d*) indicate the pressure of the stigmatic lobes within, and in many of the buds these lobes force their way through the tip some time before anthesis, a feature seldom, if ever, observed in *biennis*, and favoring cross-pollination instead of the close pollination characteristic of *biennis*. The plant, about 6 dm. tall at maturity, produced flowers, fruit, and seed in great abundance. The leaves were of a slightly darker green than is usual in *biennis*, and both they and the bracts are

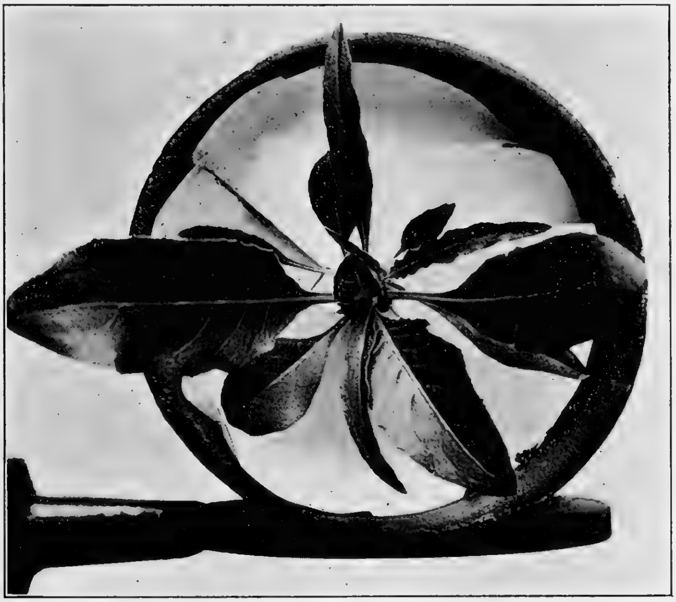

FIG. 72. *Onagra biennis*. Radium Culture, No. 15*a*. Two Rosettes, one narrow leaved, and one broad leaved, on one Tap-Root. Cf. FIGURE 73, and PLATES 13 and 14.

strikingly thick and almost brittle. The radium was of 1,500,000 activity, and the period of exposure 24 hours. With pollen thus treated the stigma of an unexposed pistil was pollinated, and this plant was from one of the resulting seeds. Below is the taxonomic description.

Stem 6 dm. tall, with several erect-ascending branches from near the base, nearly as tall as the stem; stem-leaves spreading; blades lanceolate to oblong-lanceolate, acute at the apex, undulate-sinuate; bracts similar to the stem-leaves, but shorter and relatively broader; hypanthium about 35 mm. long; sepals about 15 mm. long, or shorter, fully one half as long as the free portion of the hypanthium, the free tips in the bud stout, about 1.5–2.5 mm. long; petals nearly 15 mm. long, notched at the apex; capsules columnar, more or less tapering at the apex, nearly 20 mm. long, much narrower than the bracts. (See PLATE 12, FIGURES *f–k.*)

The most interesting and novel result of all was the plant shown in FIGURES 72 and 73, and in PLATES 13 and 14. The antecedent treatment was an exposure of the ovary for 24 hours to the rays from radium bromide of 10,000 activity, after which the stigma was pollinated with unexposed pollen. Among the young seedlings in the seed-pan, I thought I had detected one of the narrow-leaved variety growing close to a more typical plant. But when I started to separate the two plants for re-potting, I found that, instead of two plants, I had only one, this one bearing two rosettes on the same root (FIGURE 72). The anomaly cannot be called a bud-sport, as that term is generally used, unless, keeping in mind that the plumule is a bud, we decide that there was an early bifurcation in the developing embryo, of such a nature that, after the cotyledons were laid down, there was a division of the growing-point, accompanied by a separating out of antagonistic characters, and resulting in the formation of two morphologically as well as physiologically different shoots.

The seedling was very carefully freed from soil, and after thorough examination, the above conclusion seemed to be the only one warranted. There was absolutely no evidence that a lateral bud had formed early on the main stem. There were to all appearances two epicotyls, one in the axil of either cotyledon. This plant was carefully protected, and after it was transplanted into the experimental garden both rosettes sent up cauline stems, in which the differences, so marked in the rosettes, were continued (FIGURE 73). One half of the plant, as is clearly shown in the illustration, was a typical *O. biennis* in every respect. The other and narrower leaved half, in general appearance and behavior, was similar to the narrow-leaved form described above, and illustrated in PLATE 8. This plant, in light of its method of treatment, offers evidence, in addition to what we already possess, that mutation is not confined to the period during and after fertilization, but that it may occur, or at least be initiated previous to fertilization, and in either the male or the female gamete. Taxonomic descriptions of the two halves of this plant (15*a* broad and 15*a* narrow of my cultures) are appended.

15*a* broad

Rosette leaves finely and sparingly pubescent, the larger ones about 15 cm. long; blades spatulate to elliptic-spatulate, sinuate-dentate especially below the middle, often sharply or prominently so near the base, much longer than the petioles; stem 7 dm. tall, with elongate ascending branches on the lower part; stem-leaves mostly spreading; blades narrowly elliptic, somewhat acuminate at the apex, sinuate-dentate; bracts similar to the stem-leaves but smaller and usually broadest below the middle; hypanthium about 35 mm. long; sepals about 25 mm. long, slightly shorter than the free portion of the hypanthium, the free tips in the bud subulate, 2.5–3.5 mm. long; petals about 15 mm. long, nearly truncate at the apex; capsules columnar, slightly narrowed to the apex, about 35 mm. long, much narrower than the bracts.

15*a* narrow

Rosette leaves finely and sparingly pubescent, the larger ones 10–12 cm. long; blades almost linear, acuminate at both ends, undulate, somewhat longer than the petioles; stem 6.5 dm. tall, with relatively short ascending branches throughout; stem-leaves mostly drooping; blades narrowly linear-lanceolate to almost linear, tapering to both ends, undulate-sinuate; bracts similar to the stem-leaves but smaller; hypanthium about 45 mm. long; sepals about 30 mm. long, fully as long as the free portion of the hypanthium, the free tips in the bud long-subulate, 3–4 mm. long; petals about 20 mm. long, nearly truncate at the apex; capsules almost columnar, about 25 mm. long, slightly narrower than the bracts.

A functional difference between the two halves is shown by the fact that after the broad-leaved half was entirely through flowering, the narrow-leaved portion still bore opening buds and flowers. The equivilancy of the two shoots was ultimately obscured by the more vigorous and rapid development of the broad-leaved half.

Let us briefly consider this plant in light of the morphologically asymmetrical plant described on page 243. Here is also morphological asymmetry, more perfectly accomplished than in the earlier mentioned case, and participated in, not by the leaves alone, but by the entire shoot. In the former instance the entire main axis of the plant is presumably physiologically double, though structurally one, but in the latter plant the difference is expressed in the splitting up of the entire shoot-system into two main axes, each with its own secondary branches and characteristic foliage.

FIG. 73. *Onagra biennis.* Radium Culture, No. 15*a*, showing two distinct Shoots, morphologically unlike, on one Root. Cf. FIGURE 77, and PLATES 13 and 14.

As with the earlier described plant, this one may be interpreted as a kind of bud-sport, but calling it a bud-sport does not explain it. Bud-sporting in pedigreed plants is not unknown, and has been described for pedigreed evening-primroses by MacDougal,[9] who says that a possible hybrid ancestry is indicated by the bud-sporting of

"*Oenothera*"* *ammophila* into characters of *O. biennis*. But where a seed-mutant of *biennis*, from carefully guarded seed, produced by a pedigreed plant, gave a bud-sport bearing the characters of the ancestral type or true *biennis*, the significance of the bud-sporting as pointing to the hybrid nature of the plant seems to be excluded.

However, in the cases of the morphologically asymmetrical plant and the plant with two diverse shoot-systems we know that, in the first case the sperm-cell and, in the second the egg-cell were subjected to a treatment (exposure to radium rays) which has the power to affect marked change in the chromatin, as is shown in Chapter XVII. We further know that such exposure has, in another experiment been followed by the appearance of a plant similar to a type that has previously appeared by spontaneous mutation.

If one of the altered gametes takes part in an act of fertilization, the resulting zygote, as already suggested (p. 245), is fundamentally of hybrid nature; as truly so, indeed, as when, without experimental treatment, a female gamete of one elementary form is fertilized by a male gamete of another. Whether the characters, represented potentially in the gametes by certain factors, have previously found expression in a zygote in the direct ancestral line is immaterial, and, for the purpose in hand, of wholly secondary importance. A zygote is a hybrid, not because the parents of the fusing gametes are different, but because the gametes themselves differ. A mature plant of a mutant of *O. biennis* has been known to bud-sport into the parental ancestral form. If this mutation had taken place in one of the sperm-cells of a pollen-grain of this mutant, instead of in the primordium of a bud, and if subsequently a "true" *biennis* egg had been fertilized by that sperm-cell, the resulting zygote would, in reality, not have been a hybrid† at all, though its parents were distinct elementary species. Such a zygote might, however, be classed as a "cryptohybrid" of Tschermak.[14] At any rate we see that the two cases of the bud-sporting of *Oenothera ammophila* into a true *biennis* branch, and the production, by a seed-mutant of *Onagra biennis*, of a bud-sport bearing the ancestral characters, are capable of the same interpretation, viz., that the sporting plant in each instance was, *in*

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*This species (*Oenothera ammophila* Focke, Abh. Nat. Ver. Bremen **18**: 182. 1904) is closely related to *Onagra biennis*, but does not appear to have received a binomial name in *Onagra*, and it does not seem advisable to rename it here.

†In the customary sense of the term.

reality, a hybrid in which the unlike characters separated out in the course of the cell-divisions involved in the formation of a bud.*

The nature of any germ-cell may be altered in one of three ways:

1. By the acquisition of one or more new factors not previously present.

2. By the change of any factor from the domination to the recessive condition, or from the recessive to the dominant.

3. By the complete loss of one or more factors.

It is hardly probable, reasoning from other known facts, that the acquisition of a new factor could be accomplished by exposure to the radium rays, but it is quite conceivable that, by such treatment, a factor might be changed from the dominant to the recessive condition, or that a complete loss of a factor might result. Shull's[12] experiments with hybrid beans led him to suggest the hypothesis that unit characters are determined by the simultaneous action of two or more dominant factors or units in the germ-cells, and "that the later specific or varietal derivatives were produced by the disappearance of one or more of these original units as a dominant characteristic." Thus, if the original character is determined by the dominant units *ABCDEFGH*, "the later derivatives may be *ABCDEFGh*, *ABCDEFgH*, *ABCDEfgH*, etc., through all the possible permutations . . . This conception results in an interesting paradox, namely, the production of a new character by the loss of an old unit."

