VII.—*A Quantitative Investigation of the Bacterial and Protozoan Population of the Soil, with an Account of the Protozoan Fauna.*

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(From the Protozoology Department, Rothamsted Experimental Station.)

Contents.

Introduction.

Since the first half of the nineteenth century, it has been known that certain species of protozoa will grow in a suitable medium inoculated with soil. In spite of this, however, some later workers have asserted that such organisms did not occur in normal soils, while others, though recognising their presence, denied the existence of active forms.

To Martin and Lewin (28) must be ascribed the distinction of first showing that amœbæ and flagellates are present in field soil in a trophic condition; and owing to the labours of numerous investigators, it is now known that most soils contain these organisms; but unfortunately there is little or no knowledge as to their behaviour in natural conditions.

Earlier work (8) showed, however, that an investigation of the micro-population of field soil would yield interesting and important results, especially as it was evident that certain views held by soil biologists required modification; thus it has always been thought that the bacterial numbers in the soil changed very slowly, and investigators have not hesitated to take counts at monthly or even longer intervals. This assumption is incorrect, since the numbers of both protozoa and bacteria vary enormously from day to day.

These large changes appeared so important, both from the point of view of general biology and that of soil economy, that the present investigation was undertaken with the purpose of finding in what way the changes in any one group or species are related to those in another; also what influence external conditions, such as temperature, moisture and rainfall, have upon the soil micro-organisms.

The method adopted was to make systematic counts of the numbers of bacteria and of six species of protozoa in soil samples taken daily direct from the field, and, by statistical methods, to correlate these counts one with another and with data for external conditions.

Observations at shorter periods than 24 hours could not be made owing to the labour involved; but it was found possible to organise a team of workers so that soil samples were taken at the same hour (10 o'clock) for 365 consecutive days (July 5, 1920–July 4, 1921).* In this way a large body of data was obtained, which is presented in the following pages. It is recognised that with so much material there are probable interesting deductions to be made which have been overlooked by us. In an appendix, therefore, the salient figures are given to allow those interested to apply further statistical tests.

Species of Protozoa.

Of the species of protozoa recorded, the following six (two amœbæ and four flagellates) occurred constantly in sufficient numbers to admit the application of statistical methods to the results :—

Dimastigamœba gruberi (Schardinger).—This species has been described in detail by Wilson (36) under the name of *Nægleria gruberi*. It is also identical with the *Amœba punctata* of Dangeard (10) and *Vahlkampfia soli* of Martin and Lewin (27).

Species α.—A very small limax amœba, which has, up to the present, not been identified with any species previously described. Even when fully extended it rarely exceeds 10 μ in length. Its movements are slow, and it is never observed to travel far in any one direction. More frequently short, knobby pseudopodia are formed first at one point and then at another, and withdrawn again without any progression having been effected. In the living animal the protoplasm is refractile, and does not appear to be vacuolated or to show any structure, while the nucleus is not visible and no contractile vacuole has been observed. Staining reveals the

* Our thanks are due to Dr. M. Laurie and to Mr. P. H. H. Gray for their valuable assistance in the examination of the culture plates.

nucleus as composed of a karyosome and a very thin nuclear membrane, with no peripheral chromatin. Nuclear division is promitotic, resembling that described for *A. froschi* (29), and therefore corresponding to the second limax type in GLÄSER's (16) classification.

The cyst is large in proportion to the size of the active form, the diameter being about 8 μ. The outer wall is thick and has numerous pores, with the result that the cyst contents have a pentagonal or polygonal appearance in optical section. A very thin inner wall is closely applied to the cyst contents, which contain a large refractile granule similar to that occurring in many flagellate cysts. A full account of this species will be published later.

Heteromita sp.—The differentiation between the various species of *Heteromita* is in many cases not easy, and owing to the large number of cultures that had to be examined each day, it has not been attempted. Much the commonest form is one agreeing closely with the description of *Bodo repens* (KLEBS, 22), but the published descriptions do not make the distinctions between this species and *Bodo globosus* (PUSCHKAREW, 30) at all clear, and possibly both species have been present.

Heteromita lens (*Bodo lens*, MÜLLER, *Heteromastix lens*, WOODCOCK, 37) occurred in the cultures, and was also recorded under the name *Heteromita*, but it was not sufficiently numerous to affect the results.

Species γ.—This is an hitherto undescribed species. It is the smallest of the soil flagellates, varying from 3 μ to 6 μ in length and from 2 μ to 3 μ in greatest breadth. The body is slightly curved and a single flagellum arises on the concave side just behind the anterior end. Its movements are very like those of *Heteromita*, since the single flagellum adheres to the glass slide and is trailed quite passively, while the hind end of the body vibrates rapidly from side to side. The nucleus is near the anterior end and no contractile vacuole has been found. A further account of this species will be published separately.

Cercomonas sp.—Both *C. longicauda* and *C. crassicauda* were apparently present, but no attempt was made to distinguish between them.

Oicomonas termo (EHRENB.).—This is probably identical with *Monas termo*, found by MARTIN in sick soils. MARTIN (25), however, was unable to find a contractile vacuole, whereas in the present species a very distinct contractile vacuole is often present at the base of the flagellum.

SPECIES NOT COUNTED DAILY.

The dilutions employed in counting were chosen to give the maximum accuracy for the most numerous species, and although several other protozoa were recorded regularly in small numbers, the results obtained are not sufficiently accurate to justify detailed discussion. These less numerous species are the following :—

Amœba sp.—A binucleate amœba which occurred almost every day in numbers up to fifty per gramme. In appearance it is identical with *A. diploidea* (HARTMANN and NÄGLER, 18), but its life-history has not yet been completely worked out.

Nuclearia simplex (CIENKOWSKY).—This species is not included by CASH (2) in his list of British fresh-water Rhizopoda. It is distinguished from *N. delicatula* by its smaller size, the diameter of both active and cystic forms being only between 15 μ and 25 μ. It was found every day, and at times its numbers rose to as many as 3,000 per gramme.

Chlamydophrys stercorea (CIENK.).—A much less numerous form than the preceding.

Monas sp.—This species occurred constantly in small numbers.

Proleptomonas fæcicola.—Described by WOODCOCK (37) as a coprozoic species. It occurred rather less regularly than the preceding forms, but occasionally in considerable numbers.

Colpoda cucullus (O. F. M.).—Constantly present, but rarely exceeding fifty per gramme.

Other forms recorded occasionally in the course of the year were :—

Rhizopoda.

Amœba glebæ (DOBELL, 14).

Amœbæ limicola (RHUMBLER, 29A).—Rarely found.

Hartmannella hyalina (DANGEARD, 9).

Amœba sp.—This is probably *Arachnula impatiens* (CIENK., 13). Only living forms were examined and these resembled DOBELL'S description very closely in their habits and in the absence of any apparent nucleus.

Difflugia sp.—This would probably have been found much more frequently had it been practicable to incubate the plates for a longer period.

Flagellata.

Copromonas subtilis (DOBELL, 12), *Spiromonas angusta* (KENT), *Tetramitus rostratus* (PERTY), *Tetramitus spiralis* (GOODEY, 17A), *Helkesimastix fæcicola* (WOODCOCK), *Phyllomitus amylophagus* (KLEBS), *Ophidomonas* sp., *Bodo edax* (KLEBS) has been obtained from the same plot of land, but was not observed at all during the course of the count.

Ciliata.

Colpoda steinii, a much less abundant species than *C. cucullus*, *Colpidium colpoda*, *Pleurotricha* sp., *Gastrostyla* sp., *Balantiophorus* sp., *Enchelys* sp.

INFLUENCE OF EXTERNAL CONDITIONS ON PROTOZOA AND BACTERIA.

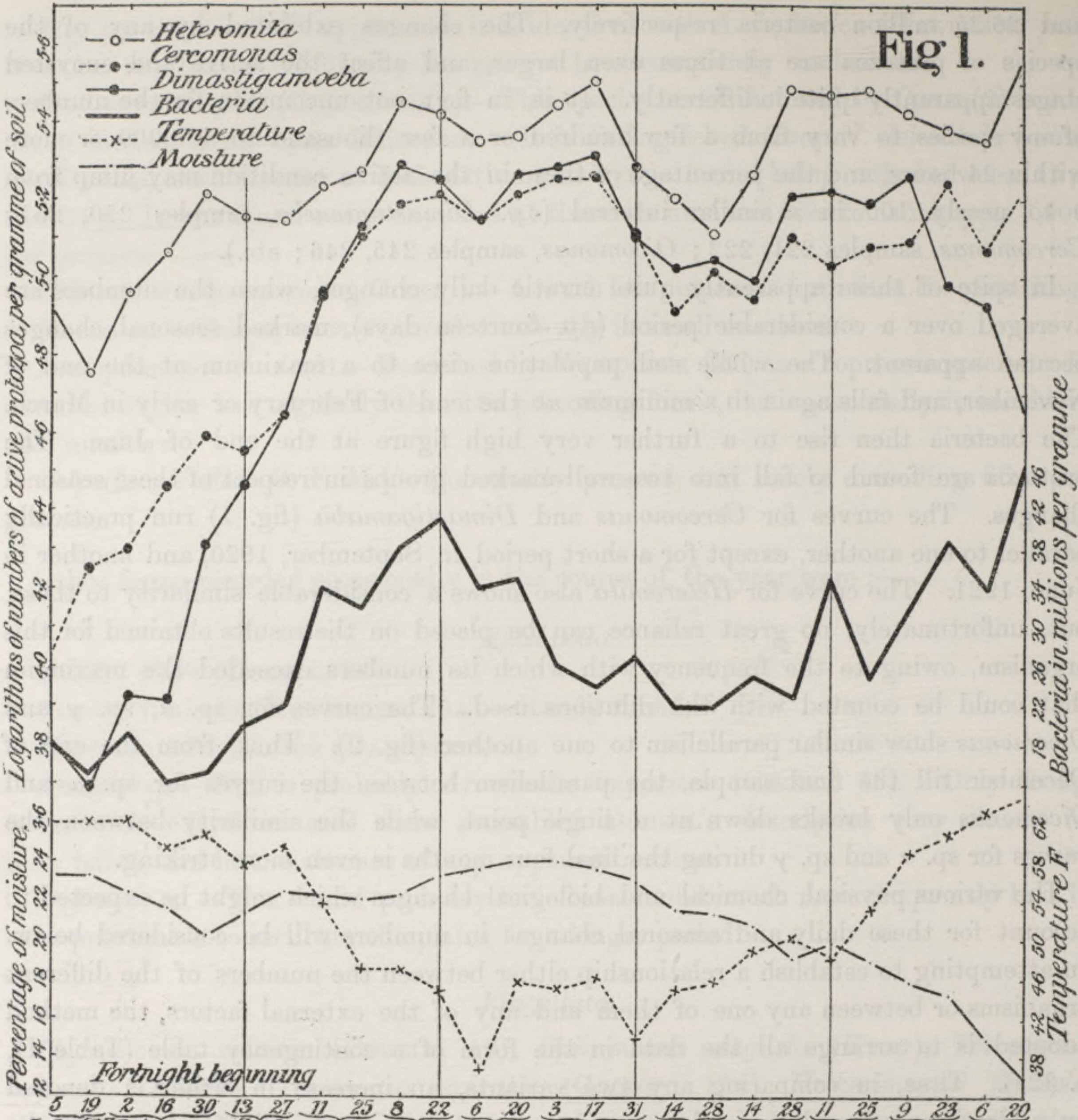
The numbers of all the organisms counted show large fluctuations of two kinds, viz., daily and seasonal. The size of the changes that take place within so short a time as 24 hours is perhaps the most surprising fact that the experiment has revealed. Thus the three consecutive samples 246, 247 and 248 give 58·0, 14·25 and 26·25 million bacteria respectively. The changes exhibited by any of the species of protozoa are at times even larger, and affect the active and encysted stages apparently quite indifferently. It is, in fact, not uncommon for the numbers of any species to vary from a few hundred or a few thousand to 400,000 or more within 24 hours, and the percentage of these in the active condition may jump from 0 to nearly 100 in a similar interval (*e.g. Dimastigamœba*, samples 250, 251; *Cercomonas*, samples 221, 222; *Oicomonas*, samples 245, 246; etc.).

In spite of these apparently quite erratic daily changes, when the numbers are averaged over a considerable period (*e.g.* fourteen days), marked seasonal changes became apparent. The whole soil population rises to a maximum at the end of November, and falls again to a minimum at the end of February or early in March. The bacteria then rise to a further very high figure at the end of June. The protozoa are found to fall into two well-marked groups in respect of these seasonal changes. The curves for *Cercomonas* and *Dimastigamœba* (fig. 1) run practically parallel to one another, except for a short period in September, 1920, and another in June, 1921. The curve for *Heteromita* also shows a considerable similarity to these, but, unfortunately, no great reliance can be placed on the results obtained for this organism, owing to the frequency with which its numbers exceeded the maximum that could be counted with the dilutions used. The curves for sp. α, sp. γ and *Oicomonas* show similar parallelism to one another (fig. 2). Thus, from the end of December till the final sample, the parallelism between the curves for sp. α and *Oicomonas* only breaks down at a single point, while the similarity between the curves for sp. α and sp. γ during the final four months is even more striking.

The various physical, chemical and biological changes which might be expected to account for these daily and seasonal changes in numbers will be considered below. In attempting to establish a relationship either between the numbers of the different organisms or between any one of them and any of the external factors, the method adopted is to arrange all the data in the form of a contingency table (Table II, p. 328). Thus, in comparing any two variants, an increase in either is denoted by a + sign and a decrease by a − sign. ++ will then indicate that both the variants are increasing together, −− that they are decreasing together, and +− or −+ that they are changing in opposite directions. If there is no relationship between the two, the total number of like signs (++ added to −−) throughout the year should be equal to the total number of unlike signs (+− added to −+). Any departure from equality will indicate a correlation between the two.

