MICROBIOLOGY OF AIR-DRIED CULTIVATED SOILS

THE MICROBIOLOGY OF AIR-DRIED, CULTIVATED SOILS.

By

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A Thesis

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G E N E R A L I N T R O D U C T I O N

The beneficial effect of the drying of soils upon their fertility has been a recognised fact in agricultural practice for many centuries. Russell, (16., p.p. 343-344), points out that this effect has been brought about, either by the exposure of the soil to the baking heat of the sun -- as in India and in Egypt, (Prescott (15)), or by deliberate burning of the soil. The increasing interest in soil science within recent years has stimulated investigation of this phenomenon from various viewpoints: evidence has accumulated to show that the beneficial effects of drying are due to changes in both the physical and the microbiological conditions of the soil. Lebediantzef (11) considers that

> 'the process of drying is a factor controlling to a large extent the fertility of the soil, and as such must play an important rôle in all processes of increasing the soil fertility.'

From the point of view of the microflora, it must be borne in mind that changes in the soil which are beneficial to bacterial activity are similarly beneficial to plants and that estimations of such activities, therefore, may be taken as indicative of the fertility of a soil with respect to the crops which it may yield.

HISTORICAL

Gedroiz (4) found increases in yields of oats grown on soils which had been dried for a number of years, the increase being relatively much greater in the unmanured than in the manured soil. Lebediantzef (11) also reports that 'the response of a soil to drying increases as the fertility decreases'. Klein (9) found, in pot-tests, that the increase of yield was proportional to the degree of desiccation. This fact was corroborated by Lebediantzef (11) who found that remoistening of partially-dried, cultivated, Russian soils causes decreases in yield, until the moisture content falls to, at most 6 per cent, after which marked increases in fertility occur.

The effect of drying upon the water-soluble, and colloidal, constituents of soils has been extensively studied by various workers, among whom may be mentioned Gustafson (6), Steenkamp (17) and König and others (10). Gustafson (6) who gives an extensive bibliography reports that oven- or air-drying increases the water-soluble constituents, but tends to decrease the nitrate content. Steenkamp (17) found that

> 'drying of a soil is a most powerful natural factor in helping in the transformation of plant nutritive substances from a potential to an active form; and the resulting

'increased fertility of the soil can hardly be dissociated from the improved physical condition due to flocculation of the soil colloids'.

König and his associates (10) found that the effect of drying was a partial destruction of the colloidal state and a consequent release of the adsorbed nutrients.

Klein (9) tested fertility increases on drying both by pot-tests with plants, and by estimating bacterial activity by means of carbon dioxide evolution and nitrate formation; he found that increases occurred in both of these estimations of bacterial activity. Further periods of drying showed further increases. Waksman and Starkey (20) report similar results with carbon dioxide evolution, together with increases in bacterial numbers; they do not report estimations of nitrification. Khalil (8) remoistened soil to 70% of its water-holding-capacity and concluded that the availability of added organic matter had been increased.

With regard to the effects of drying upon different layers of soil, Lebediantzef (11) is apparently the only worker who has studied this point, but as his work was done on layers of 0 to 20 cm. 20 to 40 cms., etc. it has little bearing on this present investigation. In concluding the reference to Lebediantzef's work, however, he reports that the

upper 5 cm. of soil is the most fertile due to its continual exposure to natural drying and wetting. Finally, he points out that drying of soil has effects upon fertility essentially similar to, though somewhat less pronounced than, those produced by volatile antiseptics, i.e., partial sterilization.

The sum of these investigations tends to show, therefore, that increases in periods of drying of soils increase the fertility resultant on remoistening and that the percentage increases of the fertility of dried, remoistened soils, are the greater, the lower the initial fertility.

One further point, with reference to previous work, lies in the lack of uniformity of the degree to which soils are remoistened after drying. Many workers give no definite figures while others express the value for moisture as percentage of soil mass. The wide range of values shown by the water-holding-capacities of various soils, emphasises the fact that the moistening of different soils to an equivalent percentage of their mass does not imply that the moisture available for crops is the same in each soil. In other words that one soil containing say 40% total moisture may be 100% saturated while another soil of an equal percentage moisture is only 50-60% saturated. Greaves and Carter (5) have

shown that the value 60% of the water-holding-capacity of a soil, which many investigators have proven to be the optimum for crop yield, is also the optimum for certain types of bacterial activity. This value has been used throughout in the studies of remoistened soils, with which this investigation was concerned.

OBJECT OF INVESTIGATION

The purpose of this study was to ascertain the effects, under controlled conditions, of moisture reaching soils which had received different manurial treatments, after they had been air-dried for varying lengths of time; with especial reference to changes in numbers of bacteria and ray-fungi, (Actinomycetes), carbon dioxide evolution and nitrate-nitrogen.

EXPERIMENTAL WORK AND DISCUSSION

SOILS USED

The five soils used for this investigation were obtained from the Central Experimental Farm, Ottawa, Ont. Their cultural history for the last eighteen years consisted of a four-year rotation of mangels, oats, clover hay and timothy hay. Each of the four sections of the field used for this rotation was subdivided on the basis of manurial treatment (see Fig. 1). The sections growing mangels were used for sampling; the data concerning the samples obtained are listed in TABLE 1. The letters M, N, X, Y and Z were those used by the Division of Field Crops at the Experimental Farm as distinguishing marks and they have been retained, for convenience, as laboratory indexes.

The method of sampling was as follows:- a trench was dug with a spade, the top inch of the adjoining soil removed and samples taken from the sides of the trench at 2"-5" and 6"-8" inclusively. Samples were taken in this manner at four different spots on each soil and thoroughly mixed to form a composite sample from each level. On arrival at the laboratory the samples were divided, one half being placed in air-tight bottles and stored in a cool dark place, the other half being allowed to dry in the air for 14 days; when thoroughly dry these units were kept in 'Kraft' paper-bags. At any one time of sampling, therefore, four units

FIGURE 1.

Showing relative position of soil samples in rotation field.

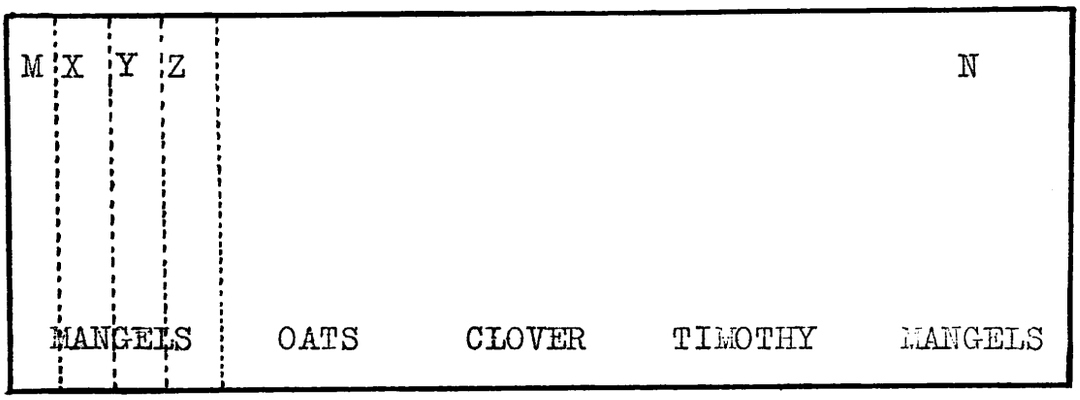


TABLE 1.

Laboratory Index.	Type of Soil.	Treatment per acre for the last 18 years.
Soil M	sandy clay	No manure or fertilizer (for 10 years only.)
Soil N	sandy loam	No manure or fertilizer
Soil X	" "	15 tons of manure every 4th. year of the rotation.
Soil Y	" "	no manure, 100 lbs. sodium nitrate)(applied 300 lbs. superphosphate)(every 75 lbs. muriate of potash)(1st.year. 100 lbs. sodium nitrate every 2nd., 3rd., and 4th. year.
Soil Z	clay loam	7.5 tons of manure per acre every 4th. year. 50 lbs. sodium nitrate)(150 lbs. superphosphate)(every 1st. 32.5 lbs. muriate of potash)(year. 100 lbs. sodium nitrate every 2nd., 3rd., and 4th. years.

were obtained from each of the five soils, i.e., composite samples from two levels, further divided into moist and air-dried units.

Samples of the soils were taken on the following dates:-

July 15, 1930
September 5, 1930
October 12, 1930

There were thus sixty units of soil on hand at the commencement of the experimental work, which was undertaken in the Bacteriology Department of Macdonald College, Que., under the supervision of Prof. P. H. H. Gray.

METHODS

The experimental work falls into three phases, namely, studies on the a.) moist units, b-1.) air-dried units and b-2.) remoistened units of the soils.

a.) Estimations were made of the total moisture, nitrate-nitrogen and numbers of bacteria and ray-fungi in moist units. The amount of carbon dioxide evolved during a period of from 300 to 500 hours was also determined.

b-1.) Determinations of the hygroscopic moisture and the water-holding-capacity were made on air-dried units.

b-2.) The studies on air-dried units, after remoistening to 60% of their water-holding-capacity, consisted of daily estimations of bacteria and ray-fungi for 5 days after treatment and of nitrate-nitrogen at intervals. The evolution of carbon-dioxide for a period of 14 days was also determined.

The methods employed for these studies are described in full below.

Methods Employed in the Studies on Moist and Air-dried Soils.

Hygroscopic moisture was determined by drying 10 gm. of air-dried soil, after passing it through a 3 m.m. sieve, at 105°C. for 24 hours; the loss of weight being expressed as a percentage of the air-dried soil. The

results of these determinations are given in TABLE 2.

Water-holding-capacity: 20 gm. of air-dried soil were placed in a small filter-funnel containing a Whatman No. 2 filter paper saturated with moisture. Distilled water was added, to duplicate samples, until the saturation point was reached, as evidenced by the exudation of moisture between the filter paper and the glass and its collection at the apex of the filter-cone. The average amount of water required was expressed as a percentage of the air-dried soil (See TABLE 3).

Total moisture was determined similarly to the hygroscopic moisture except that the soil was not sieved. The results were expressed as a percentage of moist soil and are shown in TABLE 4.

Microflora: The numbers of bacteria and ray-fungi were estimated by the plating method, using Thornton's asparagine-mannite-agar count medium (18).

The medium had the following composition in gm. per litre:-

K_2HPO_4	1.0
$MgSO4.7H_2O$	0.2
$CaCl_2$	0.1
NaCl	0.1
$FeCl_3$	0.02
KNO_3	0.5
Asparagine	0.5
Mannitol	1.0
Agar	15.0

pH adjusted to 7.4 before sterilisation.

The method of making dilutions was a modification of that described by Fisher and others (2), and may be outlined briefly thus:- 10 gm. of soil were placed in 100 c.c. of sterile physiological salt solution (NaCl-0.5%, $MgSO_4.7H_2O$-1.0%) and shaken for 4 minutes to obtain a suspension of the soil. 1 c.c. of this suspension was placed in 99 c.c. of sterile saline solution and shaken for 1 minute. 1 c.c. of this second dilution was placed in another 99 c.c. of saline and shaken for 1 minute. From the lowest dilution (1/100,000) five plates were seeded with 1 c.c. and 10 c.c. of count medium added.

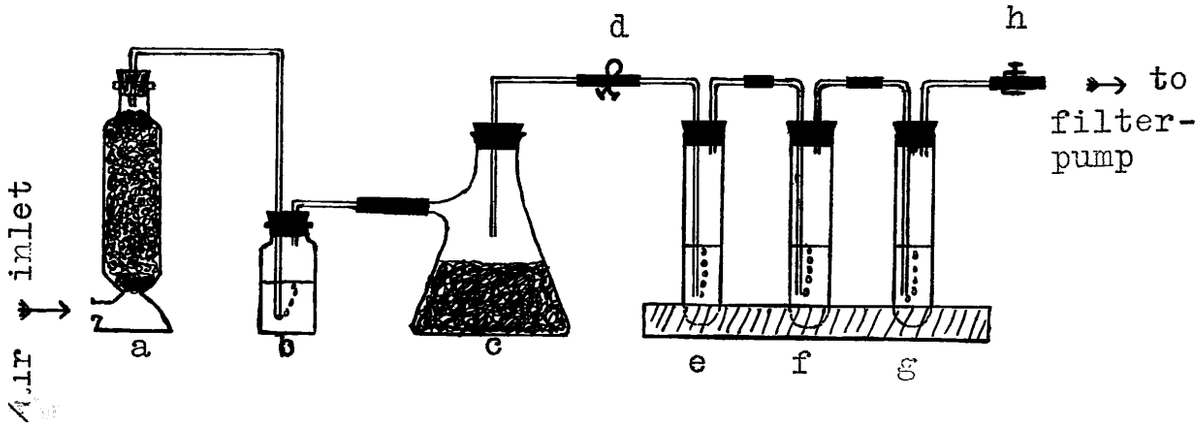
After all plates had been poured the liquid remaining in each dilution vessel was measured in a cylinder, due allowance being made for quantities transferred to plates and vessels. In this way inaccuracies in dilutions were avoided. The plates were incubated at 25°C. and the colonies counted after five days. The results were expressed in millions per gm. of oven-dried soil.

