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U. S. DEPARTMENT OF AGRICULTURE,
BUREAU OF PLANT INDUSTRY—BULLETIN NO. 173.

B. T. GALLOWAY, *Chief of Bureau.*

SEASONAL NITRIFICATION AS INFLUENCED BY CROPS AND TILLAGE.

BY

C. A. JENSEN, AGRICULTURIST.

ISSUED APRIL 18, 1910.

WASHINGTON:
GOVERNMENT PRINTING OFFICE.
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LETTER OF TRANSMITTAL.

U. S. DEPARTMENT OF AGRICULTURE,
BUREAU OF PLANT INDUSTRY,
OFFICE OF THE CHIEF,
Washington, D. C., December 23, 1909.

SIR: I have the honor to transmit herewith and to recommend for publication as Bulletin No. 173 of the series of this Bureau the accompanying manuscript, entitled "Seasonal Nitrification as Influenced by Crops and Tillage," by Mr. Charles A. Jensen, Agriculturist, of this Bureau.

The Office of Dry-Land Agriculture, of this Bureau, is carrying out extensive investigations in crop rotation and cultivation methods at thirteen stations located at widely different points in the Great Plains area. The purpose of these investigations is to ascertain the best methods of agriculture in these semiarid regions. To do this intelligently it is necessary not only to study the immediate effect of these methods on crop yields, but also to study the resultant changes occurring in the physiological properties of the plant nutritive solution, which is recognized as one of the fundamental factors in crop yields. It is therefore necessary to supplement the field work with laboratory investigations having for their aim the solution of the problems presented by the crop returns from the rotation systems mentioned.

The accompanying manuscript shows the seasonal changes in the water-soluble nitrates in summer-fallowed land, wheat land, and corn land and the rates and extent of the seasonal removal of the nitrates by these crops. The investigations were made by Mr. Jensen while he was superintendent of the Bellefourche substation of the Office of Dry-Land Agriculture. The work was conducted in cooperation with the Physical Laboratory of this Bureau, which has general supervision over the physical measurements carried out at the dry-land stations of the Bureau.

The results described in the paper are, I believe, sufficiently significant to demonstrate the importance of a more extended development of this line of investigation at the stations.

Respectfully,

B. T. GALLOWAY,
Chief of Bureau.

HON. JAMES WILSON,
Secretary of Agriculture.

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SEASONAL NITRIFICATION AS INFLUENCED BY CROPS AND TILLAGE.

INTRODUCTION.

The attempt to bring the vast areas of prairie lands of the Great Plains under a profitable system of agriculture has been undertaken with renewed vigor during the last few years, both by the older settlers and by many new ones entering the Plains. These new settlers come mainly from the Middle West, where the climatic and soil conditions are very different from those in the newer areas where they settle. On the Great Plains the rainfall is likely to be too little rather than too much for good crop returns and, moreover, it does not always come at the time most convenient for the farmer; besides, the wind movement, especially in the spring, when the crops are being planted and when the surface of the soil is usually exposed in a rough condition, saps the soil of its moisture very rapidly.

Under such conditions it becomes necessary to control the quantity and the movement of the soil moisture, and as this moisture constitutes the plant nutritive solution it is obvious that not alone the absolute amounts of soil moisture present but the nutritive character of this solution become of importance, so that any cultural system inaugurated for the purposes of moisture conservation should be such that the quality as well as the quantity of the nutritive solution can be controlled.

It has often been observed that of two contiguous plats on the same type of soil one may yield considerably higher than the other, although the amount of soil moisture may be the same in both. In such cases it is often found that the properties of the nutritive solutions from the two plats are markedly different in their behavior toward plant growth, and often the history of the plats will show that the cultural methods have been different and that these may explain the differences found in the character of the soil moisture constituting the plant nutritive solution. The mere storing up, then, of soil moisture is not always sufficient for the purposes of good crop returns.

During the last few years a number of substations have been established by the Bureau of Plant Industry throughout the Great Plains

area for the purpose of finding out, if possible, the best methods of handling the prairie lands there so that profitable crop returns might be realized. At all of these stations a large number of rotations have been established, and other plats have been set aside for experiments with various methods of cultivation without regard to crop rotations.

In the spring of 1907 the writer was put in charge of the substation at Bellefourche, S. Dak., where experiments in dry-land agriculture were inaugurated. The station site was on virgin prairie lands, so nothing was done that year beyond breaking up and preparing land for use in 1908. In the spring of that year an elaborate series of rotation systems was established, and a large number of other plats were set aside for use in the studies of soil-moisture conservation by different methods of cultivation, the success of the methods being measured finally by the crop yields, though moisture determinations were made during the entire season.

The work to be described was done on some of the plats used for the purpose of trying out different cultural methods, it being thought that different crops and different tillage methods would influence the nutritive properties of the soil solution. Owing to the press of field duties and to the inconvenient location of the station as regards supplies, it was found necessary to limit the laboratory work to a study of the natural activity of nitrification in the soil as measured by determinations of nitrates, nitrites, and ammonia in three plats receiving different treatments and bearing different crops.

The literature on nitrifying organisms and on nitrification is very voluminous, and while the author does not by any means claim to have read it all, it is believed that most references which have any direct bearing on the present subject matter have been cited. In no case was there found reported work on nitrification in the field as influenced by crops and tillage that had been carried on continuously throughout the crop season. The few references found which might have a bearing were generally reports on work carried on in humid regions or in the laboratory, the conclusions of which could hardly be extended to the semiarid regions. Much of the literature, however, was helpful in formulating suggestions explaining the phenomena found.

DESCRIPTION OF THE SOIL IN WHICH THE EXPERIMENTS WERE CONDUCTED.

