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Annual Report for 1914 With the Supplements to the Guide to the Experimental Plots Containing the Yields per Acre, Etc.



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The Plant Nutrition Work

Rothamsted Research

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If this view is correct it follows that acids need not be used in soil analysis at any rate for the extraction of bases: any agent capable of being absorbed by the soil would serve equally well. In our laboratory Mr. J. A. Hanley has recently been using a solution of ammonium nitrate and finds it dissolves at least as much potassium, calcium, etc., as an acid, and in some respects is more satisfactory. This method of extracting the bases from soils was devised by Prof. Ramann, under whom Mr. Hanley worked for a time.

These investigations promise to clear up many of the difficulties in soil analysis.

THE PLANT NUTRITION WORK.

The fundamental problem in plant nutrition is the production of sugar in the green leaf, but the investigation is seriously hampered by analytical difficulties. During the past three years considerable improvements in method have been effected by Mr. Davis and his assistants, Messrs. Daish and Sawyer, and this year satisfactory proof has been adduced of the presence of free pentoses in the leaf and a method has been elaborated for determining them.

The complete method now allows of the determination of cane sugar, maltose, dextrose, levulose, pentoses, starch, and pentosans in the leaf. It has been used for numerous analyses of leaves and stalks plucked at regular intervals during the day and night, the results of which will be discussed later.

Equally important from the standpoint of crop production is the formation of starch in the grain. Analytical difficulties have hitherto prevented a satisfactory investigation, but this year the determination of starch has been put on a satisfactory basis. The existing methods may give rise to an error equal to 20 per cent. or more of the starch present: in the ordinary diastase method, for example, a loss of this magnitude may arise from the precipitation of dextrin during the preliminary processes of precipitation. A new method has therefore been devised, based on the fact that the enzyme taka-diastase transforms starch quantitatively into maltose and dextrose, no dextrin being formed: thus the errors of the ordinary method are obviated. This is apparently the only trustworthy way of estimating starch in plant tissues, and it is now being used for wheat and barley grown on the Rothamsted plots.

During the year Dr. Brenchley has been engaged on an extended survey of the grass plots; a complete botanical analysis is being made, and the results are compared with the last survey of 1903 and the earlier ones of 1862, 1867, 1872 and 1877 in order to follow the change brought about in the herbage. A survey of the weed flora of certain of the fields, and of the flora of Geescroft and the Wilderness, has also been made and compared with that of earlier years. All these results are being worked up both from the agricultural and the ecological standpoints.

The investigations on the effect of certain organic substances on plant growth have been continued, and progress has been made by Miss Adam with the study of the effect of potassic manuring on the anatomical structure of a grass (*Dactylis glomerata*).

At the present time eleven of the laboratory staff and research

workers are on active service, but their places are gradually being temporarily filled by volunteer workers.

The following papers have been published during the year :—

- I. “*On the action of certain compounds of Zinc, Arsenic, and Boron on the Growth of Plants.*” WINIFRED E. BRENCHLEY. *Annals of Botany*, 1914. 28, 283—301.

Experiments made in water cultures with certain compounds of zinc, arsenic, and boron go to uphold the conclusions arrived at four years ago with compounds of copper and manganese, *i.e.*, that very small quantities of some inorganic poisons stimulate the growth of certain species, but this action is by no means universal. With some poisons the stimulative action is evident, with others it has not been observed; also, a poison that increases growth in one species does not necessarily do so in another, however small a quantity of the reagent that may be applied. A summary of the conclusions arrived at is as follows :—

- (1) Zinc sulphate in high concentration is very toxic to barley and peas, and no evidence of stimulation has been obtained with any strength of the poison down to a lower limit of 1/200,000,000.

- (2) Arsenious acid is more toxic in its action on peas and barley than is arsenic acid, peas being particularly susceptible. This distinction holds good for sodium arsenate and sodium arsenite, though in a less degree. Again no stimulation is evident with the smallest quantities so far tested.

- (3) Boric acid is less poisonous than zinc sulphate or arsenic compounds, especially with peas. Barley shows stimulation to the eye with some of the weaker strengths of poison, but this is not borne out by the dry weights. Peas, on the other hand, are definitely stimulated with relatively high concentrations of boric acid. The action of the greater strengths of the poison is well marked in the leaves, which tend to become brown, and to die in a characteristic manner.

