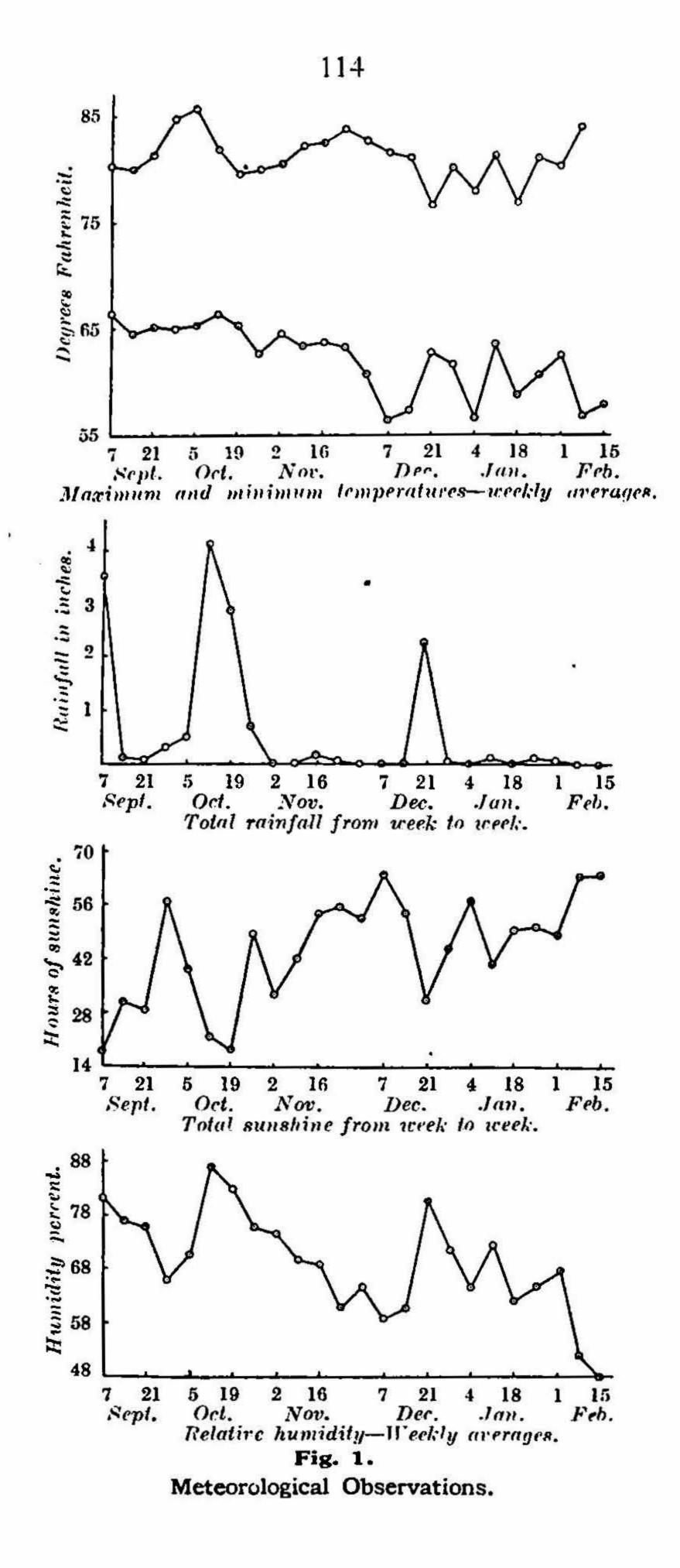
# ON THE NATURE AND EXTENT OF PERIODIC FLUCTUATIONS IN CERTAIN SOIL CONSTITUENTS.

By A. Sreenizasan and V. Subrahmanyan.

During recent years, several investigators have drawn attention to periodic fluctuations in different soil constituents such as organic matter, total nitrogen, nitrates and microbial numbers. Russell (J. Agric. Sci., 1914, 6, 18) observed that the nitrate content of arable land fluctuates regularly, the changes being more marked in loamy soils than in clays or sands. Prescott and Piper (J. Agric. Sci., 1930, 20, 517) noted that such variations depend on season, temperature, rainfall, moisture content of soil and other factors. Waksman (Soil Sci., 1916, 1, 363) found that bacterial numbers varied from time to time, in different seasons and at different depths of soil. Russell and Appleyard (J. Agric. Sci., 1917, 8, 385) observed similar fluctuations in English soils. Cutler and his associates (Ann. Appl. Biol., 1920, 7, 11; Phil. Trans., 1922, B 211, 317) recorded striking variations in the daily counts of bacteria and certain species of flagellates. Smith and Worden (J. Agric. Res., 1925, 31, 501) observed similar changes in American soil. Further evidence in support of daily and even hourly changes in bacterial numbers and nitrate contents was adduced by Thornton and his associates (Soil Sci., 1927, 23, 253; Proc. Roy. Soc., 1930, B 106, 399). Annet, Iyer and Kayasth (Mem. Dept. Agric. India, Chem. Ser., 1928, 9, 155) studied the economy of nitrogen in the black cotton soil during different seasons and observed that although the annual loss in the form of nitrate amounted to about 160 lbs. per acre, the total nitrogen still remained unaffected and continued to be practically the same over a period of 25 years. They concluded, therefore, that there is fixation of nitrogen to compensate for the above loss. The same authors also noted that the total nitrogen contents of dry soils vary from month to month. Sahasrabuddhe and his co-workers (Mem. Dept. Agric. India, Chem. Ser., 1927, 8, 53; Ind. J. Agric. Sci., 1931, 1, 631; ibid., 1932, 2, 455) observed that recuperation of nitrogen in the soils of the Bombay Deccan proceeds continuously and is facilitated by a number of factors, such as wetting followed by dry weather, better cultivation or addition of organic manures. They hold the view that the nitrogen content of soil is not a stable or constant quantity; there is a range for every soil which is determined by various factors such as moisture, temperature and aeration.