This hypothesis seems to offer a plausible explanation of the possible induction of mutation by exposing either one or both of two conjugating gametes to the rays of radium: and furthermore it discloses a possible mechanism, such as is demanded by the theory that the morphologically asymmetrical plant (FIGURE 71) and the specimen with two shoot-systems (FIGURES 72 and 73), though falling under the head of bud-sports, are fundamentally hybrids. Here also lies the warrant for at least one interpretation of the significance of bud-sporting, in general, as pointing to the fundamentally hybrid nature of the organism thus sporting. This, by no means, excludes the other interpretation of a bud-sport as a mutation, pure and simple, taking place in the somatic cells during the formation of a bud, rather than at some stage in the formation of the germ-cells.

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* This discussion was written in December, 1907, before the appearance of East's[6] paper on bud-variations (April, 1908). Metcalf[11] has also recognized that bud-variation and seed-variation are practically identical.

In conclusion then, the double plant may be interpreted as, in reality a hybrid between elementary forms, the characters of one of which had had expression only potentially in the conjugating gamete. In normal cases of hybridity every cell in a given plant of the first generation resulting from the cross is of hybrid nature. The characters of the two parents are diffused throughout the entire organism, though, in Mendelian cases, they may segregate in the F_2 and subsequent generations, according to the well-known laws of Mendel. But, in the case of the double plant, segregation of characters occurred before the embryo was complete. Not every cell in the mature plant, then, is of hybrid nature (considering merely the characters that distinguish the two shoot systems), though hybridity may be postulated of the organism as a whole.

Discussion of the possible causes that might produce a plant with two different shoot systems on one root should not be closed without reference to the case of anomalous mitosis illustrated in PLATE 6, FIGURE 1, and described on page 233. There it is seen that two mitotic spindles have developed within one cell. Without going into details, it is merely noted here that such a result in nuclear division suggests that some such departure induced in a fertilized egg-cell by radium rays, might have been the first step in the production of the double plant of *Onagra biennis*.

HERITABILITY OF INDUCED CHANGES: Carefully guarded, and also unguarded* seed, of the aberrant forms 10*a*, 11*b*, 15*a* broad, and 15*a* broad × 15*a* narrow were collected, and the second generation of plants is now (June, 1908) under observation. Attempts to secure seed from 15*a* narrow were unsuccessful, as were also attempts to cross this form with pollen from 15*a* broad. Seed was obtained, however, by crossing 15*a* broad with pollen from 15*a* narrow, but only one seedling from this cross appeared in the seed pan.

Thus far inheritance of the variations is manifested in only one instance, viz., 11*b*. Two plants in the second generation, from unguarded seed resemble the parent in every fundamental character, while a third resembles it in every way except in the flower-bud, where the character of the stout, spreading tips of the sepals is lacking, being replaced by the *biennis* character. It is proposed to con-

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* My absence from these plants at the time the seed was maturing resulted in the failure to secure carefully guarded seed in some instances.

tinue the observations over the second generation of both 11*b* and 15*a* broad × 15*a* narrow. It must be concluded, then, that most of the variants were not true mutations, and that further evidence is needed before we may say with entire confidence that mutation may be induced by the stimulus of radium rays.

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CHAPTER XIX

THEORETICAL CONSIDERATIONS

It has been clearly shown that radium rays act as a stimulus to plants, and are therefore able to modify their life processes. In certain cases the reaction to the rays is an excitation of function, in other cases a depression. But as to the method by which the stimulation is brought about, as to just how the protoplast is affected by the rays, we are in almost complete ignorance. Nor is the writer bold enough to essay an answer to these questions here. The final solution of the problem, however, will involve a careful consideration of certain facts and theories which may now be reviewed.

THE TERM STIMULUS is employed here and throughout in the sense emphasized by Verworn,[1, *] as any "change in the external agencies that act upon an organism." With reference to any individual protoplast this change will involve factors outside the protoplast, but not necessarily outside the organism of which it is a part. These external agencies are forms of energy,[†] of which Verworn[1, ‡] lists eight, viz., chemical, molecular, mechanical, thermal, photic, electrical, magnetic,[§] and energy of gravitation. In Chapter II of this Memoir, it was pointed out that to these forms of energy, hitherto commonly recognized, must be added that of radioactivity. It was furthermore shown that radioactivity is a factor in the normal environment of probably all plants. Any change, therefore, in this factor becomes a stimulus.

THE BIOGEN HYPOTHESIS: Before we can form any conception of the *modus operandi* of any stimulus we must have some sort of picture of the constitution of the cell. That protoplasm is not a chemical entity, but a morphological one, is generally accepted. It is also recognized that that which fundamentally distinguishes it from lifeless matter is its power of metabolism. One of the most thor-

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[*] Loc. cit., p. 348.

[†] Or modify some form of energy, as when one plant is shaded by another.

[‡] Loc. cit., p. 209.

[§] Verworn[1] later (l. c., p. 348–349) states that the only ones of the above classes that come into relation with the organism are the first six and the last, but Ewart[59] succeeded in modifying the rate of streaming of protoplasm in living cells and the rate of motion of spermatozoids by the influence of strong magnets.

oughly worked out theoretical conceptions of protoplasm is the "biogen hypothesis" of Verworn,[1, 2] and the details of this hypothesis are sufficiently tangible to be of service in an attempt to form some provisional conception as to how radium rays, or any other stimuli, produce their effects. I will first briefly state some of the fundamental notions of the biogen hypothesis and their bearing upon radium stimulus, and then, in a similar manner, some of the facts concerning a few physiological processes, made known by recent investigation.

According to Verworn's conception, "The metabolism of living substance, in last analysis, depends upon the continual destruction and the continual reconstruction of a very labile chemical compound." This "hypothetical compound, because of its fundamental relation to the genesis of life-processes," Verworn[2] designates * as "biogen," and, since in different forms of living substance there doubtless occur very different compounds of this sort, he designates "the entire group of them in a chemical sense as the group of the 'biogens,'" and proposes the term "biogen molecule" to supplant that of "living protein molecule" of Pflüger. The biogen is designated * as a "most highly complex, labile compound that develops at an intermediate point in metabolism, and by its construction and destruction comprehends the sum total of the processes of metabolism." It is not a protein body, nor would the author call it a living protein. It is not alive, for a molecule cannot be alive.

The essence of metabolism, then, is the construction and destruction of biogen molecules, and, under normal conditions of equilibrium $\frac{\text{Construction}}{\text{Destruction}} = 1$. On the basis of this hypothesis, "the irritability of living substance depends upon the lability of the biogen molecule."

"In metabolism as a whole two different series of processes are to be distinguished: 'functional metabolism,' in which the absolute number of biogen molecules remains unaltered and only certain nitrogen-free groups are involved in functional destruction and restorative construction; and 'cytoplastic metabolism,' which governs the absolute number of biogen molecules, and thereby the phenomena of growth, propagation, development, atrophy, regeneration, etc., since it extends over the destruction and reconstruction of the entire molecule. In case of a disturbance of their metabolic equilibrium, a compensatory self-regulation underlies both series of processes, and

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* Loc. cit., p. 25.

the biogen molecule offers a simple mechanical explanation for individual cases of this sort. . . ."

Analyzing the phenomena of stimulation on the basis of his hypothesis, Verworn[2] says:* "Irritability is the capacity of living substance to react to a stimulus by an acceleration of the metabolism of biogens." Both the dissimilatory and the assimilatory phases of metabolism may be stimulated. The degree of dissimilatory stimulation is, for equally intense stimuli, dependent upon the following factors:

(*a*) The degree of lability of the biogen molecule.

(*b*) The rapidity of the process of restitution after the functional destruction of the biogen.

(*c*) The absolute number of biogen molecules present.

(*d*) The conditions for the propagation of the effects of stimulation.

A dissimilatory stimulation, or depression, may therefore be brought about by influencing any one of these individual factors.

On the other hand, the degree of assimilatory irritability is dependent upon:

(*a*) The quality of the raw materials available for nutrition.

(*b*) The means for working up the raw material into a suitable form of elaborated matter.

(*c*) The quantity of the suitable elaborated matter.

(*d*) The rapidity of the transformation of the elaborated matter from the reserve depots into the biogen molecules.

An assimilatory irritability or depression may arise through influencing each of these individual conditions.

Radium rays, by acting on any one of the eight factors enumerated above, may therefore excite or depress processes of either assimilation or dissimilation. Now Verworn[1] has earlier explained † that the atoms of his hypothetical biogen molecule are in active vibration. "As a result of this, certain atoms come occasionally into the sphere of attraction of others, and, becoming united with them into a more fixed combination, separate off as an independent molecule. In this way the spontaneous dissimilation of the biogen molecule results." The residues of biogen molecules thus formed may combine with constituents of the food, and thus be rebuilt into

* Loc. cit., p. 89.

† Loc. cit., p. 489.

a whole biogen molecule. "Thus spontaneous regeneration of the biogen molecule follows spontaneous dissimilation."

It is evident, as Verworn points out, that any factor that increases or decreases the vibration of the atoms within the biogen molecule will correspondingly modify metabolism, causing either excitation or depression in response to the stimulus. It is conceivable that the radium rays, through their power of ionization, which alters the electrical charge of the atom and the electrical relations within the molecule, may modify this intramolecular atomic vibration, and so produce either an acceleration or retardation of any given process or processes, or an acceleration of one or more, and, at the same time, a depression of the others.

