

FERTILIZER TREATMENT AS AFFECTING NITRATE PRODUCTION.

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Nitrate production is one of the most important problems of soil fertility. The ability of a soil to produce sufficient amounts of nitrate nitrogen for desirable plant growth over and above the natural losses, as denitrification and leaching, depends primarily upon soil management and treatment.

The problem of the farmer is to know the methods that can be employed to furnish and maintain this element in available form most economically. Natural losses occur more readily than in the more stable compounds of potassium and phosphorus.

There are many factors entering into the production and utilization of this important plant food and it has been the purpose of this investigation to try to throw some light upon a few of them.

REVIEW OF PREVIOUS WORK.

It has been known for a long time that nitrates are formed from organic nitrogenous substances in the soil. Investigators were discussing the process as far back as the middle of the nineteenth century. At that time they considered it as a purely chemical process. The great chemist Liebig held this view and his support was probably the reason that the actual cause of nitrification was not discovered at an earlier period. Boussingault 1860 showed that the nitrogen of nitrate was not derived from the air.

It was demonstrated by Schloessing and Muntz 1878 that microorganisms in the soil oxidized ammonia to nitrate. His conclusions were drawn from the work he did on sewage disposal. Since this time many attempts were made to isolate the organism in pure culture and it was not until about 1890 that this was accomplished.

King and Whitson (2) found that nitrates were produced more rapidly in stirred soil due to better aeration.

Brown (4) concluded that media prepared from soil extracts permitted fewer organisms to develop than the modified synthetic agar. Fresh soil offers conditions as closely approximating field conditions as possible.

Lyon, Bizzell and Conn (5) state that a very definite relation exists between the crop yields and nitrate contents of the soil. Higher yielding plots show a larger accumulation of nitrates before planting than do the very low yielding plots. Evidently higher yields in these plots are associated with a more rapid formation of nitrates.

Brown (7) ran nitrification tests to find the nitrifying power of the soil. He treated the soils with dried blood and with ammonium sulphate. His tests show agreement to crop producing power of the soil, that is, the high nitrifying soils produced large crops.

Brown and Halversen (10) concluded that the number of molds present in the soils fluctuated from one sampling to the next but was

apparently unaffected by moisture, temperature or soil treatment. Some factors as yet uninvestigated probably account for the fluctuation. The small number of molds in soil compared with bacteria may not necessarily mean that they are less important and certainly will not prove that they are unimportant.

Greaves and Carter (12) found in their study of twenty-two soils that each one gave a maximum ammonification when its water content was sixty per cent of its water holding capacity. Nitrification was at its maximum at fifty or sixty per cent and varied with specific soils.

Whiting and Schoonover (13) conclude that soil treatment is a very important factor in nitrate production.

HISTORY OF THE PLOTS.

The field where this experiment was carried on is a part of the Purdue Experimental plots and is located on a brown silt loam underlaid with gravel at a depth of about two feet. It has been classified by the United States Department of Soils as a Sioux Silt Loam.

The field consists of thirteen one-sixteenth-acre plots. The first, fifth, ninth, and thirteenth plots are untreated or check plots and the other nine received the treatments shown in Table 1. The crop rotation of the field consists of corn, oats, wheat, clover and timothy. In 1920, the year this experiment was conducted, the field was in oats followed by fall sown wheat.

This field was laid out in 1889 and the different treatments were begun in 1890. A different system of treatment was used at first and it was not until 1918 that the present treatment was started, the field having received no treatment during 1917. The object of the change of treatment, which involved only the amount and method of application, was to secure more efficient use of the nitrogen applied.

TABLE 1
Series IV East—Field 6—Purdue Farm
Fertilizer Treatment in Pounds Per Acre

Plot No.	Corn	Oats	Wheat	Clover	Timothy	Treatment
1	None	None	None	None	None	Check
2	12,000	6,000	6,000	None	6,000	Horse Manure
3	12,000	6,000	6,000	None	6,000	Cattle Manure
4	N 30	N 15	N 15	None	N 15	N. P. K.
	P 30	P 15	P 15		P 15	
	K 30	K 15	K 15		K 15	
5	None	None	None	None	None	Check
6	N 30	N 15	N 15	None	N 15	N. P. —
	P 30	P 15	P 15		P 15	
7	P 30	P 15	P 15	None	P 15	
	K 30	K 15	K 15		K 15	— P. K.
8	N 30	N 15	N 15	None	N 15	N — K
	K 30	K 15	K 15		K 15	
9	None	None	None	None	None	Check
10	P 30	P 15	P 15	None	P 15	— P —
11	N 30	N 15	N 15	None	N 15	N —
12	K 30	K 15	K 15	None	K 15	— K
13	None	None	None	None	None	Check

Rotation Corn, Oats, Wheat, Clover and Timothy—

P=lbs. P₂O₅ per acre.
K=lbs. K₂O per acre.
N=lbs. N per acre.

Although there is now more total nitrogen applied to the manure plots than is applied to the plots receiving nitrogen in commercial form, the nitrogen in the manure must be converted into soluble nitrate while the commercial nitrogen is applied in the readily available form of nitrate of soda, so that the available nitrogen on these plots is probably comparable.

OBJECT OF THE INVESTIGATION.