Although the foregoing observations are of considerable practical interest, yet very little is known regarding the chemical and biological



mechanism of the related processes. Some of the changes, particularly those reported to occur at short intervals, are almost phenomenal, being more rapid than those that could be normally brought about by any combination of chemical and biological agencies known to be associated with the soil. Further knowledge of the nature and extent of the different changes is also needed if they are to be adequately controlled in field practice.

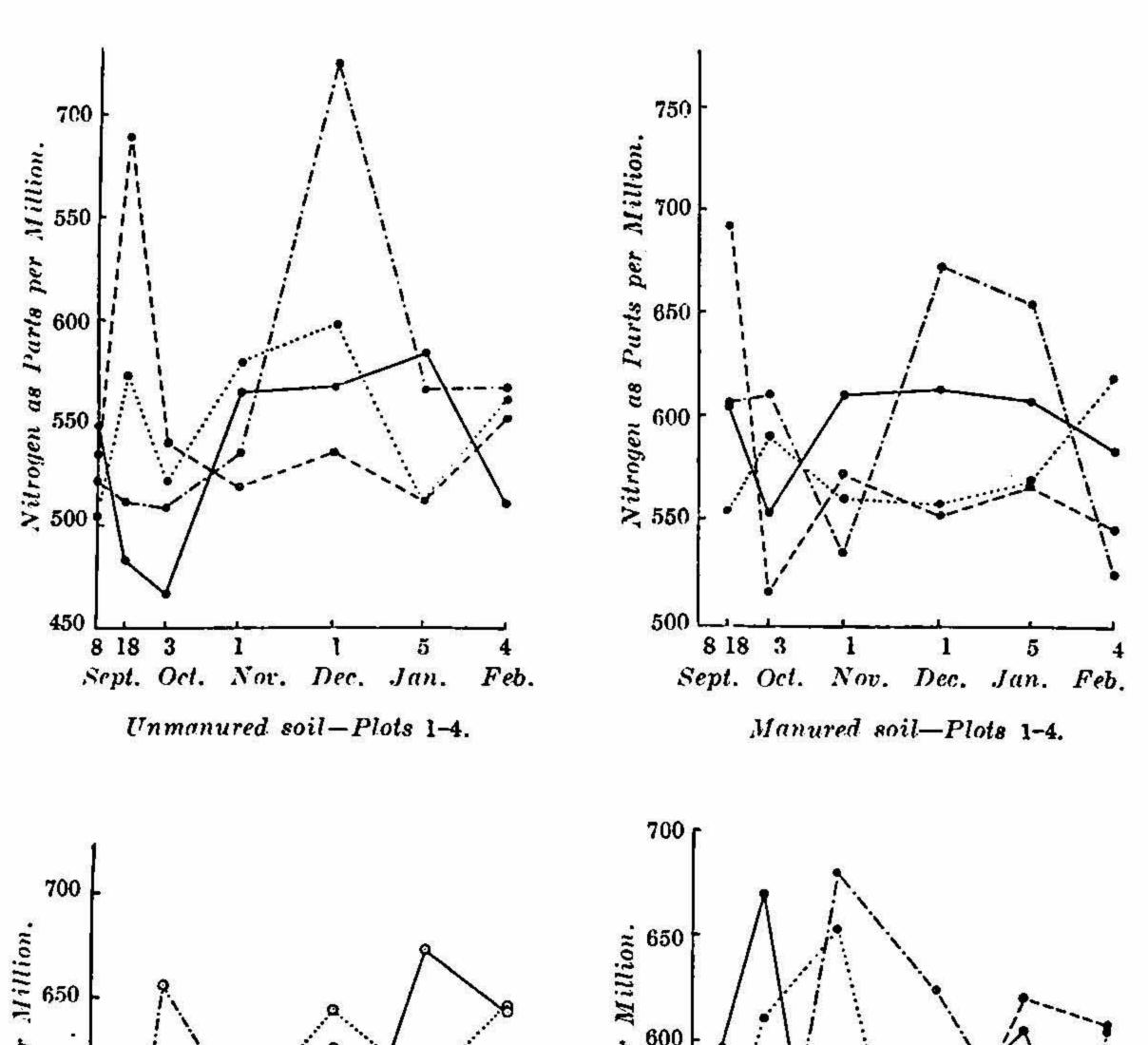
### EXPERIMENTAL.

A uniform plot of land measuring  $98' \times 49'$  was opened up and re-made so as to ensure thorough distribution of soil matter. The weeds were carefully removed and the area divided into 24 plots, each measuring  $11' \times 11'$  and separated by ridges each 1' wide and 1' high. A trench 2' wide and 2' deep was dug around the whole area to prevent (a) flooding from adjacent areas during rain, and (b) encroachment of roots from neighbouring vegetation. The plots were then numbered and given different treatments, the selections being made at random. — (1) Unmanured (control); (2) farmyard manure applied on 9–9–1933 at 3 tons per acre; 5 days later, this was followed by application of superphosphate at 2 cwts. per acre; (3) manured as in (2) and treated with potassium permanganate at 100 lbs. per acre. The last treatment was given as part of a main scheme of investigations on the rôle of oxidising agents as fertilizers. The results of those studies will form the subject of another communication.