But it is quite probable that radium rays, and doubtless other stimuli also, may not produce their stimulatory effects by acting directly upon the biogen molecule, or whatever the reality may be that corresponds to this term, but by acting upon some other substance in the individual cells, or by modifying some process either preceding or following the elaboration of the biogen molecule. In other words, the rays may act, not upon the more immediate physical basis of life, but upon some non-vital constituent other than the biogen, or upon some purely chemical process, thus producing their effects indirectly. The possibility of this is evident on the basis of Verworn's hypothesis, as may be seen by referring to the factors involved in his analysis of dissimilatory and assimilatory stimulation. It is more clearly evident when we analyze various physiological processes in the light of recent investigations. To this end let us consider first the process of photosynthesis.

PHOTOSYNTHESIS: The work of Usher and Priestley[11] may first be referred to. These investigators adduce experimental data which lead them to the conclusion that the process called by them "carbon assimilation" consists of at least three steps, as follows:

1. The conversion of CO_2 and H_2O into CH_2O (formaldehyde) and H_2O_2. In this process the chlorophyll acts as an optical sensitizer, and the vitality of the cell is not involved.

2. The formaldehyde is removed and condensed to a sugar *by the protoplasm.*

3. The H_2O_2 is removed by being split up by an enzyme into H_2O and O, and the O set free as a gas. Thus all the reducing processes are non-vital in character, and the living protoplasm functions only in condensing the formaldehyde to sugar.

This second step has been accomplished in the laboratory and outside the organism by Meldola,[10] but the gap between CO_2 and formaldehyde was not bridged until Fenton[5] reduced the former to the latter in one step, with the aid of metallic magnesium, without the intervention of the formic acid stage. At about the same time (January, 1908) Gibson[6] and his collaborators, by means of a feeble electric discharge, succeeded in synthesizing the formaldehyde from CO_2 in the presence of water. The securing of this result forms part of the experimental demonstration of Gibson's photoelectric hypothesis of photosynthesis. This hypothesis, in brief, is, "that the light rays absorbed by chlorophyll are transformed by it into electric energy, and that this transformed energy effects the decomposition of carbonic acid (H_2CO_3) within the cell, with the concomitant formation of an aldehyde and the evolution of oxygen.

The following facts in confirmation of this hypothesis have been demonstrated in Gibson's laboratory: (1) That formaldehyde is present in all actively photosynthetic tissues; (2) that the amount of formaldehyde present in the leaf bears a definite relation to the intensity of illumination; (3) that formaldehyde may be synthesized from CO_2 in the presence of water by feeble electric discharge; (4) that electric discharges of sufficient intensity occur in photosynthetic tissues when they are adequately illuminated; (5) that the light rays absorbed by chlorophyll are those specially concerned in the generation of the electric currents which Kunkle,[9] Haacke,[7] and others * have demonstrated to exist in chlorophyll-bearing tissues. Gibson's hypothesis varies from all others in regarding the electric currents as due to the transformation of the energy of the light rays, and in attributing the formation of the formaldehyde from CO_2 and water to the electric energy thus derived.

Whether further experimentation shall confirm these results or not, it is certain that several steps are involved in photosynthesis.† Several possibilities are open, therefore, as to the way or ways in which radium rays or any other stimulus may affect the process. The rays may affect any one or all, or any combination of two or more of these steps, or they may modify the power of the plastids to produce the necessary chlorophyll, or to convert sugar into starch.[3]

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* See citations Nos. 8, 49, 50, 51, 53–58.

† Kimpflin [28a] (1908) considers that there are two distinct acts in the assimilation of carbon: 1. The production of electricity, which, by ionizing the water will increase the amount of hydrogen in the nascent state. 2. The reduction of CO_2 by that

We know, from other experiments, that the rays can affect living protoplasm, and it therefore seems certain that their effects on photosynthesis are brought about, in part at least, in that way.*

As to the effects of radium rays on chemical analysis and synthesis we know next to nothing. If, as now seems highly probable, the conversion of carbon dioxide and water into formaldehyde and hydrogen peroxide is the first step in photosynthesis, and if radium rays can modify this reaction, then suitable tests for the formaldehyde and peroxide in exposed leaf tissues ought to reveal the fact. Fenton[4] has shown that the rays can decompose H_2O_2, and, therefore, *a priori*, we might expect the evolution of oxygen in photosynthesis to be accelerated under their influence.

In his presidential address before the Chemical Society of London, Meldola[10] urged the view that several organic substances besides sugars may possibly result from the photosynthetic activity of the green cell. If this shall be demonstrated, then failure to detect starch or sugar in leaves exposed to radium rays will not necessarily indicate that all photosynthetic activity has been inhibited.

FERMENTATION: In studying the effect of radium rays on fermentation by yeast we have to consider their effects, not only on the living yeast-cells, but also upon the sugar solution, and upon the enzymes secreted by the yeast and acting as the immediate cause of the fermentation. The literature dealing with the discovery of enzymes in yeast and the proposal of the enzyme theory by Moritz Traube,[24, 25] in 1858, with the isolation of the alcohol-producing enzyme, zymase,† by Eduard Buchner,[15] and with other early discoveries

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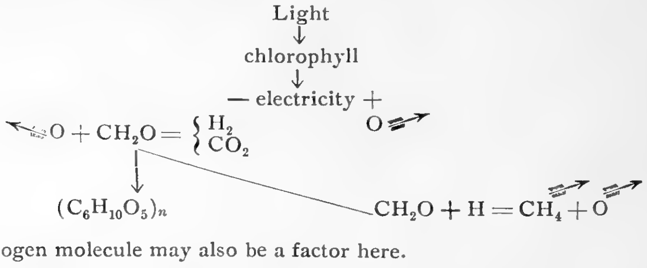
hydrogen, according to the known formula, thus setting free formaldehyde and oxygen, together with, in some cases, the transitory formation of H_2O_2. All these phenomena he expresses by the following scheme:



* The biogen molecule may also be a factor here.

† Jost[19] suggests the desirability of employing the term zymase in a generic sense for all substances produced by organisms and having the power of causing fermentative decompositions. To replace zymase he proposes the term *alcoholase*. This change seems unnecessary as it merely involves the substitution of "zymase" for "enzyme," which is now used generically.

is too well known to be reviewed here. A few of the more recent contributions will be referred to, as they serve to bring clearly before us some of the possible factors to be considered in discussing the stimulation of the process of fermentation.

Shortly after Buchner's fundamental discovery of the glycolytic zymase, he and Meisenheimer[16] announced that the process of fermentation consists of a number of successive steps, the products of which are, in order, glucose, some hypothetical intermediate product and water, lactic acid, and CO_2 and alcohol. The experiments of Brown and Glendenning[14] led them to believe that, in the transformation of starch to sugar, the process of hydrolysis is preceded by a combination of the hydrolyte with the enzyme, and that "this compound is much more unstable and less able to withstand the action of the active ions or dissociated molecules of the electrolyte than the original cane sugar. . . . According to this view these active ions are the true hydrolytes, not the enzyme itself, which has only an intermediate action." The enzyme is regarded figureatively "as the *vice* which holds the sugar molecule in a position favorable for the splitting agent to act."

In 1906 Slator[22] stated that the velocity of fermentation of dextrose varies with the amount of yeast present,* and is independent of the concentration of the dextrose. This latter fact is explained on the assumption that a compound is formed between the enzyme and the sugar, as Brown and Glendenning had previously suggested. Slator[22] states that, in the fermentation of sucrose enough sugar is almost instantaneously hydrolyzed for the fermentative reaction to attain its maximum velocity at once. From previous work and his own investigations he conceives that fermentation of dextrose by yeast involves the following steps in order:

1. Diffusion of sugar into the yeast cell.
2. Combination of dextrose and enzyme.
3. Decomposition of this compound, forming an intermediate compound.
4. Diffusion of the products from the cell out into the solution.

It is the third step, he says, which proceeds slowly, and whose velocity governs the rate of fermentation.

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* This, says Slator[22] (p. 130), confirms the work of O'Sullivan,[21] but the latter distinctly says that, "The rule laid down by Dumas[18] and supported by Brown[13] (for dextrose) holds good also for maltose," viz., that the time taken to ferment solutions of dextrose and maltose varies with the amount of the sugar present.

With reference to the stimulation of fermentation Dumas[18] stated in 1874, that the rate is more gradual in darkness and in a vacuum, and could be accelerated or retarded or destroyed by acids, bases and salts. Acceleration, he said, is very rare. In 1875 Becquerel[12] announced that fermentation was not retarded by the voltaic current "as Gay-Lussac observed." Schulz and Biernacki are said by Slator to have stated that very dilute solutions of poisons accelerate the process, while large doses have the opposite effect (*e. g.*, mercuric chloride, iodine, potassium iodide). Slator[23] was unable, under the conditions of his experiments, to secure this acceleration by any such reagents, and thinks that the effect previously interpreted as acceleration of alcoholic fermentation is due to an acceleration of the growth of the yeast, or to some other reaction. "We have not yet succeeded," he says,* "in finding a substance which will appreciably accelerate fermentation by fresh living yeast." And later,† "The velocity of such fermentation may be easily lessened by the addition of certain inhibiting agents, but cannot be appreciably raised." Among other conclusions, Slator[23] infers that, in the fermentation of dextrose, laevulose, mannose, and galactose, "the enzyme combines completely with the sugar, and that the velocity of formation of carbon dioxide is determined by the rate of decomposition of this compound."‡

At least three enzymes produced by yeast are to be considered; maltose and invertase, early recognized, and amygdalase, discovered by Caldwell and Courtauld[17] in 1907. Early in the present year (1908) we learn from the investigations of Trillat[26, 27] and of Kayser and Demalon[20] that acetic aldehyde is a normal product of alcoholic fermentation resulting from a further oxidation of the alcohol by the living yeast cells that exist aërobically near the surface of the fermenting mixture.

The above brief survey of the literature only emphasizes how impossible it is now, and how difficult it will be in the future to explain the acceleration by radium rays of the evolution of gas in alcoholic fermentation. Referring to the four steps hypothecated by Slator, it may be that the radium rays increase the ionization of the sugar solution and thus its rate of diffusion into the yeast cell; or the velocity of the reactions in the second and third steps; or the rate of

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* Loc. cit., p. 234.

† Loc. cit., p. 238.

‡ Loc. cit,, p. 241.

the fourth step. Possibly, in the light of Slator's work, the radium experiments are to be interpreted as showing acceleration, not of fermentation itself, but only of the metabolism and growth of the ferment-organism.