Although a great many investigations have been conducted in a study of nitrification, few pertain to comparisons of the efficiency of different fertilizer treatments for nitrate production. The following points were deemed important in this study and they express the aim of this work.

1. The comparison of the amount of nitrate nitrogen produced in the same field but with different fertilizer treatment.
2. Correlation between amount of nitrate production and crop yield.
3. Correlation between nitrates found in the soil under natural conditions with growing crops and amount accumulating under optimum conditions.
4. Nitrifying power of a soil compared to crop growth and nitrate content in a fresh soil.
5. Comparison of the effect of soil treatments on bacteria and molds.

TECHNIC.

There was no effort made to discover or try out new methods in this work and the technic employed was adapted to the needs and conditions of this experiment from methods already in common use in soil nitrate and bacteriological studies.

The monthly sampling time varied from the twentieth to the end of the month, but all samples for each month were taken on the same day. The time chosen for taking samples was when all conditions were most favorable thereby lessening the possibility of denitrification occurring during the incubation period. Sampling was done with a soil auger, ten borings made to a depth of ten inches were taken from representative parts of a plot. Judgment was exercised in taking the samples to make them as representative as possible of the soil of the plots.

The samples were taken from the field to the laboratory and all work performed with the fresh samples was done immediately, thus not allowing time for any material bacterial action to take place before the tests were started.

The soil from each plot was used for the following five tests:

1. Fresh nitrates;
2. Nitrates after two weeks incubation;
3. Nitrates after two weeks incubation plus ammonium sulphate;
4. Plate count of bacteria and molds;
5. Moisture content of the fresh soil.

The colorimetric method employing the phenol-di-sulphonic acid color reaction, as modified by Noyes (11) was used in determining the

amount of nitrates present. Although the accuracy of this method has been severely criticized it is the one most widely used and most practical for this type of work where comparative rather than absolute results are sought.

A one hundred gram aliquot of soil was weighed into tumblers marked for the respective plots 1A to 13A. Each tumbler was covered with a petri plate lid and set away in a locker. After two weeks incubation nitrates were determined as before.

One hundred grams of each sample were placed in tumblers marked 1B to 13B and one cubic centimeter of a ten per cent solution of ammonium sulphate was dropped over the soil in each tumbler. They were covered and incubated two weeks then tested as in the case of the fresh nitrates. In the case of both incubated samples when too dry equal amounts of distilled water were added to each tumbler or if too wet the covers were left off of each tumbler for equal periods until of the proper moisture content.

Duplicate plates were made of dilutions 1 : 100,000 and 1 : 1,000,000 from each sample. A 1 : 10,000 dilution was also plated for a few of the tests but the colonies were too crowded to make the count accurate. Several different media recommended by soil bacteriologists were tried in an effort to determine which of them would give the best growth of bacteria and not encourage the spread of molds over the plates. The following synthetic agar media seemed most satisfactory and was used through the major part of the investigation:

- 5 grams of sodium potassium tartarate;
- .5 gram of di-basic potassium phosphate;
- 1 gram of peptone;
- .2 gram of magnesium sulphate;
- 15 grams of agar;
- 1 liter of distilled water.

The plates were incubated for one week at room temperature before counting colonies of bacteria and molds. However, in two instances low temperature in the room deterred growth so that they were incubated longer.

TABLE 2.
Moisture Percentage.

Plot No.	Mar.	Apr.	May	June	July	Aug.	Sept.	Oct.	Nov.	Total	Ave.
1	23 65	21 25	23 25	18 96	10 60	13 24	14 49	18 89	20 83	165 21	18 35
2	25 15	22 77	18 65	11 70	14 39	16 34	13 01	20 17	22 60	164 59	18 29
3	24 26	22 95	19 45	11 00	12 48	15 68	15 93	24 82	21 79	168 36	18 70
4	24 38	22 05	18 45	15 48	12 65	15 55	15 84	20 97	29 11	174 48	19 39
5	23 32	21 60	17 55	10 95	8 86	15 55	13 93	21 61	20 21	153 59	17 07
6	24 00	20 75	17 80	10 65	12 88	15 13	15 25	20 92	21 53	158 91	17 66
7	23 62	21 55	19 68	11 70	12 30	15 74	16 46	21 53	21 01	163 59	18 17
8	23 84	21 05	18 35	10 70	12 40	14 17	14 17	20 73	20 52	155 93	17 32
9	23 40	21 85	18 27	10 92	11 65	14 35	14 32	20 02	20 03	154 84	17 20
10	23 14	20 45	18 42	10 15	12 75	14 17	14 01	20 23	20 38	153 30	17 03
11	21 76	20 40	14 67	10 40	10 75	13 76	13 70	20 01	19 92	145 37	16 15
12	23 15	20 45	17 55	10 00	10 70	13 57	12 94	20 26	19 80	148 42	16 50
13	22 85	20 25	17 72	10 80	11 58	13 48	13 15	20 78	20 77	151 38	16 82
Total	306 52	277 37	239 81	153 41	153 90	190 77	187 20	270 94	278 50		226 43
Ave..	23 58	21 33	18 44	11 80	11 84	14 67	14 40	20 84	21 42	158 32	17 45

Ten gram samples were weighed into tarred crucibles and dried in an electric oven at a temperature of one hundred degrees centigrade for moisture determinations.