The soil of the station and of the surrounding country is known as "gumbo" and is a heavy, black or gray, clay loam to a depth of 2 to 3 feet, and below this depth heavy clay usually occurs, extending to 6 feet or deeper, overlying the partially disintegrated cretaceous shale which is the geological formation of the whole surrounding country.

FIELD HISTORY OF THE PLATS.

The prairie sod, covered originally with western wheat-grass (*Agropyron occidentale*), buffalo grass (*Bulbilis dactyloides*), and gramma (*Bouteloua oligostachya*), with minor quantities of sedges (*Carex filifolia* and *C. stenophylla*) and other grasses, together with a few vetches, was broken in the spring of 1907 to a depth of about 3 to 4 inches, backset (replowed) in the fall of 1907 to a depth of about 5 inches, and left open until the spring of 1908, when it was disked and harrowed until a good seed bed was secured. Such was the treatment of the spring-wheat plat and the corn plat used in the work. A plat reserved for summer-fallow used in this work was spring-plowed to a depth of about 8 inches, smoothed with a disk, and harrowed. This plat was cultivated during the summer to keep weeds down and to conserve the moisture.

The determinations on the corn plat were not begun as early as on the wheat and fallow plats. As this plat was not planted as early as the wheat plat, it was taken for granted that the soil conditions would be the same as those of the fallow plat until the time of planting the corn.

METHODS OF MAKING DETERMINATIONS.

The chemical methods used were those described in Bulletin No. 31 of the Bureau of Soils. The soil samples were collected in duplicate from 6-inch layers to a depth of 2 feet—that is, in 0 to 6, 6 to 12, 12 to 18, and 18 to 24 inch depths—and the two cores from each soil layer made into a composite sample. The samples as bored up with the auger were transferred to air-tight tin cans, taken immediately to the laboratory, weighed, dried in an oven at a temperature of about 100° C., and again weighed. In preparing the solution it was found necessary to adopt this drying method in order to insure thorough mixing of the soil and the water, as it was practically impossible to break down the moist gumbo pieces directly. The ratio of soil to water was 1 to 5 (100 grams of soil to 500 c. c. of water). After drying, the soil particles readily broke down and after thorough stirring in the water would settle out quite clear in about 20 to 30 minutes. The supernatant liquid was then poured into Chamberland filters and filtered. Such a procedure of drying the soil could, of course, not be employed were determinations to be made of such soil elements as are derived directly from the soil minerals, as potassium, phosphoric acid, etc., but owing to the origin and nature of the nitrates and nitrites in the soil the solubility is not increased; neither is it likely that decomposition of a disturbing nature would take place. Experiments were made to compare the two methods of obtaining the

soil solutions from soil that was first dried and from undried samples direct from the field. The results agreed within the limits of experimental error, and as better mixing was possible by using the dried samples this procedure was an advantage.

The determination of nitrates under ideal conditions does not present any serious difficulty if the procedure is carefully followed out and precaution against errors observed. As is well known, however, difficulties are likely to be encountered when nitrates are to be determined in soil extracts, especially if the extract is highly colored with soluble organic matter or if it contains considerable soluble inorganic salts, especially chlorids. There are thus three main sources of error in nitrate determination in a soil extract: Foreign organic color, which is superimposed on the yellow color developed on nitration of the phenoldisulphonic acid, which renders the colorimetric reading more difficult; the soluble organic matter may be acted upon by the nitrates, or possibly by the acid, thus introducing a chemical error; the presence of considerable amounts of alkali salts, of which chlorids seem to be the most serious ones.

The soil extract obtained in the work here reported did not contain enough chlorids to cause error, as the amounts were too small to measure accurately by means of titration; only traces were ever observed, and frequently none at all.

The errors which might be introduced because of the presence of soluble organic matter are believed to be too small to seriously consider, as the only soil samples which gave appreciable organic colors were those from the fallow plat, and no serious difficulty was encountered in making the colorimetric readings of these extracts, the organic color being of quite a different shade from the yellow color due to nitration. The error which may have been introduced through the chemical disturbance of the organic matter was neglected, it being considered too small to be of serious consequence; as stated above, the organic color was not strong. All points considered, it was decided that under the circumstances and conditions less error would be involved in using the solutions directly as obtained than by trying to free them of organic matter by oxidation or reduction.

It should be kept in mind that the chief value of the results obtained lies in their relativity more than in the absolute amounts found. This weekly relation of the amounts of nitrates in the various soil layers in the various plats could not be seriously disturbed by any slight error which might possibly have been introduced because of the presence of soluble organic matter in the soil extracts.

It has been quite well made out that no appreciable error was introduced in quickly drying the soil samples in the oven at 100° C.

This was established at the time and has been also found in the bacteriological laboratory of this Bureau to be a safe procedure for purposes of comparison.

A comparison of results obtained by the various methods of extraction is given in Table I.

TABLE I.—*Nitrites, nitrates, and ammonia found in dry soil treated in various ways.*

Soil treatment.	Parts per million in dry soil.		
	Nitrites (NO ₂).	Ammonia (NH ₃).	Nitrates (NO ₃).
Fresh untreated soil, using undistilled water.....	0.55	1.5	26.7
Fresh untreated soil, using distilled water.....	.55	.8	22.2
Fresh untreated soil plus chloroform, using undistilled water.....	.55	2.5	22.2
Fresh untreated soil plus chloroform, using distilled water.....	.55	2.0	25.0
Soil dried in oven at 100° C.....	.55	3.33	36.4

The oven-dried sample in this case gave a higher proportion of ammonia and nitrates than the other samples. It is possible, though, that nitrification went on to some extent before the sample was properly dried, as the gasoline drying oven was troublesome, this being its first use. The soil sample was in the oven most of the day before the trouble with the burner was located, and it was warmed most of the day without quickly drying.