- II. “*Mapping as an Ecological Instrument.*” WINIFRED E. BRENCHLEY. *Transactions of the Norfolk and Norwich Naturalists’ Society*, 1914. 9, 723—733.

The progress of ecological science during the last few years has necessitated the elaboration of special methods of representing and correlating the available facts, one of the most useful of such methods being that of the map, which may now be regarded as a graphical representation of facts from which other facts and hypotheses may be deduced and upon which theories established on a firm basis can be built. A description is given of the various applications of mapping in general use, illustrated by reference to ecological surveys at Erquy (Brittany), and Blakeney Point (Norfolk). The general field map, blocked-in map, grid, transect, and quadrat are discussed and illustrated.

Some of the methods here described are being applied, with the necessary modifications, to field investigations at Rothamsted.

III. "*Partial Sterilisation of Soil by Volatile and Non-volatile Antiseptics.*" WALTER BUDDIN. *Journal of Agricultural Science*, 1914. 6, 417—451.

Nearly all the antiseptics used cause increases in bacterial numbers after treatment as Hiltner has already pointed out, and they also mostly produce an initial increase in the amount of ammonia. Here, however, the uniformity ceases, and closer examination shows that two very distinct classes of antiseptics exist—

(1) those which are completely volatile and disappear entirely from the soil once their work is done;

(2) those which remain in the soil for a considerable period or else leave decomposition products and so exert a prolonged action on the bacterial flora and on the plant.

1.—The easily volatile antiseptics are all similar in their action but differ as regards their potency. When a particular strength is reached all the usual partial sterilisation effects observed by Russell and Hutchinson show up together: an initial depression in the numbers of bacteria, the killing of all protozoa, except a few flagellates, the killing of nitrifying organisms, and a small initial production of ammonia; followed later by a large sustained increase in bacterial numbers and in the rate of production of ammonia. The methods used for the examination of the soils did not disclose any substantial difference in result with any higher dose once partial sterilisation had occurred. On the other hand the weakest doses have no effect on the numbers of bacteria occurring in the soil, nor on the rate of production of ammonia and nitrate, and as far as can be judged from the ordinary one per cent. hay infusion culture method, no appreciable action on the protozoa.

The investigation included a group of compounds so mild that they can hardly be considered as antiseptics—the open chain hydrocarbons. These give an initial depression in bacterial numbers and a very distinct increase in the initial ammonia content, but nitrifying organisms are not killed nor are the larger protozoa suppressed. There is no distinct and permanent increase in the numbers of bacteria: there is, however, a marked increase in nitrate production. A similar but weaker action occurs after merely spreading out the soil in a thin layer for 24 hours so that it dries down on the average to about five per cent. water. In this case there is after moistening a similar increased rate of production of nitrate over that in the soil which has been kept in a moist state after being bottled fairly fresh from the field, although there is no noticeable effect on the numbers of bacteria. The decreased action is indicated by the absence of marked initial effects.

2.—The non-volatile antiseptics all produce a permanent depression in bacterial numbers with the highest doses. The more potent such as quinone and hydroquinone show an initial depression in numbers of bacteria developing on gelatine plates even with the weakest dose used (approximately 0.05 per cent.), while the less potent show no initial effects with such strengths excepting the liberation of a small amount of ammonia. With all the antiseptics the dose which is sufficient to produce the full initial depression in numbers also kills the larger protozoa and checks the action of nitrifying organisms. The traces of substance left behind in the

soil exert a very distinct action on the bacterial flora. The usual result is to produce at varying periods after treatment an enormous rise in the number of certain special organisms. This is seen not only in cases where the substance is known to be attackable by certain bacteria, *e.g.*, alcohol and phenol, but in others such as quinone, where such action was not expected. The new flora is not the normal soil flora, although with the very mildly acting antiseptics the old flora may persist in approximately its original complexity but certainly in no greater numbers. The predominant part of the new flora is very much simpler than that remaining after treatment with the easily volatile but potent antiseptics. The colonies are all very slowly growing and consist of only two or three species of bacteria. Determinations of the nitrogen present in the soil as ammonia and nitrate show that the new flora does not produce ammonia. When the abnormally high numbers fall off the condition finally attained depends on the intensity of the initial action: phenol and cresol in weak doses leave a flora which is similar in character to that arising after normal partial sterilisation and produces more ammonia than that of the untreated soil. None of these non-volatile antiseptics, however, leads to such a marked increase in the amount of ammonia and nitrate present after an incubation period as do the volatile antiseptics.