Here, as always, the living matter, or the biogen molecule, the rate of its construction and decomposition, or of its formation of enzymes or other substances, must always be considered as a possible, and I believe as a very probable and essential factor, delicately sensitive on account of its extreme lability, to the changes of energy produced by the rays. Further work may demonstrate that fermentation by unorganized ferments may be capable of modification by radium rays, but this would not in the slightest degree argue against the hypothesis that, with living yeast, the effects are due, in part at least, to the direct influence of the rays on the living matter. The most probable truth is that the rays influence both the vital and the non-vital steps in the process.

RESPIRATION: Respiration is no longer considered a simple oxidation, but as a series of both vital and non-vital processes beginning with atomic changes within the protoplast, and terminating with the evolution of carbon dioxide, or, in anaërobic respiration, of carbon dioxide and ethyl alcohol.

Since the publication, in 1876, of Pasteur's[34] classic *Études sur la bière*, it has been customary to regard respiration and anaërobic respiration as essentially alike. The correctness of this view was more fully established by the researches of Stoklasa and Čzerny,[39, 40] and Stoklasa[37, 38] also showed that, in last analysis, normal, aërobic, as well as anaërobic respiration, was of the nature of fermentation. According to this author, an enzyme similar to Buchner's zymase of yeast occurs in the cells of higher and lower plants, in the case of both normal and aërobic respiration. It is secreted by living protoplasm. Plant cells contain, in addition to an enzyme that produces alcoholic fermentation, one which causes the fermentation of lactic acid. Aërobic respiration he considers as a secondary process.* The primary process is the motion of the atoms in the "living mole-

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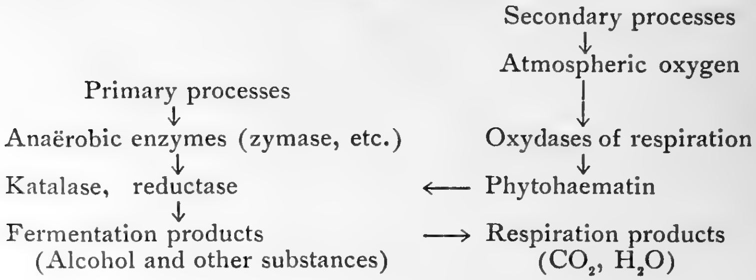
*Kostytschew[29] takes issue with the results reported by Stoklasa, and says, among other things (p. 215), that it is premature to regard anaërobic respiration as a first step in aërobic respiration.

Ideas quite similar to those of Stoklasa were presented in 1905 by Barnes,[28] who proposed the term *energesis* "to designate the disruptive processes by which energy is released, leaving respiration to designate the more superficial phenomena of aëration. . . ."

cule," accompanied by a rearrangement of oxygen within the molecule. By these processes, by which the necessary kinetic energy is secured for the maintenance of life, CO_2 and alcohol are split off, so that there arise within the "living molecule" reduced atom-groups, which have a great affinity for oxygen. In aërobic respiration there is no possibility for these reduced atom-groups (alcohol) to become fixed by the taking in of oxygen, therefore the alcohol is given off in addition to carbon dioxide. When oxygen is plentifully present, as in aërobic respiration, the alcohol in the nascent state is so combined that, under the influence of oxygen it serves for the formation of new parts of living protoplasm, through the agency of the oxydases of the air.

Maximow's[30] experiments harmonize with the essential identity of aërobic and anaërobic respiration. He found that sap, expressed from mycelium of *Aspergillus niger*, showed, on standing, a gaseous exchange analogous to that of normal respiration. This exchange was found to result from the activity of enzymes in the sap, independently of each other, and causing, the one an absorption of CO_2, the other a giving off of oxygen. The first is similar to zymase, the second belongs to the group of the oxydases. The enzyme which splits off CO_2 acts, as does Buchner's zymase (alcoholase), equally energetically in air and in hydrogen.

In April, 1908, Palladin[30-33] published the results of his investigations on the respiration-pigment in plants, detected several years ago by Schönbein[36] and later studied by Reinke.[35] Palladin considered that the physiological significance of these pigments is identical with that of the haemoglobin of the blood, and proposed for them the generic term "phytohaematin." It is the rôle of these substances to receive the oxygen from the air through the intermediation of the oxidases of respiration, and pass it on to the catalases and reductases. He gives the following diagram to illustrate, not only



the rôle of the phytohaematin, but also the relation of aërobic respiration, anaërobic respiration, and alcoholic fermentation, and the essential identity of the respiratory process in plants and animals. Doubtless further researches will make necessary certain modifications of this diagram, but it serves to present concisely our present knowledge of respiration, and to emphasize the complexity of the process.

For our purpose three points in these conceptions are to be emphasized: first, that the act of respiration is complex; second, that some of the steps apparently do not involve living matter at all, but are purely non-vital, chemical changes; third, that the processes not only of normal, aërobic respiration, but of anaërobic respiration and fermentation as well, involve the action of enzymes. Thus it is clearly evident that radium rays, or any other stimulus, may affect respiration in a variety of ways by modifying any of the steps involved. Not only may the living protoplasm (or some intermediate biogen molecule) be stimulated, but the action of the various enzymes may be accelerated or retarded or completely inhibited, or the phytohaematin may be similarly affected or completely destroyed. Also, under certain conditions, the protoplasm may be stimulated to produce these substances in greater or less quantity, or not at all.

GERMINATION: The mystery of the so-called "resting" seed is yet to be solved. We understand a few things, however, about its physiology. We know it is not dead, for it is constantly, at least at ordinary temperatures, slowly undergoing certain changes which characterize metabolism everywhere. These changes in time are sufficient to destroy the power to resume the normal rate of the life processes which, with the ripening of the seed, became reduced to their lowest terms.

The resting seed consists, in addition to its integuments, of at least three essential things: (1) The embryo; (2) the nourishment stored in or around the embryo; (3) enzymes, secreted largely if not wholly by the embryo.* Investigations were made by Albo[41, 42] for the purpose of finding out why seeds, apparently normal so far as structure and chemical composition are concerned, lose their vitality, even though the stored food remains in abundance. He found that

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* Pond's[48] studies on the capacity of the date endosperm for self-digestion indicate that the enzymes active in the germination of seeds originate, not as was formerly believed, within the endosperm, but wholly within the embryo. I have reviewed the literature on this subject elsewhere.[45]

diastatic activity in resting seeds is very feeble, varying with the species, and with the conditions under which the seeds are kept. Their diastatic power varies with their power of germination, being lowest in old seeds. In seeds which have lost their ability to germinate the diastatic power is diminished or entirely annulled. External agents, such as temperature, light, and chemical stimulants, affect enzymes and the germinating power of seeds alike. He concludes that the energy for the changes going on in seeds originates in the action of enzymes, but that the changes take place more gradually in resting seeds than in those that are germinating.

In 1907 Brocq-Rousseu and Gain[43] reported the existence of a peroxydiastase in dry seeds of Nymphaeaceae, Ranunculaceae, Malvaceae, Umbelliferae, Cupuliferae, Juglandaceae, Liliaceae, Gramineae, Coniferae, and other families of plants. This substance colors blue a tincture of guiacum in the presence of oxygenated water. It is stated that there may be more than one peroxydiastase in dry seeds, but the ferment does not exist in them indefinitely. Later the same authors[44] investigated the occurrence of the peroxydiastase in seeds of authentic ages of from 2 years to 5,000 years, taken from museums and herbaria. The oldest seeds were from the *Collections pharaonique du Musée de Boulaq*. They found that the peroxydiastases may disappear in a few years (in *Galium*, *e. g.*, 20 years); and that they may also be preserved for very long periods. The oldest seeds in which they determined its survival belonged to the 18th century. Seeds which could germinate, however, always contained the peroxydiastase, but those which have lost that faculty may still conserve it.

In connection with this last point may be mentioned the researches of Gain[46, 47] who found that the embryos of cereals from the tombs of the ancient Pharaohs, notwithstanding the seed's external appearance of good preservation, do not possess a cellular organization suitable for germination. The stored food is well preserved and may be utilized by a living embryo, but the embryo of the ancient seeds has undergone a chemical transformation, and is no longer viable. The relative age (and thus viability) of the embryo may be detected by a peculiar coloration which grows darker with age.

With reference to the effects of radium rays on dry seeds and on germination, the possibilities, suggested by the above discussion, hardly need a formal statement. The rays may act directly on the biogen molecule, or on the living matter of the embryo, on the food substances stored in and around the embryo, or on enzymes present in the seed, or on any two or all four of these factors. The molecules of the biogens, of the food, or of the enzymes may be shattered and utterly destroyed, either wholly or in part, or the rays may only hasten metabolic processes in such a way as either to accelerate germination in the active seed, or to cause a premature ageing and ultimate death in the resting seed. If only a peroxydiastase or other enzyme, normally present in the resting seed, were destroyed it is possible that the embryo, even if it were still alive, would not be able to reproduce the ferment fast enough to supply the needs arising with the imbibition of water and the reawakening of the protoplasts. Or again, the molecules of the stored food might be so altered as not to be capable of being acted on by the enzyme.

How do radium rays affect solutions, starch, oil, aleurone, enzymes and other substances stored in seeds? Until these questions are answered we cannot expect much light on the way in which the rays affect resting and dry seeds.

Studies that have so far been made of the effect of radium rays on solutions have yielded contradictory results. Soon after the discovery of Röntgen rays, Thomson[57] (1896) ascertained that, when these rays pass through a dielectric, they make the latter, during the time of their passage, a conductor. All substances, he says, when transmitting them, are conductors of electricity. "The passage of these rays through a substance seems thus to be accompanied by a splitting up of its molecules, which enables electricity to pass through it by a process resembling that by which a current passes through an electrolyte." The experiments of Graetz,[51] four years later, led him to believe that radium rays act in a similar way, while the experiments of M. Curie,[50] in 1902, clearly indicated that both radium and X rays act on liquid dielectrics as on air, communicating to them a certain conductibility. The investigations of Henning,[52] and of H. Becquerel,[49] led to the same conclusion. If X rays have such a property, then we should theoretically expect radium rays to possess it as well.