(1) *Influence of Soil Moisture.*

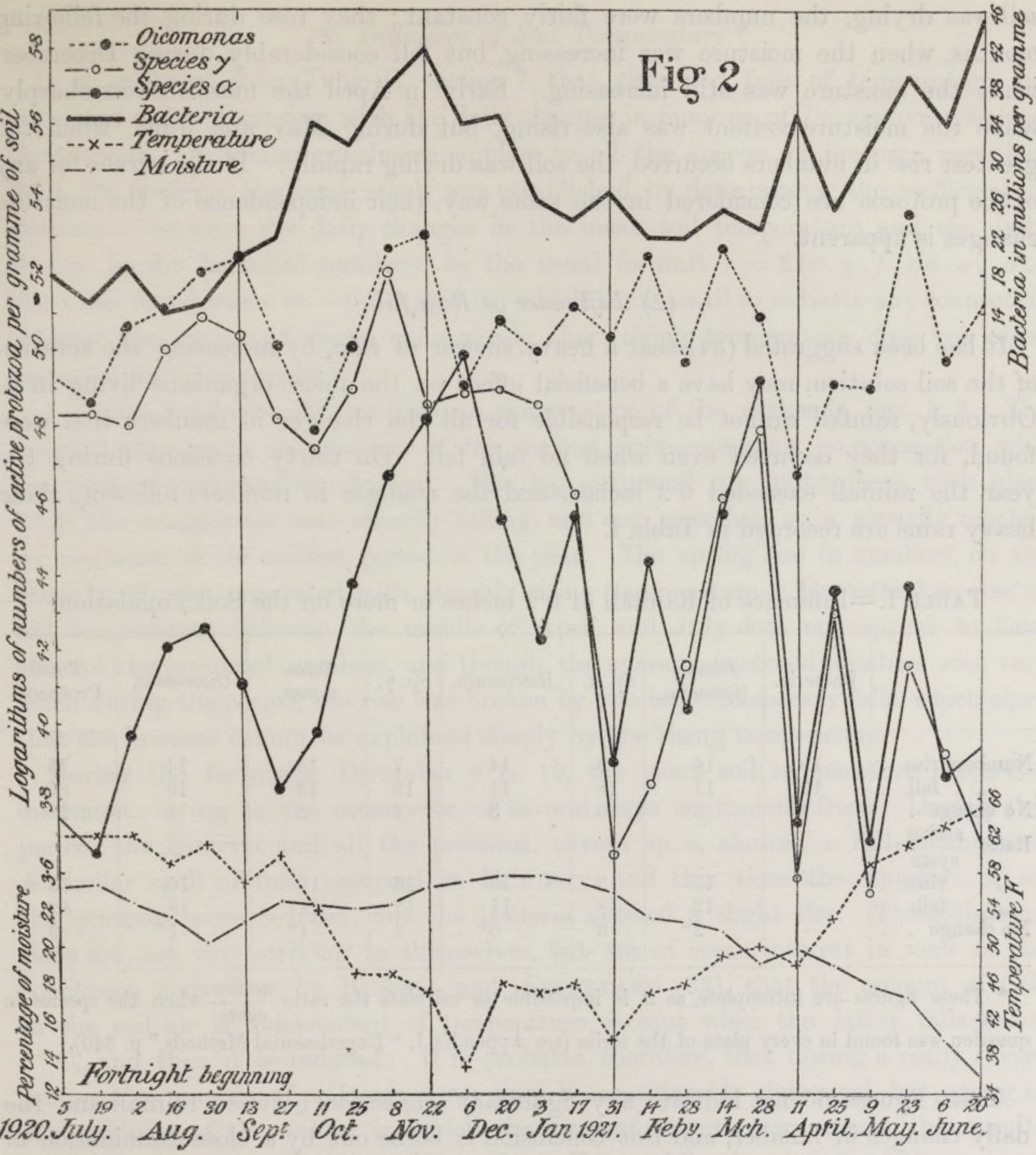
It has sometimes been claimed that the numbers of soil organisms are very closely linked up with the soil moisture. Thus Engberding (14*a*) states: "Im gewachsenen Feldboden steigt und fällt die Bakterienzahl in der wärmeren Jahreszeit mit seinem Wassergehalte," and Francé (14*b*) has recently extended this dictum to the whole of



Fortnightly averages of total numbers of *Heteromita*, *Cercomonas* and *Dimastigamœba*, and of bacteria, moisture and temperature.

the soil population. The present short-interval counts, however, lend no support to this assumption. The changes in moisture-content of the soil from one day to the next are invariably small, and frequently there is no appreciable change for several successive days, while the numbers of all the organisms may be fluctuating enormously (*e.g.* samples 222, 223, 224, 225). It is impossible, therefore, to explain the daily changes in the soil population by changes in moisture, and even the seasonal moisture changes have no clear influence. There is a very striking difference in the figures obtained for nearly all the organisms at the beginning

Fig 2



Fortnightly averages of total numbers of *Oicomonas*, sp. γ and sp. α, and of bacteria, moisture and temperature.

of July, 1920, from those for the end of June, 1921, and it is tempting to assume that these differences are in some way connected with the fact that the summer of 1920 was very wet, whereas 1921 was a year of unprecedented drought. Contrary to the assumptions quoted above, it is the drought which is in most cases associated with the high numbers. When the curves for average moisture and average numbers are compared in greater detail, however, it becomes impossible to draw any generalisations. Thus with the bacteria, during July and August, when the soil was drying, the numbers were fairly constant; they rose during the following months, when the moisture was increasing, but fell considerably during December, while the moisture was still increasing. Early in April the numbers rose sharply, while the moisture-content was also rising, but during May and June, when the greatest rise in numbers occurred, the soil was drying rapidly. If the curves for any of the protozoa are considered in the same way, their independence of the moisture changes is apparent.

(2) *Influence of Rainfall.*

It has been suggested (34) that a heavy shower of rain, by increasing the aëration of the soil solution, may have a beneficial effect on the micro-organisms living in it. Obviously, rainfall cannot be responsible for all the changes in numbers that were found, for they occurred even when no rain fell. On thirty occasions during the year the rainfall exceeded 0·2 inches, and the changes in numbers following these heavy rains are recorded in Table I.

TABLE I.—Influence of Rainfall of 0·2 inches or more on the Soil Population.

	Bacteria.	*Dimastigamœba.*	*Sp. α.*	*Heteromita.*	*Sp. γ.*	*Cercomonas.*	*Oicomonas.*	Total Protozoa.
Numbers rise	19	16	9	14	7	15	14	75
" fall	10	11	18	11	19	13	15	87
No change	1	3	3	5	4	2	1	18
Ratio $\frac{\text{active}}{\text{cysts}}$:—								
" rises	—	12	9	12	16	16	10	75
" falls	—	12	18	11	12	11	18	82
No change	—	2*	3	0*	2	1*	1*	9

* These figures are incomplete, as it is impossible to calculate the ratio $\frac{\text{active}}{\text{cysts}}$ when the species in question was found in every plate of the series (see Appendix I, "Experimental Methods," p. 340).

These figures do not indicate any significant connection between rainfall and the daily changes in number, and this conclusion is borne out by a close examination of the records. Thus, on April 13, when the first heavy shower for over two months occurred, the bacterial numbers fell by 8·75 millions, whereas on May 8, after three weeks of dry weather, a heavy shower was accompanied by a rise of 11·1 millions. It is also noteworthy that the greater part of the autumnal rise in numbers occurred at a time when the rainfall was low, while the numbers found at the end of November, and again at the end of December and early in January (figs. 1, 2), do not seem to be influenced by the heavy rains which occurred during these periods. Conversely large changes in the average numbers sometimes occurred (as at the end of March) without any corresponding change in the rainfall.

(3) *Influence of Soil Temperature.**

It is apparent from Table II, section 7, that rises and falls of temperature are associated quite at random with the daily changes in numbers of *Dimastigamœba* and *Cercomonas*. A similar conclusion applies to all the species of protozoa recorded. With the bacteria, the same result was established by determining the coefficient of correlation between the daily changes in the mean soil temperature and the daily change in the bacterial numbers, by the usual formula $r = \Sigma (x \,.\, y \,.\, f\,.\,)/n \,.\, \sigma_1 \,.\, \sigma_2$.† The value found was $r = -0{\cdot}149 \pm 0{\cdot}01$, which is too small to indicate any connection between the two, and shows conclusively that rising temperature does not cause increased bacterial numbers.

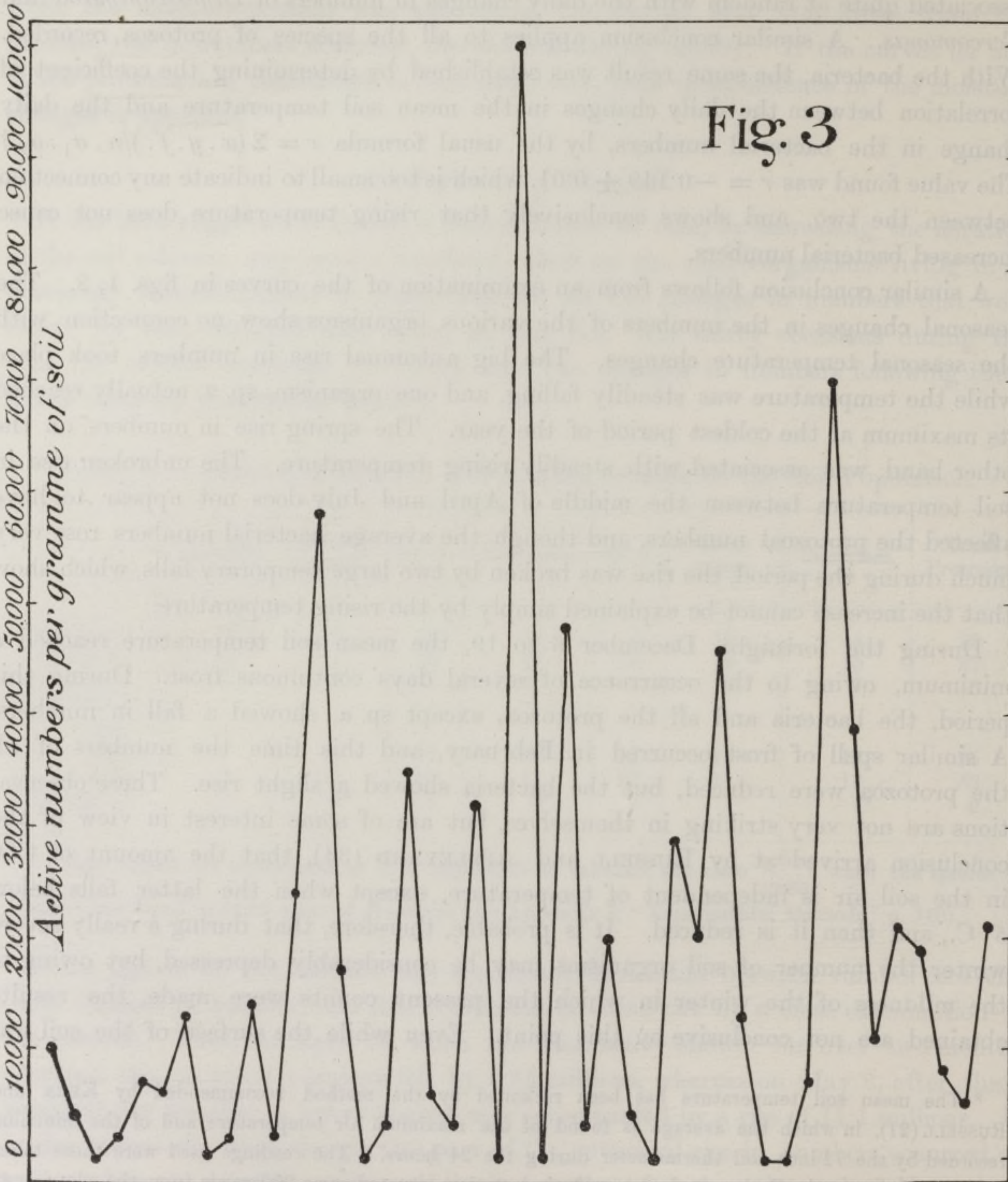
A similar conclusion follows from an examination of the curves in figs. 1, 2. The seasonal changes in the numbers of the various organisms show no connection with the seasonal temperature changes. The big autumnal rise in numbers took place while the temperature was steadily falling, and one organism, sp. α, actually reached its maximum at the coldest period of the year. The spring rise in numbers, on the other hand, was associated with steadily rising temperature. The unbroken rise in soil temperature between the middle of April and July does not appear to have affected the protozoal numbers, and though the average bacterial numbers rose very much during the period, the rise was broken by two large temporary falls, which show that the increase cannot be explained simply by the rising temperature.

During the fortnight, December 6 to 19, the mean soil temperature reached a minimum, owing to the occurrence of several days' continuous frost. During this period, the bacteria and all the protozoa, except sp. α, showed a fall in numbers. A similar spell of frost occurred in February, and this time the numbers of all the protozoa were reduced, but the bacteria showed a slight rise. These observations are not very striking in themselves, but are of some interest in view of the conclusion arrived at by RUSSELL and APPLEYARD (34), that the amount of CO_2 in the soil air is independent of temperature, except when the latter falls below 5° C., and then it is reduced. It is probable, therefore, that during a really severe winter the number of soil organisms may be considerably depressed, but owing to the mildness of the winter in which the present counts were made, the results obtained are not conclusive on this point. Even while the surface of the soil was

* The mean soil temperature has been reckoned by the method recommended by KEEN and RUSSELL (21), in which the average is found of the maximum air temperature and of the minimum recorded by the 12-inch soil thermometer during the 24 hours. The readings used were those taken daily at 10 A.M. at the Rothamsted meteorological station, situated some 200 yards from the plot; tests showed that the temperatures recorded at the two places were identical.

† r = coefficient of correlation; n = total number of cases; f = number of cases in class; x = deviation of class from mean temperature change; y = deviation of class from mean change in bacterial numbers; σ_1 = standard deviation of mean temperature changes; σ_2 = standard deviation of changes in bacterial numbers.

frozen, however, the numbers of all the organisms showed large apparently random daily fluctuations just as at other times. It was only when averages were taken that the depressions became apparent.



Active numbers of Cercomonas from July 5th to Aug 16th 1920.

(4) *Influence of Farm Operations.*

An increase in the numbers of bacteria in a field soil during the autumn has been observed by numerous workers in the past, and is commonly attributed either to the aërating effect of autumn ploughing, or to the nutritive effect of manuring. Neither of these explanations apply in the present case, for none of the farm operations (see Appendix I) were followed by any abnormal changes in numbers, and no manure was applied during the years 1920 and 1921.

(5) *Influence of Chemical Changes in the Soil.*

In preparing the soil sample each day for counting protozoal cysts, 10 c.c. of the acid was always withdrawn just before the dilutions were made, and was titrated with alkali in order to ascertain that its concentration, after neutralising the carbonate of the soil, was still sufficient to kill all the active protozoa. Slight differences were found from day to day in the amount of alkali required for the titration, and these must correspond to differences in the chemical conditions of the soil samples, but no connection could be found between them and the changes in the number of any of the organisms.

(6) *Physiological Influences—Autointoxication.*

A population of 500,000 individuals of any species of protozoa per gramme of soil represents about 2,500,000 per c.c. of soil solution, for 20 per cent. may be taken as a typical value of the water content of Rothamsted soil. If, as is not impossible, the organisms are not distributed uniformly throughout this medium, but are aggregated at some particular points among the soil particles, their density may be sufficiently high for the accumulation of their waste products, in the absence of constant renewal of the solution, to exert a depressing effect on the numbers as commonly occurs in artificial cultures. During the year, it was sometimes noticed that exceptionally high numbers of any organism on any day were followed by low numbers the next day, as if autointoxication had taken place. But an examination of the complete figures for the whole year does not substantiate this hypothesis.

Discussion.

From the foregoing sections it appears that, contrary to all expectations, no connection can be traced between climatic conditions or the other external factors considered and either the daily or the seasonal changes in the numbers of any of the soil organisms investigated. It is, however, difficult to resist the conclusion that some such connection must exist. The similarity between the seasonal curves for all the different organisms suggests that they are reacting together at any rate to the larger climatic changes. There is, moreover, small but significant correlation between the daily changes of some of the protozoa. Table II, section 2, shows that there is a definite excess of like signs when the daily changes in any two flagellates are taken together, and also when *Dimastigamœba* is taken with either *Cercomonas* or *Heteromita*.

The similarity that was observed between the seasonal changes in *Dimastigamœba*,

TABLE II.—Contingency Tables for various Soil Organisms.

(For explanation, see p. 321.)