All calculations in the tables were based on moisture free soil. Nitrate parts per million were calculated on the average for the duplicates, however there was little variation in the duplicates. The bacteria and mold calculations were based on the average of the 1 : 100,000 dilution plates, except in a few cases where development was not normal. Calculations were then based on the 1 : 1,000,000 dilution plates.

MOISTURE PERCENTAGES (Table 2).

Moisture determinations were made in order to calculate the amount of nitrates produced on a dry soil basis. Although the moisture content of a soil probably does greatly influence nitrate production it was not primarily for the study of this factor that the moisture content of the samples was made in this experiment.

Table 2 shows that the range of moisture content between the plots in any one month is small, not over four per cent except in a few instances. Plots 1 and 4 were high in June causing a range of 8.96 per cent while the range for the remaining plots was less than two per cent. The highest moisture content occurred in March with a gradual decrease to June and July, which were nearly equal and lowest for the period.

The moisture content increased from August to the end of the period and the average for November was a little more than equal to April. But these figures cannot mean very much because this factor is largely dependent on the season and weather conditions at the particular time of sampling. The plot averages for the year showed a range of only 3.24 per cent. The average deviation from the average was only a .75 per cent. The moisture content of the soils of the different plots varied so little that it was probably a very small factor in causing the difference in the nitrate production of these plots.

TABLE 3

Molds

(Millions per gram of Dry Soil Calculated on a Dry Basis)

Plot No.	March	April	May	June	July	Sept.	Oct.	Nov.	Total	Ave.
1	1.57	1.40	.60	.12	.22	.17	.30	.44	4.84	.60
2	1.34	1.03	1.35	.45	.34	.28	.00	.32	5.13	.64
3	1.58	1.44	1.47	.33	.28	.23	.20	.25	5.80	.72
4	1.19	.83	.61	.41	.28	.29	.31	.21	4.16	.52
5	1.44	1.27	.36	.28	.11	.00	.32	.88	4.67	.58
6	.72	3.79	.55	.50	.34	.17	.69	.44	7.22	.90
7	1.31	1.02	.62	.28	.40	.12	.44	.82	5.02	.62
8	1.77	.89	.37	.61	.28	.40	.44	.37	5.15	.64
9	1.31	.31	.67	.28	.22	.11	.25	.25	3.42	.42
10	.58	1.88	.35	.27	.63	.11	.50	.56	4.90	.61
11	.26	.87	.52	.27	.44	.11	.81	.43	3.75	.46
12	.71	1.57	.54	1.22	.28	.11	.44	.62	4.50	.56
13	.32	1.69	.60	.78	.34	.11	.63	.79	5.28	.66
Total...	14.10	18.02	8.64	5.85	4.20	2.27	5.40	6.42	7.98
Ave.....	1.08	1.38	.66	.45	.32	.17	.41	.49	4.99	.62

MOLD COUNTS EXPRESSED IN MILLIONS PER GRAM OF DRY SOIL (Table 3).

The results of mold counts given in Table 3 show that the mold counts of all plots averaged highest in March and April, gradually decreasing for May, June and July. The lowest count was for September, growth being very low at that time. The averages for October and November about equalled the average count for June.

The range in the counts for the different plots was wide, varying, for March, from .26 for the nitrogen plot, No. 11, to 1.77 for the N K plot, No. 8.

However, the range was usually much less as the count in September was from .11 for several plots to .40 for the N K plot, No. 8. This plot had a rather constant count, never falling below .37. This was much above the average for the July counts. The high average of plot 6 may have been due to an error since the April count was 3.79 while in March the count was only .72 and in May .55.

Mold counts for manure plots Nos. 2 and 3 were consistently above the averages for the monthly tests until October and November when the counts were much lower than the averages for these months. The cow manure plot, No. 3, had the higher count for March, April and May. But the horse manure plot, No. 2, had a little higher count for the remainder of the months, except in October when the failure of any growth to appear lowered the average count of Plot 2 noticeably below Plot 3.

Check Plot No. 9 had the low average count of .42 for the period. The N K Plot No. 8, had an average count of .64, which is .22 above this check plot. The P Plot, No. 10, had a count of .61, which is .19 above the check. But Plot No. 11, having only nitrogen treatment, has an average count of .46 which is approximately equal to the count of the check plot.

The potash and phosphorus treatments appeared to increase mold growth while nitrogen treatment had but slight effect. The average counts for check plots Nos. 1, 5 and 13 were considered equal to or higher than all the chemically treated plots. It would seem that either the source of error was very great, due perhaps to the small number of molds grown, or the various chemical treatments influenced mold growth but little.

BACTERIA COUNTS EXPRESSED IN MILLIONS PER GRAM OF DRY SOIL
(Table 4).

Bacteria counts of all plots averaged high for March, April, and October, medium for June and July, low for May and September with November lowest of all.

The range of counts for March was from 2.61 for the N plot, No. 11, to 19.60 for the cow manure plot, No. 3. But the range for July was only from 2.64 for the complete fertilizer plot, No. 4, to 5.48 for the horse manure plot, No. 2.