Nitrate-nitrogen was determined by Harper's Modification (7) of the phenol-disulphonic acid method, the results being expressed in parts per million of oven-dried soil.

Carbon dioxide evolution was determined by means of standard barium hydroxide. The method was that previously used by Potter and Snyder (14) and Neller (12) and described by Fred and Waksman (3). This is essentially the method ascribed to Pettenkofer (13) with the modification that the carbon dioxide-free air is drawn over the surface of, rather than through, the soil.
A current of air, freed from carbon dioxide, was drawn over the surface of 500 gm. of moist soil in a litre flask, into a series of 3 tubes, each

containing 33.3 c.c. of standard barium hydroxide solution (usually about 0.1 N.). The use of three tubes, rather than one, is in itself a slight modification, of the apparatus described by Fred and Waksman (3), which was found necessary for the complete absorption of carbon dioxide. The rate of aeration was approximately the same for each series of five samples. The amount of barium hydroxide remaining unchanged in the first tube was titrated at intervals with 0.2 N oxalic acid. The second and third tubes were titrated at the end of the experiment and a proportional amount of the carbon dioxide absorbed by them was added on to the values obtained from the first tubes. The results are expressed in mgm. of carbon dioxide per 100 gm. of oven-dried soil. Fig. 2, shows a diagram of the apparatus used: from time to time, tests were made of the efficiency of the absorption tower.

Figure 2.



Legend

a - soda-lime tower
b - barium hydroxide bottle
c - flask containing soil
d - spring clip
e) (tubes each containing
f)- (33.3 c.c. of barium
g) (hydroxide
h - screw-clip for regul-
-ating flow

Description of apparatus

By the use of "T"-tubes six suction-flasks, with their appropriate three test tubes, were set-up in one apparatus.

A small outlet tube containing soda-lime was also placed between the apparatus and the filter-pump in order to prevent back-suction when the pump was shut off.

Methods employed in the studies on remoistened soils.

Remoistening - each air-dried soil studied was remoistened to 60% of its water-holding-capacity. The soil was weighed into a large glass dish and the appropriate volume of distilled water added slowly, from a pipette, with careful mixing to ensure uniformity of wetting.

Microflora - after some preliminary experiments it was found that dilutions of 1/50,000 gm. and 1/100,000 gm. were the most satisfactory; four plates were poured from each of these dilutions, the first dilutions being made immediately after remoistening. Further tests were made daily for 5 days.

Nitrate-nitrogen in the remoistened soils was determined from the same series as was used for the estimation of the microflora. The initial test was made within a few hours of remoistening and further determinations made at intervals as shown in the tables.

Carbon dioxide: 400 gm. of air-dried soil were remoistened as described above; the sample was then placed in a litre suction flask and stoppered. When the five soils in a series had been treated, the flasks were connected to the carbon dioxide apparatus and estimations made as described previously.

TABLES OF RESULTS.

In the succeeding pages the results obtained from the moist and remoistened soils are given in the form of tables together with discussion.

TABLE 2.

Hygroscopic Moisture Expressed as Percentage of Air-Dried Soil.

Sample Depth	Collected	Soils M	N	X	Y	Z
2"-5"	October	2.26	1.53	4.96	4.83	1.69
	September	1.96	1.31	4.56	3.96	1.48
	July	1.41	1.07	4.40	4.36	1.55
6"-8"	October	2.03	1.69	4.83	4.98	1.49
	September	1.92	1.16	4.59	3.91	1.43
	July	1.45	0.87	3.54	4.92	1.47

TABLE 3.

Water-Holding-Capacity Expressed as Percentage of Air-Dried Soil.

Sample Depth	Collected	Soils M	N	X	Y	Z
2"-5"	October	38.0	37.5	67.5	58.0	36.0
6"-8"	October	35.0	36.5	51.0	54.0	40.0

TABLE 4.

Total Moisture Expressed as Percentage of Moist Soil

Sample Depth	Collected	Soils M	N	X	Y	Z
2"-5"	October	11.21	10.28	24.42	21.80	10.76
	September	10.07	10.51	26.86	27.46	9.52
	July	ø ..	15.26	31.50	31.07	13.30
6"-8"	October	9.60	10.74	20.87	18.78	8.52
	September	11.09	13.30	25.53	21.66	9.69
	July	ø ..	13.66	31.45	32.14	ø ..

ø Denotes samples lost in transit.

TABLE 5.

Total Moisture Expressed as a Percentage of Oven-dried Soil.

Sample Depth	Collected	Soils M	N	X	Y	Z
2"-5"	October	12.63	11.46	32.31	27.88	12.06
	September	11.19	11.71	36.73	37.86	10.52
	July	Ø	18.01	45.98	45.08	15.34
6"-8"	October	10.62	12.03	26.37	23.12	9.31
	September	12.47	15.21	34.28	27.65	10.73
	July	Ø	15.82	45.87	47.36	Ø

Total Moisture, after Remoistening to 60% of the Water-Holding-Capacity, -- Expressed as a Percentage of Oven-dried Soil.

Sample Depth	Collected	Soils M	N	X	Y	Z
2"-5"	October	25.63	24.40	47.80	41.64	23.68
	September	25.26	24.13	45.71	43.70	23.43
	July	24.57	23.82	46.97	40.95	23.53
6"-8"	October	22.26	24.01	37.24	39.18	25.87
	September	23.36	23.32	36.90	37.81	25.78
	July	24.60	21.14	48.11	54.51	21.28

ø Denotes samples lost in transit.

With regard to TABLE 3, it should be noted that the water-holding-capacity was determined on the October samples only. These determinations were used as the basis for the remoistening of all the other samples, since it was at first assumed that a physical property such as this would be unaltered in the same soil at different times of sampling. Further tests, however, showed that such was not the case and that there were definite - though slight - differences in the water-holding-capacity of the units from different sampling dates.

It may be pointed out, however, that the figure 60% of the water-holding-capacity is essentially an arbitrary one, the actual range of optimum moisture conditions being as wide as 55% to 65% for bacterial activity. It would appear, therefore, that any error which has been thus introduced into the remoistening of the July and September samples by this omission is not of sufficient significance to invalidate comparisons made between such samples and those of October sampling.

In order to institute a comparison between moisture contents of the same soil at different times, it is essential that the values be expressed on a comparable, constant basis. In TABLE 5 are listed the values for total moisture expressed as a percentage

of oven-dried soil, together with similar values for the same units remoistened to 60% of the water-holding-capacity.

It is interesting to note that these units, coming as they did from the same field, and hence having been exposed to the same climatic conditions, yet show significant differences in their respective degrees of saturation, both in the upper and the lower layers.

The July unit of the fertilized soil "Y" in the upper layer has a moisture content greater than the optimum, while that of the manured soil "X" is not significantly less than its optimum. All the units of soils "X" and "Y" are closer to their respective optima than are the unmanured soils "M" and "N", and the manured and fertilized soil "Z". The moisture contents of these last three soils in their natural condition are all in the region of 30% saturation.

Attention will be again drawn to this point in later discussion.

The first series of experiments were made with the fresh soil collected on the three dates previously mentioned. This work occupied the time from October to December. The air-dried soils were then remoistened and similar determinations made upon these; there was in

consequence an interval of three months between the commencement of the work on the moist soils as collected and the samples that had been dried. The results obtained from the moist and remoistened soils are shown together for each date of sampling.

Samples Collected on October 12, 1930

The data for all determinations made upon the samples collected at this time are shown in TABLE 6 below.

TABLE 6.

October Samples

a) CARBON DIOXIDE produced from soils, on the basis of mgm. per 100 gm. of oven-dried soil, for a period of 336 hours.

Soil Unit		Days Stored	Soils M	N	X	Y	Z
Moist	(2"-5")	10	27.74	20.32	61.83	53.87	25.63
Remoistened	do.	88	54.94	43.74	106.00	100.70	45.03
Moist	(6"-8")	16	19.35	13.07	33.01	40.68	18.07
Remoistened	do.	115	54.44	54.19	100.80	90.26	29.24

b) NITRATE-NITROGEN expressed as parts per million of oven-dried soil.

Moist	(2"-5")	3	8.33	5.80	7.08	6.11	3.90
Remoistened	do.	212	8.15	8.16	9.88	8.88	5.82
do.		(7)#	32.66	15.30	54.88	40.68	23.87
do.		(40)	80.41	20.32	132.10	113.10	56.98
Moist	(6"-8")	23	4.85	5.35	5.79	6.14	4.28
Remoistened	do.	218	7.81	7.54	9.71	7.70	6.47
do.		(7)	32.90	33.49	48.65	41.48	11.20

c) MICROFLORA: Bacteria and ray-fungi, in millions per gm. oven-dried soil.

Moist	(2"-5")	5	25.05	25.19	33.67	17.44	20.08
do.		25	12.77	10.86	17.74	15.24	23.34
Remoistened	do.	212	9.52	4.26	7.97	6.59	8.59
do.		(1)	4.62	3.43	6.52	6.11	9.99
do.		(2)	7.71	5.25	12.16	6.00	3.89
do.		(3)	4.31	6.11	22.04	5.23	5.85
do.		(4)	4.66	6.03	10.47	6.83	8.45
do.		(5)	8.28	7.43	7.66	6.16	5.62
Moist	(6"-8")	18	o....		9.50	8.27	10.87	7.84
do.		33	5.78	7.44	10.75	7.79	5.21
Remoistened	do.	218	4.65	1.94	3.43	2.52	5.89
do.		(1)	12.27	5.82	11.76	13.23	18.51
do.		(2)	14.26	14.83	28.77	8.22	29.85
do.		(3)	29.43	12.14	21.15	18.65	25.93
do.		(4)	9.69	13.10	112.10	26.70	46.99
do.		(5)	34.69	38.67	65.09	35.34	53.84

Figures in parenthesis denote days after remoistening.
o No value obtained due to contamination of plates.

From TABLE 6 there are several points to consider, with reference to soil depth, laboratory treatment and manurial treatment of the soils. One point which should be made clear with regard to this and the two similar Tables following is that the figure for days stored after the word 'remoistening' refers to the number of days in which those units were stored in the air-dried condition. The determinations were made, or started, within a few hours of remoistening, and the figures therefore serve as a basis of comparison both of the effects of drying and of subsequent changes on remoistening.

Carbon dioxide (See also Figs. 3-17)

Remoistening of the 2"-5" samples which had been dried and stored for 88 days considerably increased the amount of carbon dioxide evolved in 336 hours. In the case of the 6"-8" samples, remoistening has had a greater effect, except in the manured and fertilized soil "Z". A comparison between the values obtained for the two depths of different soils, shows that the greater amount of carbon dioxide is produced in the top layer of the moist soils, but that there is little difference, if any, between the two layers of the remoistened soils,

with the exception of the manured and fertilized soil "Z".

Nitrate-nitrogen

The amount of nitrate-nitrogen was low in both the upper and lower layers of the moist soil, at the time of sampling and after 23 days of storage, respectively. After 212 and 218 days storage in the dry state the nitrates had not increased appreciably. Within one week after remoistening, however, nitrification had proceeded rapidly, producing increases up to 6-fold the amount found on the day of remoistening; values equivalent to this rate of increase are shown for the upper layer of soil "X" (manure only) and the lower layer of soil "Y" (fertilizer only).

In the upper layer nitrification was least in soil "N" and in the lower layers, least in soil "Z".

Microflora

In the moist soil from the upper layer, after 5 days storage, the numbers of bacteria and ray-fungi ranged from 17,440,000 to 33,670,000 per gram; these numbers fell to between 10,860,000 and 23,340,000 after a further 20 days. The air-dried samples of these soils were remoistened after 212 days storage at which date

numbers ranged from 4,260,000 to 9,520,000. The effect of remoistening varied in the different soils. In no case did the numbers reach the original level and in most cases remoistening appears to have had no effect in 5 days, with the exception of the manured soil "X" in which a distinct rise in numbers occurred.