	+ +	− −	+ −	− +	Total No. of Like signs.	Total No. of Unlike signs.	Probable error.
1. Between active amœbæ and bacteria—							
Inverse relationship—							
Dimastigamœba and bacteria	47	54	124	114	101	238	± 9
Sp. α and bacteria	79	87½	95	98½	166½	193½	± 9½
Dimastigamœba + *Sp. α* and bacteria	39½	44	128½	127	83½	255½	± 9
2. Between numbers of active individuals of different species of protozoa on the same day—							
Direct relationship indicated—							
Oicomonas and *Cercomonas*	56	46½	40	32½	102½	72½	± 6½
,, ,, *Heteromita*	43½	39	27½	19	82½	46½	± 5½
,, ,, *Sp. γ*	82	78	66	43	160	109	± 8
Sp. γ and *Heteromita*	78½	77½	47½	45½	156	93	± 8
,, ,, *Cercomonas*	92	98	66	75	190	141	± 9
Heteromita and *Cercomonas*	60	73	42	54	133	96	± 7½
Dimastigamœba and *Cercomonas*	104¼	109¾	76¼	69¾	214	146	± 9½
,, ,, *Heteromita*	87	77½	61	64½	164½	125½	± 8½
No relationship apparent—							
Dimastigamœba and *Sp. α*	86½	91½	84½	78½	178	163	± 9
,, ,, *Sp. γ*	92	91	91½	86½	183	178	± 9½
Sp. γ and *Sp. α*	89½	91½	92½	86½	181	179	± 9½
Sp. α and *Oicomonas*	52½	37½	45½	43½	90	89	± 6½
3. Between numbers of cysts of different species of protozoa on the same day—							
Direct relationship indicated—							
Oicomonas and *Cercomonas*	102	114½	61	72½	216½	133½	± 9½
,, ,, *Heteromita*	74	93	41	57	167	98	± 8
,, ,, *Sp. γ*	98½	123½	67½	71½	222	139	± 9½
Sp. γ and *Heteromita*	56	63	45	48	119	93	± 7
,, ,, *Cercomonas*	89	100½	85½	77	189½	162½	± 9½
Heteromita and *Cercomonas*	80	80	45½	53½	160	99	± 8
Dimastigamœba and *Sp. α*	98¼	102¼	75¾	82¾	200½	158½	± 9½
No relationship apparent—*Dimastigamœba* and *Sp. γ*	83¾	95¼	87½	95½	179	183	± 9½

4. Between total numbers (active and cystic) of different species of protozoa on the same day—							
Direct relationship indicated—							
Dimastigamœba and *Sp. α*	96	95½	77½	79	191½	156½	± 9½
" " *Cercomonas*	110¾	110¼	71¾	69¼	221	141	± 9½
No relationship apparent—*Dimastigamœba* and *Sp. γ*	93½	96½	89½	85½	190	175	± 9½
5. Between numbers of a single species of protozoa on successive days—							
Inverse relationship—							
Oicomonas:							
Actives of 1st day and actives of 2nd day	20¾	33¾	151¾	152¾	54½	304½	± 9
" " total "	43¾	57¼	129¾	128¼	101	258	
Total " " "	41½	56	130½	130	97½	260½	± 9
Dimastigamœba:							
Active of 1st day and active of 2nd day, etc.	43½	48½	130½	129½	92	260	± 9
Direct relationship—							
Oicomonas:							
Active of 1st day and cystic of 2nd day, etc.	88½	110½	85	75	199	160	± 9½
Heteromita:							
Active of 1st day and cystic of 2nd day, etc.	71½	58½	44½	36½	130	81	± 7
No relationship apparent—							
Oicomonas:							
Cystic of 1st day and cystic of 2nd day, etc.	46¼	73¼	119¾	118¾	119½	238½	± 9
Cercomonas:							
Active of 1st day and active of 2nd day, etc	49¼	62¼	120¾	117¾	111½	238½	± 9
" " cystic "	86	91	85	93	177	178	± 9½
Heteromita:							
Active of 1st day and active of 2nd day, etc.	41¼	31¼	74¾	94¾	72½	149½	± 7
Sp. γ:							
Active of 1st day and active of 2nd day, etc.	54¼	57¾	123¼	125¾	112	249	± 9
" " cystic "	83¼	93¾	98¼	85¾	177	184	± 9½
Dimastigamœba:							
Active of 1st day and cystic of 2nd day, etc.	93½	93	89	84½	186½	173½	± 9½
Cystic " " "	63½	65½	120	115	129	235	± 9
Total " total "	63	61	118	115	124	233	± 9
Sp. α:							
Active of 1st day and active of 2nd day	52	61	124½	126½	113	251	± 9
" " cystic "	79¼	89¼	97¾	94¾	168½	192½	± 9½
6. Between numbers of active and cystic *Oicomonas* on the same day	65¾	89¾	107¾	98¾	155½	206½	± 9½
7. Between the total numbers (active and cystic) and temperature—							
No relationship apparent—							
Cercomonas and temperature	98	75½	85½	104	173½	189½	± 9½
Dimastigamœba and temperature	96½	75½	85½	104½	172	190	± 9½

Cercomonas and *Heteromita* therefore extends also to some extent to the daily changes both in their cystic and active numbers. On the other hand although the seasonal curve for sp. α is so strikingly parallel to those for sp. γ and *Oicomonas* the daily fluctuations of this amœba are quite independent of the other two species. The absence of a preponderance of unlike signs, however (except in the case of the amœbæ and bacteria which will be considered separately, p. 331), shows clearly that there is no antagonism between any of the different organisms. It appears, therefore, that, though all the organisms respond alike to seasonal environmental changes, their responses to the more temporary environmental changes are much less uniform. From the available data, however, it is impossible to say what is the actual connection between the changes in numbers of any of the soil organisms and the changes in their environment. In view of the magnitude of the changes in numbers that occur from one day to the next, it is obviously desirable to obtain some knowledge of the changes that take place within the course of a single day and night; but this is at present impracticable, and probably the only way of advance at present is through an extensive series of laboratory experiments under rigidly controlled conditions.

It is interesting to note that the seasonal variations in the numbers of the soil organisms is very similar to those which have frequently been recorded in the case of many aquatic organisms. Miss Delf (11), for instance, found that in the ponds at Hampstead the algæ are most numerous in the early spring (February to the beginning of May) and again in October and November, some species disappearing entirely during the intervals. Somewhat similar seasonal changes have been recorded in a pond by Fritsch and Rich (15), in the British lakes by West and West (35), in the Illinois River by Kofoid (22a), and elsewhere.

In the sea also the planktonic diatoms have a great pulse of activity between March and June, and the dinoflagellates attain their maximum about a month later. Both groups have a smaller second maximum in the late autumn (19).

Thus the maxima for the soil population coincide closely with those of the marine and fresh-water plankton, except that whereas the aquatic forms are most active in spring and only show a minor phase of activity in autumn, the reverse appears to be true in the soil. It is difficult to resist the conclusion that the annual variations are produced by similar causes in each case. If this assumption is correct, it follows that the seasonal changes in protozoal numbers in the soil are not determined solely by the number of bacteria present, in spite of the fact that bacteria are probably the only food supply of the species in question, for the aquatic algæ which show such closely similar seasonal changes are not dependent on such a food supply. This conclusion is supported by the fact that the number of all the protozoa except *Oicomonas* rose during March, whereas the corresponding increase in the bacteria was delayed till the beginning of April.

The annual fluctuations in aquatic organisms agree further with those in the soil organisms in having little if any close relation with temperature. A marked annual periodicity was found by Apstein (1) even in a lake in Ceylon, where the total range of temperature during the year was from 25·94° C. in November to 28·28° in May. On the other hand, the fresh-water organisms are influenced very considerably by rainfall and general hydrographic conditions (see especially Apstein (1) and Kofoid (22a)), whereas no such influence could be found in the soil. Solar energy also undoubtedly influences the development of the phyto-plankton (15), and Herdman (19) regards it as being probably the chief cause of the great increase of life in the sea in the spring.

It is interesting to note that seasonal changes very similar to those found in the soil bacteria have been recorded for sewage bacteria. At the Dorking sewage works, Dr. (now Sir) A. C. Houston (31) found the highest numbers during October, November and December, while during February, March and April the numbers were consistently lower than at any other time of the year.

Relation between Protozoa and Bacteria.

Comparison of the numbers of bacteria with those of the active amœbæ (*Dimastigamœba* and sp. α) shows a definite inverse relationship between them. A contingency table (Table II, sec. 1) formed from these numbers shows that they vary inversely 255·5 times and together 83·5 times, with a probable error ±9·2 (figs. 4, 5, and 6, Samples 320–327). If the numbers of active individuals of *Dimastigamœba* and sp. α are considered separately with the bacteria, the result is as follows :—

	Like.	Unlike.
Dimastigamœba and bacteria . . .	101	238 (70 per cent. unlike).
Sp. α and bacteria	166·5	193·5 (54 per cent. unlike).

In both cases the difference in numbers is significant, but obviously *Dimastigamœba* is very much more effective in its action on bacteria than is sp. α; but in fifty-three cases it has either remained unchanged or varied with bacteria, while sp. α has varied inversely with them. Such cases occur at Samples 34, 41, 49, etc. (fig. 6, Samples 162, 163). An examination of the data shows that generally, where the influence of sp. α on the bacterial numbers is apparent, the numbers of *Dimastigamœba* are either changing very slightly (in twenty of the fifty-three cases they have not changed at all), or else the changes in sp. α are large (*e.g.* Samples 137, 275).

Out of the 339 days considered in the formation of these contingency tables, *Dimastigamœba* shows a controlling influence on the bacteria 70 per cent. times, and sp. α another 16 per cent. There remain then 14 per cent. or forty-eight days on which the changes in numbers in the bacteria cannot apparently be attributed to the action of the amœbæ, since they vary directly and not inversely with each other. Certain of these can be explained by reference to the amœbæ if the cysts as well as the active forms are taken into consideration. At Sample 28 the numbers of sp. α are practically unchanged, but the bacteria and *Dimastigamœba* have risen ; in the

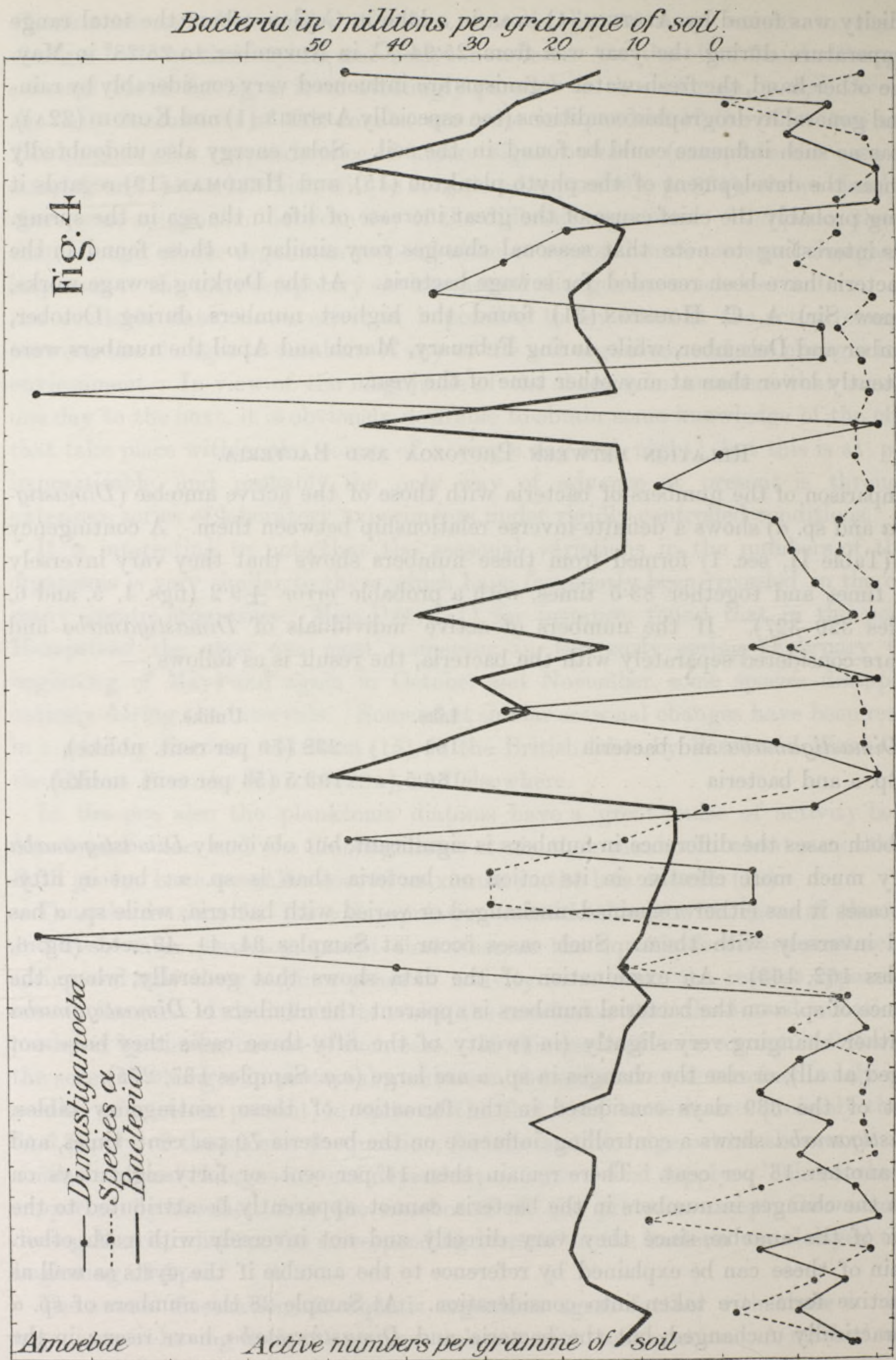
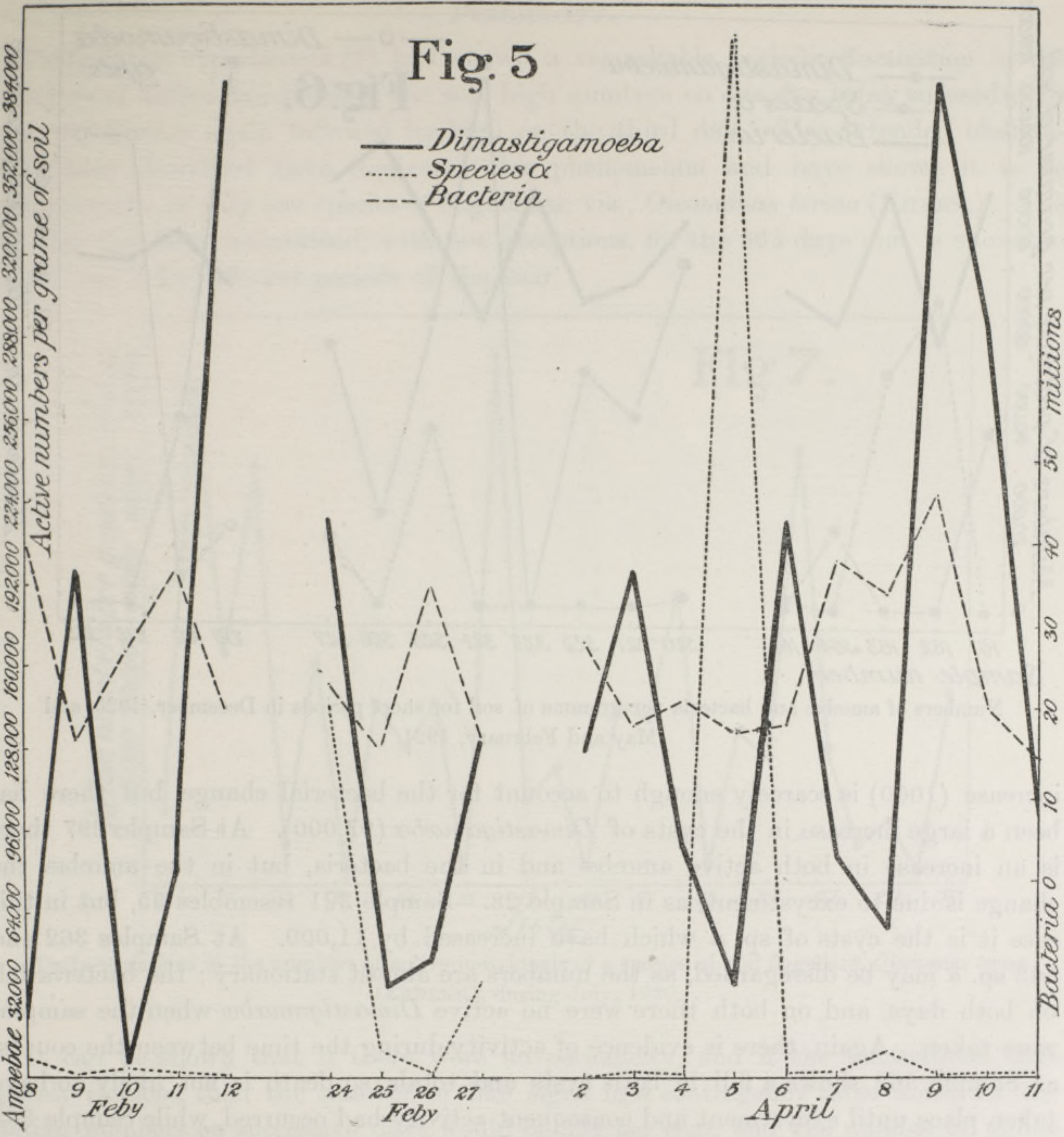