Kohlrausch,[53] however, concluded, in 1903, that such effects, if they exist at all, were to be attributed, not to the ionizing effect of the rays, but to their accelerating effect on the dissolving of the glass of the resistance vessel. Later, he and Henning[55] found that the conductivity of solutions of radium bromide of 1/12,000 to 1/20 N concentration was similar to that of analogous, but non-radioactive, salts of elements related to radium, *e. g.*, barium, strontium, and calcium, and in 1906 Kohlrausch[54] definitely stated that the conductivity of water is not increased by drawing through it a current of air that has first passed over radium. The quantity of ions thus formed, he said, is exceedingly small. Experimenting with NaCl, CaCl, BaCl, $MgSO_4$, $ZnSO_4$, K_2CO_3, HCl, and NaOH, in percentages of from 20 to 2.5, he found that radium rays increase the conductivity of electrolytes only in so far as they raise the temperature of the solution. He inferred that, if the β and γ rays do increase the degree of ionization of the electrolyte, they do so to a degree too small to be detected by the delicate means he employed.*

GROWTH: As previously stated,† the term growth is here taken to mean increase in size or increase in mass, one or both, with or without an accompanying change of form. Increase of size without increase of dry weight involves increased turgor in elastic walled cells. Increase of mass involves an excess of constructive over destructive metabolism. Radium rays may affect both processes.

Since turgor is an expression of the internal osmotic pressure of the cell, and since osmotic pressure is a function of the number of particles of the solutes present in the cell-sap, radium rays, by their power of ionization, may possibly increase this turgor, and thus accelerate or favor an increase of size of the individual cells and of the tissues and organs composed of them. Theoretically the rays would have this power. That they do alter solutions in some manner seems clearly evident from the effects of exposed water on germination and growth, as described in Experiments 48–51 on pages 158–173.

Growth, in the sense of increase of mass, is an expression of construtive metabolism. Not only is the number of biogen molecules increased, but also the amounts of various substances produced by them. Here, as in respiration, fermentation, and other metabolic processes, the activity of enzymes is involved. The digestion of the food is accomplished by various enzymes, and energy for metabolism is set free by enzymotic action. Verworn ‡ believes that a molecule of an enzyme is the substratum of the metabolic process. It is the

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* Cf. foot-note, pp. 220–221.

† p. 223.

‡ 2, p. 15.

same story of multiplicity of factors and complexity of function. Therefore, with reference to the influence of radium rays, we must consider as many possibilities as there are steps and combinations of steps. First and foremost we must take into account the living substance itself. Then the rays may affect the enzymotic digestion of the food, partly outside and partly inside of the cells to be nourished. Furthermore the rays may operate so as to increase or decrease the amount of energy available for the work, and, lastly, variations in growth may be, either wholly or partly, expressions of the influence of the rays on cell-division.

CELL-DIVISION: No one has yet succeeded in accelerating the rate of cell-division or in increasing its amount in a given tissue by means of radium rays. The only results recorded are the introduction of irregularities and complete inhibition. A brief résumé of the well known life-history of a cell may serve as the first step in attempting to explain this effect.

On the basis of Verworn's theory, the active metabolism of a young cell involves, fundamentally, the continual destruction and reconstruction of biogen molecules. The latter process is conditioned, in part, by the supply of suitable food material, and upon its preponderance over the breaking down of the biogens depends the growth of the cell. With cell-enlargement by growth the area of the cell-surface in proportion to the mass becomes finally too small to permit of the entrance of enough food and energy to maintain the reconstruction of the biogen molecules faster than they are decomposed. If the surface area relative to the mass can be increased, rejuvenescence will result, but otherwise the biogen molecules will continue to break down faster than they can be built up again, and the cell enters upon a period of senescence. In growing old either certain catabolic products are produced in relatively larger quantities than occurs in the pre-senescent period, or new catabolic substances are elaborated, or both. Normally cell-division postpones the approach of senescence.*

The evidence at hand, and presented in Chapter XVI, indicates that radium rays may hasten the approach of senescence. In other words, they may accelerate the breaking down of biogen molecules and interfere with their reconstruction. This is doubtless accomplished by

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* Of course cell-division cannot indefinitely postpone senescence, nor are the above circumstances regarded as the only condition or stimulus of cell-division.

ionizing the atoms of all the various substances (biogens and others) that are involved in the metabolism of the cell, the effects being produced both directly and indirectly. Thus we should expect, *a priori*, a retardation and finally a complete inhibition of cell-division in all tissues exposed to rays of sufficient activity and for suitable periods of time. And this is what has been observed to occur. Theoretically we ought also to be able to accelerate the process by suitable conditions of exposure, but such conditions have not yet been discovered.

The irregularities produced by radium rays in karyokinesis do do not seem to call for any special explanation in addition to that suggested in discussing the abnormalities of tissues and organs in Chapter XVI. Such irregularities are only a morphological expression of physiological disturbance, and it may be seriously questioned whether we are justified in expecting the morphological appearance and behavior of chromosomes to explain things, any more than do variations in leaf-margins, or other purely structural facts. The problem of the causes of variation and inheritance lies deeper than morphology, and the behavior of the chromosomes, even in sexual cells, instead of explaining other and grosser facts of structure, quite possibly only presents another feature to be explained.

RECOVERY FROM STIMULUS: It has been frequently observed in the study of stimulation, not only by radium rays but by other stimuli as well, that a depression of function, caused by certain conditions of exposure, may be followed, after a period, by recovery of the normal rate of functioning, provided that the stimulant is discontinued. In like manner excitation is followed by a return to the normal condition. This recovery of tonus probably means that the stimulus has not been injurious to the biogens, or that it has produced its effect largely, if not wholly, by its influence upon the non-vital steps in metabolism. Thus, if the rays of radium acting on a resting seed destroyed a large percentage of some enzyme necessary for germination, but affected the biogens only slightly or not at all, or if they destroyed only a relatively small number of the latter, germination and early growth would be retarded, but recovery would follow in a short time, due to a replenishing of the necessary enzyme by metabolic processes. A similar course of reasoning would apply to recovery from excitation.

RADIUM STIMULUS AND PLANT-ELECTRICITY: The literature dealing with the existence of electric currents in plants is far too voluminous to be reviewed here. Vassali-Eandi,[66] in 1804, was probably the first to detect evidence of such currents, and stated that the so-called "vital principle" was only "galvanic fluid." Half a century later A. C. Becquerel [58, 59] and Wartmann [67] clearly demonstrated that such currents exist in plants, and several years afterwards the phenomenon was quantitatively studied by Burdon-Sanderson,[62-65] and Munk.[60] These currents doubtless have their source in the chemical changes going on within the tissues,* and Pfeffer † states, not only that respiratory metabolism (*athmungsstoffwechsel*) contributes in an important manner to their formation, but that at present we have no clear proof that they originate otherwise.‡ Also, as Pfeffer says, it is unknown whether the electricity is merely a necessary by-product of chemical transformations that have taken place, or whether it plays a special rôle in the organism, affecting chemical or other processes.

The rôle of these currents is too little understood to make profitable any attempt to discuss them further as a factor in the response of plants to radium rays, but, since the particles of the beta and alpha rays carry charges of electricity, we should not fail to recognize the fact that the normal electric currents in plants may be a factor involved in the reactions of the plants to the stimulus of the rays. Their effect must be either explained or explained away.

CONCLUSION: § This rather involved consideration of the possible mode of action of radium rays upon the life-processes of plants has served chiefly to indicate the nature of the problem, and to suggest the direction that future researches should take. If the living matter itself is directly affected by the rays it is difficult to conceive how any one function could be modified without the others being affected, for, with long periods of exposure (24 hours or more) to radium of high activity (1,500,000 × or more) it is certain that the protoplasm will have its vitality partially or wholly destroyed, and all "vital" processes correspondingly modified or stopped. But, on

* Becquerel.[58]

†61, p. 192.

‡ Gibson's hypothesis that these currents result from the transformation of the energy of light-waves by chlorophyll is referred to on page 261. If his photoelectric hypothesis of photosynthesis shall be confirmed, then it is possible that radium rays may affect photosynthesis, in part at least, by exerting an influence on these currents.

§ Theoretical considerations with reference to tropistic response, histological effects, and the effects of exposing germ-cells are discussed in Chapters XV, XVI, and XVIII, and are therefore omitted from this chapter.

the other hand, the modification or total inhibition of any one process does not necessarily indicate that the living matter has been directly affected, for such a condition would result if, as in the case of the resting seed, the rays destroyed an enzyme essential to the completion of some function.

The broadest, and at the same time the most definite generalization warranted by the work so far done is that the rays of radium act as a stimulus to metabolism. If this stimulus ranges between minimum and optimum points, all metabolic activities, whether constructive or destructive, are accelerated; but if the stimulus increases from the optimum toward the maximum point it becomes an over-stimulus, and all metabolic activities are depressed and finally completely inhibited. Beyond a certain point of over-stimulus recovery is impossible, and death results.