In the moist soils from the lower layer, after 18 and 33 days storage, numbers were appreciably lower than in the upper layer. Remoistening seems definitely to increase the microflora within 5 days. The numbers in the manured soil "X", and in the soil receiving manure and fertilizer "Y", reached a higher level than did those of the other soils.

Samples Collected on September 5, 1930

The data for determinations made upon these samples are given in TABLE 7 below.

TABLE 7

September Samples

a) CARBON DIOXIDE produced from soils, on the basis of mgm. per 100 gm. of oven-dried soil, for a period of 336 hours.

Soil Unit		Days Stored	Soils M	N	X	Y	Z
Moist	(2"-5")	64	28.02	25.02	74.46	70.08	30.31
Remoistened	do.	159	60.66	46.52	125.90	90.07	54.01
Moist	(6"-8")	81	21.85	12.05	58.38	52.46	23 20
Remoistened	do.	175	57.29	33.03	110.70	81.93	11.34

b) NITRATE-NITROGEN expressed as parts per million of oven-dried soil.

Moist	(2"-5")	28	16.31	5.31	16.63	7.71	6.76
Remoistened	do.	260	21.95	10.34	27.97	19.16	10.28
do.		(16)#	31.61	00.00	69.08	50.08	29.45
Moist	(6"-8")	41	5.67	7.84	19.32	9.34	2.03
Remoistened	do.	272	8.35	4.39	9.74	6.19	0.00
do.		(17)	35.26	00.00	60 87	46.54	0.00

c) MICROFLORA: Bacteria and ray-fungi in millions per gm. oven-dried soil.

Moist	(2"-5")	56	15.02	16.64	13.72	23.85	10.76
do.		82	8.57	1.63	11.24	8.83	7.11
Remoistened	do.	260	No count due to low dilution used.				
do.		(1)	23.40	15.86	43.44	24.67	32.85
do.		(2)	14.79	9.34	13.45	9.16	14.00
do.		(3)	23.49	13.32	45.75	20.79	20.92
do.		(4)	17.35	11.49	7.48	7.72	12.93
do.		(5)	15.82	12.13	7.96	11.08	13.55
Moist	(6"-8")	60	8.59	4.12	10.05	8.17	5.47
do.		95	6.12	5.02	8.07	6.16	8.57
Remoistened	do.	272	2.56	1.39	4.57	2.74	3.47
do.		(1)	21.10	10.77	22.44	19.16	11.69
do.		(2)	11.55	5.85	11.70	4.31	18.46
do.		(3)	9.15	16.18	19.82	13.65	5.72
do.		(4)	19.45	6.52	21.00	7.40	11.87
do.		(5)	10.21	10.97	11.53	6.05	14.64

Figures in parenthesis denote days after remoistening.

Carbon dioxide (See also Figs. 3-17)

Samples from the upper layer of these soils were stored in the moist state for 64 days. Soils "X" and "Y" produced the greatest amount of carbon dioxide, about two and one-half times that produced by the average of the other three soils. The effect of remoistening, in the case of the upper layer is to increase carbon dioxide in all cases, though the increase in soil "Y" was only of a small order. Remoistening the samples of the lower layer produced a greater increase in all except soil "Z", which showed a decrease.

Nitrate-nitrogen

Soils "M" (unmanured) and "X" (manured) both contain, in the upper layer, slightly more nitrates than the other three soils. Nitrates increased in all the soils during 260 days of storage. The effect of remoistening was to increase nitrates, within 16 days, in all soils except "N". No nitrates were found in "N", in either the upper or the lower layer, after remoistening.

Of the other samples taken at this date, from the lower layer, soil "Z" showed only 2.03 p.p.m. after storage for 41 days, no trace after drying and storing for 272 days, and none 17 days after remoistening. In

the case of the remaining three soils, nitrification increased after remoistening to a relatively greater extent in the lower layer than in the upper.

Microflora

The table shows that though there are appreciable differences between the numbers in the two layers after 56 to 60 days storage, continued storage reduced numbers to a narrower range of differences. Remoistening of the dried samples from the upper layer, after 260 days, caused increases in all cases except those of soils "N" and "Y". Numbers, however, fluctuated considerably during the 5 days, in any one soil. Numbers increased in all samples from the lower depth, but showed considerable fluctuation.

Samples Collected on July 17, 1930

The data for determinations made upon these samples are given in TABLE 8 below.

TABLE 8

July Samples

a) CARBON DIOXIDE produced from soils, on the basis of mgm. per 100 gm. of oven-dried soil, for a period of 336 hours.

Soil Unit		Days Stored	Soils M	N	X	Y	Z
Moist	(2"-5")	131	ø ..	31.15	202.60	199.30	58.33
Remoistened	do.	228	59.10	60.37	168.70	152.50	94.84
Moist	(6"-8")	172	ø ..	16.17	80.69	97.25	ø ..
Remoistened	do.	264	53.22	46.74	171.60	184.70	67.90

b) NITRATE-NITROGEN expressed as parts per million of oven-dried soil.

Moist	(2"-5")	138	ø ..	29.22	7.19	4.08	No test
Remoistened	do.	327	15.36	6.07	10.00	6.43	5.54
do.		(9)#	10.73	17.33	61.13	53.64	33.14
Moist	(6"-8")	259	ø ..	26.76	78.54	78.11	ø ..
Remoistened	do.	333	11.65	6.30	13.76	10.74	3.95
do.		(7)	00.00	12.73	32.15	34.27	31.13

c) MICROFLORA: Bacteria and ray-fungi, in millions per gm. oven-dried soil.

Moist	(2"-5")	110	ø ..	15.16	11.90	8.54	5.77
Remoistened	do.	327	15.69	6.44	15.82	3.54	10.73
do.		(1)	9.91	10.02	29.39	4.95	9.03
do.		(2)	16.22	12.95	28.15	8.89	27.10
do.		(3)	37.95	23.95	23.68	42.80	30.12
do.		(4)	18.91	7.63	21.25	5.72	13.54
do.		(5)	14.55	10.12	19.04	5.12	8.68
Moist	(6"-8")	112	ø ..	4.12	6.29	8.46	ø ..
Remoistened	do.	333	3.46	1.71	1.58	4.33	3.68
do.		(1)	4.45	13.16	6.06	5.68	4.86
do.		(2)	4.80	4.07	7.23	7.42	8.44
do.		(3)	6.16	8.38	10.64	6.71	6.67
do.		(4)	4.07	5.05	21.23	11.70	5.82
do.		(5)	13.00	13.98	7.93	16.06	6.24

Figures in parenthesis denote days after remoistening.
ø Samples lost in transit.

Carbon dioxide (See also Figs. 3-17)

The samples from the upper layer of these soils, which were stored in the moist state, were tested after 131 days and from the lower layer after 172 days. Of the four soils tested from the upper layer soils "X" and "Y" produced by far the greater amount of carbon dioxide. Remoistening resulted, however, in decreased amounts in these two soils and increases in "N" and "Z", respectively. In the lower layer soils "X" and "Y" produced more than "N" and remoistening caused further increases.

Nitrate-nitrogen

138 and 259 days elapsed between collecting the samples and the determination of the nitrates therein. The amounts appear to be low in the upper layer of soils "X" and "Y" but considerably higher in that of "N". Storage of the air-dried soils for 327 days appears to have reduced the nitrates to a similar low level in this latter soil, while the figures for soils "M" and "Z" show similar values. Remoistening caused considerable nitrification in soils "X", "Y" and "Z", but less so in "N". Nitrification did not occur in soil "M".

Storage of the samples from the lower layer resulted in considerable accumulation of nitrates in soils "X" and "Y". Nitrates were low in the air-dried, stored samples. Seven days after remoistening there was an increased amount of nitrate in all soils except "M", in which there was no trace of nitrate. The relative increase was greater in the samples from the upper layer, except in the case of the soil "Z", where the increase in the lower layer was the greater.

Microflora

The samples were stored moist for 110 and 112 days. Such differences as are shown in TABLE 8 may not be significant in either layer. Storage in the dry state appears likewise to have had little influence, although the numbers given by the lower layer of soils "N" and "X" suggest significant differences.

The effect of remoistening the samples from the upper layer has been to cause an increase in numbers in each soil, at the second or third day after treatment. A reduction in numbers occurred in all soils after the third day. Increases occurred in the soils from the lower depth, with the exception of soil "Z", generally at the third or fourth day. Additionally there was an

increase of nearly 8-fold in soil "N", 24 hours after re-moistening. There is not much evidence of wide fluctuations of numbers in each soil.

COMPARATIVE EFFECTS OF STORAGE

In comparing the results obtained from the soils sampled at the different periods, it must be borne in mind that the length of time of storage of the moist soils, prior to testing, might mask the effects of remoistening the soils which had been dried for increasing lengths of time. In other words, if circumstances had allowed, the tests on the moist soil units should have been made immediately after collection. Such comparisons as have been made between remoistened and moist soil units have referred only to the moist units of the respective sampling times. It was not the intention of this investigation to study the effects of storage of moist soils, but some reference to such effects has been found necessary.

As an illustration of the point referred to above, reference to TABLES 9 and 10 shows that there is relatively little increase of carbon dioxide evolution with increased periods of drying, when the values are compared on the basis of their respective moist units.

In TABLE 11, however, the percentage increase after remoistening has been expressed for each soil unit on the basis of the amount of carbon dioxide produced by the moist unit from the October sampling. In this

way the masking effect brought about by the accumulation of carbon dioxide during the storage of the moist units, has been eliminated. The values obtained are discussed after each of the Tables referred to above.

Carbon dioxide (See also Figs. 3--17)

TABLE 9

Carbon dioxide from moist soils stored.

2" - 5"

Collected	Days Stored	Soils M	N	X	Y	Z
October	14	27.74	20.32	61.83	53.87	25.63
September	64	28.02	25.02	74.46	70.08	30.31
July	131	ø ..	31.15	202.60	197.50	58.33

6" - 8"

October	16	19.35	13.07	33.01	40.68	18.07
September	81	21.85	12.05	58.38	52.46	23.20
July	172	ø ..	16.17	80.69	86.14	ø ..

ø Samples lost in transit.

In TABLE 9 are shown the values obtained for carbon dioxide from the five soils. Storage of the units in the moist state resulted, in the case of the upper layer, in a slight increase in the amount of carbon dioxide evolved in 64 days as compared with 10 days; similar increases occurred in the lower layer soils after 16 days except that there was none in the case of soil "N" and a rather larger increase in the case of soil "X". Continued storage up to 131 and 172 days resulted in much higher values except in the case of soil "N".

TABLE 10

Carbon dioxide from dried, stored and remoistened soils.

2" - 5"

Collected	Days stored before remoistening	Soils M	N	X	Y	Z
October	88	54.94	43.74	106.00	100.70	45.03
September	159	60.66	46.52	125.90	90.07	54.01
July	228	59.10	60.37	168.40	152.10	94.88

6" - 8"

October	115	54.44	54.19	100.80	90.26	29.24
September	175	57.32	33.03	110.70	81.93	11.34
July	264	53.31	47.08	176.20	184.70	67.92

From TABLE 10 it will be seen that the amount of carbon dioxide evolved from the remoistened soil units of the upper layer increases with length of time of storage, with the exception of soil "M".

In the case of the units from the lower layer the increases occur only in soils "X", "Y" and "Z".

It is interesting to note that the amount of carbon dioxide evolved from each remoistened soil unit is greater than that of its corresponding moist unit, with the exception of the July units of the upper layers of soils "X" and "Y". It will be remembered that reference has already been made to the fact that these

two soil units contained their optimum moisture content when stored, and that presumably the accumulated carbon dioxide after 131 days storage interferes with a valid comparison between these moist and remoistened units. A similar decrease has taken place in the September unit of soil "Z" in the lower layer, for which no explanation can be given.

TABLE 11

Increase of carbon dioxide evolution, due to air-drying, expressed as a percentage of the amount produced by each soil in the October sampling.

2" - 5"

Collected	Days Stored Dry	Soils M	N	X	Y	Z
October	88	99.13	115.30	71.44	86.92	75.71
September	159	118.70	128.90	103.70	69.04	110.80
July	228	113.10	197.10	172.40	182.30	270.20

6" - 8"

October	115	181.30	314.50	205.40	121.19	61.80
September	175	196.20	152.80	235.40	101.40	(36.92)#
July	264	175.50	260.20	433.80	354.40	275.80

The September unit of soil "Z" in the lower unit yielded a decreased amount of carbon dioxide from that produced by the moist, October unit in this layer.