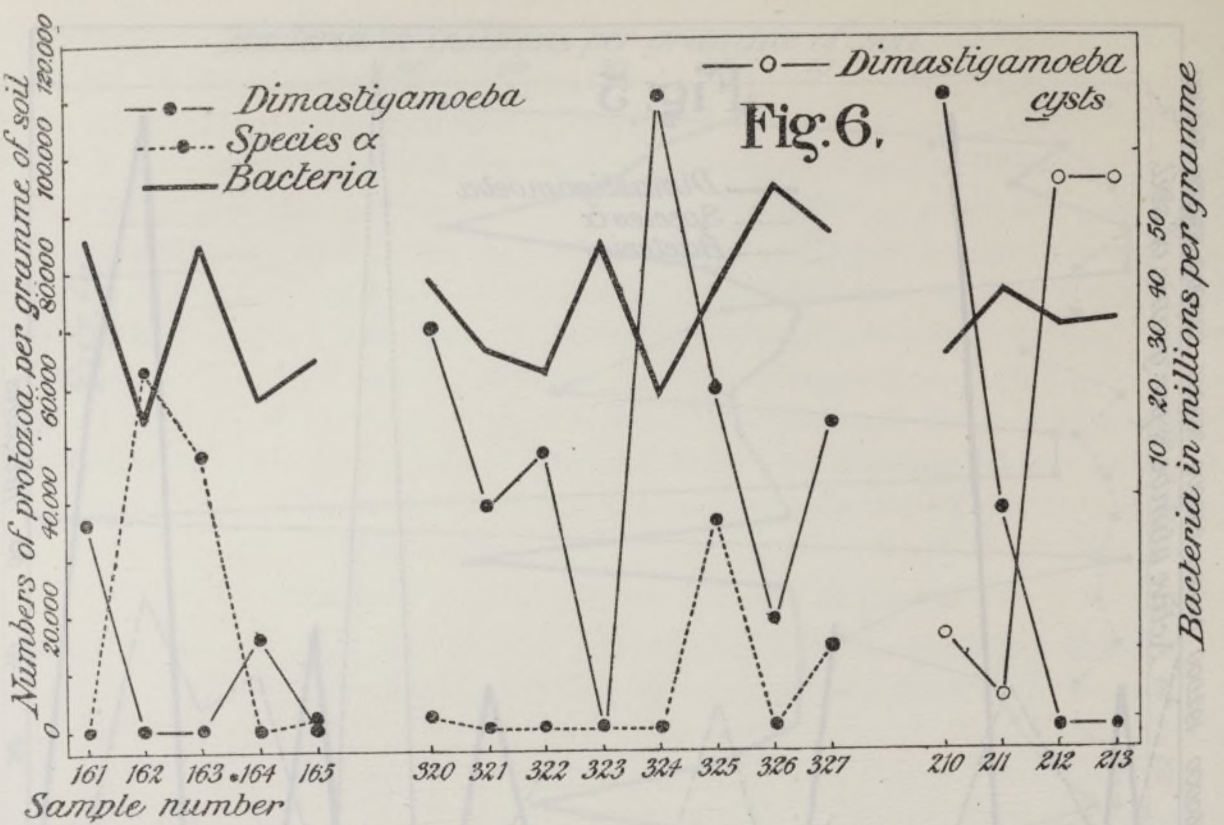
Fig. 4.

Daily numbers of active amœbæ (*Dimastigamœba* and sp. α) and bacteria in 1 grm. of field soil, from August 29 to October 8, 1920.



Numbers of active amœbæ (*Dimastigamœba* and sp. α) and bacteria in 1 grm. of field soil for typical periods in February and April.

latter, however, the rise is entirely due to excystment, so that the influence upon the bacteria would naturally have been slight, especially if the excystment did not occur until near the time of sampling. At Sample 35 sp. α is again unchanged, the bacteria fall, and in *Dimastigamœba* the active numbers fall slightly, though the totals rise by 8000, showing that considerable activity has gone on in the preceding twenty-four hours, terminating in cyst formation. At Sample 212 (fig. 6) there is a similar case; the bacteria and active *Dimastigamœbæ* fall, and though sp. α rises, the



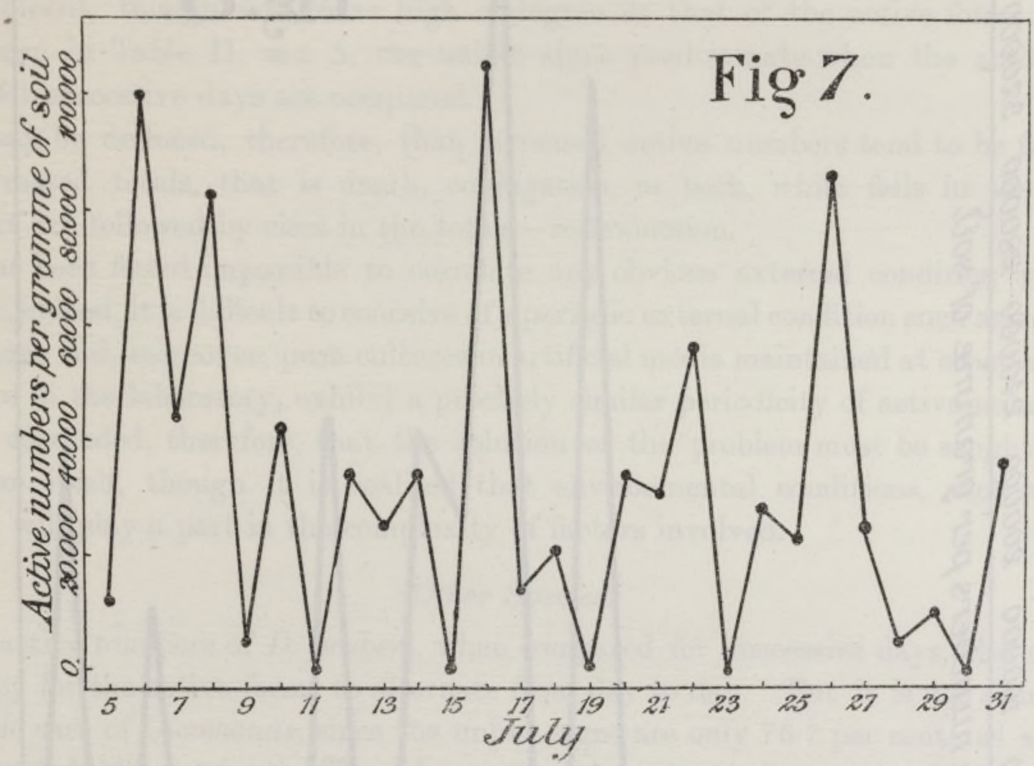
Numbers of amœbæ and bacteria per gramme of soil for short periods in December, 1920, and May and February, 1921.

increase (1000) is scarcely enough to account for the bacterial change, but there has been a large increase in the cysts of *Dimastigamœba* (97,000). At Sample 297 there is an increase in both active amœbæ and in the bacteria, but in the amœbæ the change is due to excystment, as in Sample 28. Sample 321 resembles 35, but in this case it is the cysts of sp. α which have increased by 11,000. At Samples 362 and 363 sp. α may be disregarded, as the numbers are almost stationary; the bacteria fell on both days, and on both there were no active *Dimastigamœbæ* when the samples were taken. Again, there is evidence of activity during the time between the counts, as Sample 362 shows a fall in both cysts and totals, so death is not likely to have taken place until excystment and consequent activity had occurred, while Sample 363 shows a rise in cysts and totals, again implying that there has been an intervening period of activity. There are numerous other cases where the same thing has occurred, but in these the active numbers at the time of sampling are sufficient to account for the bacterial changes.

From an inspection of the figures it is not possible to decide whether the interaction is primarily one in which the amœbæ influence the bacteria or *vice versâ*. The possibility of the bacterial changes initiating those in the amœbæ is, however, entirely ruled out by the fact that the contingency tables show no positive correlation between the changes in numbers of the active forms in the two amœbæ.

PERIODICITY.

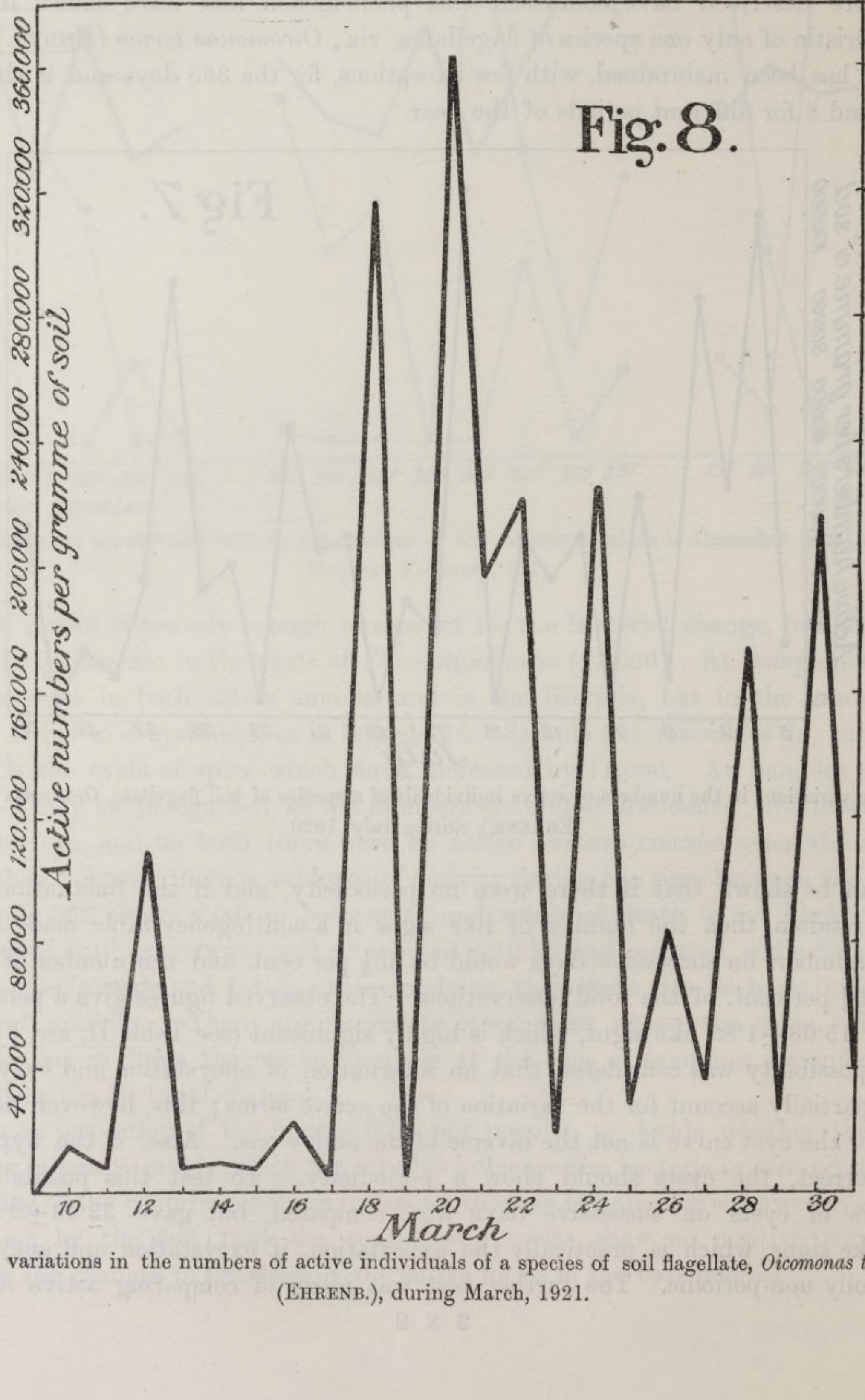
Preliminary experiments (8) had shown a remarkable periodic fluctuation in the numbers of active flagellates in the soil, high numbers on one day being succeeded by low, which were again followed by high on the third day. The extended observations here described have confirmed this phenomenon and have shown it to be characteristic of only one species of flagellates, viz., *Oicomonas termo* (EHREN.). The rhythm has been maintained, with few exceptions, for the 365 days and is shown in figs. 7 and 8 for different periods of the year.



Daily variations in the numbers of active individuals of a species of soil flagellate, *Oicomonas termo* (EHRENB.), during July, 1920.

It can be shown that if there were no periodicity, and if the fluctuations were purely random, then the number of like signs in a contingency table made for the active numbers on successive days would be $33\frac{1}{3}$ per cent. and the number of unlike signs $66\frac{2}{3}$ per cent. of the total observations. The observed figures give a percentage of only $15 \cdot 08 \pm 1 \cdot 89$ like signs, which is highly significant (see Table II, sec. 5).

The possibility was considered that an alternation of encystation and excystation might partially account for the variation of the active forms; this, however, does not hold, for the cyst curve is not the inverse of the active one. Also, if the hypothesis were correct, the cysts should show a periodicity. To test this possibility the numbers of cysts on successive days were compared, but gave $33 \cdot 43 \pm 2 \cdot 49$ per cent. like signs, which is practically the expectation, if excystation and encystation are wholly non-periodic. The further test was made of comparing active forms of one day with the cystic of the next. The result was 199 like, 160 unlike signs. Now, if there is no connection between the two, that is, if high active numbers do not tend to cause cyst formation, or, conversely, the expectation would be 50 per cent. like to 50 per cent. unlike signs, which is not widely different from the



Daily variations in the numbers of active individuals of a species of soil flagellate, *Oicomonas termo* (EHRENB.), during March, 1921.

numbers obtained. There is, however, a small significant excess of like signs, brought about by the tendency for a drop in active numbers, to be followed by a similar drop in the number of cysts (see Table II, sec. 5). The significance of this tendency is, however, sufficiently small to render it almost negligible. Thus the hypothesis of regular excystation and encystation will not completely account for the periodicity of the active forms. As would be expected from the above considerations, the total numbers of *Oicomonas* are periodic to a small extent. Thus the comparison of the totals for successive days gives 27·33 per cent. $\pm$2·35 of like signs, which is significant, though not to as high a degree as that of the active forms. Also, as is seen in Table II, sec. 5, the unlike signs predominate when the actives and totals for successive days are compared.