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General

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EXPLANATION OF PLATES

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HISTOLOGICAL EFFECTS OF RADIUM RAYS ON LUPINUS ALBUS

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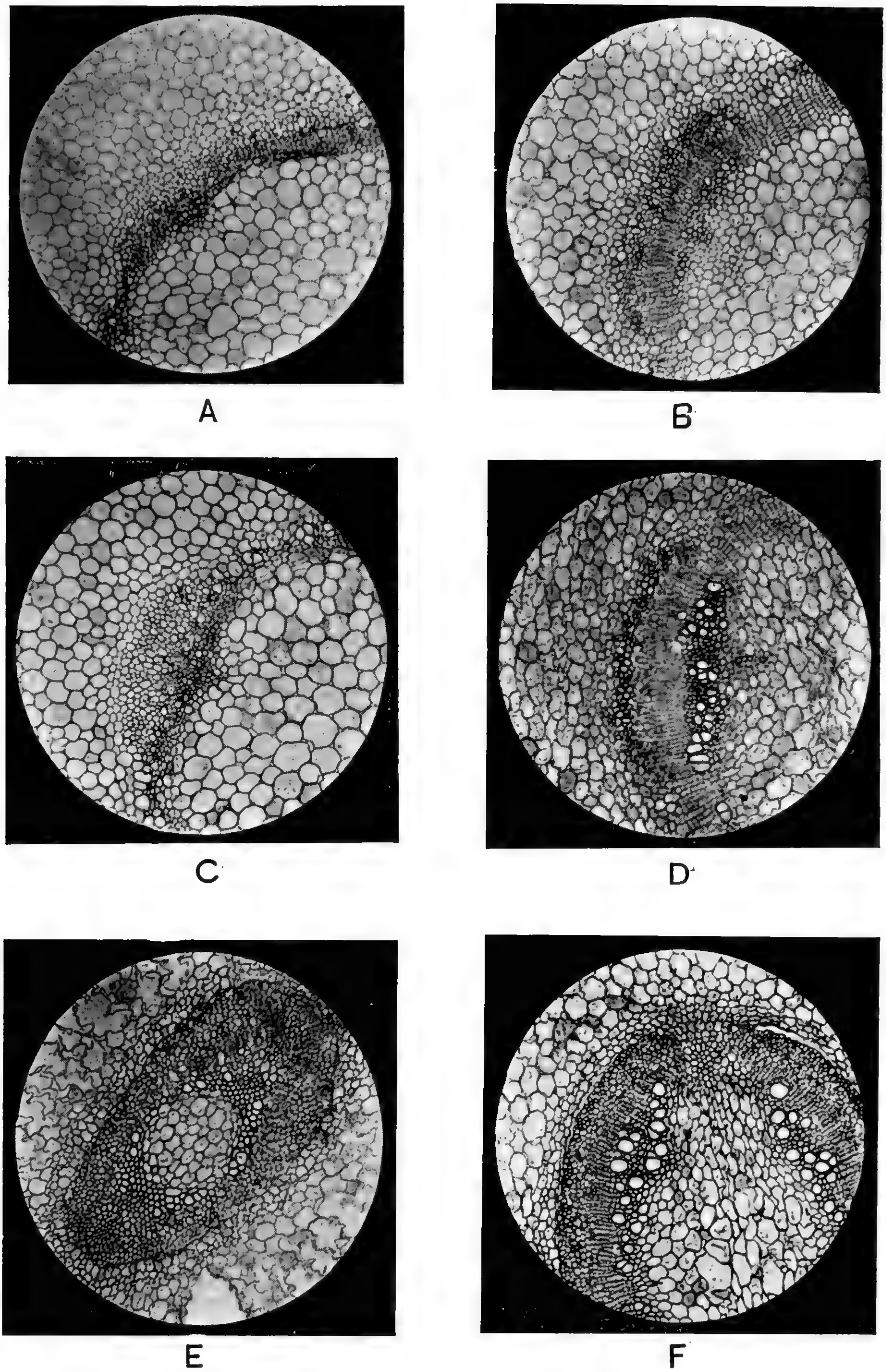

A B C D E F

HISTOLOGICAL EFFECTS OF RADIUM RAYS.

PLATE 2

HISTOLOGICAL EFFECTS OF RADIUM RAYS ON LUPINUS ALBUS AND PHASEOLUS VULGARIS

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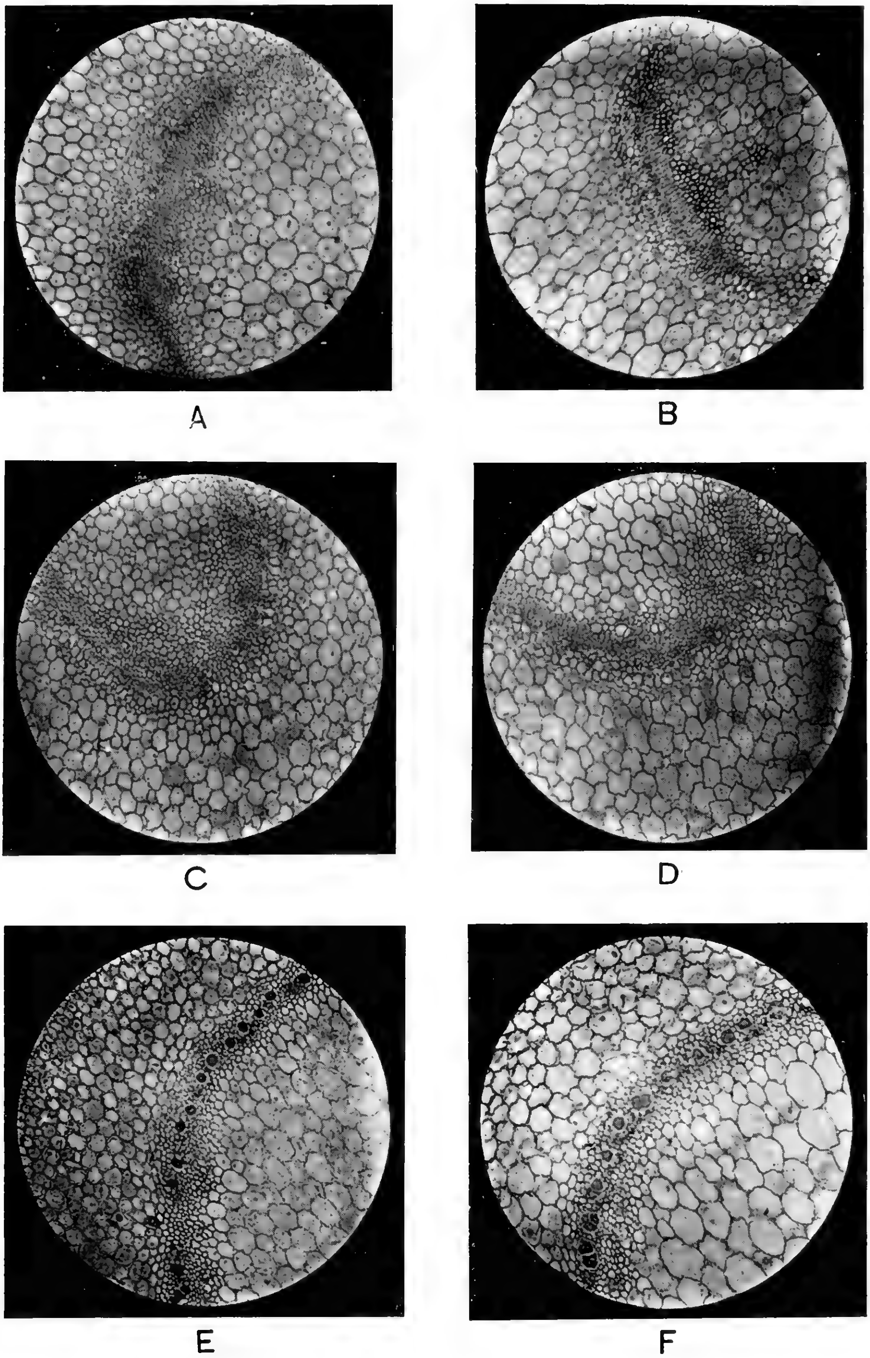

HISTOLOGICAL EFFECTS OF RADIUM RAYS.

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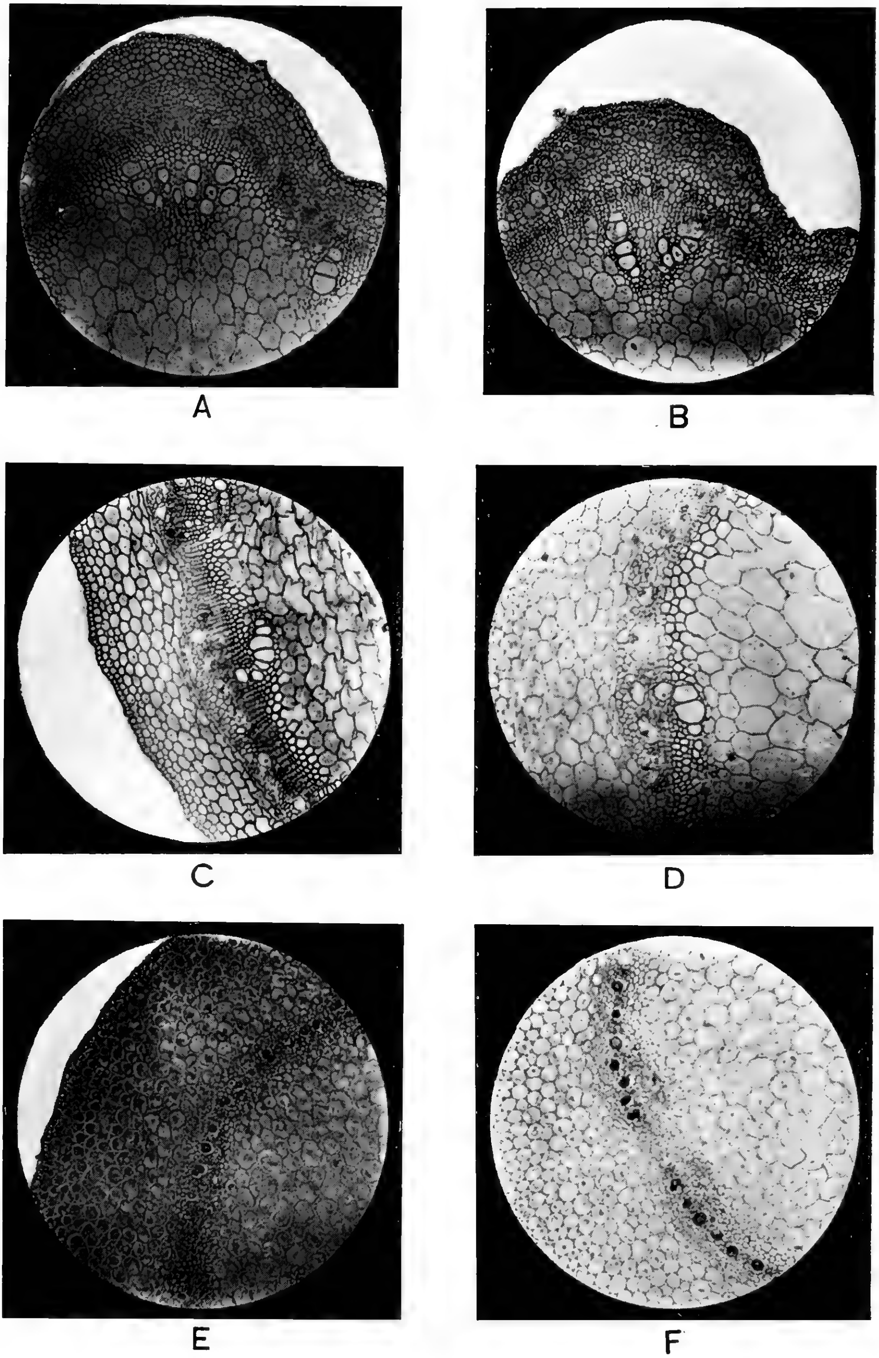

HISTOLOGICAL EFFECTS OF RADIUM RAYS.