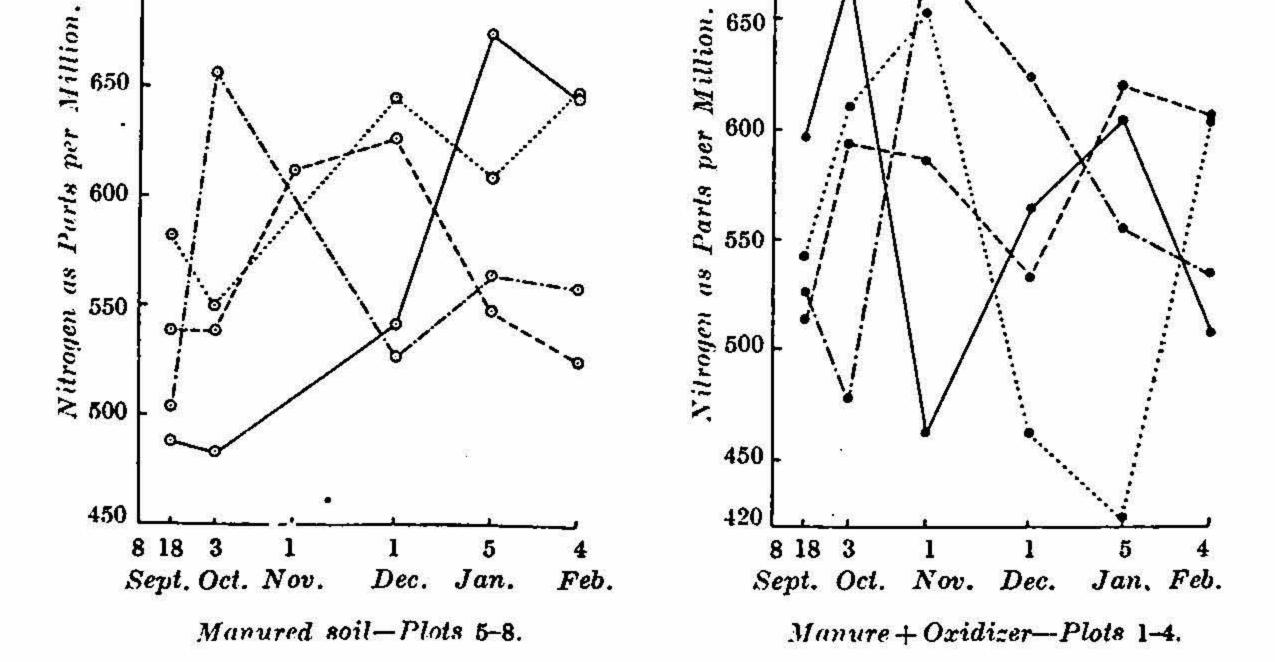
Ragi (*Eleucine coracana*) (H<sub>22</sub> variety) was sown on 18–8–1933. Samples of soil were collected from the various plots at different stages.— (1) Before manuring (8–9–1933); (2) prior to sowing (18–9–1933); (3) seedling stage (3–10–1933); (4) flowering stage (1–11–1933); (5) grain formation (1–12–1933); (6) ripening stage (5–1–1934); and (7) after harvesting (4–2–1934). The samples were collected, with a large cork borer, to a depth of 6" and the different samples thus collected from each plot, at one time, mixed together and treated as a whole specimen. They were then brought to the laboratory, dried and ground to pass the 30-mesh sieve. Determinations of the different forms of nitrogen were carried out as follows.—Total nitrogen by the Kjeldahl method as modified by Sreenivasan and Subrahmanyan (*Ind. J. Agric. Sci.*, 1933, 3, 646); ammonia and nitrate according to Olsen (*Compt. rend. Trav. Lab. Carlsberg*, 1929, 17, No. 15); and organic matter and combined moisture by determining loss on ignition.

*Meteorological Observations.*\*—Systematic daily records were maintained throughout the period of the investigation. The weekly averages have been presented in Fig. 1.

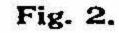
<sup>\*</sup> The data were obtained through the courtesy of the Meteorologist to the Government of Mysore to whom the authors' thanks are due.



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Changes in Total Nitrogen.