It will be seen from TABLE 11, that although a short period of drying, 88 and 115 days, causes the greatest percentage increase in carbon dioxide production in the unmanured soils "M" and "N", further periods of drying have little effect upon these two soils. In both layers, however, the most marked increase with increased length of drying is shown by the manured and fertilized soil "Z", while soils "X" and "Y" also show considerable increases over "M" and "N".

Nitrate-nitrogen

TABLE 12

Nitrate-nitrogen in moist, stored soils.

2" - 5"

Collected	Days Stored	Soils M	N	X	Y	Z
October	3	8.33	5.80	7.08	6.11	3.90
September	28	16.31	5.31	16.63	7.71	6.76
July	138	ø ..	29.22	7.19	4.08	No test

6" - 8"

October	23	4.85	5.35	5.79	6.14	4.28
September	41	5.67	7.84	19.32	9.34	2.03
July	259	ø ..	26.76	78.54	78.11	ø ..

ø Samples lost in transit.

From a study of TABLE 12 it is to be noted that, of the samples taken from the upper layer, nitrification on storage occurred only in soil "N"; while for the lower layer, nitrification occurred in this soil to about the same extent and very actively in soils "X" and "Y".

TABLE 13

Nitrate-nitrogen in dried, stored and remoistened soils.

2" - 5"

Collected	Days Stored	Days Stored Remoistened	Soils M	N	X	Y	Z
October	212	0	8.15	8.16	9.88	8.88	5.82
		7	32.66	15.30	54.88	40.68	23.87
September	260	0	21.95	10.34	27.97	19.16	10.28
		16	31.61	0.00	69.08	50.08	29.45
July	327	0	15.36	6.07	10.00	6.43	5.54
		9	10.73	17.33	61.13	53.64	33.14

6" - 8"

October	218	0	7.81	7.54	9.71	7.70	6.47
		7	32.90	33.49	48.65	41.48	11.20
September	272	0	8.35	4.39	9.74	6.19	0.00
		17	35.26	0.00	60.87	46.45	0.00
July	333	0	11.65	6.30	13.76	10.74	3.95
		7	0.00	12.73	32.15	34.27	31.13

Storage of these soils in the dry state has not resulted in any appreciable changes in their original nitrate content. Nitrification occurred rapidly in most soils after remoistening, as has been discussed previously.

Microflora

The effects of storage upon the number of bacteria and ray-fungi together, in these soils, have been previously mentioned. A comparison can also be made between the number of bacteria and ray-fungi in their relation to one another. This can be done by comparing the number of ray-fungi as given in TABLES 14, 15 and 16 below, with the total counts as shown in TABLES 6, 7 and 8 (p.p. 23, 28 and 32).

TABLE 14

October Samples

Ray-fungi expressed in millions per gm. of oven-dried soil.

Soil Unit		Days Stored	Soils M	N	X	Y	Z
Moist	(2"-5")	5	4.92	5.02	5.24	5.15	5.39
do.		25	4.01	4.52	5.65	5.17	7.27
Remoistened	do.	212	2.96	2.13	3.53	3.42	3.73
do.		(1)#	2.23	1.33	2.78	2.44	2.35
do.		(2)	1.83	1.54	2.07	2.29	1.16
do.		(3)	1.69	2.72	3.46	1.86	1.70
do.		(4)	1.95	1.70	2.96	2.64	2.28
do.		(5)	3.31	1.58	2.63	2.24	1.56
Moist	(6"-8")	18	0...	2.15	2.53	2.13	1.83
do.		33	2.17	2.99	4.55	2.77	1.91
Remoistened	do.	218	1.95	1.13	1.04	1.46	2.83
do.		(1)	3.81	2.26	3.07	3.80	3.40
do.		(2)	4.21	2.30	4.29	2.10	3.99
do.		(3)	12.20	1.67	2.30	2.84	3.48
do.		(4)	2.32	1.77	12.28	3.56	4.49
do.		(5)	5.07	4.93	5.49	5.72	8.91

Figures in parenthesis denote days after remoistening.
0 No value obtained due to contamination of plates.

TABLE 15

September Samples

Ray-fungi expressed in millions per gm. of oven-dried soil.

Soil Unit		Days Stored	Soils M	N	X	Y	Z
Moist	(2"-5")	50	5.14	4.94	5.21	6.13	4.36
do.		82	3.86	0.36	4.74	3.83	3.11
Remoistened	do.	260	No count due to low dilution used.				
do.		(1)#	5.74	3.10	7.13	5.66	6.10
do.		(2)	4.22	2.09	1.34	3.26	3.37
do.		(3)	4.89	2.44	6.67	4.11	3.64
do.		(4)	3.90	3.18	2.45	2.90	3.04
do.		(5)	3.66	2.30	2.29	2.56	3.11
Moist	(6"-8")	60	2.80	1.35	3.41	3.03	1.50
do.		95	2.63	2.03	3.31	2.40	3.39
Remoistened	do.	272	1.30	0.71	1.89	1.40	1.22
do.		(1)	5.49	1.55	3.71	3.68	2.42
do.		(2)	3.24	7.85	2.07	1.24	2.83
do.		(3)	1.63	1.17	2.28	2.16	1.22
do.		(4)	3.90	0.84	3.95	1.49	1.87
do.		(5)	1.92	0.97	2.09	0.91	1.92

Figures in parenthesis denote days after remoistening.

TABLE 16

July Samples

Ray-fungi expressed in millions per gm. of oven-dried soil.

Soil Unit	Days Stored	Soils M	N	X	Y	Z
Moist (2"-5")	110	ø..	3.53	3.51	2.45	2.35
Remoistened do.	327	2.93	1.26	3.65	0.82	3.06
do.	(1)#	2.03	1.68	2.50	0.49	2.18
do.	(2)	2.72	1.75	2.27	0.87	3.56
do.	(3)	2.64	1.25	1.39	1.39	2.13
do.	(4)	2.87	1.60	2.75	1.14	2.25
do.	(5)	2.75	1.52	2.63	0.96	1.91
Moist (6"-8")	112	ø..	1.34	2.35	2.75	ø..
Remoistened do.	333	0.90	0.48	0.47	1.36	1.37
do.	(1)	1.22	0.63	0.66	0.74	0.70
do.	(2)	1.59	0.29	1.14	1.18	1.31
do.	(3)	1.45	0.78	2.16	1.42	1.74
do.	(4)	0.47	0.51	2.04	1.66	1.15
do.	(5)	2.42	1.39	1.13	2.65	1.71

\# Figures in parenthesis denote days after remoistening.
ø Denotes samples lost in transit.

From TABLES 14, 15 and 16 it will be seen that the numbers of ray-fungi found in the upper layers of the soils collected in September and October appear to fall after drying and remain low after remoistening. In the remaining series, numbers appear to fluctuate very little.

Attention might be drawn here to the work of Waksman and Starkey (21) on partial sterilization. These workers report that ray-fungi are diminished only slightly in numbers by treatment with volatile antiseptics, as opposed to the bacteria, and that they are essentially a stable group of microorganisms. As reference has already been made to the similarity in effects, of air-drying and partial sterilization, it would appear that the essential stability in numbers, of ray-fungi, found in this investigation, conforms with expectation.

The significance of fluctuations in numbers of microorganisms in soils at short intervals of time has so far been demonstrated (19) only for the bacterial flora in field soil. No work has yet been done on similar significances for other groups of soil microorganisms.

It was thought advisable, therefore, as a check on the plating technique, to determine the

significance of the mean plate-counts of both bacteria and ray-fungi.

The statistic used was that usually referred to as 'Chi squared' (χ^2) and described by Fisher (1., p.p. 57-61); it is represented by the equation

$$\chi^2 = \frac{S(x-\bar{x})^2}{\bar{x}}$$

where $\bar{x}$ equals the mean of the plate counts, and $S(x-\bar{x})^2$ signifies the sum, of the squares of the differences from the mean. It is known that if the distribution of colonies on the plates obeys the recognized laws of random distribution then the values for χ^2 will fall within certain limits.

Following the example given by Fisher (1., p.59) the following TABLES (17 and 18) have been drawn up to demonstrate the reliability of the means for bacteria and ray-fungi.

TABLE 17

Significance of the Means of Plate-counts on Moist Soils.
(5 Plates)

χ^2	Expected		Bacteria Observed		Ray-fungi Observed	
0						
	0.38)		0)		1)	
0.247	)	0.76	)	1	)	1
	0.38)		1)		0)	
0.429						
	1.14)		0)		1)	
0.711	)	3.04	)	6	)	3
	1.90)		6)		2)	
1.064						
	3.80)		2)		9)	
1.649	)	7.40	)	4	)	15
	3.80)		2)		6)	
2.149						
	7.60)		4)		7)	
3.357	)	14.40	)	7	)	10
	7.60)		3)		3)	
4.878						
	3.80)		5)		4)	
5.989	)	7.40	)	8	)	8
	3.80)		3)		4)	
7.779						
	1.90)		1)		0)	
9.488	)	3.04	)	6	)	1
	1.14)		5)		1)	
11.668						
	0.38)		2)		0)	
13.277	)	0.76	)	6	)	0
	0.38)		4)		0)	
Total	38.0		38		38	

TABLE 18

Significance of the Means of Plate-counts on Re-moistened Soils.

(4 Plates)

χ^2	Expected	Bacteria Observed	Ray-fungi Observed
0	1.55	1	1
0.115	1.55	0	1
0.185	4.65	1	5
0.352	7.75	0	10
0.584	15.50	7	19
1.005	15.50	4	12
1.424	31.00	12	32
2.366	31.00	17	35
3.665	15.50	14	11
4.642	15.50	13	8
6.251	7.75	12	6
7.815	4.65	13	10
9.837	1.55	13	2
11.341	1.55	48	3
Total	155.00	155	155

From a study of TABLE 17 it is apparent that the distribution of the bacteria on the plates from moist soils, conforms fairly well with expectation. The tendency towards excessive variation from the mean, and to a lesser extent towards subnormal variation, can be ascribed to slight variations in the technique of diluting and plating. As these counts on moist soils were the first made, using a technique new to the investigator, such variations might be expected. The distribution of the ray-fungi conforms with expectation better than that of the bacteria.

In TABLE 18, the most significant point lies in the utter lack of conformity, with expectation, of the bacterial distribution. Fisher, and others (2) in their statistical study of Thornton's agar medium with soil samples, refer to certain organisms which when growing on a plate, 'exert an inhibitory influence on the development of colonies by other forms'. It is interesting to note that such an organism as they describe occurred in soil which had been treated with napthalene - an antiseptic. The effect of this organism was to cause excessive variance between parallel plates. In their conclusion, they state that 'any significant departure from the theoretical distribution is a sign that the mean may be wholly unreliable'. In view of these

statements, therefore, it would appear that no significance can be attached to the total counts, on the remoistened soils, in so far as the bacteria are concerned, since the variance between parallel plates of these soils is abnormally excessive. From further reference to TABLE 18, it would appear that the ray-fungi show an almost perfect agreement with expectation and that therefore the mean count of each set of parallel plates is a direct measure of the density of their population.

S U M M A R Y A N D C O N C L U S I O N S

Investigations have been made upon the microbiological activity in samples, taken from the two levels of the surface 8 inches, of soils having different manurial treatments and of varying degrees of fertility, after different periods of drying. The work included (a) determinations of microbiological activity as indicated by the evolution of carbon dioxide; (b) estimations of the number of bacteria and ray-fungi; and (c) determinations of the activity of nitrifying bacteria as indicated by the production of nitrate-nitrogen. Physical determinations were also made, viz., total moisture percentage, hygroscopic moisture percentage and water-holding-capacity.

From the results obtained the following conclusions have been drawn with reference to the effects of air-drying:-

(1) Microbiological activity, as shown by the percentage increases in carbon dioxide evolution, increases with lengths of drying only in the manured soil "X", the fertilized soil "Y" and the manured and fertilized soil "Z".

(2) The two unmanured soils "M" and "N" show a percentage increase in carbon dioxide evolution after 88 days and 115 days drying which is greater than the increases of soils "X", "Y" and "Z", but further periods of drying have relatively slight effects upon their bacterial activity.

(3) The greatest increases in bacterial activity were shown by the manured and fertilized soil "Z", after drying for more than 200 days.

(4) There were more bacteria in the upper layer of all soils than in the lower.

(5) The apparent effect of remoistening was to increase bacterial numbers within five days, but statistical evidence was produced which tends to show that the plate-counts on remoistened soils were unreliable, due, possibly, to the stimulation on air-drying of some organism which when the soils are plated on Thornton's mannite-asparagine-agar count medium, tends to inhibit

the colony-formation of other soil organisms.