The following set was run through the next day, a soil being used that had been carefully prepared for a winter-wheat seed bed the previous year but which had not been planted:

TABLE II.—*Nitrites, nitrates, and ammonia found in dry soil from a seed bed treated in various ways.*

Soil treatment.	Parts per million in dry soil.		
	Nitrites (NO ₂).	Ammonia (NH ₃).	Nitrates (NO ₃).
Fresh untreated soil immediately extracted.....	0.28		133.3
Fresh soil plus chloroform and immediately extracted.....	.28		133.3
Fresh soil after three hours' treatment with chloroform and dried quickly at 103° C.....	.20		133.3
Fresh soil dried immediately in oven without being treated with chloroform.....	.33		133.3

The ammonia was not read, as the solution contained foreign coloring matter. These and the previous results give an idea of the limits of experimental error. The standard solutions used for comparison in the colorimeter were made up fresh on the day they were used, and all readings were made within a few hours. The nitrite reagents were kept separate and small portions mixed as needed for use.

The systematic determinations were begun April 27, and from then until July 31, about ten days after harvest, the determinations were made once a week.

DISCUSSION OF RESULTS OF DETERMINATIONS.

NITRATES IN THE FALLOW PLAT.

Figure 1 shows the parts per million of nitrates calculated for the oven-dried soil in each 6-inch layer in the fallow plat from April 27 to July 31 and the precipitation for the same period. About 1 inch of rain had fallen from April 1 to April 27. The weekly periods are laid off on the abscissa, and the parts per million of nitrates on the ordinate. The precipitation scale is on the right-hand side of the figure.

The first two sets of determinations show decreasing amounts of nitrates from the surface 6-inch layer downward, the second foot con-

FIG. 1.—Diagram showing the parts per million of water-soluble nitrates in dry soil in the fallow plat in each 6-inch soil layer to a depth of 2 feet; also precipitation curve.

taining very small amounts. There were no marked changes until May 9, when the surface 6 inches of soil showed a considerable increase in nitrates, and it is also seen that at this time this soil layer contained a maximum amount of water-soluble nitrates. The nitrates in the second layer, 6 to 12 inches, had more than doubled since the week before, but there were no considerable changes in the lowest two layers. By the next week, May 16, the surface 6 inches of soil had decreased about forty parts per million, and there was also a decrease in the 6 to 12 inch layer. By the next week, on May 22, a marked increase in the amount of nitrates is evident in the 12-inch layer, it having increased about seventy parts per million during six days, and the surface 6-inch layer had decreased about ten parts per million. At this date the 12-inch layer contained the maximum quantity of

nitrates. Up to this time there were no great changes in the nitrates in the 18 and 24 inch layers, but from May 22 to June 5 these layers accumulated considerable amounts, and on the latter date the 18-inch layer contained the maximum, which remained constant until the next week, June 12. In the mean time, the 24-inch layer accumulated considerable quantities of nitrates, reaching its maximum on June 12.

It will be noticed in general that from April 27 to June 5 the maximum amount of water-soluble nitrates shifted every two weeks to the next lower layer and that once any layer had accumulated a maximum it decreased and in general never again contained as large amounts as the maximum of the next lower layer, which was reached at a later date.

From June 12 to June 19 there was an interesting change, in that every soil layer which had recently been actively accumulating nitrates suddenly showed a marked decrease in amounts of water-soluble nitrates, and all of the layers on June 19 contained nearly the same amount of nitrates. The upper 6-inch layer did not show any such decrease, the reason for which will be discussed later on. From June 19 till July 31 there were no regular changes, all of the curves zigzagging irregularly.

In view of the marked increases in the amount of nitrates in the 18 to 24 inch layer, it is to be regretted that determinations were not made in the third foot also, but time did not permit the increased work. The 18 to 24 inch layer never contained a maximum amount of nitrates in comparison with the other layers, but the results certainly indicate that nitrification is quite active at a depth below 2 feet in these prairie soils when they are brought under cultivation. Bazarewski^a found very few nitrifying bacteria, however, at a depth of 50 centimeters and reports that they were "plentiful at a depth of 10 centimeters." It will be noticed from figure 1 that the 18 to 24 inch soil layer accumulated as much water-soluble nitrate as did the 0 to 6 inch layer, so nitrifying organisms are presumably active in this prairie soil to a depth considerably greater than 2 feet. Hunt^b likewise reports that nitrification took place to but a small extent below a depth of 2 feet, and the figures given by him show that under the conditions discussed nitrification was not very active in even the second foot.

In this connection it may be of interest to mention that the unbroken prairie "gumbo" never becomes moistened deeper than about 8 to 10 inches even after the heaviest rains. Cultivation,

^a Bazarewski, L. von. Nitrification and Denitrification in Soils. Reviewed in Experiment Station Record, vol. 21, no. 1, July, 1909, pp. 20-21.

^b Hunt, T. F. The Importance of Nitrogen in the Growth of Plants. Bulletin 247, New York (Cornell) Agricultural Experiment Station.

however, soon results in the soil becoming moistened deeper than 6 feet within one year after breaking the sod and cultivating the soil. It is fair to assume, therefore, that nitrification in the unbroken land can not take place to any great extent much below a depth of 1 foot.

NITRATES IN THE SPRING-WHEAT PLAT.

Figure 2 shows the results obtained from determinations made on the spring-wheat plat. These determinations were not begun until May 9, but a vertical section made in figure 1 on May 9 will show that the order is about the same at this point on both figures.