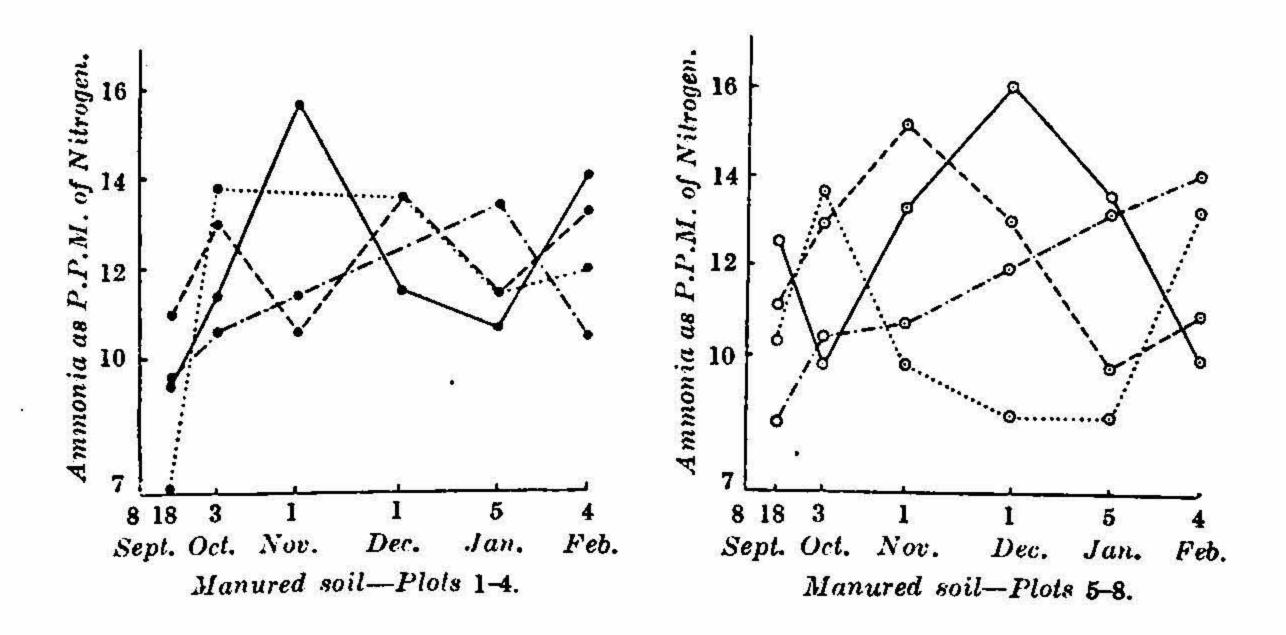
Plot Plot 5

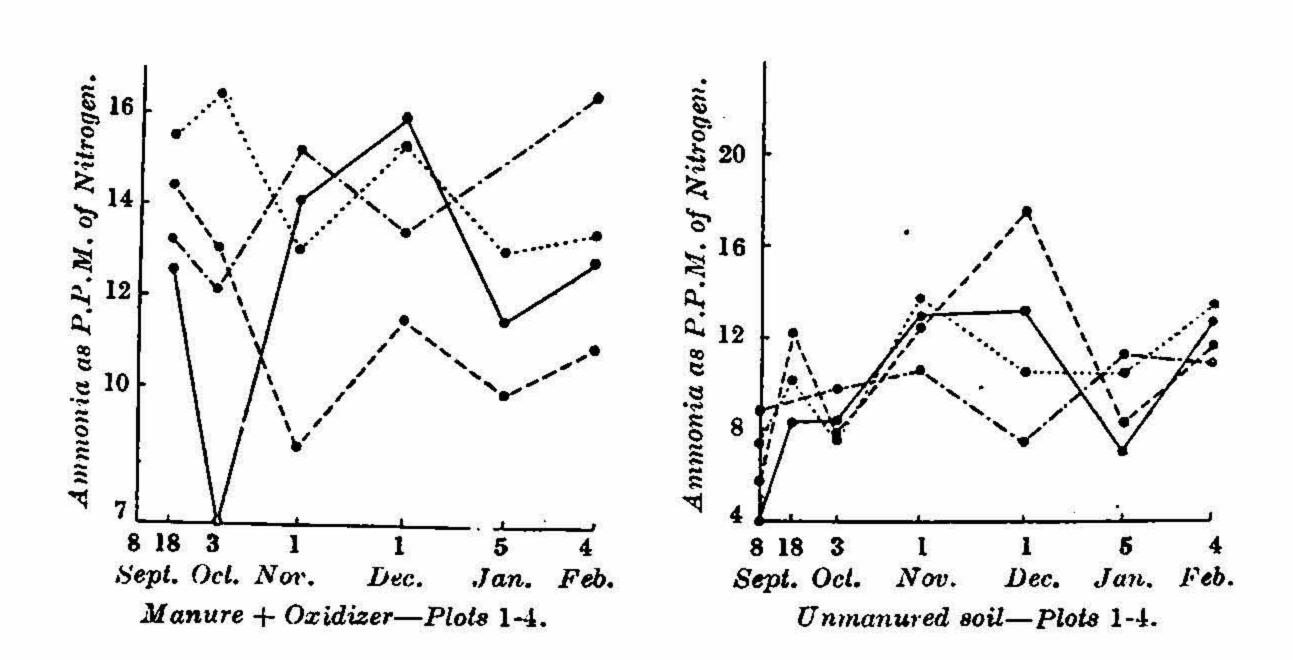
It may be noted that it was fairly warm upto the middle of November after which it turned cool and continued to remain so upto the third week in January when it became warm again. The hours of sunshine bore no relation to changes in temperature. With the exception of the first three weeks in October and a short period during the middle of December, the season was fairly bright and sunny. There were heavy showers early in September just before the commencement of the investigation. There were further periods of heavy rainfall during October and December. The remaining period had either no rains or only occasional, light showers. The relative humidity values followed nearly the same course as the rainfall. Each shower of rain was accompanied by a rise in humidity which was maintained for nearly a week. Succeeding falls caused it to rise again, so that except for a short period between the third week of November and the second week of December, and during early Feburary, the humidity remained consistently above 70 per cent. throughout the period under observation.

Taken as a whole, the observations would suggest that the period of investigation was a moderately wet and humid season with sufficiently long periods of sunshine and fair amount of warmth to favour different types of chemical and biological changes in the soil.

Total Nitrogen.-It may be noted (Fig. 2) that there was considerable fluctuation in nitrogen similar to those recorded by some previous workers. On the other hand, the observations on parallel plots show absolutely no relation to changes in season. Thus, a sudden rise observed in one plot would correspond to no change in a second one, slight depression in a third and marked fall in a fourth. The observations thus show that (a) the changes in nitrogen content do not follow any order and (b) the results obtained for individual plots at different stages are not significantly different from those in others. It may be further noted that the quantities of nitrogen in the unmanured plots are not appreciably different from those in the manured ones or treated with oxidising agents so that although the added manure enriches the soil with nitrogen, yet the estimates of that constituent have not appreciably improved. Ammonia.-The changes (Fig. 3) are as varied in character as those observed in the case of total nitrogen. Some of the figures for plots treated with manure and oxidizer are slightly higher than those for others suggesting the production of small quantities of ammonia. No significant difference could be noticed, however, between the other plots or treatments.

Nitrates.—The quantities of nitrates present at any time (Fig. 4) show no relation to either season or treatment. The data for the different plots are of about the same order so that no evidence could be adduced regarding either the production or loss of nitrate under different conditions.