It may be deduced, therefore, that increased active numbers tend to be followed by decreased totals, that is death, conjugation, or both, while falls in the active numbers are followed by rises in the totals—reproduction.

It has been found impossible to correlate any obvious external condition with the rhythm, indeed, it is difficult to conceive of a periodic external condition such as would be necessary; and, moreover, pure cultures in artificial media maintained at constant temperature in the laboratory, exhibit a precisely similar periodicity of active numbers.

It is concluded, therefore, that the solution of the problem must be sought in the organism itself, though it is realised that environmental conditions, such as food supply, will play a part in the complexity of factors involved.

Other Species.

The active numbers of *D. gruberi*, when compared for successive days, also show a tendency for the active forms to alternate from day to day. But it is not so marked as in the case of *Oicomonas*, since the unlike signs are only 76·7 per cent., as against the expected 66·6 per cent. The other species show no tendency to periodicity when tested for a two days' period, for their active numbers are found distributed in a random way. It is possible that a rhythm of a longer interval—three, four, or five days —may obtain, but this can be tested only by experiment in controlled culture media.

In no case is there evidence that cyst formation occurs regularly ; in every species considered, the results of comparisons between actives and cystic, or between the cystic numbers for alternate days, are what would be expected on the supposition of random excystation and encystation, or the departure from expectation is too small to be worth comment. Thus, although excystation and encystation play a part in the variation of the active numbers, the preponderating influence is death, conjugation and reproduction, causing variation in the total numbers of the species.

Summary.

1. The results of 365 consecutive daily counts of the numbers of bacteria and of six species of protozoa in a normal field soil are given, and the methods of counting bacteria and protozoa are described.

2. The numbers of both bacteria and protozoa rarely remain the same from one day to the next. The fluctuations are very great, but it has not been found possible to connect them with meteorological or general soil conditions.

3. Fourteen-day averages of the daily numbers demonstrate that well-marked seasonal changes in the soil population are superimposed on the daily variations in numbers. In general, both bacteria and protozoa are most numerous at the end of November and fewest during February. These changes are not directly influenced by temperature or rainfall, but show a similarity to the seasonal fluctuations recorded for many aquatic organisms.

4. There is a slight tendency for the various species of flagellates to fluctuate together from day to day, but this is not shown by the two species of amœba.

5. An inverse relationship is found between the numbers of bacteria and active amœbæ in 86 per cent. of the total observations.

6. A two-day periodicity obtains for the active numbers of one species of flagellate, *Oicomonas termo*.

APPENDIX I.

Experimental Methods.

Sampling of Soil.

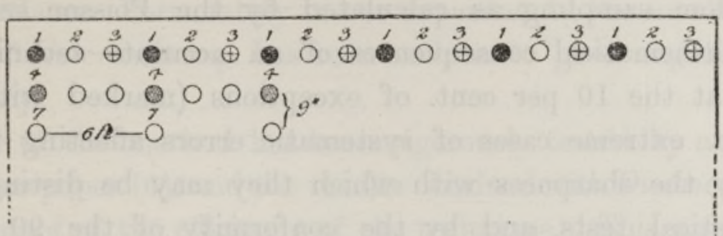
The samples of soil were taken from the plot on Barnfield, Rothamsted, which annually receives farmyard manure at the rate of 14 tons to the acre. The portion of the plot chosen was approximately 20 yards long by 12 yards broad. No crop was grown during the period of the experiment, and no manure added in the year 1920; but, in July–August, 1920, it was horse-hoed; on February 5–7, 1921, ploughed; on March 3, cultivated; on April 15–16, 27–28, rolled; and in May, June, and July, hoed.

The samples were taken at the same time (10 a.m.) every day, the method being as follows:—

Six 9-inch borings, taken at intervals of about 6 feet in a line across the plot, and thoroughly mixed together formed Sample 1, which was taken to the laboratory in a sterile tin.

The second sample was taken on the following day in the same manner and on the same line as the first, except that the borings were made about 2 feet away. The third sample borings were made about 2 feet from those of the second sample, thus completing the first line across the plot. (See Map, p. 339.) For the next three samples a second line about 9 inches from the first was used, and so on to the end of the series. This method of sampling was adopted to obviate, as far as possible, errors due to unequal distribution of the micro-organisms over the plot, also to ensure against such an error control experiments were made. On three occasions four samples were taken simultaneously across the plot, and counted. The results for the protozoa and bacteria were in no case significantly different in the four samples. Further, for the last fifteen days a duplicate sample was taken 6 yards back, and the bacteria counted;

the results of the two counts were concordant. These experiments justify the belief that unequal distribution in the field cannot account for the daily fluctuations in number of the organisms.



Map showing method of taking soil samples, each sample being formed from six borings, and each boring being denoted by the respective numeral.

Bacteria.

The nutrient medium employed is a modification, prepared by Mr. H. G. THORNTON and as yet unpublished, of one described by CONN. It consists of :—

Distilled water	1	litre.
K_2HPO_4	1	grm.
KNO_3	0·5	,,
$MgSO_4$	0·2	,,
$CaCl_2$, KCl, NaCl	0·1	,, each.
Asparagine	0·5	,,
Agar	15·0	,,
$FeCl_3$	0·002	,,

After the substances have melted in a steamer 1 grm. of mannite is added, and the P_H value adjusted to 7·3.

Since this medium contains only simple chemical substances it is not liable to the variations which often occur in successive preparations containing peptone, lemco, or gelatine. Such variations have been found to be sources of considerable error in counting.

The bacteria are counted by a dilution method, practicable direct means being yet unknown ; 10 grm. of soil, which have passed through a 3-mm. sieve, are placed in 250 c.c. of sterile physiological salt solution, contained in a 750 c.c. flask, and shaken for 5 minutes, thus giving a 1/25 dilution. 1 c.c. of this is shaken for 1 minute in 100 c.c. of saline contained in a 300 c.c. flask ; 1 c.c. of this second dilution is shaken for 1 minute in a 100 c.c. of saline contained in a 300 c.c. flask, thus giving a dilution of 1/250,000. Of this 1 c.c. quantities are placed in the bottom of five separate sterile Petri dishes ; 5 c.c. of the nutritive medium are added, and a thorough admixture ensured by rotating the dish. After incubation at 20° C. for 7 days the total number of colonies are counted, and the numbers of bacteria in millions per gramme calculated.

Strict adherence to the details of the technique is necessary, otherwise the results are not reliable.

The data obtained have been examined statistically by Mr. R. A. FISHER, and in nearly 80 per cent. of the 365 daily counts of bacterial numbers, the distribution of variation in the numbers of colonies on each plate is strictly that which should be obtained by random sampling as calculated by the Poisson series. This series represents the mathematical consequences of an accurate technique of sampling and dilution. That the 10 per cent. of exceptions (marked with an asterisk in Table IV) are not extreme cases of systematic errors affecting the whole of the data, is shown by the sharpness with which they may be distinguished from the rest by the statistical tests, and by the conformity of the 90 per cent. to the theoretical distribution. For these latter counts the probable error would therefore be approximately 2/3 of the standard error, which depends solely on the total number of colonies counted. Thus, if the total number of colonies counted on a series of plates is 291, this figure is affected by a standard error of $\sqrt{291} = 17{\cdot}1$, or by a probable error $17{\cdot}1 \div 2/3 = 11{\cdot}4$. Since this example was obtained from a four-plate series, the result in millions per gramme is obtained by dividing by 16 giving $18{\cdot}2 \pm 0{\cdot}71$.

The probable error varies with each count made, but for the whole series (excluding the exceptions) it is between 4–6 per cent.

Protozoa.

The protozoa are counted by a dilution method, the essentials of which have already been described (7). In the present experiment the three ranges of dilutions employed at different times of the year (see Table IV) were 1/25–1/102,400, 1/50–1/204,800 and 1/50–1/409,600. It has been shown (6, 7) that a more complete census of the soil population is obtained if the Petri dishes, inoculated from each dilution, are incubated for twenty-eight days and examined weekly, since a few protozoa, *e.g. Chlamydophrys*, develop only slowly; but it was impracticable to continue observations for so long owing to the great labour involved. The counts, therefore, refer only to those species which reproduce sufficiently rapidly to give accurate results after fourteen days' incubation. Even with this restriction over 100 plates were examined daily, and there were always about 700 plates in the incubator room.

The numbers of organisms in the soil sample are deduced as follows:—

For a given soil population the average number of negative plates in a series (that is plates showing no protozoan development) can be calculated, consequently any observed number of negative plates will represent a certain density of population in the soil sample investigated. Such calculations have been done by Mr. R. A. FISHER who compiled Table III for us. Thus in a series of dilutions ranging from 1/25–1/102,400 five negative plates will represent an average population of 16,000 per gramme of soil. The probable error of observations made in this way can best be regarded as one of plate difference, a two-plate difference not being significant, thus a four-negative plate result is not significantly different from a two- or six-negative plate result. Such a probable error holds so long as the number of plates falls in the intermediate portion of the series, at each extreme end (one or two negative plates or twenty-four or twenty-five negative plates) the error becomes considerable and difficult to calculate. Fortunately in our experiments the number of organisms with the exception of *Heteromita* sp. has been such as to give negative plates within the range of accuracy.

The accuracy of the method has been rigorously tested by a series of experiments given in a earlier paper (6), and the results obtained with *Oicomonas termo* (see p. 335) afford further proof of the efficiency of the technique. This animal shows a two-day periodicity of the active forms which can hardly be ascribed to experimental error.

The number of negative plates are given in Table IV and the actual figures may be obtained by reference to Table III.

TABLE III.—Tables showing the Number of Soil Organisms per gramme of Soil corresponding to the Number of Negative Plates in the various Dilution Series used.

1. Dilutions 25–102,400.		2. Dilutions 50–204,800		3. Dilutions 50–409,600.	
No. of sterile plates.	Protozoa per gramme.	No. of sterile plates.	Protozoa per gramme.	No. of sterile plates.	Protozoa per gramme.
1	110,000	1	220,000	1	420,000
2	59,000	2	118,000	2	230,000
3	36,000	3	72,000	3	140,000
4	23,000	4	46,000	4	95,000
5	16,000	5	32,000	5	64,000
6	11,000	6	22,000	6	44,000
7	7,600	7	15,200	7	30,000
8	5,300	8	10,600	8	21,000
9	3,700	9	7,400	9	15,000
10	2,600	10	5,200	10	10,000
11	1,800	11	3,600	11	7,300
12	1,300	12	2,600	12	5,100
13	900	13	1,800	13	3,600
14	640	14	1,280	14	2,600
15	450	15	900	15	1,800
16	320	16	640	16	1,300
17	230	17	460	17	900
18	160	18	320	18	640
19	110	19	220	19	450
20	79	20	158	20	320
21	56	21	112	21	220
22	38	22	76	22	160
23	25	23	50	23	110
24	15	24	30	24	77
25	6·8	25	13·6	25	51
				26	30
				27	14

TABLE IV.—Summary of Complete Data (to be used in Conjunction with Table III).

The asterisks against certain bacterial numbers denote that such figures do not conform to the Poisson Series (see p. 340) and have therefore an unknown experimental error. From Sample 1–22 the range of dilutions employed for counting total numbers of Protozoa was 1/25–1/102,400; from Sample 23–123 it was 1/50–1/204,800; and from Sample 124–365 it was 1/50–1/409,600. The range used for estimating the number of cysts throughout the experiment was 1/25–1/102,400.