The curves obtained for the numbers of bacteria present after the treatment of the soil with non-volatile antiseptics are remarkably similar to those obtained by Hutchinson and MacLennan with Woburn soil treated with various doses of quicklime. The increased numbers are attributed to an increased supply of food: in these experiments it is difficult to avoid the conclusion that the bacteria are actually feeding on the antiseptics.

The alcohols proved curiously ineffective: formaldehyde also was less potent than was anticipated, behaving in weak doses very much like the open chain hydrocarbons.

- IV. "*The increased Nitrate Content of a Soil subjected to Temporary Drying in the Laboratory.*" WALTER BUDDIN. *Journal of Agricultural Science*, 1914. 6, 452—455.

A remarkable effect of drying a soil was observed in the foregoing experiments. Soils spread out to dry were found on moistening to have undergone practically no change in bacterial numbers or in nitrate content and to resemble in every point tested similar soils that had remained moist. But at the end of a month it was found that the nitrates had increased to a considerably greater extent in the soil that had been dried although there was no difference in bacterial numbers. The effect closely resembles that of treatment with volatile mild antiseptics.

- V. "*Methods of Estimating Carbohydrates.*" II. "*The Estimation of Starch in Plant Material.*" W. A. DAVIS and A. J. DAISH. *Journal of Agricultural Science*, 1914. 6, 152—168.

A careful study of the existing methods of estimating starch in plant material has shown that they are unreliable and a new process has been worked out which gives more accurate results.

The Sachsse method, based on the hydrolysis of the starch with dilute acid, fails because the presence of pentosans falsifies the results by giving rise to reducing sugars (pentoses) which count as dextrose, and also because actual destruction of dextrose occurs during the prolonged heating with dilute acid.

O'Sullivan's method, which is sufficiently accurate when pure starch is employed, gives low results in the case of plant material, owing to a loss of dextrin during purification of the solution after the conversion with diastase, this purification being always necessary to remove tannins, proteins, etc., from the plant material.

The method now proposed is based on the fact that taka-diastase in a few hours at 38° converts starch quantitatively into a mixture of two sugars only, maltose and dextrose; no loss of these sugars is to be feared during the ordinary basic lead acetate process of purification.

The details are as follows:—The plant material immediately after picking is dropped into a large volume of 95 per cent. alcohol containing 1 per cent. by volume of 0.880 ammonia. This destroys the plant enzymes and prevents subsequent changes in the carbohydrates. The material is then extracted for 18 to 24 hours with boiling alcohol in a special large form of Soxhlet extractor, so as to remove completely the sugars and other soluble substances. It is then freed from alcohol by pressure and the press-cake is broken up, dried in a steam oven for 18 hours, rapidly ground in a small mill and bottled for analysis.

Ten grams (or a suitable proportion) is dried to constant weight at 100-110° in vacuo over phosphorus pentoxide and the results of the analysis returned on the vacuum-dried weight. To estimate starch, the dry material (previously extracted with water, if necessary, to remove gums, amylans, etc.) is gelatinised with 200 c.c. of water in a beaker flask heated for $\frac{1}{2}$ hour in a water bath at 100°, the mixture is cooled to 38°, 0.1 gram of taka-diastase and 2 c.c. of toluene added, and the mixture left 24 hours for conversion to take place; it is then heated in boiling water to destroy the diastase and the clear solution filtered through a fluted filter paper into a 500 c.c. measuring flask; the leaf residue is thoroughly washed several times by decantation until about 475 c.c. of solution is obtained. The necessary quantity of basic lead acetate solution (generally 5 to 25 c.c.) is then added to precipitate the tannins, etc., avoiding as far as possible any great excess of lead; tests should be made after each small addition so as to ascertain when precipitation is complete. When this is the case, the volume is made up to 500 c.c. at 15° and filtered; 100 c.c. of the filtrate are placed in a 110 c.c. flask, the slight excess for lead precipitated by adding solid sodium carbonate and the volume adjusted to 110 c.c. 50 c.c. of the filtrate from the lead carbonate are used to measure the reducing power and another portion polarised in a 400 mm. tube. From the values so obtained the quantity of maltose and dextrose present can be calculated and the percentage of starch obtained. The values obtained by Brown, Morris, and Millar for the reducing power of dextrose and maltose are used as a basis of calculation.