(6) The numbers of ray-fungi did not appear to be affected by air-drying.

(7) Nitrification tended to increase rapidly in both layers of all soils with the exception of the unmanured soil "M".

(8) In all cases the lower layer of the soils showed greater increases in bacterial activity, on remoistening, than did the corresponding upper layer.

A C K N O W L E D G E M E N T S

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L I T E R A T U R E C I T E D

1. Fisher, R. A. (1928).
Statistical Methods for Research Workers. 2nd Ed.
Edinburgh.

2. Fisher, R. A., Thornton, H. G. and Mackenzie, W. A. (1922).
The accuracy of the plating method of estimating the density of bacterial populations with particular reference to the use of Thornton's agar medium with soil samples.
Ann. Appl. Biol., 9, 325-359.

3. Fred, E. B. and Waksman, S. A. (1928).
Laboratory Manual of General Microbiology, with special reference to microorganisms of the soil.
London.

4. Gedroiz (1915).
Bull. Internal. Instit. Agric. Rome. p.37, cited by:-
Russell (16, p.p. 221-222).

5 Greaves, J. E. and Carter, E. G. (1920).
Influence of moisture on the bacterial activities of soil.
Soil Sci., 10, 361-387.

6. Gustafson, A. F. (1922).
The effect of drying soils on the water-soluble constituents.
Soil Sci., 13, 173-213.

7. Harper, E. J. (1924).
The accurate determination of nitrates in soil.
Jour. Indus. Engin. Chem., 16, 180-183.

8. Khalil, F. (1929).
The effect of drying on the microbiological processes in soil.
Cent. fur. Bakt., II Abt., 79, 93-107.

9. Klein, M. A. (1915).
Studies in the drying of soils.
Jour. Amer. Soc. Agron., 7, 49-77.

10. König, J. and Hasenbäumer, J. (1920).
Landw. Jahrb., 55, 185-252 cited by:-
Waksman, S. A. (1927).
Principles of Soil Microbiology,
London.

11. Lebediantzef, A. N. (1924).
Drying of soil, as one of the
natural factors in maintaining
soil fertility.
Soil Sci., 18, 419-447.

12. Neller, J. R. (1918).
Studies on the correlation between
the production of carbon dioxide
and the accumulation of ammonia
by soil organisms.
Soil Sci., 5, 225-242.

13. Petterkofer, M. V. (1858).
Ueber eine Methode die Kohlensäure
in der atmosphärischen Luft.
zu bestimmen.
Chem. Soc. Trans., 10, 292-?.

14. Potter, R. S. and Snyder, R. S. (1916).
Carbon dioxide production in soils
and carbon and nitrogen changes in
soils variously treated.
Iowa Agr. Exp. Sta. Res. Bul. 39.

15. Prescott, J. A. (1920).
A note on the Sheraqi soils of
Egypt. A study of partial
sterilization.
Jour. Agr. Sci., 10, 177-181.

16. Russell, E. J. (1927).
Soil Conditions and Plant Growth.
5th Ed., London.

17. Steenkamp, J. L. (1928).
The effect of dehydration of soils
on their colloid constituents.
Part III. Soil Sci., 25, 327-332.

18. Thornton, H. G. (1922).
On the development of a standardised agar medium for counting soil bacteria, with especial regard to the repression of spreading colonies.
Ann. Appl. Biol., 9, 241-274.

19. Thornton, H. G. and Gray, P. H. H. (1930).
The fluctuations of bacterial numbers and nitrate content of field soils.
Proc. Roy. Soc., B., 106, 399-417.

20. Waksman, S. A. and Starkey, R. L. (1923).
Partial Sterilization, microbiological activities and soil fertility, Part II. Soil Sci., 16, 247-268.

21. _____ (1923).
Part III. ibid., 16, 343-357.

The following graphs (Figs. 3-17) show the total evolution of carbon dioxide from all soil units.

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Figure 3.

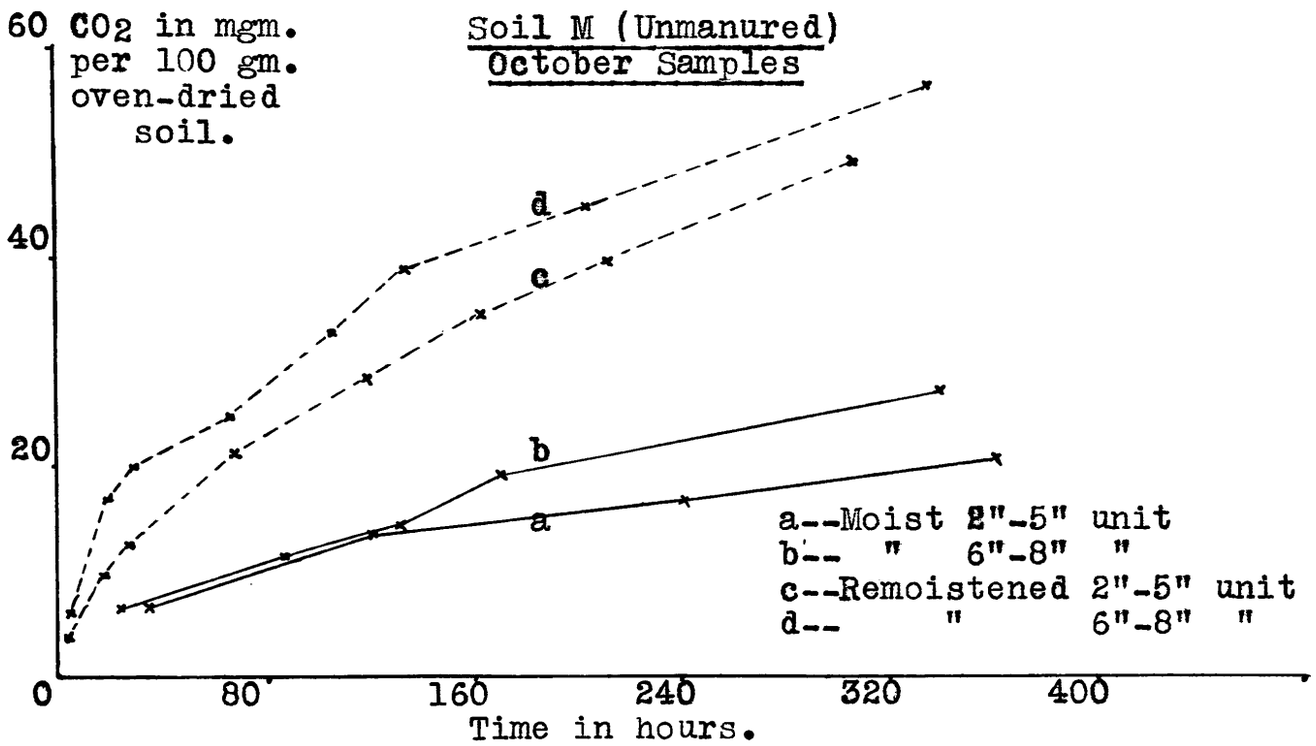


Figure 4.

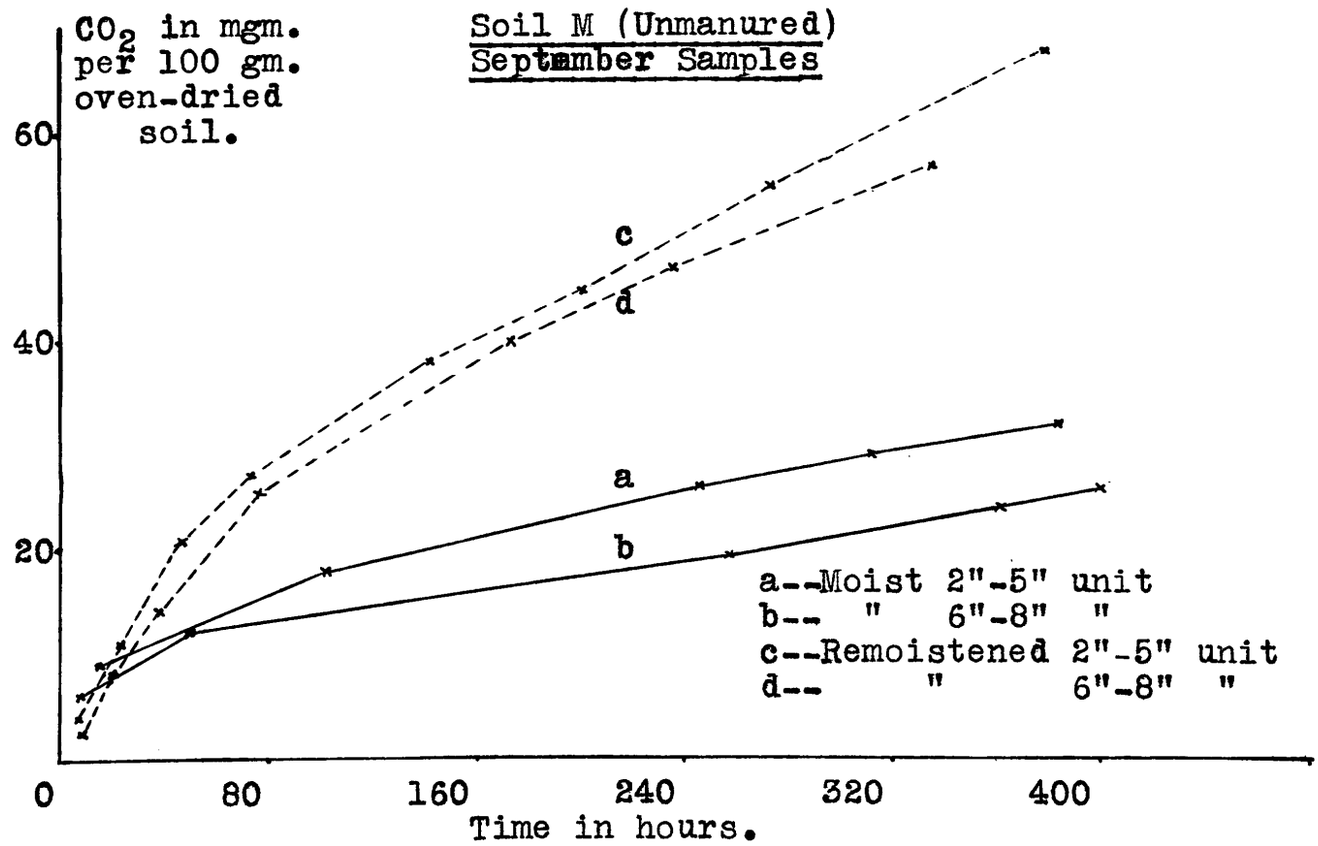


Figure 5.