Figure 2 does not contain the precipitation curve, but in addition to the curves showing the amounts of nitrates in the 6-inch soil layers,

FIG. 2.—Diagram showing the parts per million of water-soluble nitrates in dry soil in the spring-wheat plat in each 6-inch soil layer to a depth of 2 feet; also weekly increase of wheat plants.

it shows the curve representing the weekly increase in the dry weight of the wheat plants. The increase was determined each week by cutting four 3-foot squares, two near each end of the 2 by 8 rod plat, both green and air-dry weights being obtained. Only the dry weights are plotted in the figure.

A comparison of figure 1 and figure 2 shows the same general fact, namely, that the maximum amount of water-soluble nitrates in any soil layer is shifted every two weeks to the next lower layer. The detailed variations are about the same in the two plats, but there is one general difference—the date of the maximum accumulation of nitrates in the respective soil layers in the two plats is shifted one week forward in the case of the spring-wheat plat—that is, each of the 6, 12, and 18 inch layers, respectively, attained its maximum

accumulation of water-soluble nitrates just one week later in the wheat plat than in the fallow plat. It will be remembered that it was stated that the fallow plat was plowed to a depth of about 8 inches in the spring of 1908 and that the wheat plat had not been plowed since being backset in the previous fall to about 5 inches deep. There was thus a difference in the mechanical condition of the two plats; the summer-fallow plat being more open and stirred to a greater depth, there was presumably a more rapid access of air, and in consequence a more rapid rise in temperature. Hunt ^a also found that early plowing promoted nitrification more rapidly than late plowing, and presumably early plowing would promote nitrification more rapidly than no plowing at all.

It was expected that some information would be obtained in regard to the rate at which the growing wheat plants removed the nitrates from the different soil layers, that being one of the main points sought for in the work. As a matter of fact, it will be noticed that the curves of the two figures are so much alike in the general trend that were they not scaled or labeled in any way it would be difficult to say which figure represented the fallow plat and which the wheat plat. The shifting of the dates of the maximum accumulation of nitrates in the individual soil layers one week back in the case of the fallow plat might be explained by the difference in the tillage. By using the fallow plat as a check on the wheat plat, which is really what it is, it will be noticed that aside from this shifting already mentioned the only general differences are the smaller amounts of nitrates found in the wheat plat and that after June 19, when the general decrease in nitrates took place—as in the fallow plat—the nitrates in each soil layer in the wheat plat were more constant in both total amounts and amplitude of variation. Had the results on the fallow plat not been obtained as a check on the results obtained on the wheat plat, it might have been supposed that during the time immediately following June 12, when the wheat plants were making rapid increase in growth, the crop was removing nitrates much more rapidly than they were being formed. Even thus one would hardly expect the nitrates to be removed so spontaneously and almost completely from each 6-inch soil layer. One can not, then, from the amounts of water-soluble nitrates found, determine the soil layers in which the wheat roots were feeding most rapidly or demonstrate any progressive downward ranging of the roots as the season advanced. The apparent “removal” of the nitrates from the soil is due, then, to some other factor than the presence of the crop.

Even after June 19, when the marked decrease in the amounts of water-soluble nitrates occurred, there is no consistent correlation

^a Hunt, T. F., loc. cit.

between the weekly increase in the dry weight of the wheat crop and the amounts of nitrates present. On the contrary, from June 24 to July 3 there was a general increase in the amounts of nitrates corresponding to the greatest weekly increase in the growing crop.

COMPARISON OF NITRATES IN FALLOW, SPRING-WHEAT, AND CORN PLATS.

Figure 3 shows the amounts of nitrates—averages of the four 6-inch soil layers—found during the season in the wheat plat, the fallow plat, and the corn plat; also the precipitation curve and the weekly crop-increase curve. It is to be regretted that the same amount of data could not have been obtained from the corn plat as from the wheat plat, but time did not permit. Nothing much can

FIG. 3.—Diagram showing the average parts per million of water-soluble nitrates in dry soil to a depth of 2 feet in the summer-fallow, spring-wheat, and corn plats; also weekly increase of wheat plants and precipitation curve.

be said about this curve beyond the statement that the rapid decrease in nitrates corresponds quite closely to the period of most rapid growth of the corn as recorded in the notes at the time.

The corn plat had evidently accumulated quite large amounts of nitrates before the plants were large enough to materially draw upon these salts, for the corn curve shows that on June 19 the accumulation of nitrates in the corn plat had reached about the same point as that in the fallow plat. The direction of the corn-plat curve from June 19 to June 24 also suggests the general drop found in the fallow and wheat plat curves. As the corn was not planted until May 7, the plat was evidently in a state of summer fallow until determinations were begun. During the most active growing period of the corn these plants evidently reduced the amounts of nitrates to as low a

content as did the wheat plants in the wheat plot. Apparently the corn plants were as fond of the nitrates as were the wheat plants. The general field observation that a corn crop renders the ground more productive for grain than a previous grain crop is apparently not due to the leaving of greater amounts of nitrates in the soil.

WITHDRAWAL OF NITRATES BY CORN ROOTS.

As already mentioned, the general decrease in the amounts of water-soluble nitrates in both the fallow and the wheat plots masked the effect of any progressive withdrawal of the nitrates by the wheat-plant roots in the successive soil layers. In the case of the corn plot, however, there was the advantage of the fact that the nitrates had already been accumulated, and so the corn offered an opportunity for observing the removal of nitrates from the various soil layers as the

FIG. 4.—Diagram showing the parts per million of water-soluble nitrates in dry soil in each 6-inch soil layer in the corn plot to a depth of 2 feet.

season progressed. Figure 4 shows the probable effect of the feeding of the corn roots and indicates that the surface 6-inch layer early lost considerable amounts of nitrates and that very shortly thereafter the 12-inch layer was drawn upon most heavily. From July 3 to July 17 the corn plants made their most rapid growth, and the curves also show that at this period the nitrates were most heavily drawn upon. The 18-inch layer contained most nitrates and lost most. All of the layers to a depth of 2 feet were probably reduced to the limit of the powers of the corn roots to withdraw nitrates from this heavy soil.