Several examples are given of the application of this method to plant material, and the necessity is emphasised of removing optically active substances soluble in water, such as gums. Special care is

also necessary in taking the sample for analysis, as the material which falls to the bottom of the stock bottle is often far richer in starch than the lighter fibrous material that rises to the top.

- VI. "*Some Notes on the Chemistry of Starch and its Transformations.*" W. A. DAVIS. *Journal of the Society of Dyers and Colourists*, July, 1914.

It is shown that taka-diastrase, the enzyme of *Aspergillus Oryzae* converts starch first into dextrans, then into maltose and finally into dextrose. After a few hours all the dextrin has disappeared and the product consists of maltose and dextrose only; the amount of dextrose gradually increases with the time. It is considered that taka-diastrase contains maltase as well as the ordinary enzymes of malt diastrase and that this converts the maltose into dextrose. No evidence could be obtained that taka-diastrase converts starch directly into dextrose.

The departure of the hydrolysis from a simple logarithmic curve, is ascribed to the occurrence of several successive stages.

- VII. "*The Action of Cold Concentrated Hydrochloric Acid on Starch and Maltose.*" A. J. DAISH. *Transactions of the Chemical Society*, 1914. 105, 2053.

The action of cold fuming hydrochloric acid ($D^{15} 1'210$) on starch appears to be essentially the same as that brought about by taka-diastrase. The first stages—the formation of soluble starch, dextrose, and maltose—are, however, passed through very rapidly and after 135 minutes 86 per cent. of the theoretical quantity of dextrose has been formed. The action is apparently limited by the rate of hydrolysis of the maltose, since this is hydrolysed to dextrose not much more rapidly than starch itself.

When dextrose is dissolved in ordinary concentrated or fuming hydrochloric acid a certain amount of synthetic action always occurs, even when only 1 per cent. of the sugar is present. This fact, and the destruction of some of the dextrose, makes it impracticable to utilise cold fuming hydrochloric acid in the accurate estimation of starch or cellulose.

- VIII. "*The Velocity of Hydrolysis of Starch and Maltose by Cold Concentrated and Fuming Hydrochloric Acid.*" A. J. DAISH. *Transactions of the Chemical Society*, 1914. 105, 2065.

When fuming hydrochloric acid acts on maltose at the ordinary temperature the velocity coefficient is practically constant throughout the change; the action is unimolecular. But when either the fuming or ordinary concentrated acid acts on starch or soluble starch the velocity coefficient is not constant but rises continuously throughout the change; the departure from the ordinary logarithmic law is ascribed to the occurrence of successive stages. The transformation of cellulose by fuming hydrochloric acid is an extreme instance of an action occurring in successive stages, and the velocity co-efficient also shows considerable variation at different periods.

- IX. "*Methods of Estimation of Carbohydrates III. The Cupric Reducing Power of the Pentoses—Xylose and Arabinose.*" A. J. DAISH. *Journal of Agricultural Science*, 1914. 6, 255—262.

In the scheme of analysis of plant extracts outlined in last year's report (1913, pp. 12-14) allowance must be made for the pentoses present; it therefore became necessary to ascertain the exact value of the cupric reducing power under the standard conditions of Brown, Morris, and Millar. Pure specimens of the xylose and arabinose were therefore prepared and the reducing power at different degrees of concentration accurately determined. The results are given in tables and curves, enabling one to read off directly the weight of pentose corresponding to different weights of cupric oxide. The specific reducing power of the pentoses differs only very slightly from that of dextrose.

- X. "*The Estimation of Carbohydrates IV. The Presence of Free Pentoses in Plant Extracts and the Influence of other Sugars on their Estimation.*" W. A. DAVIS and G. C. SAWYER. *Journal of Agricultural Science*, 1914. 6, 406—412.

The existence of free pentoses in plants has not hitherto been recognised. Evidence is adduced to show that they are usually present in the alcoholic extracts of leaves of turnips, mangolds, potatoes and other plants. This also explains their accumulation in the "vinasses" of distilleries employing the molasses of the beet sugar industry.