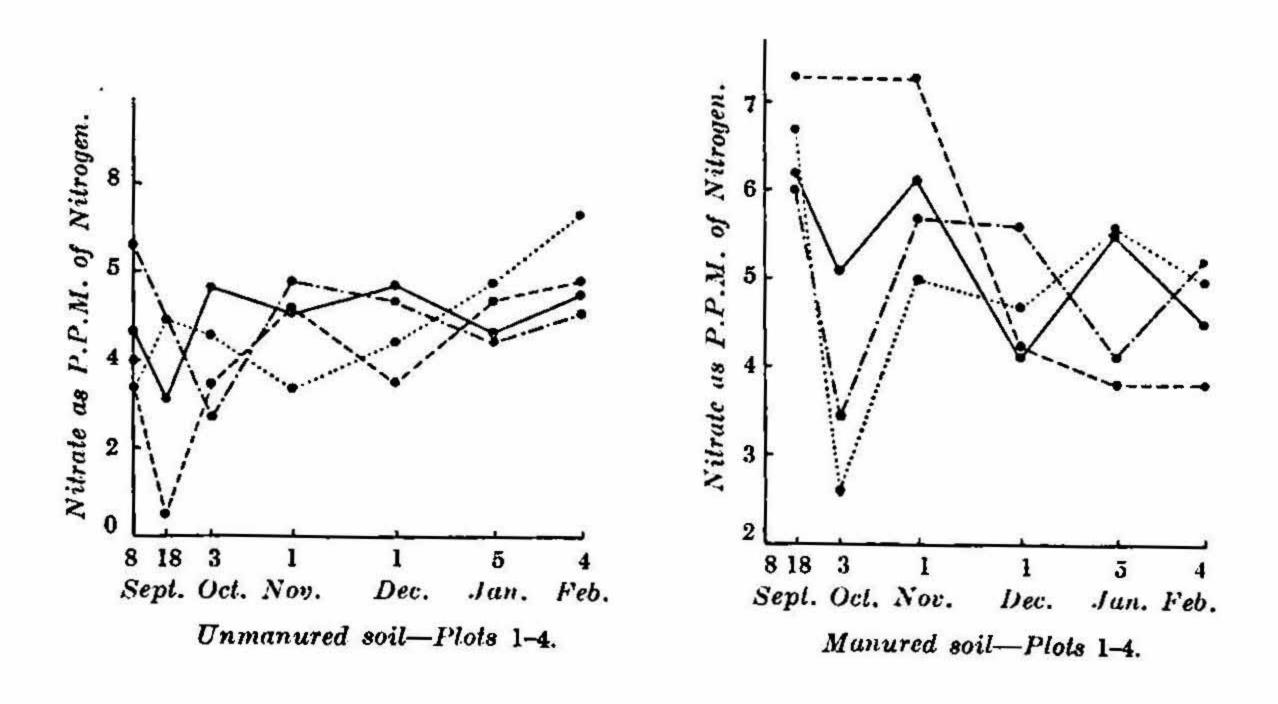


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Fig. 3.

Changes in Ammonia Content.

<b></b>	Plot 1	00	Plot 5
<b>****</b>	., 2	00	6
	., 3	00	
<b></b>	,, 4	00	., 8



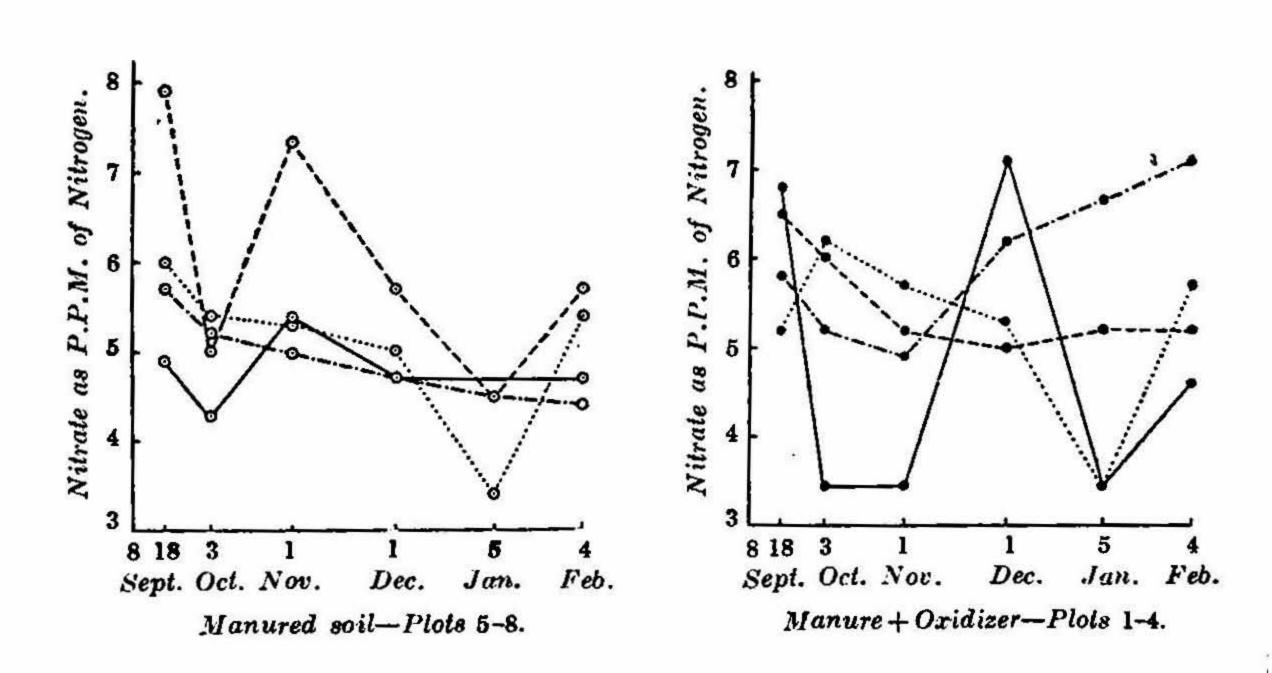
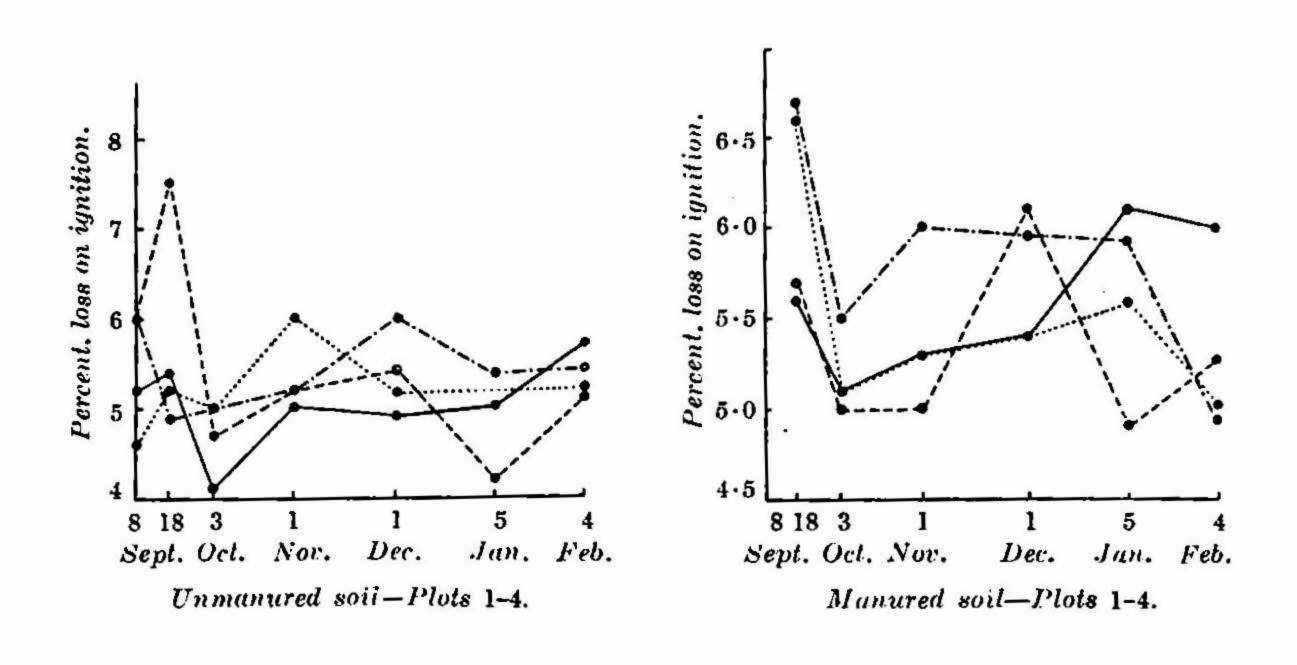