The curves in figure 4 also show variations in the activity of nitrification, especially an increase from June 24 to July 3, but the

progressive downward feeding of the corn roots seems clearly shown. The characteristic manner of surface feeding of corn roots would early draw upon the plant food in the surface soil, as indicated in figure 4 by the curves of the 6 and 12 inch layers. The soil samples, however, were taken not between the corn rows, where the lateral roots only were abundant, but as near the corn plant as they could be taken, not more than 3 or 4 inches away. Such sampling would encounter the primary roots of the corn, as well as penetrate the soil area in which the lateral roots were feeding most actively, and it seems probable that the reduction of the amounts of nitrates in the 18 and 24 inch layers was due to the penetration of these primary roots to that depth. This would account for the tardy removal at the lower depth, as the primary roots would be longer in reaching these lower layers. The moisture determinations showed that the moisture was being removed quite rapidly from the 18 and 24 inch soil layers at the same time.

The most striking feature about figure 3 is the general parallelism of the nitrate curve of the wheat plat with the nitrate curve of the fallow plat. The seven-day lagging of the maxima in the wheat plat is not, however, as clearly shown here as in figures 1 and 2, but is evident in the early part of the season. The only other point of difference in the two curves is in the period from May 29 to June 12, during which time the summer-fallow plat steadily accumulated nitrates, while there was a bare increase in these salts in the wheat plat. This is the period during which the fallow plat laid in its increased stock of water-soluble nitrates, and from June 12 on there is a constant difference of about twenty to thirty-five parts per million in favor of fallowing. It is probable that this difference represents in general the continuous amount of soluble nitrates used up by the plants. It is, however, also likely that had more nitrates been formed the plants would have used them and that the curves represent in a general way the minimum amount to which the corn and wheat roots can reduce the nitrates in this heavy soil. Traps ^a found that the nitrogen content of crops was increased with an increased nitrifying activity in the soil.

In figure 3 no close relation is evident between the weekly crop-increase curve and the decrease in the nitrates, using the fallow curve as a check.

RELATION OF NITRIFICATION TO FIELD FACTORS.

The nitrification in the soil depending, as it does, on so many factors—temperature, moisture, aeration, organic matter, kind of crop, cultivation, etc.—it is difficult in the field to run down the

^aTraps, G. S. Bulletin 106, Texas Agricultural Experiment Station, p. 4.

specific physical or chemical factors most influential in the promoting of the formation of nitrates in a given soil type. An attempt was made to correlate the variations in and the amounts of the nitrates found during the season with some of the factors known to influence their formation. As some of the most important ones from a chemical or bacteriological standpoint could not be taken up at the time the attempt at correlation had to be confined to a consideration of some of the physical factors. As, however, the soil of the different plats was similar and probably as nearly uniform as it is possible to obtain soil in different plats, and, furthermore, as the past history of the plats was exactly the same up to the time of beginning the work, it is not likely that there were any fundamental chemical or bacteriological differences in the plats at the time of planting. The wheat and corn plats were adjacent and were separated from the fallow plat by only 6 rods, and certainly no differences were apparent. Continuous air-temperature records were kept by means of a thermograph,

FIG. 5.—Diagram showing the average parts per million of water-soluble nitrates in dry soil in the fallow plat to a depth of 2 feet; also soil-moisture curve and precipitation curve.

but no evident relation could be obtained between the variations in amounts of nitrates and in temperature, whether the mean maximum, mean minimum, or general mean temperature was considered.

Figure 5 shows the curve of average amounts of nitrates to a depth of 2 feet in the fallow plat, the average soil-moisture curve to the same depth, and the precipitation curve. It is evident that neither the soil-moisture curve nor the precipitation curve shows any close agreement with the nitrate curve. The moisture curve is nearly straight, varying only from 24 to 29 per cent during the whole season. The other two curves parallel from June 19 to the end of the season, but as the formation of nitrates had already taken place and as the amounts thereafter remained constant, it is likely that this agreement is only incidental. It might be mentioned, incidentally, that the

mean temperature of the air did not reach nearly its maximum until some time later than June 19; in fact, the maximum temperature occurred in September.

In figure 6, which gives the corresponding data obtained on the wheat plat, there is a closer relation between the moisture and the nitrate curves, but as the growing crop has removed much of the moisture and the nitrates from the soil it is apparent that no close causal relationship could be insisted on in this case either. There is, on the other hand, a close agreement between the moisture curve and the weekly crop-increase curve. The hygroscopic coefficient for this soil is very high (about 12 per cent), and judging from the general appearance of the crop and of the soil at the time it is likely that the moisture curve from the first part of July represents about the minimum to which the wheat plants could reduce the moisture content of the soil, and the same holds true in regard to the nitrate

FIG. 6.—Diagram showing the average parts per million of water-soluble nitrates in dry soil to a depth of 2 feet in the spring-wheat plat; also soil-moisture, precipitation, and weekly crop-increase curves.

curve, as mentioned earlier. This same removal of moisture would, of course, also check nitrification, so both this and the plant feeding would keep the nitrates from increasing.

Figure 7 brings out more clearly than figures 1 and 2 the phenomenon of the general parallelism in the maximum nitrate accumulation in the individual soil layers in the wheat and fallow plats. This figure does not show the absolute amounts of nitrates found in the soil, but shows simply the period during which the respective 6-inch soil layers in the fallow and wheat plats contained an amount of nitrates greater than any other 6-inch layer. It will be noticed that there is a very close agreement between the two plats in the depth

at which nitrates were most actively accumulating at different periods and that other factors than cropping evidently determine the seasonal activity of nitrification.