It is shown that the pentoses may be estimated quantitatively with a fair degree of accuracy by the ordinary distillation process or from the reducing power of the purified liquor after other sugars have been fermented away with yeast.

When the amounts of pentoses are small, relative to the other sugars, it is advisable, as suggested by Kluyver, to ferment away these sugars completely before applying Kröber's process.

- XI. "*The Hydrolysis of Maltose by Hydrochloric Acid under the Herzfeld Conditions of Inversion.*" W. A. DAVIS. *Journal of Agricultural Science*, 1914. 6, 413—416.

It is shown that maltose undergoes slight hydrolysis (about 2 per cent. when 1 per cent. solutions of maltose are employed) when heated with hydrochloric acid under the Herzfeld conditions of inverting cane sugar. It is preferable, therefore, to adopt 10 per cent. citric acid as suggested in last year's report, as this acid causes no hydrolysis of maltose under the conditions prescribed.

- XII. "*Estimation of the Surface of Soils.*" J. A. HANLEY. *Journal of Agricultural Science*, 1914. 6, 58—62.

The method of König, Hasenbäumer, and Hassler for estimating soil colloids by means of dyes was considerably modified.

Methyl violet was used, as this colour undergoes only a slight change even after prolonged contact with the soil; but it was shown to be useless to employ dye solutions of the same strength for soils of the same type, e.g., for sands, 1 gram; for loams, 2 grams; for clays, 3 grams of dye per litre.

The reaction is a typical absorption and the quantity of dye taken up depends on the strength of solution used, an equilibrium setting up between dyed soil and dye solution. For comparing surfaces of soils it is therefore necessary to estimate the dye absorbed when the *dyed* soils are in contact with supernatant dye solutions of the same strength. A series of dye solutions has therefore to be used for each soil and the appropriate concentration picked out by trial.

- XIII. "*The Humus of Acid and Alkaline Peats.*" J. A. HANLEY. *Journal of Agricultural Science*, 1914. 6, 63—76.

Experiments were made to determine whether acidity or alkalinity of peats depends on the presence of acid or alkaline humus.

In every case part of the organic matter could be extracted without previous treatment with acid, and when the soils were arranged in order of the ratio of humus obtained without, to humus obtained with, previous acid treatment, alkaline fen peats, some of which contained carbonate, were found high in this "acidity" list.

Five typical peats were examined in more detail.

The humus determinations were repeated with ammonia and with caustic soda, and the nitrogen in the extracts was estimated, but the results, which were probably affected by the solubility of neutral organic matter, showed less connection with the other characteristics of the peats than those obtained by studying the inversion of sucrose.

Acid peats have a greater power of inverting sucrose than neutral peats. In order to eliminate the effect of varying organic matter content it was found necessary to compare the ratios of inversion after, to inversion before, treatment of the soil with dilute hydrochloric acid. For acid peats the ratio approached 1, for normal peats it was very low; in the case of a Cambridge Fen peat, 0.062.

- XIV. "*The Relative Effect of Lime as Oxide and Carbonate on certain Soils.*" H. B. HUTCHINSON and K. MACLENNAN. *Journal of Agricultural Science*, 1914. 6, 302—322.

Further evidence is adduced to show that caustic lime acts as a partial sterilising agent, as pointed out in the previous paper dealt with in last year's Report. The amount of lime required for this purpose is found to vary with different soils. Each soil absorbs a certain amount of lime; only the excess over and above this absorbed quantity produces any sterilising effect. In addition, the lime liberates a certain amount of ammonia. The available nitrogen in the soil as found by pot experiments was found to be comparable with the amounts of ammonia and nitrate produced in the laboratory experiments.

Chalk, on the other hand, has no partial sterilising action.

- XV. "*The Determination of Soil Carbonates.*" H. B. HUTCHINSON and K. MACLENNAN. *Journal of Agricultural Science*, 1914. 6, 323—327.

A method is described for rapidly estimating the carbonates in soil. The apparatus consists of two round bottomed flasks of 100

and 1,000 c.c. capacity respectively, connected by a glass tube and each provided with tap funnels. The smaller flask contains the soil, the latter is to hold the alkali for absorbing the CO_2 . The apparatus is partially evacuated (a water pump suffices for this purpose), the alkali is run into the larger and the acid into the smaller flask. After the first action is over air is allowed to bubble slowly through the mixture of soil and acid in order to carry the remaining CO_2 into the larger flask. The CO_2 is then determined by titration.