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HISTOLOGICAL EFFECTS OF RADIUM RAYS ON ZEA MAYS

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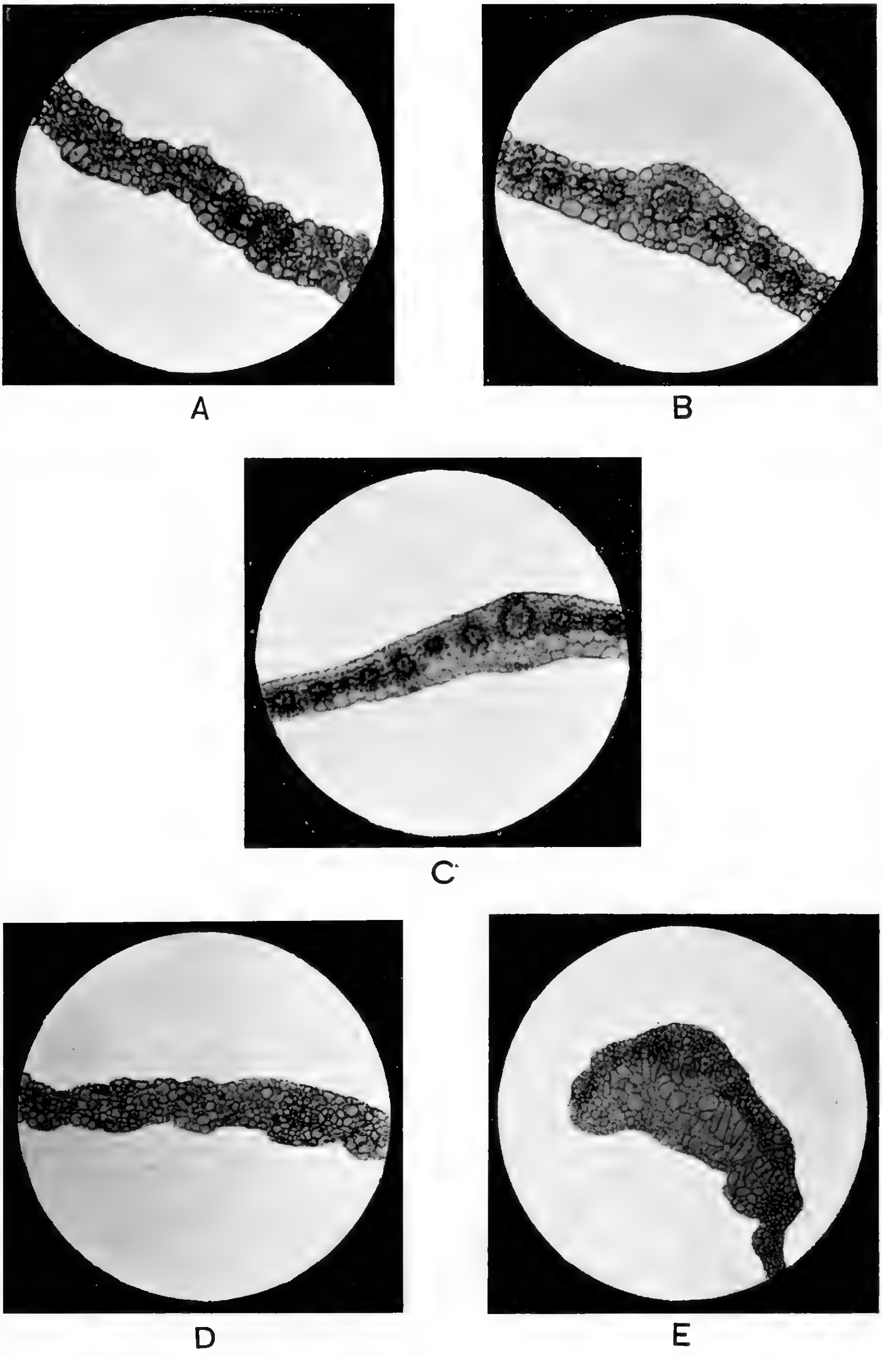

HISTOLOGICAL EFFECTS OF RADIUM RAYS.

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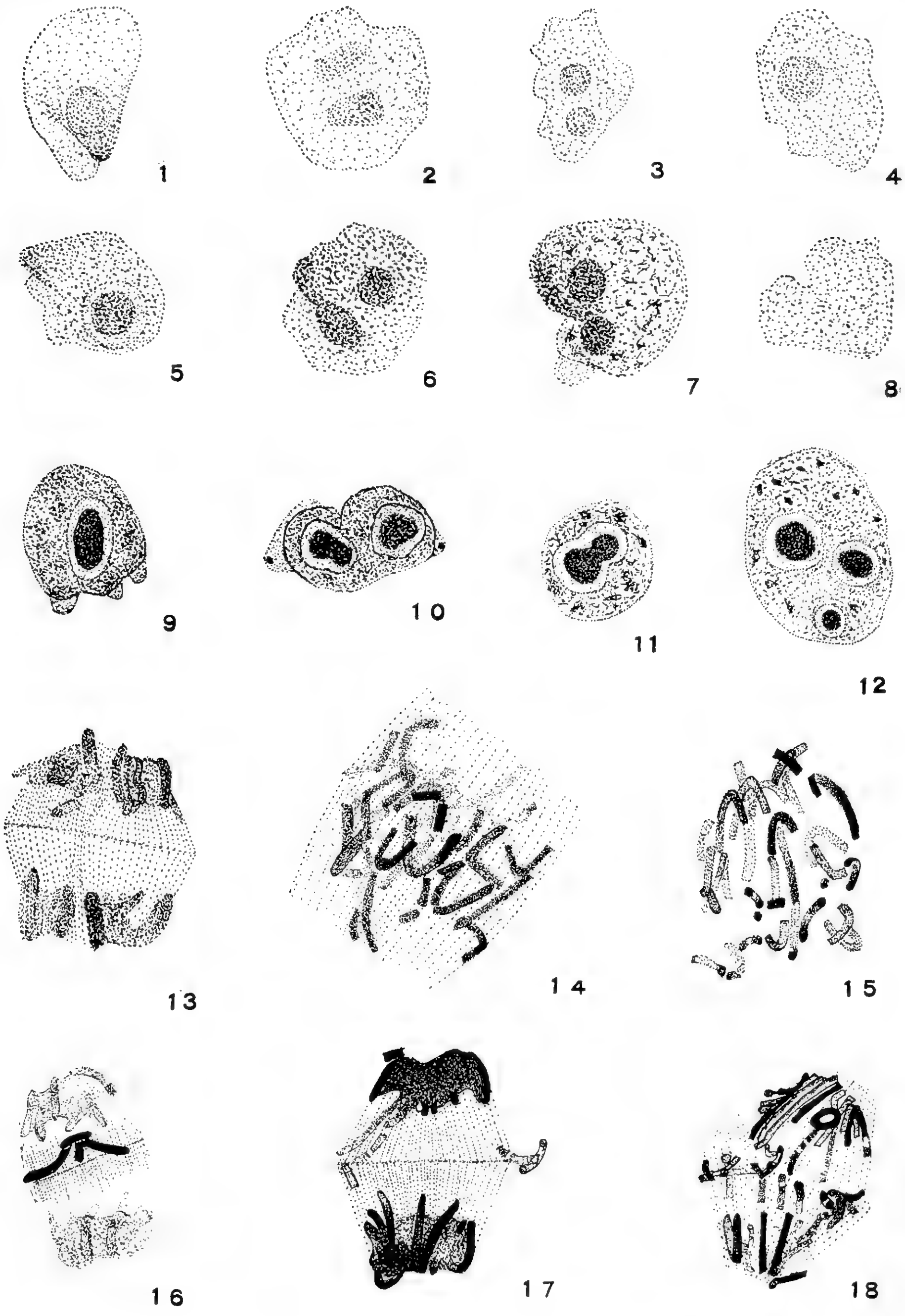

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EFFECTS OF RADIUM RAYS ON NUCLEI AND MITOSIS.

PLATE 6

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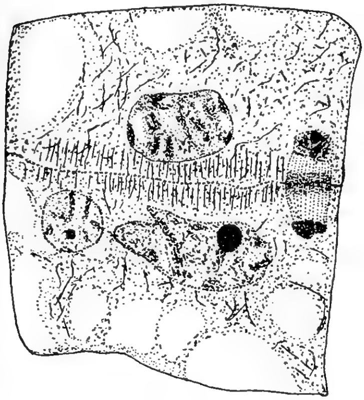
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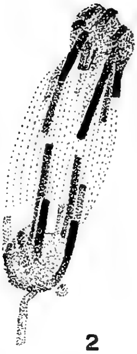
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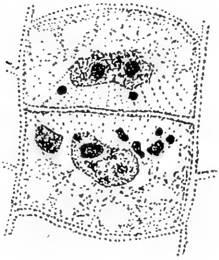
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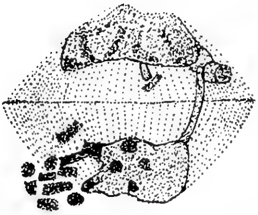
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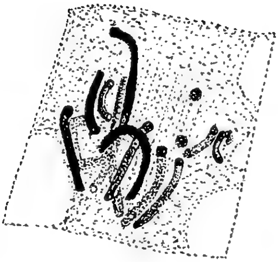
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EFFECTS OF RADIUM RAYS ON MITOSES.