THE DISAPPEARANCE OF NITRATES.

Glancing again at figures 1 and 2 it would naturally be asked what became of the water-soluble nitrates in the individual 6-inch soil layers after accumulation had reached a maximum, and why the seasonal shifting of the maximum amount of nitrates has such a distinct downward movement. It would be expected that the rainfall would tend to wash down the nitrates along with other salts, and such probably did take place to some extent. Selecting the week from May 9 to May 16, in figure 2, there would seem to have been

FIG. 7.—Diagram showing the time and location of the maximum amount of nitrates in the wheat and fallow plats.

such a movement from the surface layer to the 12-inch layer. The rainfall for that week was 1.25 inches. This is the only instance, however, in the two figures which shows the plausibility of such an explanation of the apparent downward "movement" of the nitrates. Inspecting figure 1, where no growing crop complicates the results, it would even appear that the rainfall had the opposite effect, as, for instance, during the period from May 22 to May 29. The effect seen there could not be explained by the washing down of the nitrates by the rains, as there was an increase in nitrates in all of the soil layers

except the 12-inch layer during that week. The same point is brought out during the period from June 5 to June 12, there being no rainfall at all to wash down the nitrates from the surface into the 12, 18, and 24 inch layers. And, finally, no such explanation can be given for the sudden drop in amounts of nitrates during the period from June 12 to June 19, when there was practically no rain.

DENITRIFICATION.

At the beginning of the work denitrification was expected to become an obvious factor in affecting the seasonal changes of nitrates in the soil, and ammonification was also expected to become of measurable importance; accordingly, determinations of nitrites and ammonia were made. Neither of these activities, however, assumed any importance at all during any part of the season so far as the analyses brought out, and, accordingly, toward the end of the season ammonia and nitrite determinations were not regularly made—sufficiently often, however (usually every other week), to know the amounts present of these compounds.

No conveniences were at hand for making ammonia determinations directly on the fresh soil, and this is obviously the best way of determining these compounds. The determination of the water-soluble ammonium compounds could not be expected to give reliable data. That ammonia may have been formed in larger amounts than would be suggested by the quantities found during the season would appear from the work of Stevens and Withers,^a who state that in some soils "nitrification proceeds as rapidly as ammonification, converting the ammonia to nitrate as fast as it is rendered available by the ammonifying organisms."

The amounts of nitrites found varied during the whole season from nothing to 1.8 parts per million in the dry soil. There was no regularity in even these small amounts, one week traces only or none at all being found and the next week traces to 1.8 parts per million. The same irregularities held true in the individual soil layers. The usual amounts of nitrites were from 0.1 to 0.3 part per million, and one single sample—from the 6-inch layer in the wheat plot, on May 22—had 6 parts per million, the only nitrite determination made that exceeded 1.8 parts per million at any time during the season.

The determinations likewise showed the presence of but small quantities of ammonia per million parts of dry soil, but in this case there was a slight decrease toward the end of the season. During the earlier part of the season, the amounts found averaged 6 to 8 parts

^a Stevens, F. L., Withers, W. A., et al. *Centralblatt für Bakteriologie, Parasitenkunde und Infektionskrankheiten*, pt. 2, 1909, p. 23; abstracted in *Experiment Station Record*, vol. 21, no. 2, 1909, pp. 118-119.

of ammonia per million and toward the end of the season the amounts fell to about 3.6 to 4.7 parts per million. No such gradual decrease in amounts of nitrites toward the end of the season occurred. The variations in the quantities of ammonia and of nitrites in the individual soil layers were apparently not then sufficient to account for the marked and general and consistent changes observed in the case of the nitrates.

It may be remarked that although the soil is heavy in texture and likely to compact under bad treatment, the soil on the plats was quite loose and open, it being the first year after breaking. Also, toward the middle of summer the soils cracked quite freely, so that there was fair opportunity for soil aeration all season, and, as the precipitation curve will show, there was no trouble with water-logging of the soil. All conditions considered, it is apparent that conditions were not extremely favorable for active denitrification.

SEASONAL VARIATIONS IN NITRIFICATION.

Referring again briefly to all of the figures, one is struck by the remarkable uniformity of direction of all the curves, especially in figures 1, 2, and 3. The facts that the same general variations exist in all cases and that denitrification is probably not responsible for the nitrate changes point to a general seasonal phenomenon which is affected in degree but not in kind by the presence of the growing crops.

It must be kept in mind that the nitrates found were those soluble in water and that the curves thus represent only the water-soluble nitrates. It would appear that nitrification began earliest in the surface layer of the soil and that the maximum activity of this process gradually extended downward into the successively lower 6-inch layers and reached its maximum in any one layer in general at a period of about two weeks later than in the next 6-inch layer above. In other words, conditions of temperature, aeration, etc., were favorable for nitrification in the surface layer earliest, as would of course be expected, and these favorable conditions obtained later in the lower soil layers. By June 12 these requisite conditions had in general reached their optimum throughout the 2-foot soil section studied, and it might well be conceived that the physiological activity of the bacterial flora necessary for nitrification reached its maximum in the comparatively short period of about two or three weeks in each soil layer after the conditions had once become most favorable and that the general "crop" of nitrifying bacteria matured by the middle of June.