Date.	Sample No.	H_2O content.	Rainfall 8″ gauge.	Max. air temp. in °F.	Min. soil temp., 12″ thermometer.	Bacteria in millions per grm.	Protozoa (numbers of negative plates).											
							Dimastigamœba gruberi.		*Sp. α.*		*Heteromita sp.*		*Sp. γ.*		*Cercomonas sp.*		*Oicomonas termo.*	
							Total.	Cystic.	Total.	Cystic.	Total.	Cystic.	Total.	Cystic.	Total.	Cystic.	Total.	Cystic.
1920.																		
July 5	1	21 ·64	0 ·594	55 ·0	59 ·0	8 ·05	9	10	9	12	3	4	3	4	6	13	3	4
„ 6	2	22 ·89	0 ·488	61 ·0	57 ·0	30 ·125	13	11	14	18	0	6	1	9	8	13	1	8
„ 7	3	25 ·10	0 ·158	60 ·1	58 ·0	24 ·825	9	10	12	21	0	0	4	9	14	11	1	2
„ 8	4	23 ·98	0 ·150	60 ·3	58 ·0	17 ·9	8	10	11	13	0	3	1	13	8	9	0	5
„ 9	5	23 ·26	—	63 ·1	58 ·0	24 ·95	11	10	14	15	3	5	5	7	7	13	5	6
„ 10	6	21 ·98	0 ·119	61 ·9	57 ·9	9 ·65	10	9	10	25	0	1	4	8	7	11	0	2
„ 11	7	24 ·54	0 ·115	67 ·1	58 ·3	20 ·75	10	10	10	13	2	3	3	4	5	10	3	3
„ 12	8	23 ·12	0 ·008	68 ·5	60 ·2	39 ·5	4	4	9	10	0	0	3	11	11	12	2	4
„ 13	9	21 ·3	—	65 ·8	60 ·2	13 ·825	5	10	6	13	2	7	4	7	10	13	3	6
„ 14	10	20 ·38	0 ·089	66 ·9	59 ·8	12 ·825	6	8	9	11	1	3	5	7	5	11	2	4
„ 15	11	23 ·09	0 ·005	66 ·9	60 ·0	18 ·375	6	6	5	11	1	1	0	6	7	8	3	2
„ 16	12	20 ·5	—	68 ·1	59 ·3	9 ·875	6	7	7	19	1	4	2	5	4	12	1	13
„ 17	13	21 ·68	0 ·083	69 ·5	61 ·0	8 ·5	6	10	9	22	2	6	2	13	2	13	4	7
„ 18	14	19 ·11	—	66 ·8	60 ·3	12 ·5	7	9	9	13	0	1	2	2	4	7	4	9
„ 19	15	19 ·95	—	68 ·9	60 ·7	19 ·75	8	7	12	15	0	0	3	2	16	10	1	1
„ 20	16	21 ·62	—	67 ·3	61 ·0	12 ·5	8	9	8	19	1	3	1	5	8	10	2	4
„ 21	17	20 ·44	0 ·479	66 ·1	61 ·7	12 ·75	11	11	12	10	1	2	1	7	3	13	3	8
„ 22	18	22 ·18	0 ·004	66 ·9	61 ·8	6 ·5	6	10	8	13	2	5	3	9	7	11	2	10
„ 23	19	23 ·15	0 ·776	61 ·9	60 ·8	8 ·3	6	7	10	21	1	2	7	10	9	17	3	3
„ 24	20	22 ·1	—	61 ·8	59 ·6	9 ·95	6	9	10	17	4	8	3	9	3	9	3	7
„ 25	21	23 ·0	0 ·318	61 ·7	58 ·4	9 ·25	7	10	10	18	3	5	8	6	13	12	2	3
„ 26	22	22 ·74	0 ·016	58 ·9	58 ·5	22 ·062	7	9	10	11	0	5	4	7	1	8	0	5
„ 27	23	22 ·31	0 ·024	61 ·8	57 ·2	15 ·0	11	11	12	14	4	7	9	10	7	5	4	4
„ 28	24	22 ·75	0 ·069	61 ·8	57 ·7	16 ·05	13	9	11	13	8	7	1	11	4	12	8	8
„ 29	25	23 ·02	0 ·099	67 ·0	57 ·2	3 ·7*	13	14	7	22	6	9	4	11	10	10	7	8
„ 30	26	24 ·27	0 ·280	65 ·1	58 ·8	10 ·875	11	9	15	14	4	2	5	3	6	11	4	2
„ 31	27	23 ·0	0 ·002	69 ·1	58 ·7	14 ·15	9	7	12	12	3	3	4	10	7	6	3	3
Aug. 1	28	22 ·37	0 ·005	65 ·9	59 ·5	37 ·31	9	8	13	15	4	6	4	9	8	6	3	5
„ 2	29	21 ·39	0 ·350	60 ·0	59 ·0	17 ·43	8	8	11	12	1	9	5	9	5	10	4	4
„ 3	30	24 ·18	—	65 ·0	59 ·0	16 ·65	11	10	13	21	3	7	6	13	6	11	4	6
„ 4	31	20 ·14	0 ·158	62 ·2	58 ·8	10 ·0	11	10	9	16	3	8	4	5	4	9	1	6
„ 5	32	24 ·82	0 ·055	66 ·5	58 ·8	13 ·125	12	11	14	15	3	1	3	9	7	11	3	2
„ 6	33	22 ·80	—	62 ·2	58 ·5	18 ·37	10	8	7	8	3	4	2	8	17	9	3	5
„ 7	34	22 ·22	—	65 ·9	57 ·8	22 ·57*	9	10	14	13	2	0	3	4	7	1	3	1
„ 8	35	18 ·13	0 ·099	71 ·5	58 ·0	8 ·6	7	6	14	13	2	1	4	3	9	14	3	2
„ 9	36	21 ·91	0 ·024	63 ·1	59 ·7	11 ·5	4	11	8	12	1	6	3	9	3	11	1	8
„ 10	37	22 ·16	—	64 ·6	59 ·0	10 ·0	6	10	13	18	0	3	4	5	4	6	0	3
„ 11	38	21 ·04	—	67 ·8	59 ·0	20 ·0	14	12	13	14	3	6	3	6	7	9	6	6
„ 12	39	22 ·48	—	63 ·9	59 ·5	21 ·3	7	8	10	19	5	1	6	12	5	6	7	2
„ 13	40	19 ·24	—	67 ·6	58 ·7	15 ·25	12	11	4	15	1	8	5	11	6	10	3	11
„ 14	41	20 ·27	—	69 ·2	59 ·0	20 ·05*	10	11	15	18	2	3	5	8	8	10	3	6
„ 15	42	19 ·11	—	69 ·3	60 ·3	60 ·625*	13	10	14	16	5	3	7	7	8	8	10	5
„ 16	43	19 ·17	—	64 ·1	60 ·8	15 ·07	13	9	10	19	2	1	4	8	5	6	6	12
„ 17	44	20 ·54	—	62 ·2	60 ·7	—	8	6	7	18	1	1	1	4	2	10	0	11
„ 18	45	20 ·00	0 ·520	62 ·2	59 ·8	17 ·5	9	7	5	11	0	6	1	6	9	14	1	4
„ 19	46	22 ·15	0 ·009	57 ·7	58 ·1	14 ·06	7	10	10	14	1	2	3	11	3	12	2	5
„ 20	47	21 ·97	—	57 ·5	57 ·0	20 ·83	11	8	12	13	4	6	6	6	4	9	2	5
„ 21	48	21 ·16	0 ·047	57 ·8	56 ·1	13 ·3	6	3	6	11	2	1	3	10	6	3	2	1
„ 22	49	20 ·32	—	59 ·4	56 ·1	18 ·08*	8	7	12	13	3	4	5	6	9	8	4	5
„ 23	50	22 ·11	—	61 ·7	55 ·9	10 ·18	10	7	11	19	1	5	4	10	6	12	6	16

,,	25	52	19·42	—	60·9	56·9	8·58	13	10	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
,,	26	53	20·05	—	60·3	56·9	15·625	7	4	5	10	0	2	3	4	4	7	4	6
,,	27	54	18·59	—	62·8	56·0	11·35*	10	13	14	18	3	7	3	7	9	17	4	14
,,	28	55	21·35	—	67·9	57·2	8·9												
,,	29	56	20·31	—	58·9	57·5	12·9*	11	14	12	22	3	8	5	12	8	9	3	12
,,	30	57	20·23	—	56·8	56·3	9·2	7	8	6	9	3	1	2	7	3	10	2	2
,,	31	58	20·88	—	56·3	55·5	14·875	7	6	8	10	1	1	3	7	5	3	4	1
Sept.	1	59	19·63	—	59·8	56·0	19·25	6	7	15	23	1	4	1	2	7	9	3	8
,,	2	60	18·09	0·079	62·1	56·0	17·75*	11	8	5	10	1	1	6	6	6	6	7	5
,,	3	61	19·68	0·095	68·5	56·7	15·95	10	10	7	17	2	1	1	7	3	5	1	1
,,	4	62	19·0	0·020	67·8	57·9	15·75	7	8	18	23	2	4	9	11	8	15	3	10
,,	5	63	21·86	0·039	68·1	56·8	23·65	9	12	9	8	0	1	5	4	4	8	3	4
,,	6	64	18·83	—	70·0	58·0	14·55	6	7	13	21	1	4	4	7	8	8	3	7
,,	7	65	18·83	—	64·5	57·1	13·18	7	8	14	21	2	0	7	3	8	10	5	7
,,	8	66	19·56	—	66·1	57·2	11·75	10	9	8	21	1	1	4	2	3	4	0	0
,,	9	67	18·9	—	66·3	58·0	9·325	8	8	10	14	0	1	0	7	1	6	0	3
,,	10	68	18·77	—	60·8	57·8	13·375	3	6	5	18	0	0	2	2	8	7	1	2
,,	11	69	16·17	—	64·1	56·0	9·575	2	6	7	19	0	4	2	8	4	8	2	10
,,	12	70	19·13	—	71·4	55·0	9·5	5	5	4	14	0	0	3	2	4	2	1	3
,,	13	71	18·4	—	64·2	55·0	7·325	5	5	4	13	1	1	1	6	2	3	2	0
,,	14	72	15·11	0·158	64·2	56·0	5·6875	3	8	5	25	0	5	1	12	5	7	0	6
,,	15	73	18·02	0·126	60·2	57·0	6·0625	8	10	6	22	3	1	3	1	4	5	3	0
,,	16	74	19·29	0·067	60·1	56·5	48·75	2	0	13	10	0	1	1	2	2	3	0	1
,,	17	75	18·75	0·343	62·3	55·1	35·5	7	10	11	12	2	0	5	3	5	4	3	0
,,	18	76	20·31	0·229	56·8	56·1	23·5625	4	10	13	21	1	1	6	3	4	2	2	1
,,	19	77	21·57	0·001	59·8	55·0	30·5625	10	8	11	18	0	0	2	9	2	6	0	3
,,	20	78	21·39	0·015	59·1	53·8	14·2	5	5	8	23	0	2	4	10	5	6	3	4
,,	21	79	21·43	0·386	52·3	54·7	38·1	8	7	15	22	5	5	7	11	9	14	4	3
,,	22	80	22·22	0·095	57·6	53·7	19·75	7	7	20	22	4	5	6	9	14	11	2	6
,,	23	81	24·09	—	62·3	54·1	11·625	6	6	10	16	2	1	3	7	8	5	2	2
,,	24	82	22·13	—	60·3	55·0	12·375	3	2	10	17	2	2	6	7	8	7	1	2
,,	25	83	22·59	—	57·3	55·1	11·5625	5	9	13	14	1	2	4	4	5	4	2	1
,,	26	84	20·89	0·005	63·5	55·0	12·6	6	10	11	16	0	2	2	3	8	5	3	5
,,	27	85	21·00	0·004	66·0	54·9	45·45	7	5	11	16	2	2	8	13	8	6	6	5
,,	28	86	21·64	—	62·7	54·5	13·0	2	6	13	13	1	0	2	8	5	7	3	2
,,	29	87	20·31	0·002	62·9	55·4	14·7	5	4	13	19	1	5	8	9	10	5	12	10
,,	30	88	20·21	0·362	65·0	55·9	18·15	5	4	10	16	2	0	3	11	5	6	2	2
Oct.	1	89	22·43	0·376	57·1	55·8	19·1875	3	5	17	18	4	2	12	11	5	4	4	3
,,	2	90	22·5	0·009	58·8	55·1	13·9	3	4	8	18	0	0	6	6	3	8	1	4
,,	3	91	22·5	0·043	60·1	54·0	12·25	4	6	10	16	2	5	4	10	5	7	7	5
,,	4	92	22·06	0·308	62·8	54·7	21·0	4	2	8	8	6	1	6	7	7	4	3	2
,,	5	93	22·27	—	68·0	55·0	47·1	3	1	12	10	0	1	5	9	2	3	7	2
,,	6	94	22·33	0·003	67·1	56·0	31·2	6	6	10	9	0	0	0	0	1	4	1	1
,,	7	95	21·79	—	68·3	55·0	24·9375	6	5	6	10	0	1	6	7	4	4	3	1
,,	8	96	21·19	0·008	68·8	55·9	15·125	3	8	13	19	5	2	9	12	8	5	5	8
,,	9	97	22·33	—	68·5	56·2	21·1875	3	2	13	19	1	2	6	3	3	0	5	6
,,	10	98	18·3	—	58·2	56·1	14·625	3	6	13	12	0	2	3	10	3	3	1	7
,,	11	99	21·57	0·008	61·0	55·5	20·95	9	7	11	12	2	4	8	14	5	8	4	4
,,	12	100	21·57	—	61·5	54·3	21·0	3	2	11	14	1	1	4	3	2	3	4	5
,,	13	101	19·05	0·005	61·0	53·5	36·312	5	4	13	18	0	0	5	4	6	6	12	9
,,	14	102	21·48	0·236	59·4	53·0	37·3125	4	2	13	13	1	0	9	10	1	0	5	4
,,	15	103	22·02	0·083	58·9	53·5	57·9375	4	2	15	13	0	0	5	10	7	2	7	4
,,	16	104	21·35	0·079	58·9	54·0	36·3125	2	1	12	14	2	2	6	3	3	3	4	7
,,	17	105	23·17	—	54·8	54·1	21·125	2	5	16	19	2	4	5	11	2	6	7	10
,,	18	106	21·16	—	50·3	51·8	29·9375*	0	0	11	10	0	1	3	2	3	3	3	4
,,	19	107	21·48	—	51·9	50·1	16·8	0	0	4	14	1	0	2	2	1	1	5	3
,,	20	108	22·71	0·004	53·8	50·1	38·8125	3	2	15	12	2	0	4	5	3	1	3	2
,,	21	109	21·62	—	52·1	48·8	36·6	1	4	9	19	0	1	6	3	2	2	7	4
,,	22	110	20·07	0·004	53·3	48·7	53·9*	2	1	10	9	0	1	5	4	1	1	2	7
,,	23	111	20·96	0·004	53·4	49·7	41·16	2	2	7	10	0	0	8	5	0	1	4	2
,,	24	112	19·59	0·008	51·6	48·0	25·1	3	6	8	9	1	1	2	5	3	3	2	6
,,	25	113	20·95	0·004	56·9	48·0	23·5*	2	3	5	14	1	1	7	10	1	2	3	4
,,	26	114	21·64	0·008	56·0	47·4	43·3125	2	1	7	8	0	0	9	10	0	1	9	7
,,	27	115	20·29	0·008	52·2	46·5	21·2	2	5	9	14	2	2	1	3	1	2	1	2
,,	28	116	21·23	0·008	53·7	45·3	83·1	0	1	6	9	1	1	8	7	1	2	3	4
,,	29	117	21·21	0·004	50·0	45·1	14·6875*	1	9	8	12	0	0	6	1	2	2	3	6
,,	30	118	19·56	0·003	47·8	44·3	34·125	0	1	8	12	0	2	5	5	0	2	1	6
,,	31	119	21·19	0·235	51·5	43·2	36·0625	1	7	10	13	0	7	7	13	6	7	7	8