PLATE 7

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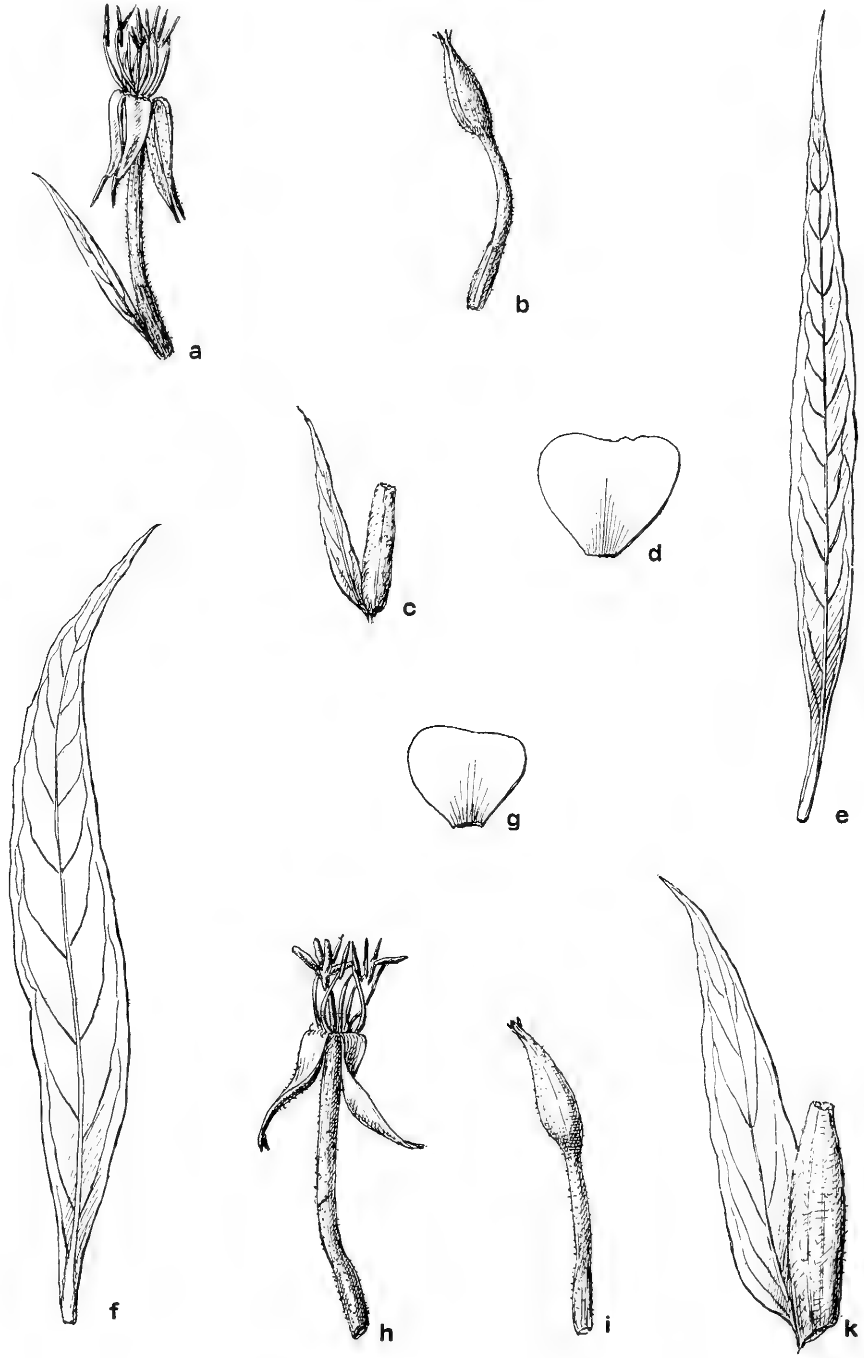

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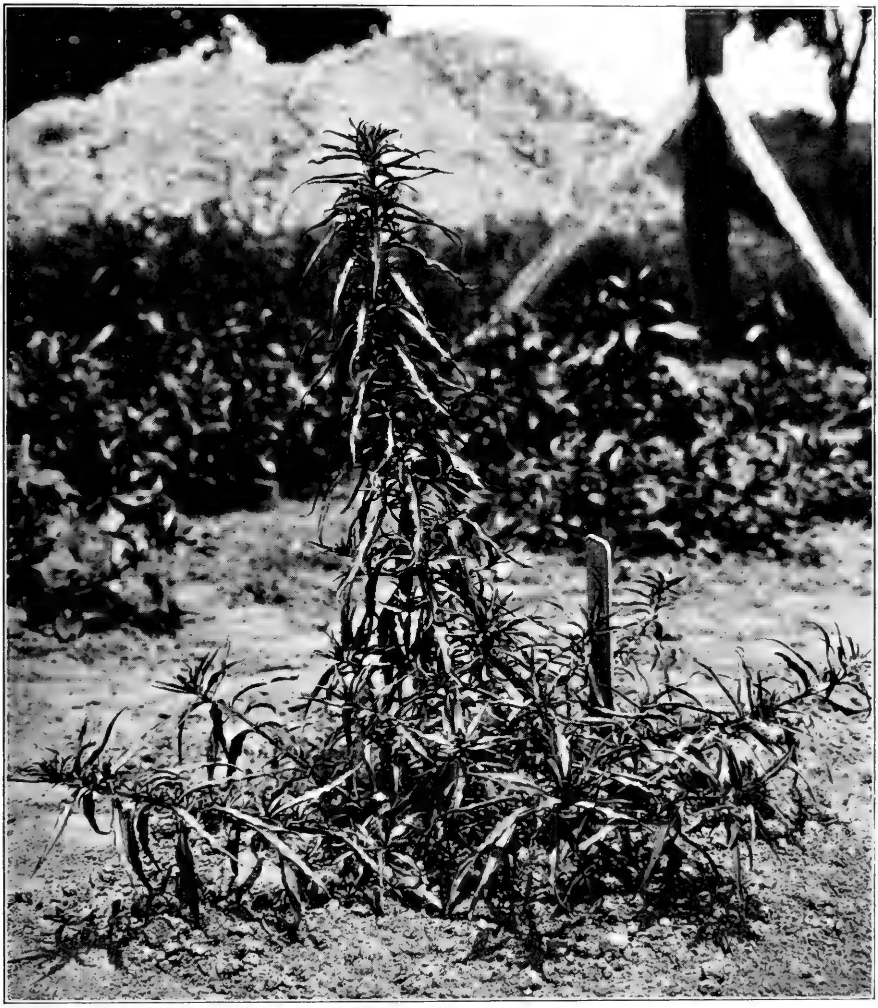

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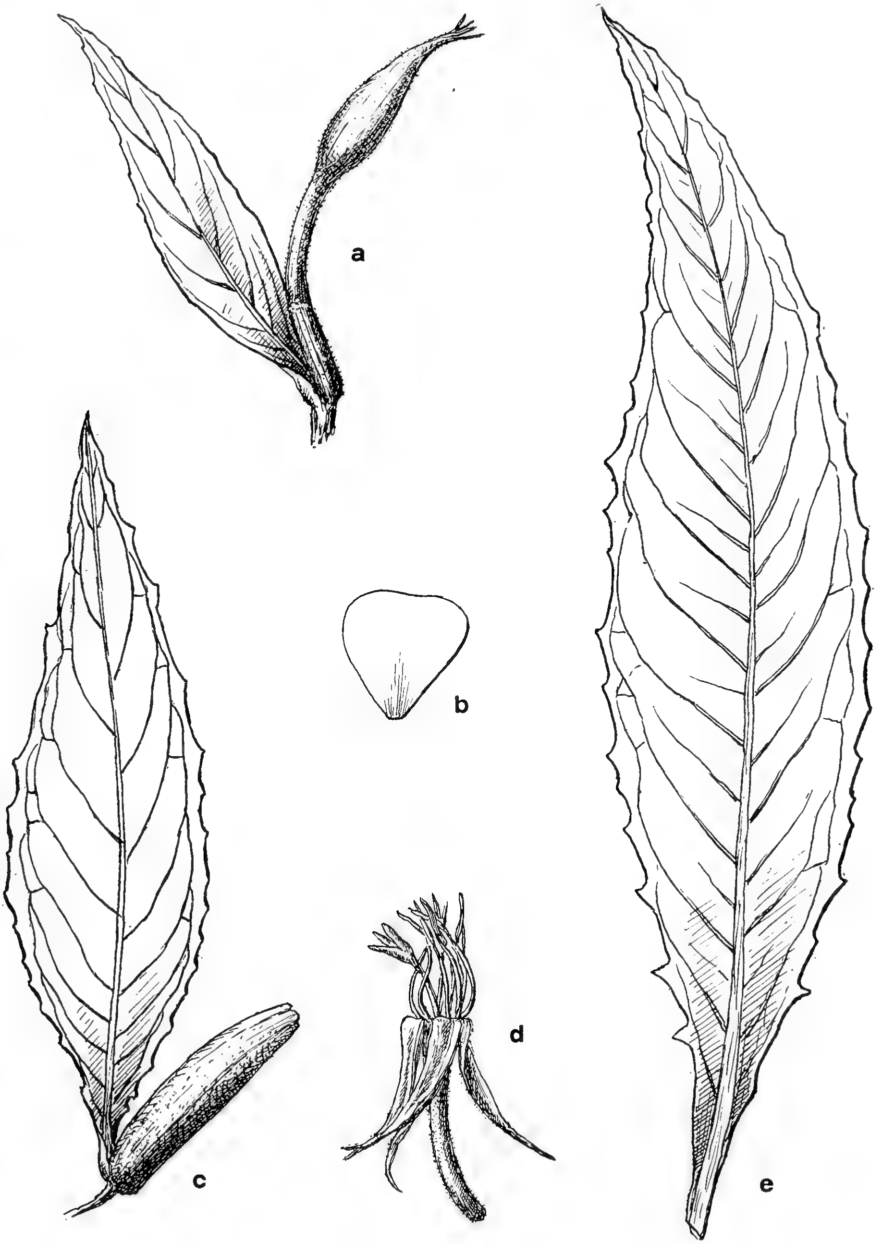

ONAGRA BIENNIS. RADIUM CULTURE, No. 15a, Broad Leaf.

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