Fig. 4.

Changes in Nitrate Content.

 Plot 1	00	Plot 5
., 2	00	
 ., 3	00	
 4	00	8



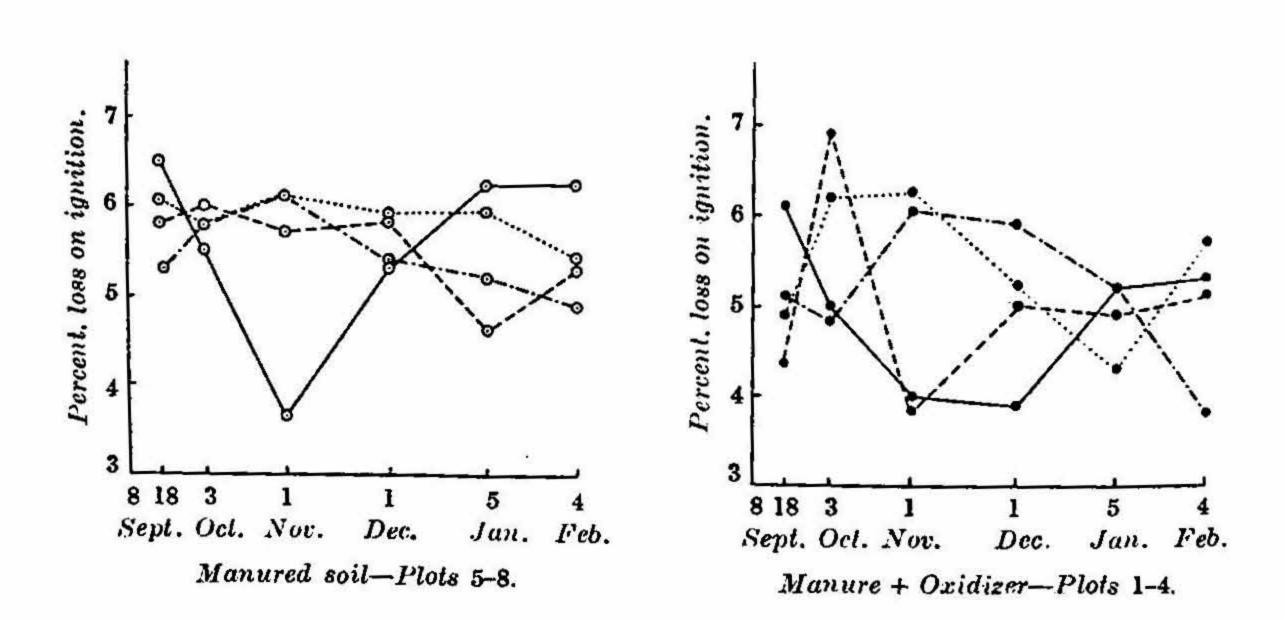


Fig. 5.

Loss on Ignition.

 Plot 1	0-0	Plot 5
 ., 2	00	
 3	00	
 . 4	00	. 8

Loss on Ignition.—With the exception of a few individual specimens which have yielded abnormally high or low values, all the others come within a narrow range and show no response to either manurial treatment or seasonal changes (Fig. 5).