TABLE IV.—*continued.*

Date.	Sample No.	H_2O content.	Rainfall 8″ gauge.	Max. air temp. in ° F.	Min. soil temp., 12″ thermometer.	Bacteria in millions per grm.	Protozoa (numbers of negative plates). *Dimastigamœba gruberi.* Total.	*Dimastigamœba gruberi.* Cystic.	*Sp. a.* Total.	*Sp. a.* Cystic.	*Heteromita sp.* Total.	*Heteromita sp.* Cystic.	*Sp. γ.* Total.	*Sp. γ.* Cystic.	*Cercomonas sp.* Total.	*Cercomonas sp.* Cystic.	*Oicomonas termo.* Total.	*Oicomonas termo.* Cystic.
1920. Nov. 1	120	21 ·39	0 ·004	45 ·8	45 ·1	12 ·85	1	4	6	9	1	0	1	1	1	3	1	2
,, 2	121	21 ·35	—	42 2	44 ·3	31 ·5*	2	3	5	14	1	1	10	10	1	2	7	5
,, 3	122	22 ·22	—	42 ·2	44 ·5	52 ·5	2	4	9	15	1	0	10	9	4	8	9	10
,, 4	123	20 ·50	0 ·004	46 ·7	44 ·0	53 ·75	2	4	5	12	1	4	6	8	3	3	3	5
,, 5	124	20 ·75	0 ·008	43 ·8	42 ·9	43 ·9	2	6	12	15	2	4	3	10	4	4	6	7
,, 6	125	21 ·39	—	48 ·3	42 ·5	40 ·45*	3	2	8	10	3	1	6	16	4	2	4	3
,, 7	126	21 ·52	0 ·004	50 ·9	42 ·5	59 ·41	4	1	6	3	1	3	3	2	3	5	6	5
,, 8	127	21 ·00	—	49 ·8	43 ·9	59 ·375	4	1	10	12	1	0	0	2	6	6	0	2
,, 9	128	21 ·06	—	52 ·6	45 ·1	18 ·4375	3	2	8	14	0	2	2	11	4	2	9	5
,, 10	129	20 ·73	0 ·004	53 ·9	46 ·0	52 ·775	7	3	16	11	1	0	4	6	3	3	2	8
,, 11	130	20 ·59	0 ·005	50 ·3	46 ·4	67 ·1875*	1	1	8	12	1	1	9	4	2	1	8	5
,, 12	131	20 ·80	0 ·035	51 ·7	44 ·2	49 ·875	0	0	8	4	0	0	4	2	1	2	4	2
,, 13	132	20 ·36	0 ·080	55 ·1	45 ·0	30 ·375	3	2	4	10	4	1	9	7	2	2	10	2
,, 14	133	21 ·28	0 ·375	56 ·4	44 ·7	25 ·9375*	2	5	12	12	1	3	5	10	3	4	7	4
,, 15	134	21 ·48	0 ·087	56 ·5	47 ·1	23 ·625	3	3	8	11	3	2	3	3	6	8	4	1
,, 16	135	21 ·39	0 ·002	48 ·9	46 ·5	41 ·625*	1	1	9	14	4	1	11	6	6	2	6	4
,, 17	136	21 ·70	0 ·004	49 ·1	44 ·7	28 ·8125*	3	4	10	12	1	5	7	7	1	3	5	5
,, 18	137	21 ·75	0 ·004	47 ·5	42 ·7	25 ·85	4	1	2	11	3	1	15	12	4	2	9	5
,, 19	138	20 ·73	0 ·007	48 ·1	43 ·0	25 ·875	3	3	8	12	1	1	3	2	1	0	0	1
,, 20	139	22 ·25	0 ·007	38 ·5	43 ·8	58 ·0*	1	0	7	8	0	0	7	7	2	0	8	2
,, 21	140	22 ·59	0 ·007	40 ·0	41 ·9	25 ·6	0	6	8	17	0	2	1	8	3	3	1	4
,, 22	141	21 ·66	0 ·010	42 ·9	40 ·0	53 ·666	5	3	17	16	2	1	12	9	7	4	6	6
,, 23	142	20 ·89	0 ·004	45 ·1	39 ·0	56 ·5*	2	2	6	7	2	1	7	9	5	3	9	7
,, 24	143	21 ·26	—	46 ·1	38 ·7	60 ·25*	2	2	9	14	1	0	8	6	1	4	1	8
,, 25	144	21 ·50	0 ·004	51 ·7	39 ·7	49 ·7	2	2	5	13	1	0	10	9	2	0	3	4
,, 26	145	22 ·64	0 ·005	48 ·3	41 ·1	30 ·4*	2	3	13	17	1	0	9	6	2	2	5	10
,, 27	146	23 ·3	0 ·673	49 ·3	41 ·6	31 ·5625*	1	0	3	10	2	0	13	13	0	1	4	5
,, 28	147	21 ·95	0 ·008	47 ·2	43 ·4	44 ·825*	4	7	15	10	0	3	1	4	1	3	5	3
,, 29	148	22 ·04	0 ·166	47 ·6	43 ·7	52 ·65	4	4	15	11	4	1	9	1	2	3	0	4
,, 30	149	23 ·37	0 ·209	49 ·4	44 ·0	21 ·0625	1	2	9	14	2	1	14	13	3	2	12	9
Dec. 1	150	23 ·42	0 ·331	47 ·2	43 ·4	30 ·6	0	2	9	14	0	2	3	14	3	3	4	4
,, 2	151	22 ·62	0 ·150	53 ·2	42 ·8	45 ·375	5	8	13	16	0	1	8	16	8	8	2	5
,, 3	152	23 ·40	0 ·016	54 ·2	43 ·1	49 ·68	8	3	10	12	4	2	13	8	7	2	6	3
,, 4	153	22 ·66	0 ·047	44 ·8	42 ·8	12 ·35*	0	0	1	8	2	1	11	16	2	2	7	6
,, 5	154	21 ·80	0 ·130	43 ·7	42 ·9	44 ·43*	6	4	14	13	0	3	4	11	3	3	0	5
,, 6	155	22 ·24	0 ·024	39 ·8	42 ·2	8 ·6	2	1	7	2	2	1	7	4	0	1	5	3
,, 7	156	22 ·62	0 ·016	39 ·9	41 ·7	54 ·18	1	0	2	4	4	0	5	5	4	4	5	4
,, 8	157	23 ·22	0 ·006	36 ·1	41 ·5	49 ·6	3	1	8	7	1	3	6	2	8	5	10	7
,, 9	158	21 ·77	0 ·008	38 ·2	40 ·1	41 ·45*	3	1	5	7	1	2	5	13	3	5	5	7
,, 10	159	22 ·24	0 ·016	37 ·3	39 ·0	25 ·0*	3	3	8	8	2	0	10	5	5	2	7	3
,, 11	160	22 ·15	0 ·028	34 ·4	38 ·9	17 ·5	2	2	0	9	4	1	8	12	4	0	4	4
,, 12	161	23 ·42	—	28 ·2	38 ·3	46 ·375	4	2	12	7	1	4	1	4	5	7	5	5
,, 13	162	22 ·29	0 ·012	31 ·2	37 ·3	14 ·15	4	1	5	12	7	0	10	8	6	3	5	2
,, 14	163	22 ·82	0 ·024	34 ·0	36 ·6	44 ·69*	4	1	5	5	0	2	9	10	4	3	6	2
,, 15	164	23 ·07	0 ·016	30 ·0	36 ·3	18 ·1875	8	8	15	11	3	1	9	8	2	4	9	8
,, 16	165	23 ·44	0 ·002	33 ·5	36 ·0	25 ·5*	4	0	14	17	2	4	4	14	5	3	8	4
,, 17	166	21 ·93	0 ·030	35 ·3	35 ·9	24 ·625	2	2	5	10	1	2	12	12	1	3	4	3
,, 18	167	22 ·64	0 ·039	37 ·3	36 ·0	55 ·25*	2	0	14	12	2	4	6	9	0	3	6	7
,, 19	168	23 ·21	0 ·002	36 ·1	37 ·0	46 ·00*	2	2	10	16	0	0	8	5	4	1	2	2
,, 20	169	22 ·93	0 ·012	43 ·3	37 ·3	21 ·125	3	3	3	5	2	1	10	6	6	6	7	3
,, 21	170	22 ·65	0 ·087	45 ·2	38 ·5	31 ·35*	3	2	10	9	2	3	4	13	6	4	6	5
,, 22	171	23 ·17	0 ·099	43 ·6	38 ·7	17 ·25	2	1	7	16	2	0	15	12	6	1	6	3
,, 23	172	23 ·77	0 ·422	48 ·5	38 ·4	29 ·3125	3	0	17	12	1	1	3	6	4	1	0	3
,, 24	173	23 ·85	0 ·052	53 ·1	38 ·2	40 ·575	4	0	16	13	3	0	8	8	5	1	5	5

,, 26	175	23·37	0·032	53·0	41·3	32·2	3	0	16	22	1	1	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
,, 27	176	22·22	0·134	51·5	41·1	40·5	4	2	16	18	0	2	0	10	3	3	2	1
,, 28	177	22·59	0·004	52·0	42·6	45·25	0	0	5	14	2	1	16	15	1	1	7	5
,, 29	178	22·44	0·259	52·3	42·8	27·69	2	1	14	18	2	1	16	17	6	1	10	2
,, 30	179	22·04	0·256	50·9	44·0	40·19	1	1	9	10	3	0	6	2	1	0	5	1
,, 31	180	24·02	0·099	53·9	43·8	52·30*	3	0	5	15	1	1	9	5	2	1	6	4
1921.																		
Jan. 1	181	22·25	0·256	52·8	44·2	40·00	2	1	9	12	1	0	10	14	1	2	5	1
,, 2	182	23·39	0·063	51·9	45·1	35·31	2	1	6	13	2	1	8	5	2	1	4	2
,, 3	183	23·70	0·002	52·1	45·0	30·05	3	3	10	11	2	3	9	11	2	1	6	4
,, 4	184	22·53	0·292	52·6	45·0	46·12	4	0	12	10	2	1	15	13	4	1	10	8
,, 5	185	23·12	—	45·8	45·9	36·19	2	1	16	19	0	0	10	16	1	1	7	3
,, 6	186	22·38	0·083	47·5	44·0	38·95	3	3	14	18	0	2	13	15	2	6	4	5
,, 7	187	22·68	0·488	42·9	44·1	25·13	2	5	15	20	1	0	1	11	1	7	10	9
,, 8	188	23·44	0·028	52·1	42·6	22·81	2	4	9	16	2	1	6	17	4	3	3	3
,, 9	189	23·24	—	54·2	43·0	33·25	7	3	5	19	0	2	3	18	3	1	6	10
,, 10	190	23·58	0·209	53·0	44·9	14·70	2	4	15	10	0	0	3	12	1	1	3	2
,, 11	191	22·36	0·087	45·7	44·0	15·94	3	4	11	17	3	1	14	17	6	5	7	3
,, 12	192	23·73	0·182	46·3	43·0	14·38	2	2	5	12	2	2	10	16	1	3	4	5
,, 13	193	23·77	0·020	42·3	42·2	29·70	2	0	17	13	1	1	12	23	2	1	5	6
,, 14	194	23·19	—	38·1	41·1	23·56	1	5	18	19	0	0	17	17	4	6	2	5
,, 15	195	22·33	0·002	40·8	39·2	22·88	0	2	13	18	2	0	13	14	4	1	7	3
,, 16	196	21·61	0·138	47·5	38·1	16·00	0	2	10	9	2	1	17	15	3	1	3	2
,, 17	197	23·48	0·059	51·8	39·1	22·25	1	2	15	17	1	0	4	12	1	2	5	4
,, 18	198	23·09	0·170	51·9	41·8	22·38	1	4	14	15	1	4	18	24	1	3	8	4
,, 19	199	22·82	—	48·2	40·1	20·95	5	0	16	11	0	0	7	13	6	2	5	6
,, 20	200	21·68	—	52·2	40·1	32·13	4	1	17	13	0	1	3	20	3	1	6	2
,, 21	201	22·57	—	49·2	41·6	24·40	3	2	7	13	1	1	7	15	1	4	3	5
,, 22	202	23·02	—	49·3	42·3	38·05	5	6	14	9	2	1	15	2	4	1	5	2
,, 23	203	22·41	—	50·7	41·3	12·69	1	2	7	14	0	0	8	15	0	3	0	3
,, 24	204	22·36	0·005	49·8	42·1	11·60	1	3	16	14	0	2	18	16	4	5	7	5
,, 25	205	22·47	0·012	50·8	43·0	22·15	5	2	15	12	3	1	18	16	6	2	8	6
,, 26	206	22·86	0·012	48·9	43·5	15·50	0	1	19	13	0	1	11	16	4	1	4	7
,, 27	207	23·14	0·024	49·7	43·1	41·60	3	1	4	10	1	0	17	20	1	1	7	3
,, 28	208	22·93	—	52·4	42·9	22·06	1	1	2	12	0	1	9	9	2	0	2	2
,, 29	209	22·18	0·035	49·9	44·0	23·13	2	2	13	12	2	1	4	11	5	2	5	4
,, 30	210	21·30	0·114	49·5	44·8	24·81	3	5	14	14	2	3	8	16	3	7	4	6
,, 31	211	22·13	0·008	43·8	44·2	36·10	6	8	13	12	3	1	12	9	4	4	5	3
Feb. 1	212	21·35	0·004	44·8	42·0	30·05	5	0	12	11	1	0	13	18	4	3	11	8
,, 2	213	22·71	0·007	43·8	40·7	30·85	4	1	16	19	1	0	19	15	3	1	9	5
,, 3	214	21·80	0·002	41·2	39·8	32·75	1	4	11	12	1	1	15	20	1	6	7	5
,, 4	215	22·18	—	40·8	38·5	21·45	3	5	16	15	3	7	14	18	2	3	1	5
,, 5	216	22·55	—	40·1	38·7	24·10	4	5	20	18	3	3	12	21	2	4	1	1
,, 6	217	22·49	—	35·9	39·0	27·62	2	2	19	17	2	2	17	17	4	4	10	4
,, 7	218	22·53	—	34·8	38·2	21·95	5	3	7	16	7	3	8	3	5	3	4	1
,, 8	219	21·70	—	37·8	38·1	40·50	5	3	11	15	0	0	16	20	4	3	10	5
,, 9	220	22·53	0·012	42·2	38·2	16·00	2	3	15	19	1	1	16	21	4	5	8	5
,, 10	221	22·57	0·012	43·2	38·1	27·25	5	2	12	12	4	3	15	19	4	1	6	2
,, 11	222	21·14	0·020	41·3	38·2	36·60	3	2	14	15	2	5	14	18	0	6	7	7
,, 12	223	21·02	—	42·5	39·0	18·80	1	3	17	20	7	1	19	22	8	1	11	7
,, 13	224	21·50	—	44·7	39·2	21·20	3	4	8	14	4	4	15	20	5	6	7	8
,, 14	225	21·73	—	47·9	40·0	12·65	2	6	11	12	2	2	11	9	2	4	5	2
,, 15	226	20·95	—	47·1	39·5	27·40	6	2	6	6	1	1	11	19	4	5	2	4
,, 16	227	21·30	—	51·2	40·1	16·88	3	1	10	14	2	3	18	21	4	2	3	6
,, 17	228	19·53	—	49·8	41·7	45·38	4	1	17	12	5	5	14	7	3	7	1	5
,, 18	229	21·02	—	44·9	41.0	26·55	5	2	13	16	5	3	15	14	4	7	7	9
,, 19	230	21·59	—	39·6	41·0	15·90	3	8	8	18	2	7	12	21	4	10	5	11
,, 20	231	20·18	0·008	48·1	40·5	25·40	4	3	15	15	0	0	8	1	5	1	1	0
,, 21	232	19·03	0·004	50·7	39·8	12·88	1	4	10	5	1	2	14	16	3	3	3	1
,, 22	233	20·02	0·008	54·2	39·0	20·60	6	1	19	14	7	4	14	16	7	5	3	5
,, 23	234	21·02	—	53·1	39·9	12·38	5	8	11	17	4	2	19	19	8	7	5	7
,, 24	235	21·00	0·016	58·1	40·0	23·05	2	5	3	16	4	4	9	17	3	5	4	8
,, 25	236	21·61	0·091	48·3	40·5	15·55	6	7	10	17	3	6	15	13	4	8	7	12
,, 26	237	20·91	0·004	45·4	40·3	35·50	5	5	15	19	4	5	15	18	4	3	5	5
,, 27	238	19·13	—	48·0	39·1	15·95	3	5	6	8	3	3	11	15	5	3	5	2
,, 28	239	21·00	—	45·9	40·0	15·35	2	2	16	16	7	0	15	17	3	1	5	8
Mar. 1	240	20·09	0·092	50·0	40·1	20·56	4	6	6	11	5	2	10	8	5	7	8	4
,, 2	241	21·23	—	46·5	40·2	17·19	7	6	20	25	6	7	13	22	7	7	7	9