The check plots Nos. 1, 5, 9, and 13 showed a lower average count for the period than the intervening treated plots. Check plot, No. 1,

TABLE 4.
Bacteria
(Millions per gram of Dry Soil Calculated on a Dry Basis)

Plot No.	March	April	May	June	July	Sept.	Oct.	Nov.	Total	Ave.
1	9.95	7.25	3.39	4.75	3.92	2.40	5.80	1.58	39.04	4.88
2	12.70	10.05	9.21	4.93	5.48	4.61	2.25	3.11	52.34	6.54
3	19.60	11.40	4.92	6.10	4.28	5.05	7.58	3.38	62.36	7.79
4	6.60	13.85	4.41	6.34	2.64	2.85	5.82	2.31	44.83	5.60
5	7.05	8.18	1.46	1.74	3.62	1.86	5.87	1.38	31.16	3.89
6	4.94	5.41	2.32	4.76	4.70	1.88	5.69	3.94	33.64	4.20
7	6.15	10.20	2.48	2.84	4.90	2.52	7.00	3.10	39.19	4.89
8	5.81	11.14	4.55	2.01	5.51	8.40	6.95	1.51	45.88	5.73
9	10.92	1.50	3.37	2.02	3.84	1.63	6.50	2.50	32.28	4.03
10	10.11	9.82	1.04	3.90	4.30	2.21	6.27	2.32	39.97	4.99
11	2.61	6.90	3.22	11.10	4.58	1.62	10.00	2.12	42.15	5.27
12	4.96	3.77	3.64	6.68	4.64	2.64	5.14	2.37	33.84	4.23
13	5.82	4.34	4.26	6.40	2.66	1.27	4.05	2.91	31.71	3.96
Total....	107.22	103.85	48.27	63.58	55.07	38.94	78.92	32.53	66.05
Ave.....	8.24	7.98	3.71	4.89	4.23	2.99	6.07	2.50	40.64	5.07

had an average count of 4.88 which was lower than the average count for plots Nos. 2, 3 or 4.

The manure plots Nos. 2 and 3 had the highest average counts. The count for the cow manure plot, No. 3, was higher, for March, April, June, September, October, and November, than the horse manure plot No. 2. The average count for the period was 1.25 greater.

Three of the four plots having nitrogen in their treatments had higher average counts for the period than plots receiving no nitrogen in their treatments. The N K plot No. 8, was high with 5.73, the complete fertilizer plot, No. 4, was next with 5.60 and the N plot, No. 11, was lower with 5.27. However, the N P plot, No. 6, shows a slightly lower count than the P plot, No. 10, and the P K plot, No. 7, but nearly equal to the average count of the K plot, No. 12.

The results seem to indicate that manure treatment causes greatest bacterial growth. The nitrogen in commercial fertilizer treatments usually encourage bacterial growth more than phosphorus or potassium. All treatments increased bacterial numbers over the no treatment plots.

FRESH NITRATE EXPRESSED IN PARTS PER MILLION PER GRAM OF DRY SOIL (Table 5).

The nitrate found in the fresh soil is not a real test of the amounts being produced. The amount of water present, due to physical condition of the soil, the amount lost by leaching, and the amount and rapidity of crop growth cause the nitrate content to vary unequally. The averages for the months indicate periods of consumption and excess production. The plot averages for the entire period probably indicate roughly the nitrate producing ability of the soil.

The amounts of soluble nitrates found to occur in the soils of the different plots at the monthly sampling times vary greatly. The plots average highest for March, medium for April, July, September, and November, low for October, May and August and lowest for June. The

TABLE 5
Fresh Nitrates
(Parts per million calculated on a Dry Basis)

Plot No.	Mar.	April	May	June	July	Aug.	Sept.	Oct.	Nov.	Total	Ave.
1	21 00	8 15	20 90	15 80	10 70	12 91	16 82	7 89	14 15	128 32	14 26
2	29 00	9 30	8 80	7 25	35 80	15 31	30 20	16 10	29 00	170 76	18 97
3	29 60	11 40	8 91	6 30	18 60	22 80	50 90	20 22	24 50	193 23	21 47
4	16 90	12 30	8 80	3 79	28 10	9 50	17 20	10 10	18 08	124 77	13 86
5	17 80	10 40	12 61	3 60	7 01	10 42	11 10	6 12	10 02	89 08	9 90
6	21 80	20 10	8 79	5 48	11 00	11 32	15 15	7 08	17 31	118 03	13 11
7	23 80	14 30	7 92	3 63	7 31	9 51	15 40	10 70	13 15	105 72	11 74
8	24 20	13 20	15 71	3 58	18 52	12 12	13 08	11 10	13 60	125 11	13 90
9	25 00	16 20	11 70	4 50	10 91	13 10	13 09	7 00	10 00	111 50	12 39
10	26 00	17 10	7 85	3 57	22 00	12 10	13 03	8 55	14 08	124 28	13 80
11	16 40	16 10	10 31	5 94	14 31	8 35	14 85	10 00	16 00	115 22	12 80
12	21 90	10 00	7 78	3 56	28 70	11 10	16 50	8 04	10 96	118 54	13 17
13	20 80	12 00	9 75	5 38	18 21	7 40	12 81	5 05	12 12	103 52	11 50
Total	294 20	170 55	139 83	75 38	281 37	155 94	230 13	127 45	203 07	181 87
Ave..	22 63	13 12	10 76	5 80	17 79	12 00	17 70	9 80	15 62	126 22	14 00

low average nitrate content for June was probably due to the rapid crop growth and dry weather occurring at that time. But the low average for October was probably influenced by heavy rains just preceding the taking of the samples.