The average values for the different constituents at various stages as also the corresponding standard deviations were each calculated from 16 sets of determinations in the case of the manured plots and 8 each in those of the others (Table I). The response of the individual plots to different treatments as shown by changes in the chemical constituents was calculated from the results of 14 sets of determinations in each case (Table II). It may be seen that the figures for each constituent are so close and the corresponding standard deviations so large that no significant difference between either different treatments or various plots can be observed.

### TABLE I.

Date $\rightarrow$		8—9—193	3	1	8-9-193	33	3	-10-193	33
Treatment->	U	м	м+0	U	м	м+-0	U	M	м+о
Total Nitrogen (as p.p.m.)	526-8	No treat- ment	No treat- ment	563.8	570-8	558·1	508.5	562 • 1	593.2
Std. devia- tion ± ···	16-3		•••	79.2	61.0	44.9	24 • 3	51.4	69.5
Ammonia nitro- gen (as p.p.m.)	7.1			10-2	9-9	13.9	8-4	11.9	12.1
Std. deviation $\pm$	1.7			1.6	1.6	1.1	0.8	1.5	3-3
Nitrate nitrogen (as p.p.m.)			• •	2.8	6.3	6-1	4.1	5.9	5.2
Std. deviation $\pm$	1.8			1.8	0-9	0.6	1.2	0.8	1.1
Loss on Igni- tion (as per- centage)	5.4			5.8	6.0	5-1	4.7	5.5	5.7
Std. devia- tion ± ••	0.6			1.3	0-5	0.0	0-4	0-3	0.9

Effect of different treatments on certain soil constituents.

U = Unmanured.

M = Manured.

M + O = Manure + Oxidizer.

TABLE I.—(Contd.)

Date $\rightarrow$	1-	-11-19	33	1-	-12-19	933	5-	-119	34	4-	-2-193	34
Treatment→	U	м	M+O	U	M	м+о	. <sup>v</sup>	м	м+о	U	м	м+0
Total Nitrogen (as p.p.m.)	549.5	556.6	600.5	605·4	591-1	551.0	541 • 8	599·3	557.7	542.7	581-4	569 . 2
Std. devia- tion ± •••	24.2	92.3	84 - 4	72.0	49.6	58.4	31.9	42.9	78.7	25.8	47.3	43-5
Ammonia nitro- gen(as p.p.m.)	12-5	12.3	12.8	12.2	12.7	14.0	9-4	11.5	11.5	12.0	12.2	13-4
Std. devia- tion ±	1.2	2.2	2.5	3.7	2.3	1.7	1.7	1.1	1.2	1.5	1.5	2.0
Nitrate nitrogen (as p.p.m.)	4.8	4-9	4.8	4.6	4.9	5.9	4.9	4.4	3.9	5.7	4.8	5-7
Std. devia- tion ±	0.9	0.6	0-9	0.9	0.2	0.8	0.2	1.0	0.8	0.8	0.6	0-9
Loss on Igni- tion (as per- centage)	5.3	5.4	5.0	5.4	5.7	5.0	4-9	5.6	4.9	5-4	5.0	5-(
Std. devia- tion $\pm$	0.4	0-8	1.1	0.4	e e	1	N I	0.6	0.4	0.2	0.5	0.1

U = Unmanured.

M == Manured.

M + O = Manure + Oxidizer.

The foregoing observations may be due to either inadequacy of the methods or uneven distribution of the different constituents in the soil. The standard errors of the average values obtained at each stage as also the errors of duplicate determinations of different constituents on individual plots have been given in Tables III and IV and would show that the errors due to analytical methods are very small and do not account for the large variations observed. It may be inferred therefore that the composition of independent specimens taken either from the same area or parallel plots receiving the same treatments are not similar; that there are large variations between parallel samples which account for all the discrepancies observed.

# **DISCUSSION.**

A critical enquiry into the results obtained by many of the previous workers would show that at least some of their conclusions would require modification in the light of certain errors inherent to the methods employed by them. Most of the estimates regarding the changes occurring in the field are based on results obtained with small samples of a few TABLE II.

Response of individual plots to different treatments.

Treatment		UNNANURED	IURED					MANURED	RED		÷.		MAS	MANURE + OXIDIZER	OXIDI	ZEK
Plot numbers	-	5	e	4		2	~	4	5	8	7	œ	1	63	e	-
Total nitrogen (as p.p.m.)	529.8	553.6	548.7	561 · 1	595.4	574-3	574.9	600.4	530.4	563.1	606-7	578-0	578-2	581.4	553 • 9	571.6
Std. deviation ±	42.6	56.6	34.1	6.69	21-0	55.8	23.5	56.1	108.4	36.8	37.2	59.2	20-8	39-1	84.0	66.5
Ammonia nitrogen (as p.p.m.)	9.6	10.7	10-5	9.9	12.1	12.1	11.6	11-1	12.5	12.2	10.7	11.4	12-3	11.4	14.4	1.4.1
Std. deviation ±	3.2	3.6	2.4	1.3	2.1	i.	2.4	1.3	2.2	1.7	2.0	2.0	13.98 19	1.9	1.4	1.5
Nitrate nitrogen (as p.p.m.)	4	3.0	4.7	4.9	5.2	5.3	4.6	6.0	4. 8	0.9	ŏ.1	5.0	4.7	5.5	5.2	5.8
Std. deviation ±		1.6	1.2	1.2	0.8	1.6	1.3	1.0	0.4	1.2	2.6	0.5	1.6	1.7	6.0	0.8
Loss on Ignition (as percen- tage)	<u> </u>	5.4	5.2	5.4	5.5	5.4	5.5	5.8	ũ • 7	5.5	5.9	5.4	4.9	5.0	5.4	5.2
Std. deviation ±	13-0 	1.0	0.4	0.4	0.3	0.4	0.5	0.5	1.0	0-5	0.8	0.4	0.7	1.0	2.0	0.8

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### TABLE III.