TABLE IV.—*continued.*

Date.	Sample No.	H_2O content.	Rainfall 8″ gauge.	Max. air temp. in °F.	Min. soil temp., 12″ thermometer.	Bacteria in millions per grm.	Protozoa (numbers of negative plates). *Dimastigamœba gruberi.*		*Sp. a.*		*Heteromita sp.*		*Sp. γ.*		*Cercomonas sp.*		*Oicomonas termo.*	
							Total.	Cystic.	Total.	Cystic.	Total.	Cystic.	Total.	Cystic.	Total.	Cystic.	Total.	Cystic.
1921. Mar. 3	242	20·68	0·063	48·9	39·3	14·05	2	6	18	14	5	3	14	9	6	4	8	3
,, 4	243	20·95	—	51·1	40·8	26·30	4	9	17	13	1	2	11	8	8	4	8	6
,, 5	244	20·70	0·002	49·1	42·0	8·88	2	4	7	19	2	1	9	20	2	8	6	3
,, 6	245	20·98	0·158	50·7	41·9	15·75	5	8	17	19	3	8	21	22	6	10	12	6
,, 7	246	20·84	—	41·9	41·1	58·00	9	5	14	17	1	3	4	25	0	6	1	5
,, 8	247	20·44	0·012	45·0	39·9	14·25	3	1	17	22	3	0	11	15	5	4	3	4
,, 9	248	19·37	—	51·0	40·3	26·25	6	2	9	5	2	3	13	18	6	2	8	4
,, 10	249	20·79	—	55·2	41·0	16·81	9	6	16	14	8	3	15	18	8	5	8	7
,, 11	250	20·48	—	55·1	42·0	36·06	8	4	12	8	3	2	12	8	5	6	6	3
,, 12	251	20·38	0·035	50·2	43·0	14·25	0	5	12	11	3	2	16	12	4	4	3	3
,, 13	252	17·51	0·078	54·8	43·8	30·75	2	1	9	13	3	1	11	9	1	4	6	3
,, 14	253	20·40	0·002	53·2	44·0	16·35	1	1	15	16	6	2	15	16	9	6	9	8
,, 15	254	19·85	—	53·0	43·3	22·85	4	3	11	12	4	0	14	14	9	4	6	3
,, 16	255	20·20	0·051	53·9	44·7	37·80	3	2	12	15	3	0	12	15	4	5	7	7
,, 17	256	19·79	0·162	54·8	45·0	40·19	7	5	5	12	2	0	10	11	7	4	5	2
,, 18	257	21·02	—	49·6	45·8	23·10	5	6	10	13	2	3	8	3	0	3	1	1
,, 19	258	20·51	0·122	50·8	44·0	16·06	2	3	8	5	1	2	10	5	5	5	5	2
,, 20	259	19·39	0·028	47·2	44·0	30·55	8	7	4	10	2	3	6	6	3	4	1	2
,, 21	260	20·23	—	54·1	42·8	19·60	3	7	10	7	1	1	3	13	3	1	2	3
,, 22	261	20·48	—	56·8	44·7	39·19	4	1	11	12	2	1	3	15	5	3	2	6
,, 23	262	20·51	—	55·1	45·8	36·69	5	3	11	15	1	0	10	11	3	3	6	4
,, 24	263	18·63	0·002	63·0	45·5	18·31	2	5	3	10	2	3	7	13	8	9	2	7
,, 25	264	19·61	0·002	58·3	45·0	18·70	3	5	7	11	3	2	8	16	6	6	5	3
,, 26	265	17·90	0·001	47·8	45·0	15·25	9	9	12	12	3	7	13	11	9	9	4	6
,, 27	266	18·19	0·004	49·2	43·7	18·5	9	5	16	14	2	1	13	12	2	4	4	2
,, 28	267	17·01	0·099	51·0	43·7	13·75	2	3	3	12	1	3	6	4	3	6	2	2
,, 29	268	18·04	0·051	50·0	44·0	14·5	2	6	13	13	4	2	8	14	4	9	6	5
,, 30	269	17·85	—	51·1	43·4	15·625	4	3	4	9	1	2	14	15	2	4	2	5
,, 31	270	17·19	—	57·8	43·9	20·15	3	3	2	9	0	0	6	12	4	2	3	1
Apr. 1	271	20·03	0·004	61·1	45·0	28·2	2	1	13	9	3	0	13	9	6	6	11	4
,, 2	272	18·61	—	64·4	45·7	18·1875	2	3	13	17	1	1	11	13	5	2	4	7
,, 3	273	19·35	—	64·0	47·0	21·125	4	9	16	17	2	0	14	23	2	11	8	6
,, 4	274	17·17	—	51·1	47·2	17·10	6	7	1	13	0	1	0	11	4	3	3	4
,, 5	275	19·40	—	52·7	45·8	17·7	2	5	13	12	2	1	14	12	2	4	7	2
,, 6	276	20·46	—	55·1	47·0	37·625	3	2	15	18	0	0	12	15	1	7	7	7
,, 7	277	19·52	—	53·2	46·4	34·2	4	3	10	18	1	0	8	13	2	4	6	4
,, 8	278	18·66	0·107	54·9	45·2	45·825	1	3	16	15	0	2	14	18	3	4	5	5
,, 9	279	16·80	0·055	51·5	45·2	20·4	0	1	14	21	1	2	10	16	4	5	6	8
,, 10	280	14·06	—	63·3	45·1	13·6875	3	3	14	15	0	0	9	11	4	2	2	2
,, 11	281	18·08	—	49·9	47·0	28·1	2	4	9	17	0	6	14	14	5	12	11	8
,, 12	282	18·51	—	64·9	46·9	30·35	6	2	13	15	2	1	14	17	7	2	9	5
,, 13	283	18·95	0·472	65·5	47·7	21·6	1	5	16	13	0	2	16	13	5	4	12	7
,, 14	284	17·88	—	47·2	48·5	74·0*	4	3	15	13	1	0	16	19	4	3	5	3
,, 15	285	16·8	0·067	40·0	45·7	61·08	6	1	11	18	2	1	11	13	5	3	5	9
,, 16	286	17·96	0·248	42·9	44·1	69·1	2	0	14	21	0	0	14	18	4	4	5	3
,, 17	287	19·85	0·178	41·9	43·7	39·6875	1	6	18	22	0	12	18	23	2	9	10	5
,, 18	288	20·48	0·016	47·1	42·7	12·55	3	3	18	14	7	0	15	12	4	4	6	8
,, 19	289	20·03	0·002	49·9	43·0	13·875	1	3	16	15	2	0	13	17	2	5	9	6
,, 20	290	19·09	—	50·8	43·3	29·4	4	5	15	13	1	3	13	15	3	6	6	6
,, 21	291	18·34	—	54·7	44·2	28·15	5	4	11	15	1	1	13	21	5	7	7	6
,, 22	292	18·66	0·166	54·7	46·2	20·95	1	1	14	20	2	0	12	20	5	2	4	3
,, 23	293	20·88	0·103	54·4	46·9	34·8125	3	2	15	17	0	0	14	19	2	1	6	3
,, 24	294	19·31	0·055	48·9	46·0	24·75	3	4	9	12	0	0	12	11	1	6	8	6
,, 25	295	19·4	—	53·8	45·8	20·55	5	4	12	21	2	8	17	26	4	11	8	5
,, 26	296	19·95	—	52·7	46·7	22·55	4	1	10	9	6	0	18	14	8	2	8	8

,,	28	298	19 ·15	—	70 ·2	48 ·7	31 ·35	[illegible]	[illegible]	10	14	1	0	4	18	1	2	4	1
,,	29	299	18 ·64	—	65 ·0	49 ·5	22 ·65	2	5	14	16	0	1	9	20	2	7	7	7
,,	30	300	18 ·11	—	65 ·9	49 ·8	26 ·9	3	1	14	13	0	1	15	16	4	3	5	6
May	1	301	17 ·79	—	60 ·0	50 ·1	21 ·0625	2	1	6	4	0	0	10	6	2	1	4	1
,,	2	302	17 ·85	0 ·099	63 ·0	50 ·0	24 ·5625	2	7	3	0	4	1	5	7	4	1	3	1
,,	3	303	16 ·65	0 ·071	45 ·8	50 ·1	32 ·3	5	5	11	15	1	1	16	12	5	8	9	6
,,	4	304	16 ·89	0 ·039	45 ·4	48 ·0	26 ·6	4	3	12	15	3	0	15	14	4	5	4	5
,,	5	305	16 ·36	—	53 ·1	46 ·8	25 ·5	1	6	13	21	0	0	16	24	3	5	8	7
,,	6	306	18 ·88	0 ·028	56 ·1	48 ·2	29 ·95	5	4	18	22	0	0	14	19	3	3	3	7
,,	7	307	18 ·02	0 ·071	61 ·6	49 ·5	17 ·35	1	2	12	6	1	1	9	3	3	4	5	1
,,	8	308	17 ·08	0 ·268	57 ·0	50 ·8	28 ·4	5	4	14	12	0	0	11	9	4	5	6	5
,,	9	309	18 ·83	0 ·213	57 ·9	50 ·9	34 ·4*	1	0	15	9	4	7	12	9	3	7	8	3
,,	10	310	18 ·94	0 ·020	60 ·6	51 ·0	37 ·7	5	2	15	12	2	2	15	13	5	5	8	6
,,	11	311	18 ·19	0 ·002	64 ·5	51 ·1	32 ·6	3	3	15	13	1	1	14	12	3	4	6	2
,,	12	312	18 ·80	0 ·186	67 ·9	52 ·3	31 ·95	4	3	16	19	0	4	12	21	4	4	5	7
,,	13	313	19 ·11	—	69 ·9	53 ·5	22 ·85	1	3	13	17	1	2	18	23	6	4	8	10
,,	14	314	18 ·42	0 ·153	69 ·2	55 ·0	24 ·9	1	2	14	14	2	1	12	16	0	5	2	5
,,	15	315	17 ·46	0 ·020	57 ·9	56 ·0	21 ·5625	1	4	8	14	0	1	13	12	1	4	2	3
,,	16	316	17 ·31	—	63 ·0	53 ·8	66 ·4875	4	1	12	8	9	1	12	5	7	4	7	1
,,	17	317	15 ·95	—	59 ·5	54 ·1	49 ·125	2	2	14	17	2	1	15	16	5	4	6	6
,,	18	318	15 ·30	—	62 ·2	54 ·0	23 ·9	2	3	16	23	0	0	20	16	3	3	9	8
,,	19	319	15 ·82	0 ·005	66 ·0	55 ·1	12 ·2	1	7	17	21	1	0	14	23	3	4	4	5
,,	20	320	15 ·76	—	61 ·6	56 ·3	38 ·65	4	4	15	19	3	0	14	18	5	3	8	7
,,	21	321	14 ·47	—	66 ·8	55 ·9	25 ·66	5	4	11	6	1	1	16	9	5	2	7	4
,,	22	322	12 ·72	—	69 ·8	56 ·2	22 ·5	5	5	16	14	0	4	15	22	2	7	10	6
,,	23	323	14 ·61	—	68 ·7	56 ·4	43 ·6875	7	3	15	8	1	2	12	9	3	2	3	2
,,	24	324	16 ·97	—	72 ·8	57 ·0	17 ·6	3	3	12	8	3	0	16	11	8	3	8	3
,,	25	325	17 ·29	0 ·020	72 ·1	57 ·9	36 ·3125	4	3	6	7	1	2	6	7	4	2	1	2
,,	26	326	17 ·72	—	70 ·6	59 ·0	54 ·15	6	4	17	13	3	2	10	19	9	4	8	8
,,	27	327	16 ·36	0 ·004	58 ·1	58 ·7	45 ·875	5	6	9	14	2	3	12	19	1	3	0	5
,,	28	328	16 ·84	0 ·008	55 ·7	57 ·0	45 ·8125	3	2	13	17	0	1	11	14	3	2	4	6
,,	29	329	14 ·89	0 ·138	57 ·7	56 ·2	36 ·5	3	4	14	14	3	1	16	15	2	5	7	6
,,	30	330	17 ·55	0 ·028	64 ·2	56 ·4	17 ·5	2	6	3	12	2	4	7	9	2	4	2	5
,,	31	331	16 ·1	0 ·004	63 ·0	57 ·0	48 ·65	4	2	11	17	1	1	16	19	1	1	4	6
June	1	332	16 ·42	—	64 ·3	57 ·7	45 ·55	5	4	11	17	1	3	11	19	3	5	2	7
,,	2	333	16 ·23	—	68 ·3	57 ·5	51 ·35	6	6	8	18	2	1	20	18	0	5	3	6
,,	3	334	16 ·25	0 ·083	66 ·8	58 ·7	33 ·15	5	4	17	12	2	0	14	16	7	2	4	1
,,	4	335	15 ·0	—	56 ·7	57 ·6	49 ·3125	4	1	15	9	2	0	15	11	2	1	8	2
,,	5	336	15 ·69	—	63 ·2	56 ·4	14 ·85	1	9	13	13	1	4	6	7	1	5	0	3
,,	6	337	15 ·71	—	70 ·6	57 ·0	11 ·95	7	3	16	16	5	1	17	12	7	0	5	2
,,	7	338	15 ·57	—	72 ·8	58 ·0	75 ·55*	6	3	12	13	3	0	14	10	3	2	6	4
,,	8	339	16 ·57	—	69 ·1	58 ·7	14 ·5	2	3	15	17	2	3	13	20	3	3	5	5
,,	9	340	15 ·35	—	63 ·7	58 ·2	64 ·375*	5	2	8	13	1	0	7	15	2	0	1	5
,,	10	341	15 ·11	—	64 ·8	58 ·0	26 ·15	2	6	10	16	1	1	15	18	1	2	8	3
,,	11	342	14 ·83	—	64 ·9	58 ·0	34 ·85	4	2	16	16	2	1	12	14	2	3	10	6
,,	12	343	13 ·34	—	63 ·3	57 ·7	25 ·65	4	7	16	17	0	3	13	18	2	9	8	7
,,	13	344	12 ·60	—	63 ·3	58 ·5	32 ·75	6	3	14	16	2	1	14	16	6	1	6	6
,,	14	345	14 ·22	—	66 ·8	57 ·8	14 ·9375	3	3	9	16	1	0	18	14	2	4	7	3
,,	15	346	14 ·51	—	76 ·0	58 ·8	27 ·4	7	4	14	17	4	2	13	22	8	4	7	4
,,	16	347	12 ·46	—	73 ·2	60 ·2	24 ·85	5	8	10	16	5	1	14	15	5	3	9	5
,,	17	348	11 ·44	—	79 ·5	61 ·0	44 ·875	7	3	15	11	2	0	16	12	5	3	2	1
,,	18	349	13 ·30	—	61 ·0	61 ·1	39 ·7	3	2	12	12	1	0	13	7	5	2	7	2
,,	19	350	10 ·4	0 ·079	57 ·9	58 ·7	34 ·75	2	7	16	23	0	3	7	20	3	4	8	3
,,	20	351	13 ·7	0 ·004	59 ·8	58 ·0	30 ·625	6	4	15	13	0	1	17	14	5	6	6	3
,,	21	352	13 ·92	—	60 ·9	57 ·3	37 ·9375	8	4	17	24	1	1	19	19	1	3	10	6
,,	22	353	13 ·54	—	64 ·7	58 ·2	41 ·8	7	4	14	20	0	2	12	23	2	4	4	6
,,	23	354	12 ·84	—	74 ·7	58 ·7	42 ·375	6	3	13	15	0	1	13	16	4	2	4	4
,,	24	355	12 ·44	—	81 ·5	60 ·2	56 ·4*	7	4	13	13	1	0	17	17	3	0	4	5
,,	25	356	12 ·84	0 ·001	82 ·8	63 ·0	34 ·375	4	2	7	22	0	0	13	24	1	0	1	9
,,	26	357	11 ·41	0 ·006	71 ·1	63 ·7	65 ·0	10	8	11	6	1	4	9	8	2	6	5	5
,,	27	358	12 ·28	—	62 ·0	61 ·7	17 ·375	7	7	13	16	2	0	17	19	6	9	2	5
,,	28	359	11 ·72	—	68 ·8	59 ·8	31 ·7	10	9	13	17	0	6	14	21	5	11	12	10
,,	29	360	11 ·55	—	67 ·2	61 ·0	35 ·4	8	4	7	6	1	0	9	6	4	1	2	1
,,	30	361	10 ·94	—	62 ·7	60 ·3	62 ·1875	3	0	13	21	0	0	14	19	0	0	5	2
July	1	362	10 ·72	—	68 ·0	60 ·8	60 ·3125	7	2	16	14	0	1	15	15	3	1	6	5
,,	2	363	11 ·26	—	72 ·3	60 ·1	54 ·875	5	1	15	13	0	0	18	19	2	2	9	5
,,	3	364	9 ·54	—	71 ·5	61 ·3	57 ·0	2	3	14	18	0	2	16	17	2	8	5	10
,,	4	365	10 ·26	—	61 ·0	60 ·2	17 ·45	6	9	15	16	4	4	15	18	6	6	9	7

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