Manure plots Nos. 2 and 3 were highest for March and November. This was probably due to the residual effect of the manure. In contrast with these plots the N plot, No. 11 was lowest for March and low in November; this plot was higher than the manure plots for April, May, and June, and showed the least variation for the period.

The average parts of nitrate for check plot, No. 1, was .26 higher than the average for all plots. The other three check plots Nos. 5, 9 and 13 were all low with an average for the three of 11.26 or 2.74 parts lower than the average for all plots, and .48 lower than the lowest treated plot, No. 7.

The three plots Nos. 4, 6, and 8 receiving N P K, N P, and N K, respectively, had an average for the period of 13.62. But the average of the plots Nos. 7, 10, and 12 which received no nitrogen in their treatments was 12.90 for the period. This was .72 less than the average for the plots receiving nitrogen in addition to these treatments.

It seems, from these results, that check plot, No. 1, was influenced by the treatment from the manure plots next to it. Manure treatment had a tendency to produce nitrates continuously throughout the period.

Any one of the treatments increased nitrate content over no treatment. Nitrogen combined with phosphorus or potassium gave a higher nitrate content than when nitrogen was used alone or when phosphorus and potassium were used without nitrogen.

NITRATES AFTER TWO WEEKS INCUBATION EXPRESSED IN PARTS PER MILLION PER GRAM OF DRY SOIL (Table 6).

This test was intended to show the amount of nitrates that would accumulate when the soil was placed under optimum conditions. It

TABLE 6.
Nitrates After Two Weeks Incubation.
(Parts per million calculated on a Dry Basis)

Plot No.	Mar.	April	May	June	July	Aug.	Sept.	Oct.	Nov.	Total	Ave.
1	21 00	30 60	48 01	35 51	75 00	17 50	11 20	9 86	16 15	264 83	29 42
2	25 80	31 00	47 21	29 02	157 00	31 40	20 20	14 05	27 90	383 58	42 62
3	30 80	29 10	47 60	27 91	157 00	13 50	44 60	12 75	39 95	493 21	44 80
4	25 40	31 80	29 42	35 03	74 10	19 01	13 31	10 10	27 04	265 21	29 47
5	27 10	26 60	40 81	14 41	57 00	40 00	13 00	12 25	16 05	247 22	27 47
6	21 00	28 20	33 00	17 90	75 00	12 37	15 15	10 10	19 36	231 78	25 75
7	21 00	26 60	31 91	14 51	68 51	19 00	12 50	10 20	22 25	226 48	25 19
8	23 10	36 60	32 00	26 92	81 40	7 46	9 34	12 15	20 08	249 05	27 67
9	23 00	36 40	29 31	18 01	65 40	18 60	9 35	10 00	17 00	227 07	25 23
10	16 70	23 20	31 42	17 83	73 40	14 95	13 21	12 05	19 05	220 81	24 53
11	16 40	24 00	31 01	17 85	51 00	14 88	14 85	12 00	24 95	206 94	22 99
12	20 80	21 10	45 82	14 21	35 80	14 85	14 50	10 05	16 92	194 05	21 56
13	20 80	20 00	35 00	14 30	58 00	12 96	9 22	12 16	20 20	202 64	22 51
Total Ave..	292 90 22 53	365 20 28 09	482 52 37 12	284 41 21 88	1,028.61 79 12	236 48 18 20	200 43 15 72	147.71 11.36	286 90 22 07	255 79	369 21 28 41

differed from the fresh nitrate test in that moisture content was controlled and there was no loss of nitrates from crop growth or leaching.

Table No. 4 shows that the nitrate content of the soils was high for May and July, medium for April, low for March, June and November and very low for August, September and October.

The manure plots, Nos. 2 and 3, were high for March, April, and November. The averages of these plots for the period were about equal and much above the average of all plots.

The three plots 4, 6, and 8 receiving nitrogen in addition to phosphorus or potassium or both had an average nitrate content of 27.63 for the period. The average nitrate content was 23.79 for plots 7, 10, and 12 which received the same treatments except the nitrogen was left out.

The average of the three check plots 5, 9, and 13, was 25.07 for the period. The check plots in this case were slightly higher in nitrate content than the plots receiving either phosphorus or potassium or both. This difference was not marked but it may have been caused by a greater lack of nitrogen in the treated soil due to a larger crop growth. However, the average of the untreated plots was also higher than the plot receiving only the nitrogen treatment. The average of the plots 7, 10, and 12 which had no nitrogen in their treatments was slightly higher than the average for the N plot No. 11.

The increase in nitrate content of the incubated samples over that of the fresh soil samples was proportionately much less when the fresh nitrates were low as in June and October. The average on both tests was highest for the period in July. The July increase in nitrate content after incubation was 350 per cent. But in October the increase with incubation was only 15.9 per cent. The increase for the high month of July was 62.33 parts but the increase for the low month of June was only 16.08 parts.

It then would seem in this instance that periods of low nitrate content may indicate times of low nitrifying power of a soil. The results

seem to show that the addition of nitrogen to the phosphorus and potassium treatments causes the higher nitrate content of those soils.