XVI. "*The Determination of the Lime Requirements of Soils.*" H. B. HUTCHINSON and K. MACLENNAN. *Chemical News*, 1914. 110, 61.

A new method is proposed for the determination of the lime requirements of the soil. 10 to 20 grams of the soil are placed in a bottle of 500-1,000 c.c. capacity together with 200-300 c.c. of approximately N/50 solution of calcium carbonate, and the air in the bottle is displaced by a current of carbon dioxide in order to insure against possible precipitation of calcium carbonate during the period of the determination. The bottle is then placed in a shaking machine for three hours, after which time the solution is filtered and an aliquot portion of the filtrate is titrated against N/10 acid, using methyl orange as indicator. The difference in strength of this filtrate and that of the initial solution represents the amount of calcium carbonate absorbed, each cubic centimetre of N/10 acid being equal to 5 mgrms. calcium carbonate. The method has been found to provide a useful index of the lack of base or "sourness" of the soil.

XVII. "*The Evaporation of Water from Soil.*" BERNARD A. KEEN. *Journal of Agricultural Science*, 1914. 6, 456—475.

The evaporation of water from sand, silt, china clay, and ignited soil is a relatively simple phenomenon which can readily be explained by the known laws of evaporation and diffusion. The evaporation from soil is more complex, something being present which operates in making the relation between the soil and the soil water of a different and closer nature than in the case of sand, etc. The effect is not due to the soluble humus, for the removal of this material from the soil does not appreciably affect the phenomena of evaporation. Any possible effect of the insoluble organic matter is largely eliminated by the consideration that *ignited* sand or silt behaves like the *unignited* material.

But when the colloidal properties of the clay are destroyed by ignition, the evaporation curve is completely altered, and becomes identical with that given by sand or silt. Again, evaporation from china clay, which shows very feeble colloidal properties, is of the same character as that from sand. We may infer then, that the colloidal properties of the clay fraction are partly, if not mainly, responsible for the characteristic shape of the evaporation curve from soil.

Further information on the process of evaporation has been obtained by a mathematical examination of the rate curve for soil. Two factors have been distinguished, which operate over practically the whole range of water content dealt with in these experiments.

In the first place the initial proportionality between water content and time observed with sand is not seen with soil, the curve being more exponential in character. This indicates that the relationship of water to soil is quite different from its relationship to sand, a circumstance which has been traced as already stated to the colloids. This relationship has at present only been expressed empirically but it is probably connected with the relation between vapour pressure and moisture content. But there is clearly something else at work for the curve is not of a simple exponential type. It is necessary to allow for another factor: the effect on the rate of evaporation of the decreasing water surface in the soil, the surface obviously diminishing in area as evaporation continues.

The equation finally developed is:—

$$A \frac{dw}{dt} = \sqrt[3]{\left(\frac{ws}{100} + 1\right)} [2.303 \log_{10} (w + K) - \log_e K],$$

where $\frac{dw}{dt}$ = rate of evaporation.

w = percentage of water present by weight.

s = specific gravity of the soil.

A and K = constants.

XVIII. "Some Notes on Soil Protozoa." C. H. MARTIN and K. R. LEWIN. *Philosophical Transactions*, 1914. 205 B, 77—94.

The authors show that the view current among zoologists limiting the range of free living protozoa to ponds, streams, the sea, etc., is not justified by the observations of the older investigators. A method is described by which it is easy to demonstrate in field and garden soils the presence of free living protozoa in a trophic state: it is as follows:—A small quantity of the soil is added to an equal quantity of a saturated aqueous solution of picric acid; the mixture is then stirred very thoroughly, so that the protozoa which are situated on the liquid films between the soil particles are freed. The mixture is then allowed to stand for 12—24 hours, and a scum gradually rises to the surface which contains a proportion of the bacteria and protozoa of the soil. Cover slip preparations are made by floating cover slips on the surface of the mixture, then transferred to corrosive sublimate solution or 70 per cent. alcohol, stained and mounted in the ordinary way.