Standard error of the average values obtained at each stage. (32 determinations in each case.)

				DATE			
Form of Nitrogen	8-9-1933	18-9-1933	3-10-1933	1-11-1933	1-12-1933	5-1-1934	4-2-1934
Total (as p.p.m.)	4.2	3.9	5.0	4.1	5.2	4.9	6-6
Ammonia (as p.p.m.)	0•4	0.2	0+2	0.3	0.5	0.4	0-6
Nitrate (as p.p.m.)	0.3	0.5	0.7	0.5	0.2	0 · 2	0-6

## TABLE IV.

Standard error of the average values for individual plots at each stage. (14 determinations in each case.)

Treatment →	ן 	Jnma	nurea	1				Man	ured				Ma	nure-	+Oxio	lizer
Plot number $\rightarrow$	1	2	3	4	1	2	3	4	5	6	7	8	1	2	3	4
Form of nitrogen Total (as p.p.m.)	4.0	6.6	6.3	6.3	5.9	3.7	4.9	3.9	4.2	2.0	4.4	5.7	7.2	7-6	4.7	2-8
Ammonia (as p.p.m.)	0.6	0-3	0.4	0.4	0.7	0.6	0.5	0.3	0.8	0.6	0.5	0.4	0.2	0.3	0.6	0.5
Nitrate (as p.p.m.)	0.3	0.4	0.5	0.6	0.2	0.3	0.8	0.6	0-4	0-4	0.5	0.7	0.6	0-4	0-5	0.4

grams each, so that the related calculations would involve the multiplication of the original errors by hundreds of millions. The influence of certain factors that affect the accuracy of laboratory methods has also to be carefully considered. Thus, it is known that an enhanced nitrogen value can be obtained by merely moistening a specimen of soil (Sreenivasan and Subrahmanyan, *loc. cit.*) so that there will be an apparent increase in nitrogen content if the same specimen is examined just after a shower of rain. Pfeiffer and Blanck (*Landw. Vers. Sta.*, 1913, 78, 367) considered the analytical error involved in the study of nitrogen balance in the soil and have concluded that the non-homogeneous character of the soil is itself a serious factor which would cause large differences between duplicate determinations or even successive samples. Robinson and Lloyd (*J. Agric. Sci.*, 1915, 7, 144) have drawn attention to the existence of normal variations in the composition of the soil from point to point in the field. Similar observations have also been recorded by Waynick (Univ. Calif. Pub. Agric. Sci., 1918, 3, 243) and Waynick and Sharp (Univ. Calif. Pub. Agric. Sci., 1919, 4, 121) who observed large differences in the total nitrogen, nitrate and carbon contents of field samples of soils. Bear and McClure (Soil Sci., 1920, 9, 65) and, later, Prince (Soil Sci., 1923, 15, 395) have shown that the nitrogen contents of specimens of soil and sub-soil chosen from the same plot of land vary in different places and lay emphasis on the error due to sampling in field experiments.

It has been observed by the present authors that even in finely ground and apparently homogeneous specimens of soils, such as are used for analytical work in the laboratory, the distribution of nitrogen is not uniform so that it is not possible to obtain concordant estimates unless the entire specimen is mixed repeatedly and carefully resampled from time to time. It is not surprising therefore to find that independent or even mixed specimens from the same area, however apparently uniform, exhibit striking variations which might be easily mistaken for periodic fluctuations in the composition of the soil. Furthermore, the major part of the nitrogen of the soil is in organic form, being mostly derived from either added manures or plant residues decomposing in it. Since it is not possible in ordinary field practice to distribute the organic matter so evenly as to facilitate accurate sampling for analytical work, it would follow that although the total amounts of nitrogen spread over large areas may be the same, the quantities present in small samples collected as representative specimens may vary considerably. Additional error is also introduced by cattle grazing or sheep-folding during offseasons, resulting in the droppings being unevenly distributed over the surface of the soil. In assessing the influence of season on processes like nitrogen fixation, the effects of different external factors on the movement of different soil constituents would also require careful consideration. Thus, it is well known that during periods of fallow, a large part of the nitrogen, particularly the soluble forms, moves into the subsoil and does not become available until the planting of the subsequent crop. During the rainy season, the finer fractions containing the highest percentage of fertilising ingredients would be washed from the surface of a sloping or uneven soil and carried to the lower regions exposing only the less rich, coarse fractions at the surface. Even on the same plot of land considerable variations in the distribution of different soil constituents can be brought about by washing or silting as the case may be.

It would thus be seen that although it is desirable to study changes in the different soil constituents under conditions actually obtaining in field practice, there are yet serious inherent defects due to soil heterogeneity, uneven distribution of matter, erosion and silting so that no accurate estimate of the various changes can be obtained by

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carrying out studies exclusively on the field. Such investigations are therefore best carried out under the controlled conditions of the laboratory and with uniform specimens of soil, parallel samples of which agree closely in composition.

### SUMMARY.

1. The periodic changes in total nitrogen, ammonia, nitrate and organic matter contents of apparently uniform plots of land subjected to different treatments were studied.

2. It was observed that in none of the cases was there any correlation between (a) seasonal changes, (b) manurial treatment, or (c) stage of crop and the variations in the different constituents.

3. Examination of the results obtained in a large number of cases have shown conclusively that the laboratory error due to sampling and methods of analysis is negligible as compared with the field error due to variations in the composition of the soil from point to point. Attention is also drawn to external factors such as erosion, silting and movement of soluble nutrients which lead to uneven distribution of different soil constituents.

4. It is concluded therefore that in the study of biochemical transformation in the soil, experiments conducted with uniform samples and under controlled conditions in the laboratory are likely to yield far more useful results than similar studies conducted on field samples.

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