TABLE 7.
Nitrates After Two Weeks Incubation and Addition of $(\text{NH}_4)_2\text{SO}_4$
(Parts per million calculated on a Dry Basis)

Plot No.	Mar.	April	May	June	July	Aug.	Sept.	Oct.	Nov.	Total	Ave.
1	35 60	25 50	41 61	50 51	114 50	17 50	19 70	13 80	21 22	342 94	38 10
2	37 40	36 10	55 12	108 20	187 00	50 70	25 07	36 08	41 45	577 12	64 12
3	36 00	37 40	60 51	108 00	178 00	60 45	36 09	44 52	47 00	607 97	67 55
4	31 80	37 00	39 21	44 51	100 00	27 25	24 61	16 25	33 90	254 53	39 39
5	29 40	32 70	30 22	36 01	97 50	16 11	14 90	13 25	20 02	290 11	32 23
6	33 60	31 40	38 90	35 82	109 00	28 12	21 68	14 17	21 40	334 09	37 12
7	34 60	28 80	32 81	36 23	146 00	21 91	15 40	16 35	26 28	358 38	39 82
8	35 80	35 00	39 23	52 01	168 00	29 92	16 75	15 15	22 18	404 94	44 99
9	35 40	27 00	39 12	35 94	173 00	41 20	20 25	16 00	18 00	406 21	45 13
10	35 40	24 10	39 32	41 03	147 50	34 50	24 21	12 05	20 10	378 21	42 02
11	32 80	28 10	30 01	42 81	103 50	28 39	18 61	14 00	19 95	318 17	35 35
12	33 10	36 10	38 92	35 50	109 50	37 10	27 25	12 02	15 95	336 44	37 38
13	34 80	20 00	39 00	39 50	132 00	29 81	14 80	14 15	18 15	337 21	37 47
Total Ave.	445 70 34 28	422 50 32 50	523 98 40 30	666 07 51 23	1,747 40 134 41	422 96 32 53	279 32 21 48	237 79 18 28	325 60 25 04	390 05	560 67 43 23

NITRATES AFTER TWO WEEKS INCUBATION PLUS AMMONIUM SULPHATE. EXPRESSED IN PARTS PER MILLION PER GRAM OF DRY SOIL (Table 7).

This test was intended to show the efficiency of the different soils in changing a soluble nitrogen compound into nitrate nitrogen. Any lack of nitrogen was supplied and variations in the amounts of nitrates formed in the soils were dependent on their ability to change ammonia to nitrate. However, this ability cannot be directly attributed to the original soil treatments as their power may have been changed because of the influence of the nitrogen added.

The monthly averages of the nitrates for the plots in this test were high for May, June, and July, low for September, October, and November and medium for March, April, and August. The manure plots Nos. 2 and 3 were high throughout the period with a general average of 65.83. The cow manure plot, No. 3, was slightly higher, for the period, than the horse manure plot, No. 2. The most noticeable variation between these manure plots and the other soils in the study occurred in October and November when the manure plots were nearly twice as high as any other plot.

Variations among all other plots were small. The range for any month was usually less than fifteen parts per million. The range of averages for the period was from 32.23 for check plot No. 5 to 45.13 for check plot No. 9.

The average for the check plots Nos. 1, 5, 9 and 13, was 38.23 for the period. The average of the plots Nos. 4, 6, 8, and 11 receiving nitrogen in their treatments was 39.21 for the period. The average of plots Nos. 10, 12, and 7 receiving phosphorus, potassium, and phosphorus and potassium respectively, was 39.74. Although the average for the check plots was slightly lower than for the treated plots the

results as measured by this test seemed to show the nitrifying power of these soils to be very similar.

Comparing Tables 4 and 5 the results show that the nitrates in the ammonium sulphate treated samples were increased more for the fertilizer plots Nos. 7, 10, and 12 which received no nitrogen in their treatments, than were the nitrates for plots Nos. 4, 6, 8, and 11, which received nitrogen in their treatments. The average difference due to increase for the plots receiving no nitrogen in their treatments was 15.98. But the average difference for the plots receiving nitrogen in their treatments was only 12.74. The average difference due to increase was a little lower for the check plots, it being 12.12. The greatest increase occurred in the case of the manure plots which had an average difference of 22.12.

The results of this test seem to indicate that manure treated soil has the strongest nitrifying power because of the increased physiological efficiency of the bacteria. The check plot shows the lowest nitrifying power due to the lowered physiological efficiency of the bacteria. The treatment with phosphorus, potassium or both increased the nitrifying power of the soil. The nitrogen applied in the form of ammonium sulphate at the time this test was started made doubtful the effect of the original nitrogen treatments on the nitrifying power of the soils.

TABLE 8.
Table of Averages.

Plot No.	Per Cent Volatile Matter	Per Cent Moisture	Nitrate Parts Per Million on Dry Basis			Millions Per Gram of Dry Soil Calculated on Dry Basis	
			Fresh Soil	After 2 Weeks Incubation	After 2 Weeks Incubation Plus (NH ₄) ₂ SO ₄	Bacteria	Molds
1	6.10	16.13	14.26	29.42	38.10	4.880	.605
2	6.15	18.29	18.97	42.62	64.12	6.543	.641
3	5.91	18.70	21.47	44.80	67.55	7.795	.726
4	5.62	19.39	13.86	29.47	39.39	5.604	.521
5	5.75	17.07	9.90	27.47	32.23	3.895	.584
6	5.52	17.66	13.11	25.75	37.12	4.205	.903
7	5.90	18.17	11.74	25.19	39.82	4.899	.629
8	5.68	17.32	13.90	27.67	44.99	5.735	.645
9	5.75	17.20	12.39	25.23	45.13	4.035	.428
10	5.66	17.03	13.80	24.53	42.02	4.996	.613
11	5.66	16.15	12.80	22.99	35.35	5.270	.469
12	5.69	16.50	13.17	21.56	37.38	4.230	.563
13	5.86	16.82	11.50	22.51	37.47	3.964	.661

TABLE OF AVERAGES (Table 8).