At least eight organisms, some of them new, were discovered in a trophic state in a "sick" cucumber border. The thecamoebae were probably present in considerable numbers, as also were the amoebae, which contained large numbers of ingested bacteria. Neither flagellates nor ciliates were observed in any quantity in the trophic state. An organism, *Vahlkampfia soli*, appeared to be the dominant form during August and is described in some detail as it differs in certain points from *Amoeba limax*. It is very active and has a flagellate stage: in nuclear division it shows the phenomena of promitosis. A second organism, *Amoeba cucumis*, is also described; this is very sluggish and has characteristic pseudopodia.

An outdoor seedling bed was found to contain a smaller number of protozoa but in larger variety: a relationship very similar to that

observed on the Rothamsted grass plots, where the unmanured plot carries the largest number of species but a low crop, whilst the heavily manured plots carry a smaller number of species. One organism was described in some detail, *Amoeba gobanniensis*, which is of interest because it is closely allied to *Amoeba cucumis*. A *Chlamydothryx* was also found, different from the forms previously described.

NOTE.—In a later paper, which was almost ready for the Press when the authors were called up for military service, Messrs. Lewin and Martin describe another method by which protozoa in the trophic form were picked out of the soil alive. A glass tube $1\frac{1}{2}$ inches in diameter and about 2 feet long is closed at the lower end with a rubber bung through which passes a glass tube drawn out to a jet whereby a current of air can be blown through. The tube having been clamped upright, a newly made suspension of the soil in water is poured in until the water level nearly reaches the top. Three hooks are hung round the rim of the tube so as to furnish a support for the cover slip which is smeared with agar jelly and placed about $\frac{1}{4}$ inch above the water level. Air is now blown through the jet so as to produce a stream of fairly small bubbles rising through the suspension and breaking on the cover slip. After about 30 seconds the air stream is stopped and the cover slip lifted off and examined under a microscope.

XIX. "A Preliminary Communication on Three new *Proteomyxan Rhizopods from Soil*." T. GOODEY. "Archiv für Protistenkunde," 1914. 35, 80—102.

The organisms described in this paper were obtained in cultures of soil on a nutrient agar medium and the soils which yielded them were all of a rich character. They were first obtained from a cucumber house at Harpenden, and from a vinery in Hampshire; later on they were obtained from rich garden soils taken in Edgbaston, Birmingham, and Great Barr, Staffordshire. This shows that they are widely distributed. They are large amoeboid forms and are new to science.

Leptomyxa reticulata nov. gen. et. nov. spec. is the largest of the three and measures from $40\ \mu$ to 1 m.m. in greatest breadth. The protoplast spreads out into a thin sheet branching and anastomosing; pseudopodia often filose and delicate; contractile vacuoles numerous and minute, multinucleate, each nucleus small and possessing a comparatively large karyosome. Encystation into double-walled cysts; excystation of original organism.

Leptomyxa flabellata nov. spec. The protoplast is naked and spreads out into a thin sheet frequently resembling a fan in outline, measuring from $60\ \mu$ to $150\ \mu$ in breadth. The pseudopodia are filose but do not anastomose; contractile vacuoles numerous and minute. Multinucleate, possessing from 1 to 14 nuclei much larger than those of *L. Reticulata* each having a large karyosome. Encystation into double-walled cysts, the ectocyst showing well defined prominences; excystation of original organism.

Gephyramoeba delicatula nov. gen. et. nov. spec. The protoplast is naked and stretches out into long narrow arms

measuring from 60 μ to 250 μ from tip to tip. Contractile vacuoles numerous and minute. Encystation into single-walled cysts; excystation of original organism.

All three forms feed on bacteria and small amoebae and flagellates.

XX. "*The Estimation of Phosphates in Soil Extracts.*"
J. A. PRESCOTT. *Journal of Agricultural Science*,
1914. 6, 111—120.

The volumetric method originally proposed by Pemberton for the determination of phosphates has been investigated, and the conditions for obtaining accurate results with soil extracts have been worked out.

The following is the method finally adopted:—

REAGENTS. (1) Ammonium nitrate: 500 gms. to 1 litre of water.

(2) Ammonium molybdate: 150 gms. molybdate dissolved in 1,000 c.c. of water and poured into 1,000 c.c. of nitric acid (S.G. 1.2).

(3) 2 per cent. sodium nitrate.