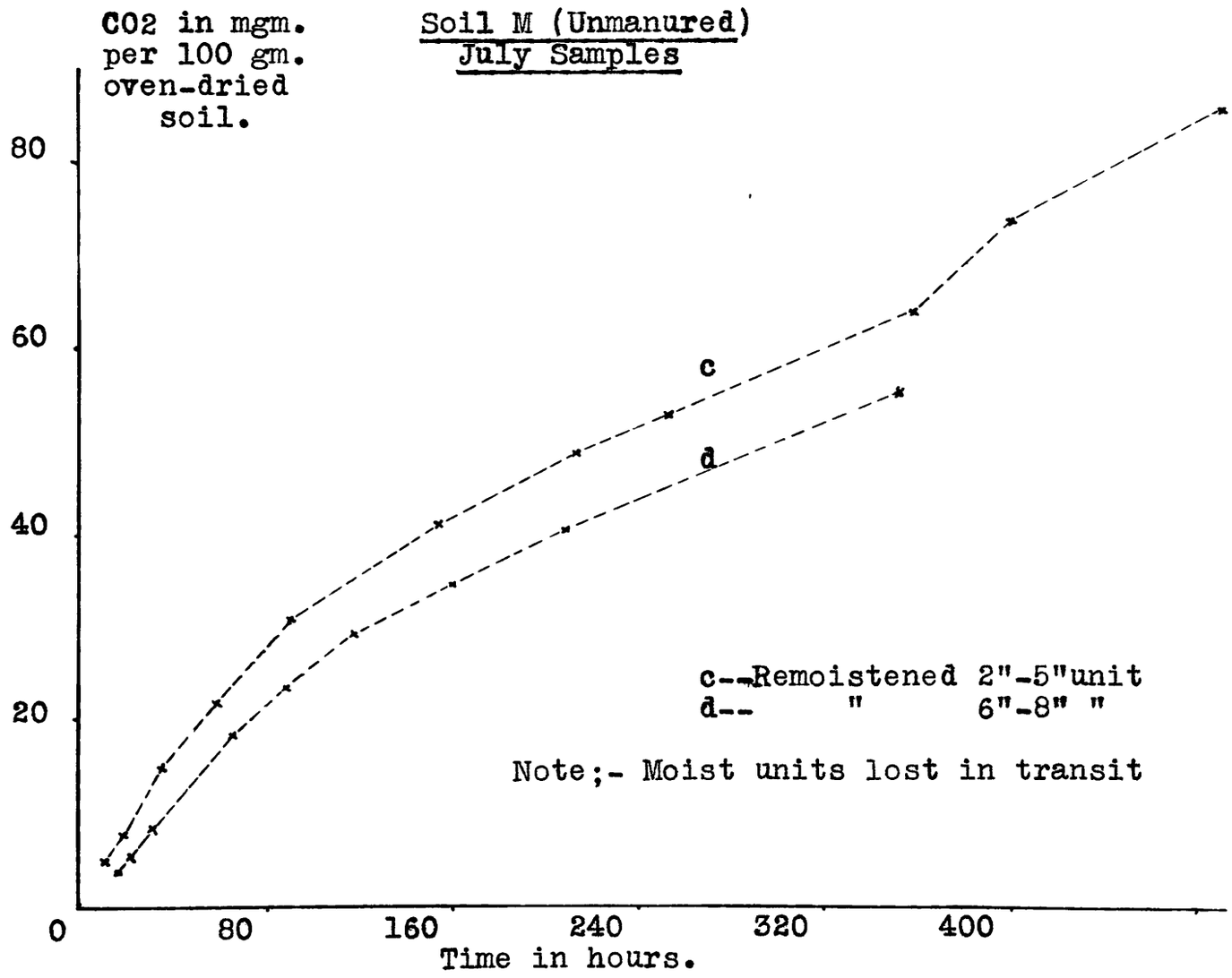

CO2 in mgm. per 100 gm. oven-dried soil.
Soil M (Unmanured)
July Samples
80
60
40
20
0
80
160
240
320
400
Time in hours.
c
d
c--Remoistened 2"-5"unit
d-- " 6"-8" "
Note;- Moist units lost in transit

Figure 6.

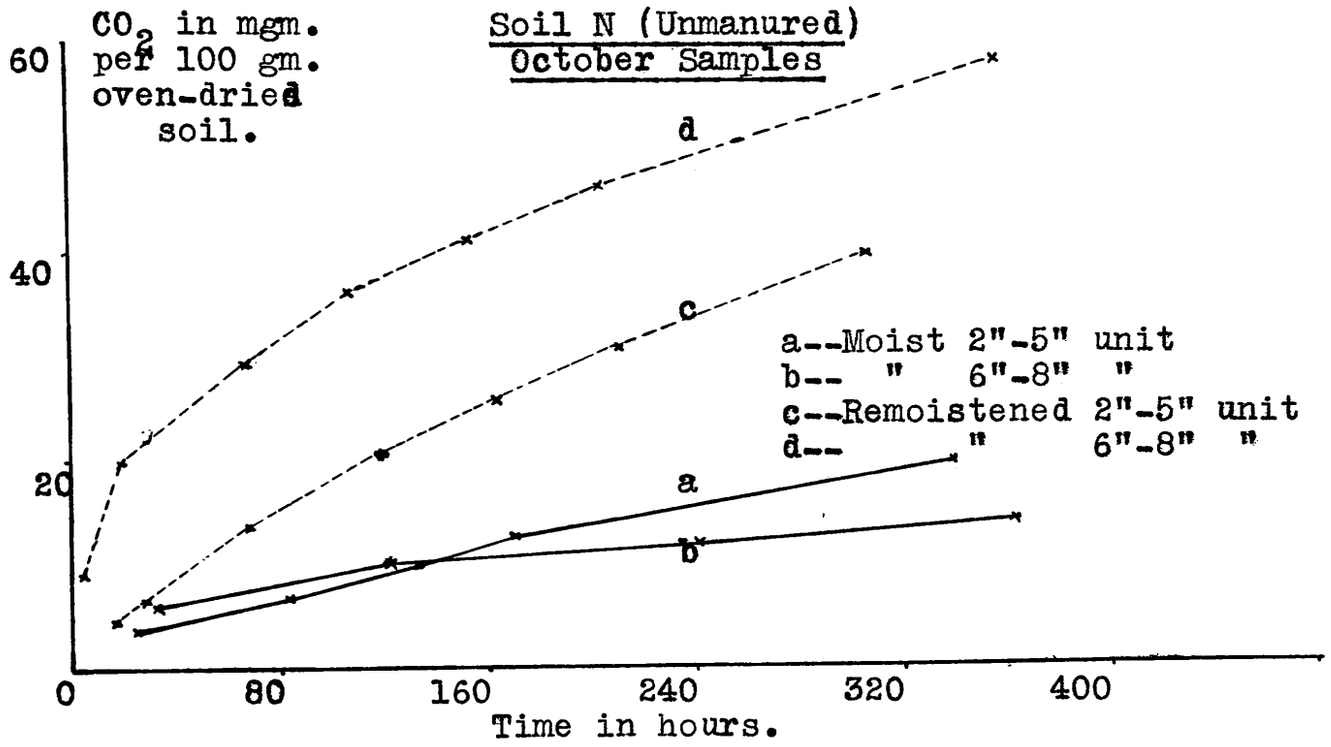


Figure 7.

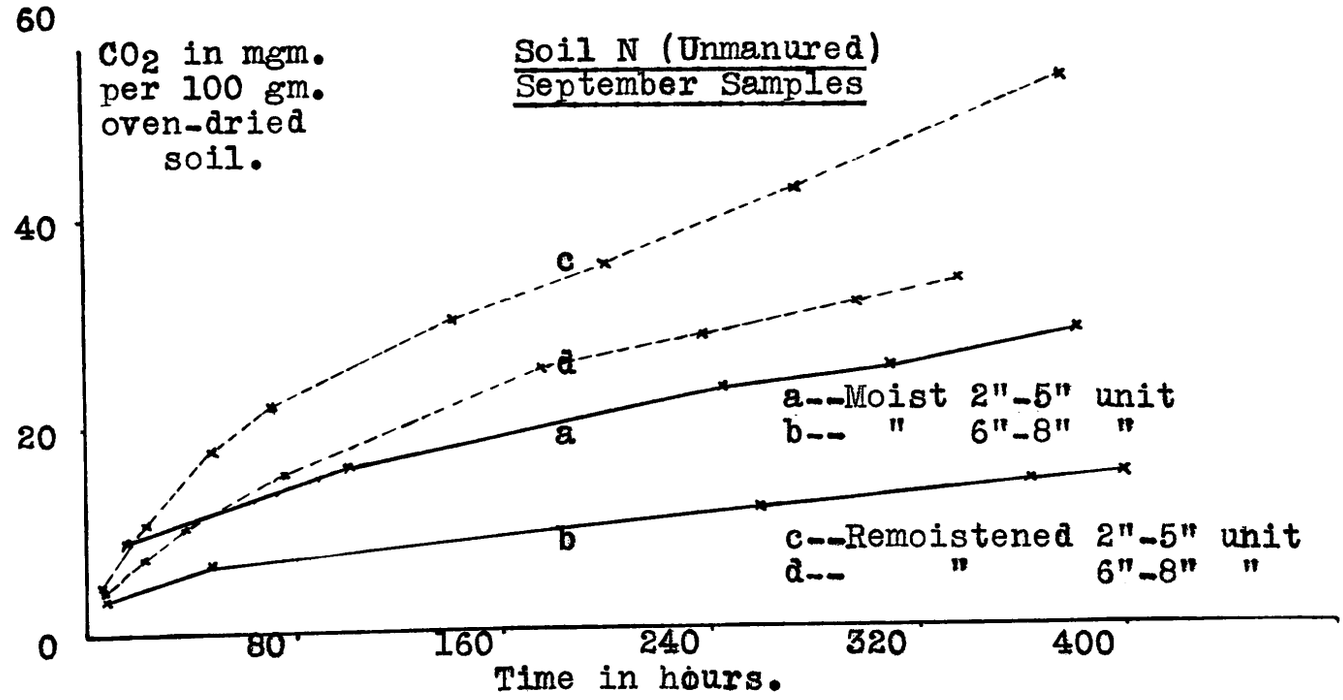


Figure 8.

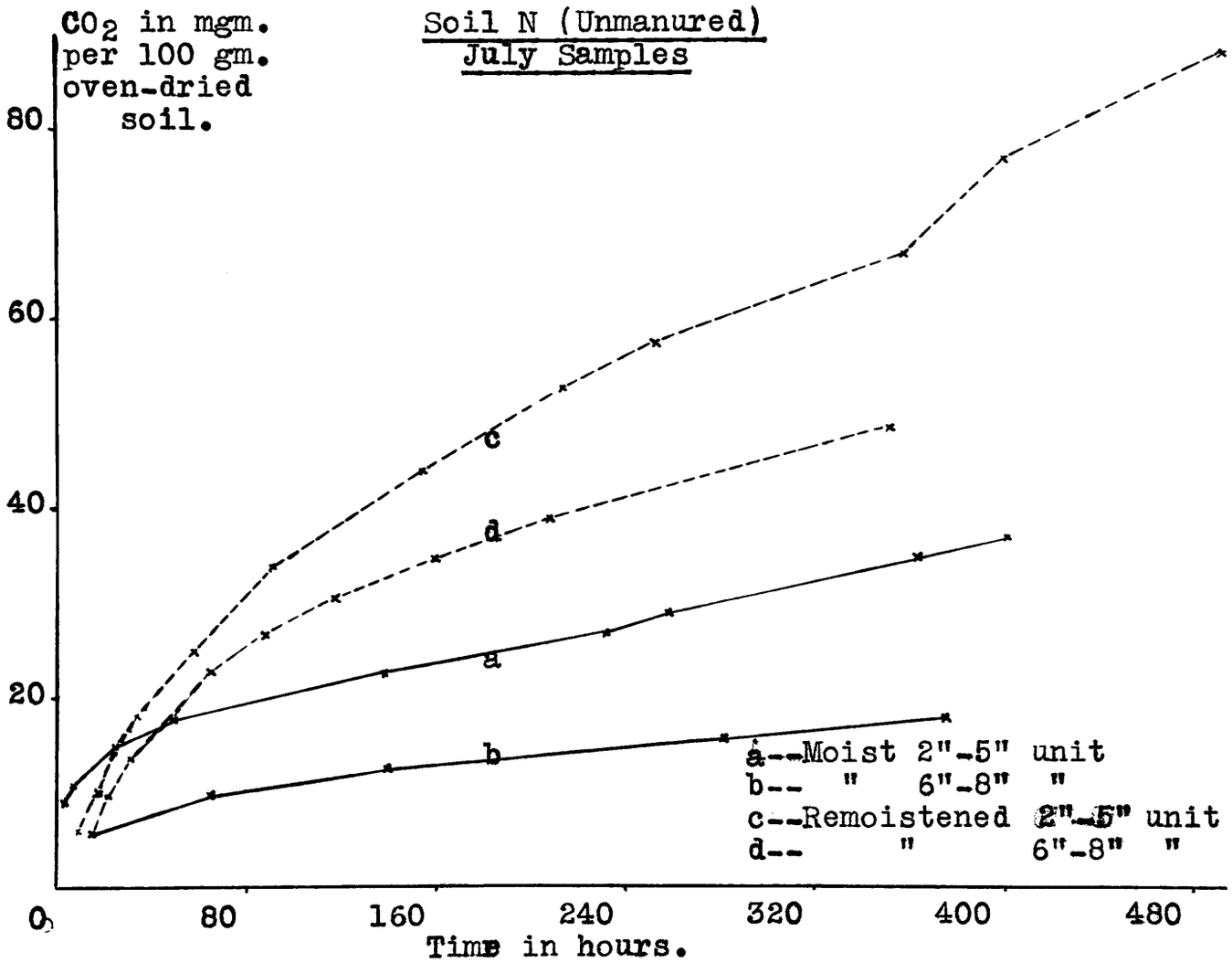

CO_2 in mgm. per 100 gm. oven-dried soil.
Soil N (Unmanured)
July Samples
80
60
40
20
0
80
160
240
320
400
480
Time in hours.
a
b
c
d
a--Moist 2"-5" unit
b-- " 6"-8" "
c--Remoistened 2"-5" unit
d-- " 6"-8" "

Figure 9.