Since it seemed that the plot averages for the period were a more accurate measure of the nitrate producing ability of the plots than figures for any one month, these plot averages for all the tests were brought together in Table 8. A study of this table as illustrated by Figure I shows that there is a direct correlation between bacterial activities and nitrate production. The nitrates were low in the fresh soil and correspondingly higher in the incubated soil. The highest nitrate production occurred in the ammonium sulphate treated soils. Bacteria and mold counts correlate closely, bacteria having the much higher count.

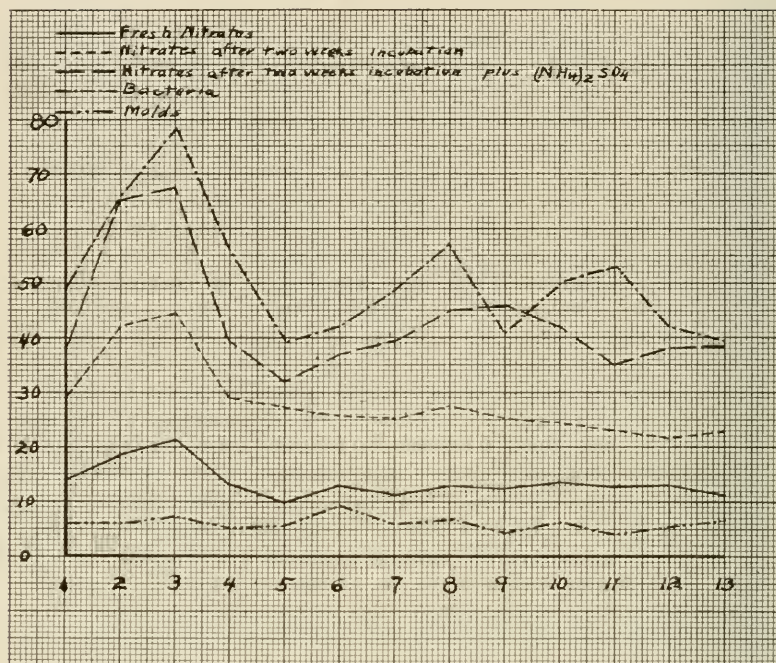


FIGURE I.

Correlation of Bacterial Activities and Nitrate Production.

CORRELATION OF BACTERIAL ACTIVITIES AND NITRATE PRODUCTION

(Figure I).

This graph is based on the figures in Table 8. It can be readily seen that there is a marked correlation between these five basic factors. One noticeable disagreement may be seen in the case of the treated incubation test on Plot No. 9, when the nitrates were comparatively higher than in the other tests. Another disagreement occurs due to high bacterial counts for plots Nos. 10 and 11. With the exception of a few other minor differences these curves follow each other very closely.

COMPARISON OF CROP PRODUCTION WITH AN EFFICIENCY FACTOR

(Figure II).

Any effort to compare the nitrate production and bacteriological efficiency of a soil with crop production makes it desirable that some common basis of comparison should be decided upon. For this purpose an efficiency factor for each of the tested plots was secured by adding together the parts per million of nitrates from the three tests with the mold and bacteria counts per million for each plot using the last five columns of figures in Table No. 8. The sums obtained for check plots Nos. 1 and 5 were added, divided by two, and the resulting figure taken