Under this interpretation it would be inferred that the nitrifying bacteria had an active growing season, so to speak, of two to three

weeks after favorable climatic conditions had been established and that after this period of growth they underwent a resting period, or at least their physiological activity was much reduced, just as is the case with an annual flora of higher plants. From this explanation one would expect that their general activity would be more intense and begin earlier in the fallow plat than in the wheat plat, as the former was spring-plowed to a good depth, while the latter was not so treated—and this is the case. As already noted, the maximum nitrate content in the respective soil layers occurred in general about one week earlier in the fallow than in the wheat plat, but the general “maturity of the crop,” to continue the analogy of the higher flora, was effected at about the same time in the two instances, viz, about June 19. This seasonal decrease in nitrification may also be caused by a decrease in food readily available for the nitrifying organisms. Whether the seasonal variation in amounts of nitrates is due to periodicity of the activity of the nitrifying organisms in the soil, unaffected by any climatic or food factor, or whether it is due to lack of sufficient food for continued activity would be hard to decide from the various curves. As somewhat similar variations may be obtained in the laboratory under controlled food conditions, this factor must be kept in mind. Schneider^a found that the fixation of nitrogen in the soil was greatly increased by the addition of organic food, as also by the addition of potassium phosphate and of lime; and Bazarewski^b found that the addition of small amounts of organic food promoted nitrification, but that the addition of larger amounts decreased the activity of the organisms.

Notwithstanding the fact that neither nitrites nor ammonia were ever found in sufficient amounts to account for the actual decrease in amounts of nitrates, other organisms active in the production of these compounds may conceivably have increased vegetatively at the expense of the nitrates already present. Whether the food supply itself could be entirely responsible for the variations in the amounts of nitrates it would be hard to decide, but certainly the prairie soil in question is considered quite fertile so far as the ordinary farm crops are concerned, and the plats on which these determinations were made had never been cropped before.

That this rather definite and comparatively short seasonal period of activity of nitrification has not been limited by lack of moisture in the soil would seem to be indicated by the moisture conditions in the summer-fallow plat. The drying up of the soil in the wheat plat, due to the demands made upon the moisture by the plants, would of

^a Schneider, P. *Landwirtschaftliche Jahrbücher*, vol. 35, sup. 4, 1906, pp. 63-83; abstract in *Experiment Station Record*, vol. 18, no. 8, April, 1907, p. 722.

^b Bazarewski, L. von. *Nitrification and Denitrification in Soils*. Reviewed in *Experiment Station Record*, vol. 21, no. 1, July, 1909, pp. 20-21.

itself inhibit nitrification, just as excessive dryness would check the growth of any other plant.

It seems evident, therefore, that the decrease in amounts of nitrates can not be wholly explained by either moisture content of the soil, denitrification, ammonification, or temperature. A shortage in organic food supply may have been one of the principal causes, but it is also possible that the amounts of nitrates accumulated by the nitrifying organisms would inhibit further increase in these salts. Instances of such inhibition are not uncommon in the case of other bacteria and fungi. It is also within the province of possibility that the nitrifying organisms may have accumulated compounds as by-products other than nitrates and that these compounds might have checked any otherwise possible increase in amounts of nitrates. Even when all these factors are considered there would appear to be a rhythmic periodicity of nitrifying activity during the season due to some unknown property of the bacteria.

CHANGE OF WATER-SOLUBLE NITRATES TO ALBUMINOID NITROGEN.

The disappearance of the water-soluble nitrates does not by any means indicate that the nitrogen was actually lost from the soil, but that it was perhaps converted into albuminoid nitrogen by other groups of bacteria. It is quite conceivable that the food requirements might be sufficiently different for the two groups to permit a considerable decrease in the activity of one group and at the same time increase the activity of another group. It may very well be that the artificial application of a food supply suitable for use by the nitrifying organisms would again increase their activity to a point beyond that which they formerly had reached. It is likely that had water-insoluble nitrogen been determined in the same soil series, no general decrease would have been found in this from June 12 to June 19; on the contrary, there would probably have been an increase. In this connection it may be mentioned that Montanari^a found least amounts of "nitric nitrogen" in February, March, and June, and most in July and August. He found that plats growing wheat, rape, and corn contained more nitrates in the lower stratum (10 to 20 centimeters) than in the upper (0 to 10 centimeters); on a plat growing Medicago, however, this relationship was reversed. This accumulation of the (probably) albuminoid nitrogen would very possibly be of use to plants the following spring and would simply be held in reserve in the soil and either break down into readily water-soluble forms or in other ways become available for use by higher plants. Whitson,

^a Montanari, C. Quantity of Nitric Nitrogen in Soils Variously Cultivated. *Le Stazioni Sperimentali Agrari Italiane*, vol. 41, 1908; abstract in *Experiment Station Record*, vol. 20, no. 8, March, 1909.

Stoddart, and McLeod^a found that the difference in the amount of nitrogen between virgin land and cropped land was not enough to account for the crops removed, and they suggest that this fact is due to the removal of nitrogen by bacteria. Their work was carried on in humid regions, however, where leaching would affect the distribution of nitrogen in the soil.

In an area such as the prairie region represented, where the amount of rainfall is rather limited, there would be an accumulation in the fallow plat of both water-soluble and albuminoid nitrogen. The crop would have the advantage of the nutritive condition of the soil solution represented on July 31 in figure 1, as compared with the less favorable conditions obtaining on April 27 in figure 2.

It should be kept in mind that the plats on which the determinations were made were "dry farmed," and it is not intended strictly to apply to irrigated lands the conclusions derived from this work. Irrigation would in all probability cause disturbances in the bacterial processes in the soil, and so, to some extent at least, mask the seasonal activities of the bacterial flora by translocation of the nitrates and by intermittent wetting and drying. It is likely, however, that by properly controlling the work the same general phenomena would be observed.

RELATION OF NITRIFICATION TO SUMMER FALLOW.