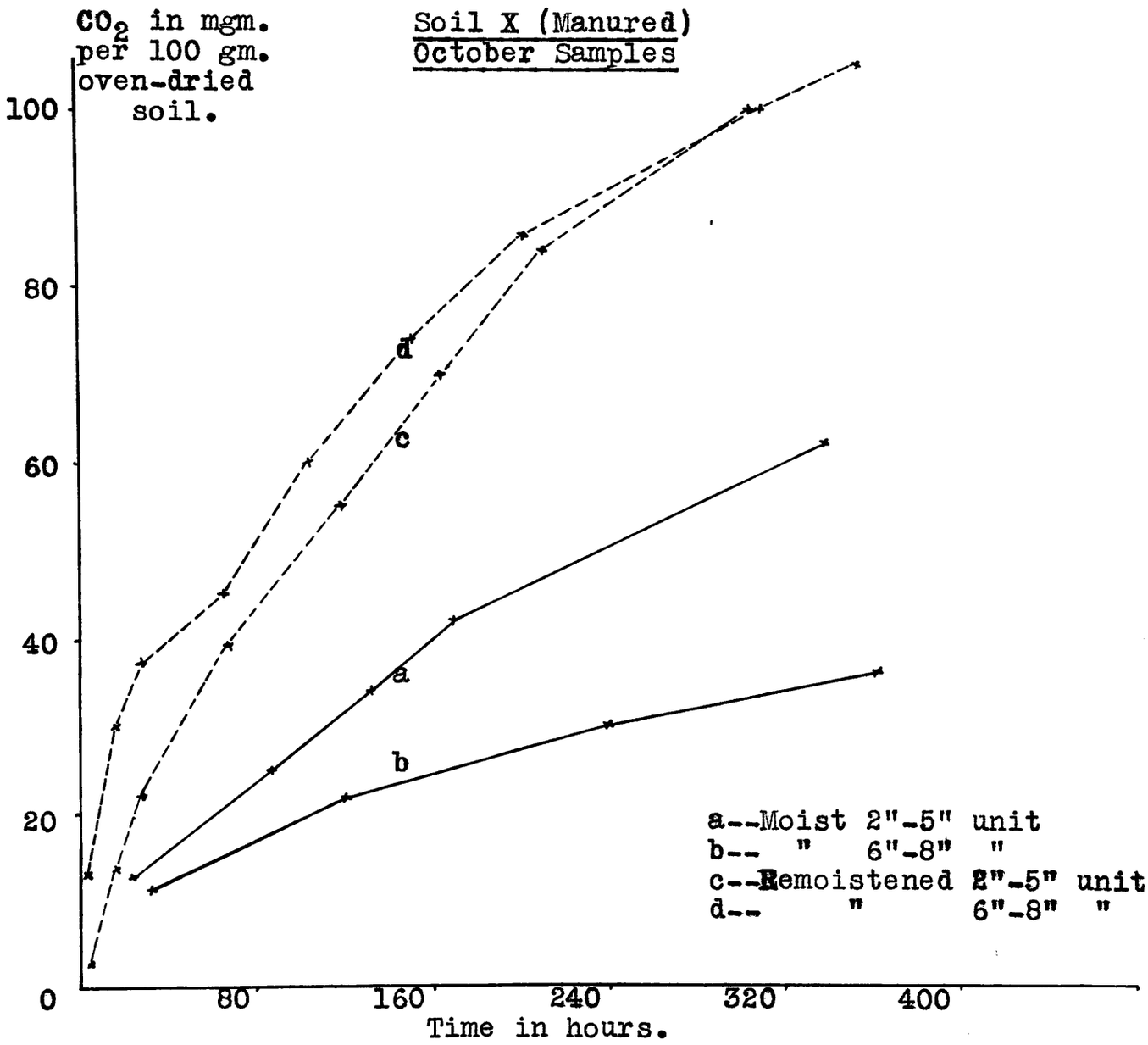

CO_2 in mgm. per 100 gm. oven-dried soil.
Soil X (Manured)
October Samples
100
80
60
40
20
0
80
160
240
320
400
Time in hours.
a
b
c
d
a--Moist 2"-5" unit
b-- " 6"-8" "
c--Remoistened 2"-5" unit
d-- " 6"-8" "

Figure 10.

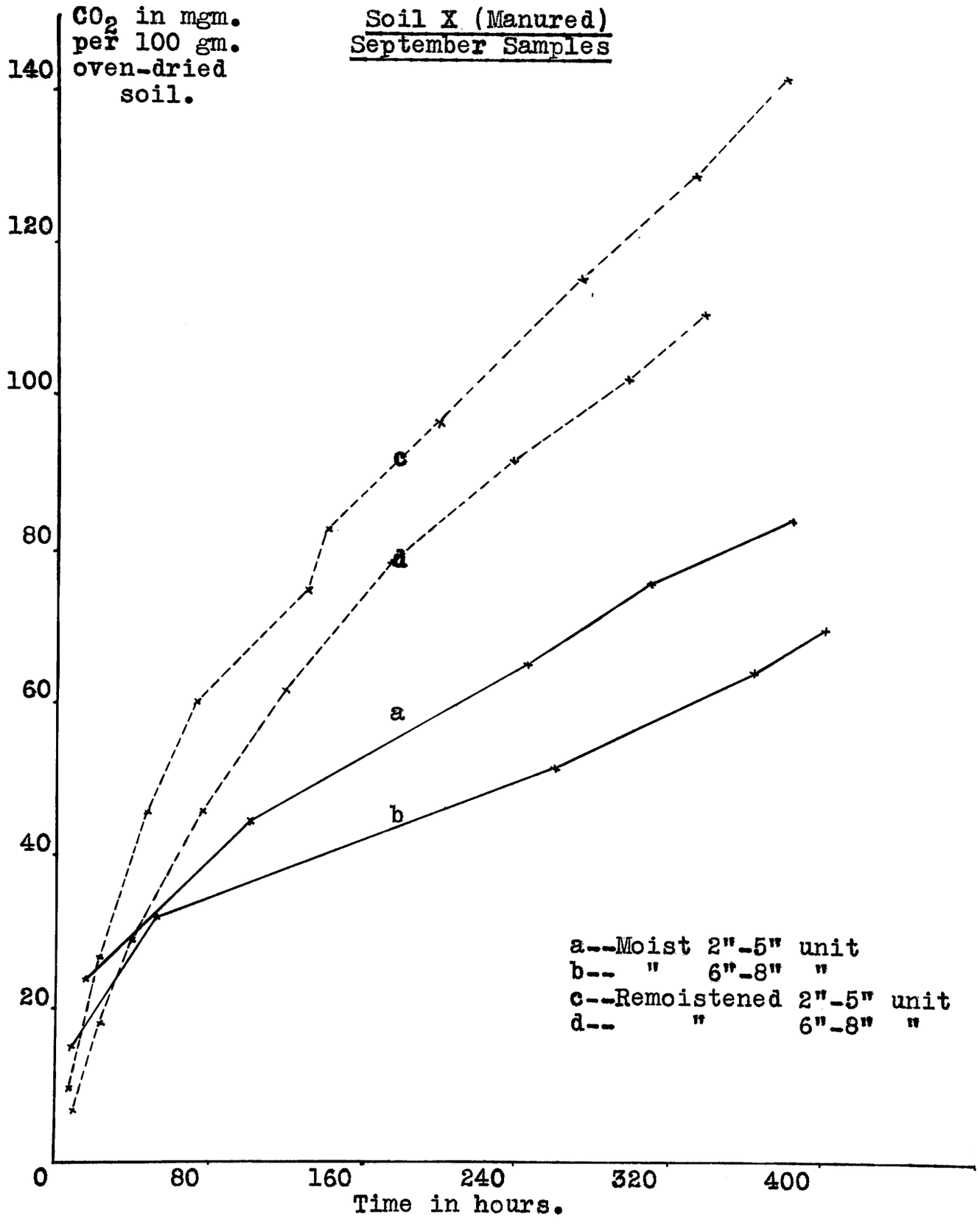

CO_2 in mgm. per 100 gm. oven-dried soil.
Soil X (Manured)
September Samples
140
120
100
80
60
40
20
0
80
160
240
320
400
Time in hours.
a
b
c
d
a--Moist 2"-5" unit
b-- " 6"-8" "
c--Remoistened 2"-5" unit
d-- " 6"-8" "

Figure 11.

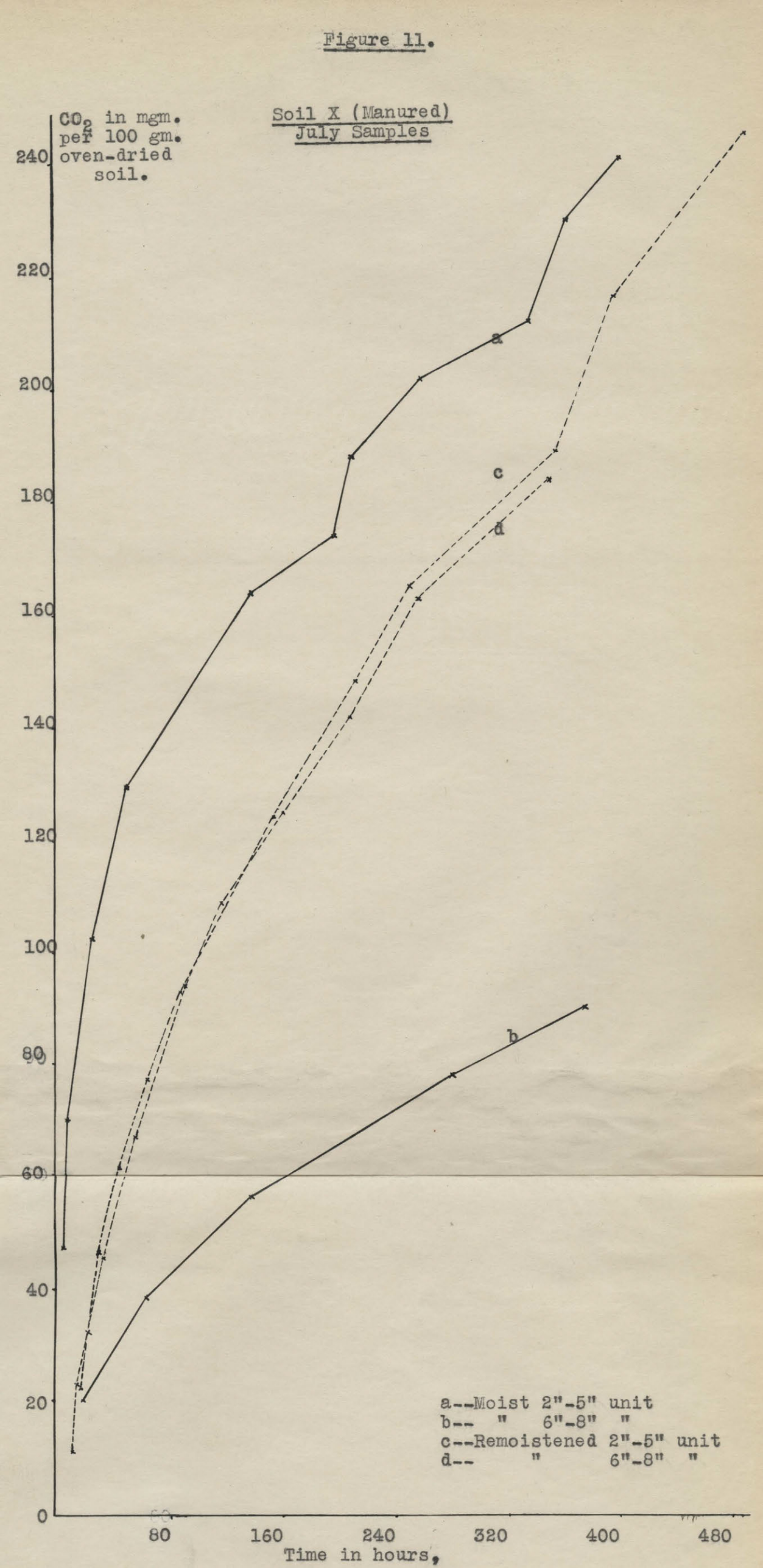

Soil X (Manured)
July Samples
CO_2 in mgm. per 100 gm. oven-dried soil.
240
220
200
180
160
140
120
100
80
60
40
20
0
a
b
c
d
80
160
240
320
400
480
Time in hours,
a--Moist 2"-5" unit
b-- " 6"-8" "
c--Remoistened 2"-5" unit
d-- " 6"-8" "

Figure 12.