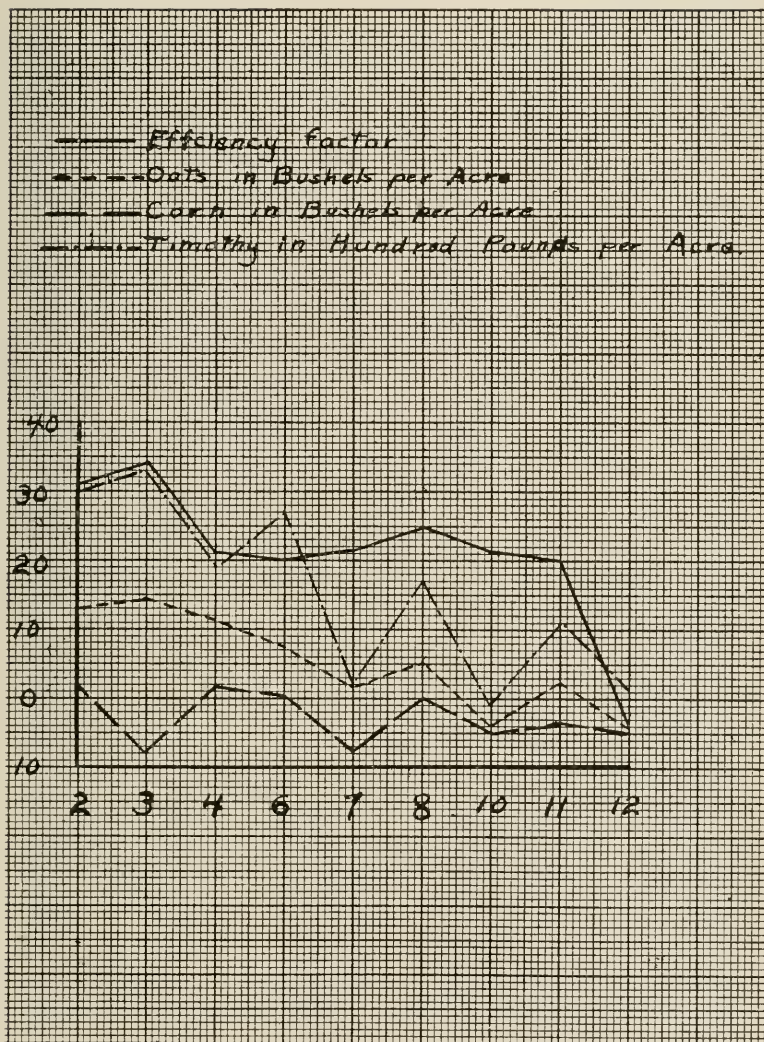


FIGURE II.
Comparison of Crop Production with an Efficiency Factor.

as 100. The intervening plots Nos. 2, 3, and 4 were then compared to this standard.

This efficiency factor was determined for all plots in a similar manner. The graphic crop yields based on a similar method of calculation were compared to the efficiency factor in Figure II. It is readily seen from this graph that there is a correlation of crop yield with biological activities and nitrate production. The closest correlation is shown by the oats and timothy yields. The yield of corn shows the

least correlation. Plot No. 3 has the highest efficiency factor and its corn yield was lowest of all the plots. Again in Plot No. 4 the efficiency factor goes down and the corn yield is highest of all the plot yields. However, for the remainder of the plots the yield and the efficiency factor show a close agreement.

SUMMARY.

A general study of the results of this experiment seem to show that the manure plots which were high in mold counts; highest in bacterial numbers; highest for fresh nitrates; equally high in incubated nitrates and very high in the ammonium sulphate treated samples, had the greatest efficiency for nitrate production. The cow manure treatment seemed to be somewhat more efficient than the horse manure treatment since the results of all tests were slightly higher in its favor.

Check plot No. 1, seemed to have been influenced by the manure treatment due to its nearness to those plots. The results from plot 1 usually were as high or higher than the average for all the plots and on the whole higher than the other check plots.

The use of nitrogen with phosphorus or potassium was superior to either of the treatments used alone for bacterial count and all nitrate tests except the ammonium sulphate treated samples where the difference was slight. Phosphorus and potassium treatments increased mold and bacteria growth, fresh nitrates and ammonium sulphate treated samples.

The results of ammonium sulphate treated samples which were least influenced by crop growth and seasonal variations seem to show that the greatest nitrifying power of a soil is in May, June and July. This power seems to decrease during the latter part of the summer and increase in the late fall and spring.

There seems to be a general correlation, when averages are taken for the entire season, between the amount of nitrate found in the soil under natural conditions with growing crops and the amount accumulating under optimum conditions.

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BIBLIOGRAPHY.

1. Gibbs, W. M. 1919
The Isolation and Study of Nitrifying Bacteria Soil. Science Vol. VIII, No. 6.
2. King, P. H. and Whitson, A. A. 1901
Development and Distribution of Nitrates and Other Soluble Salts in Cultivated Soils. Wisconsin Agr. Experiment Station Bulletin 85.
3. Voorhees, E. B. 1907
A review of Investigations in Soil Bacteriology. U. S. D. A. Bulletin 194.
4. Brown, P. E. 1913
Methods of Bacteriological Examination of Soil. Iowa State College Research Bulletin 11.
5. Lyon, T. L., Bizzell, J. A., and Conn, H. J. 1913
An examination of Some More Productive and Some Less Productive Sections of a Field. Bulletin 338, Cornell University.
6. McLean, H. C., and Wilson, G. W. 1914
Ammonification Studies With Soil Fungi. New Jersey Agricultural Exp. Sta. Bulletin 270.

7. Brown, P. E. 1915
Bacterial Activities and Crop Production. Research Bulletin 25, Iowa State College.
8. Hopkins, C. G., and Whiting, A. L. 1916
Soil Bacteria and Phosphates. Illinois Agr. Exp. Station Bulletin 190.
9. Noyes, H. A., and Voigt, E. 1917
A Technic for the Bacteriological Examination of Soils. Proceedings of Indiana Academy of Science.
10. Brown, P. E., and Halversen, W. V. 1919
Effect of Seasonal Condition and Soil Treatment on Bacteria and Molds in Soil. Iowa State College Research Bulletin 56.
11. Noyes, H. A. 1919
Journal of Industrial and Engineering Chemistry. Volume XI, No. 3.
12. Greaves, J. E., and Carter, E. G. 1920
Influence of Moisture on the Bacterial Activities of the Soil. Soil Science, November, 1920.
13. Whiting, A. L., and Schoonover, W. R. 1920
Nitrate Production in Field Soils in Illinois. Illinois Agricultural Experiment Station Bulletin 225.