PREPARATION OF SOIL EXTRACTS. A volume of the extract containing 5 to 10 m.g. of P_2O_5 is evaporated to dryness, ignited for 15 minutes at dull red heat, and where much silica is present, roasted for 2 hours at $140^\circ C$ to render the silica insoluble. The residue is extracted with 50 c.c. of 10 per cent. sulphuric acid by digestion for half an hour on a sand bath; the extract is diluted and filtered.

PRECIPITATION OF THE PHOSPHO-MOLYBDATE. To the above extract, 25 c.c. of the ammonium nitrate solution is added and the mixture brought to $55^\circ C$. 25 c.c. of the molybdate solution, previously brought to the same temperature, is then added and the mixture stirred, allowed to cool, and filtered after 2 hours. The precipitate is washed free from acid with 2 per cent. sodium nitrate solution, dissolved in standard alkali and titrated back using phenol phthalein as indicator. The factor recommended is 1 c.c. of $\frac{N}{10}$ alkali = '0003004 gms. P_2O_5 .

XXI. "*The Reaction between Dilute Acid Solvents and Soil Phosphates.*" J. A. PRESCOTT. *Proc. Chem. Society*, 1914. 30, 123.

A preliminary note showing that the reaction is more readily interpreted as a colloidal absorption phenomenon than as a simple chemical reaction (*see p.* 9).

XXII. "*The Nature and Amount of the Fluctuations in Nitrate Contents of Arable Soils.*" E. J. RUSSELL. *Journal of Agricultural Science*, 1914. 6, 18—57.

The amount of nitrate in the soil of arable land fluctuates regularly, but in these experiments it rarely exceeded the following values:—

	Per million	Per cent.	Lbs. per Acre, 0—18 ins.
Sand ...	6	'0006	28
Loam ...	23	'0023	115
(excepting on heavily dunged land, when it rose to 37 parts per million).			
Clay ...	14	'0014	60

In almost all the soils examined the accumulation of nitrate took place most rapidly in late spring or early summer. After this there was usually little if any gain, and frequently a loss. In the hot dry autumn of 1911, however, and again in 1913, the accumulation continued in some of the soils right on till September.

During the winter loss of nitrate took place. This was more marked in the wet winter of 1911—1912 than in the drier winter of 1908—1909.

The fluctuations in nitrate content are more marked on loams than on clays or sands. Clays lose less of their nitrates in winter, but, on the other hand, they accumulate smaller amounts in June and July. Sands lose much of their nitrates in winter and do not accumulate very large amounts in summer. It appears that the main loss in winter is due to leaching and not to denitrification.

On comparing the nitrate content of cropped and fallow land it is found that during late summer and early autumn the fallow land is the richer, even after allowing for the nitrate taken up by the crop. The question arises, whether the growth of a crop exerts any depressing effect on the rate of nitrate production in the soil. This is under further investigation.

The rapid rise in nitrate content in spring does not usually set in immediately the warm weather begins; there is a longer or shorter lag. There are indications of greater bacterial activity in early summer than later on, a phenomenon readily explicable on our view that the soil population is complex and includes organisms which are detrimental to the activity of bacteria, but which are, on the whole, more readily put out of action.

The supply of nitrate to the plant is known to be a factor of prime importance in plant growth. Similarly it is found that the factors which determine the accumulation of nitrates in the soil also play a great part in determining the amount of crop production. Thus heavy winter rainfall, which washes out nitrates, tends to reduce crop growth; on the other hand, hot dry summers, succeeded by dry winters, are shown to be favourable to nitrate accumulation, and therefore to crop growth.

XXIII. "*The Effect of Climate and Weather on the Soil.*"
E. J. RUSSELL. *Journal of the Royal Agricultural Society*, 1914. 74, 1—21.

Climate is shown to play a considerable part in determining the character of a soil, affecting not only the mineral particles, but also the nature and amount of the organic matter. Where definite climatic zones exist they are usually found associated with definite soil zones. Instances are quoted from North America and from Russia.

The effects of weather are of more direct interest to the cultivator, and involve the texture of the soil and also the stores of nitrates. Using the data collected in the preceding paper it is shown how a considerable saving may be effected by sowing a green crop at the end of a dry summer or autumn, which shall take up the nitrates and save them from being washed away. When the crop is ploughed in or folded, the nitrogen is returned to the soil in a form in which it is not readily lost.