As will be noticed from figure 3, the summer-fallow plat contained on July 31 about 35 parts per million more of water-soluble nitrates than did the wheat plat, which would present a more favorable nutritive condition than found in the cropped plat. In view of the fact, however, that the fundamental phenomena underlying nitrification are the same in both the cropped plats and the fallow plat, it would seem that summer fallow would be of doubtful economic value so far as the use of nitrates is concerned. The various curves distinctly show that nitrification is affected in degree but not in kind by the presence of a growing crop, and it is certainly likely, with this point in view, that nitrates were formed to as large an extent in the wheat plat as in the summer-fallow plat: interest on the investment would be lost from the fallow land while lying idle, when by suitable treatment of the soil use could as well be made of the products of the nitrifying organisms each year as to allow the land to lie idle. The bacterial groups active in producing soluble nitrates and albuminoid nitrogen, feeding largely on the organic matter in the soil, would of course reduce their food supply more rapidly in a system of fallowing than in a system of crop rotation, where organic matter is continually being added to the soil. Another serious objection to summer fal-

^a Whitson, A. R., Stoddart, W. C., and McLeod, A. F. Twenty-Third Annual Report, Agricultural Experiment Station of the University of Wisconsin, 1906, pp. 160-170.

lowing, aside from the one just mentioned, is the fact that favorable conditions for chemical oxidation of the organic matter obtain because of the more ready access of heat to the soil due to its exposed condition. So in an area such as is represented by this station where even so far as moisture is concerned fallowing is questionable as a system, the nitrifying flora would be served better by eliminating fallowing.

SUMMARY.

In the early part of spring the surface 6 inches of soil contained more water-soluble nitrates than the lower 6-inch layers, and the amount reached its maximum in the first layer in the first and second weeks in May.

From that time the amounts of water-soluble nitrates decreased in the surface layer, and at the same time the nitrates in the 12-inch layer increased rapidly for one to two weeks, when a decrease was in turn shown in that layer; similarly with the 18-inch and 24-inch layers.

Thus the successive 6-inch layers from the surface 6-inch layer downward, at periods of about two weeks apart, each contained more water-soluble nitrates than any other layer at the same date, and each layer after accumulating a maximum amount showed a decided decrease, which it never again made up.

An exception was the 24-inch layer, which, while it showed the same phenomenon as distinctly, did not at any time accumulate more nitrates than any other layer, but underwent the same general changes as the upper layers.

The respective soil layers in the fallow plat reached their individual maximum accumulation of nitrates about a week earlier than the corresponding layers in the wheat plat. From June 12 to June 19 all soil layers in both plats suffered a general decrease in amounts of water-soluble nitrates. This occurrence took place in the spring-wheat plat in the same order and in about the same magnitude as in the fallow plat.

No evidence could be brought out that the wheat-plant roots drew more heavily on the water-soluble nitrates in one 6-inch soil layer than in any other at any time. The general decrease of nitrates in all of the soil layers in the fallow plat masked any such reduction by the wheat if any occurred.

The corn plants at the end of their most active season of growth had reduced the amounts of water-soluble nitrates in the soil to the same degree of exhaustion as had the wheat plants in the corresponding period of their development. The roots of the corn plants probably used the nitrates in the surface soil layer first, then drew most heavily upon the 12-inch layer, and finally upon the 18 and 24 inch layers. This progressive downward feeding of the corn roots occupied

the time from June 19 to July 17; at the latter date the nitrates had probably been reduced by the corn roots to the lowest limit possible.

The wheat and corn plants reduced the nitrates to a constant amount—about 15 parts per million in the dry soil. The wheat removed the moisture also to a fairly constant degree, namely, about 15 per cent.

Denitrification, as measured by the nitrites found, could not account for the changes occurring in the nitrate content in the soil layers.

No correlation could be established between the amounts of nitrates and the air temperature or between the nitrates and the soil moisture.

Translocation of the nitrates by rain could not account for the seasonal changes in the amounts of nitrates in the different soil layers.

The general increase and decrease in water-soluble nitrates followed the same course in the fallow plat and in the cropped plats, the only general difference being that there were more nitrates in the fallow plat after May 16 than in the wheat plat.

It is suggested:

(1) That the marked increase in the amounts of water-soluble nitrates in the individual soil layers, followed by marked decrease in the same, and not at any time again reaching the same quantity, is due to a seasonal physiological activity of the bacterial flora, depending upon the climatic and nutritive factors in the soil, or else the seasonal changes are due to a rhythmic periodicity of activity in the nitrifying organisms during the season, the nature of which could not be made clear by the data obtained.

(2) That the general decrease in amounts of water-soluble nitrates from June 12 to June 19 may be due to the formation of albuminoid nitrogen by other groups of bacteria which may have become most active at this time; that the excessive accumulation of nitrates or the accumulation of other by-products by the nitrifying organisms may have inhibited further activity on their part.

(3) That, while the summer-fallow plat contained at the end of the season more water-soluble nitrates than did the corn and wheat plats, the same general phenomena of nitrification are found to take place in both fallow and cropped plats; hence the extended practice of summer-fallowing would seem to be of doubtful value so far as the accumulation of nitrates in the soils represented by this station is concerned.

NOTE.—Since this bulletin was put into type, a review of Kansas Station Bulletin 161, appearing in the Experiment Station Record, vol. 22, no. 1, p. 21, gives as one of the conclusions arrived at by the authors, that "bacterial life and activity seem to rise and fall with more or less regularity. These periods of maximum and minimum activity are to a certain extent independent of moisture and temperature, and are possibly due to the presence of bacterial by-products." This conclusion corresponds closely with the work herein described and shows that other bacterial groups besides the nitrifying flora have the periodicity mentioned in this bulletin.

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