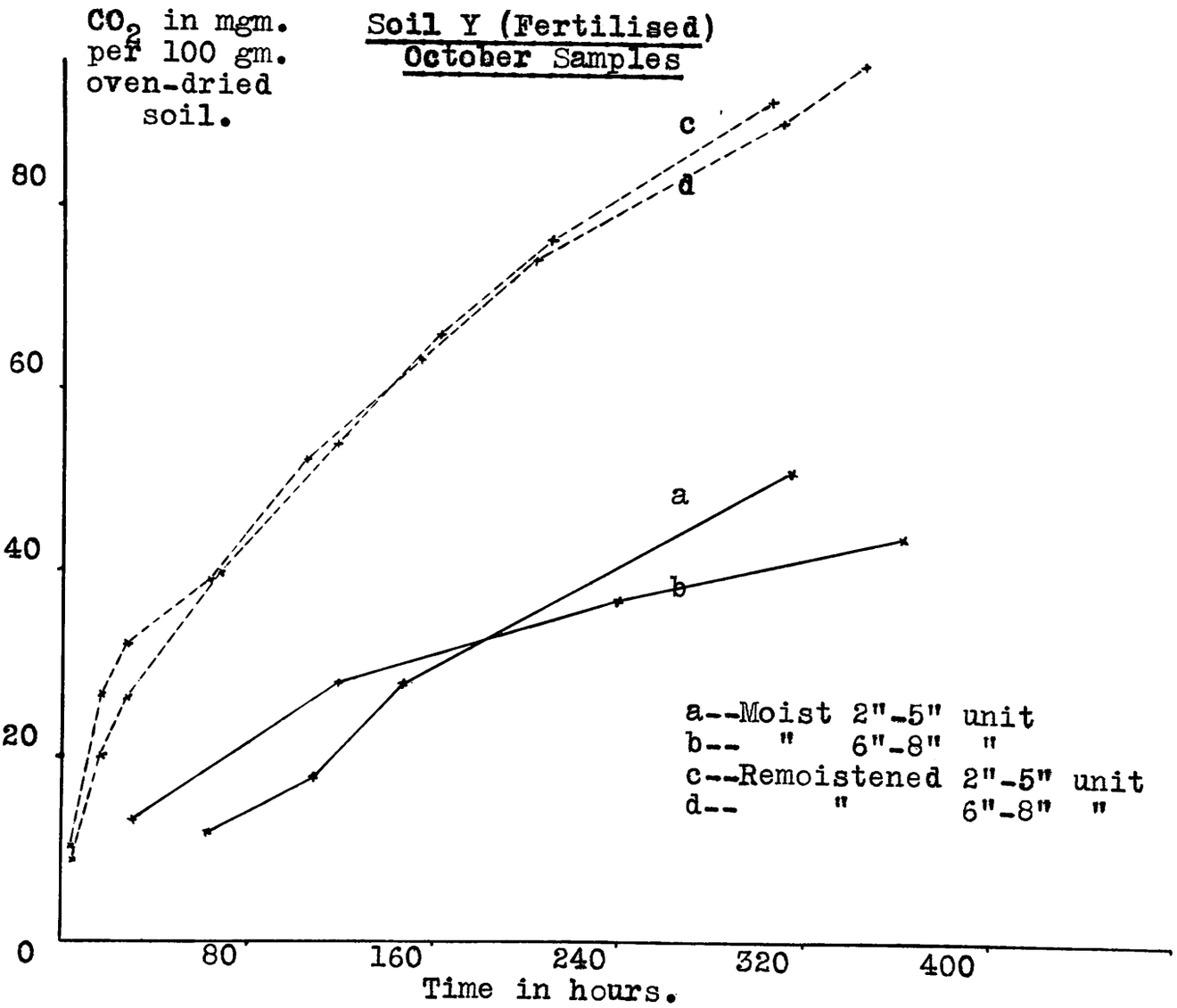

CO_2 in mgm. per 100 gm. oven-dried soil.
Soil Y (Fertilised)
October Samples
c
d
a
b
80
60
40
20
0
80
160
240
320
400
Time in hours.
a--Moist 2"-5" unit
b-- " 6"-8" "
c--Remoistened 2"-5" unit
d-- " 6"-8" "

Figure 13.

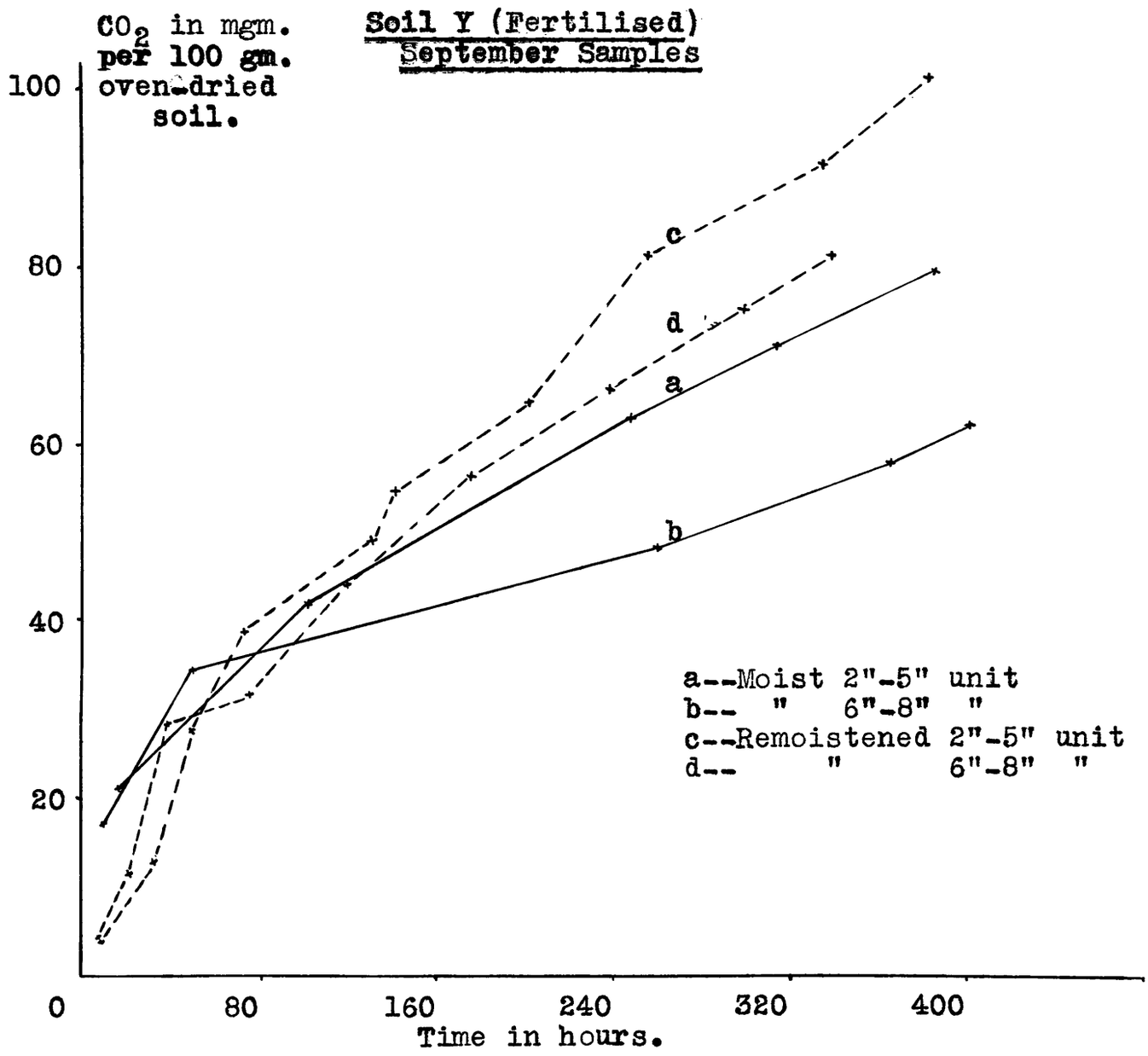

CO_2 in mgm. per 100 gm. oven-dried soil.
Soil Y (Fertilised)
September Samples
100
80
60
40
20
0
80
160
240
320
400
Time in hours.
a
b
c
d
a--Moist 2"-5" unit
b-- " 6"-8" "
c--Remoistened 2"-5" unit
d-- " 6"-8" "

Figure 14.

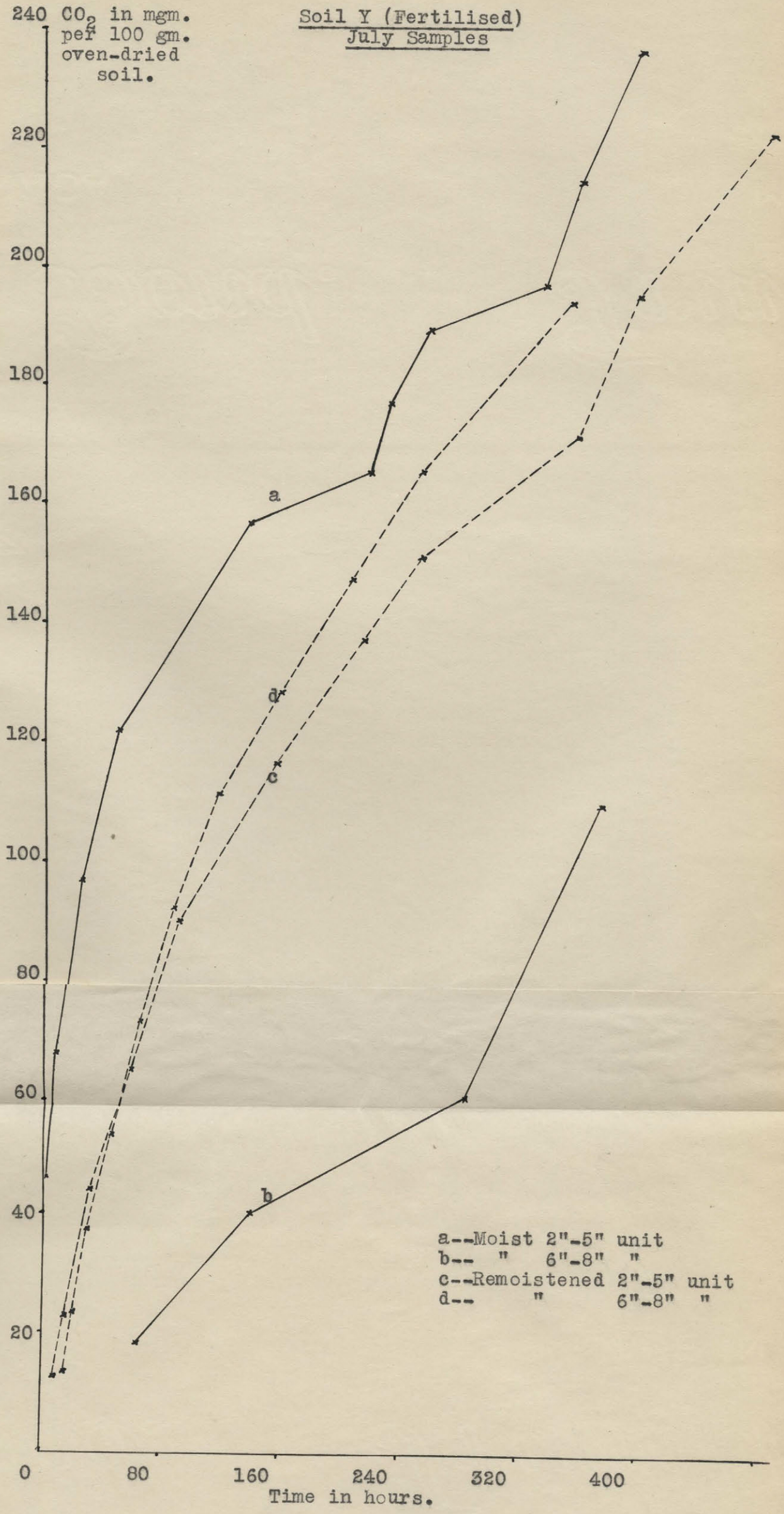

Soil Y (Fertilised)
July Samples
CO_2 in mgm. per 100 gm. oven-dried soil.
240
220
200
180
160
140
120
100
80
60
40
20
0
80
160
240
320
400
Time in hours.
a
b
c
d
a--Moist 2"-5" unit
b-- " 6"-8" "
c--Remoistened 2"-5" unit
d-- " 6"-8" "

Figure 15.

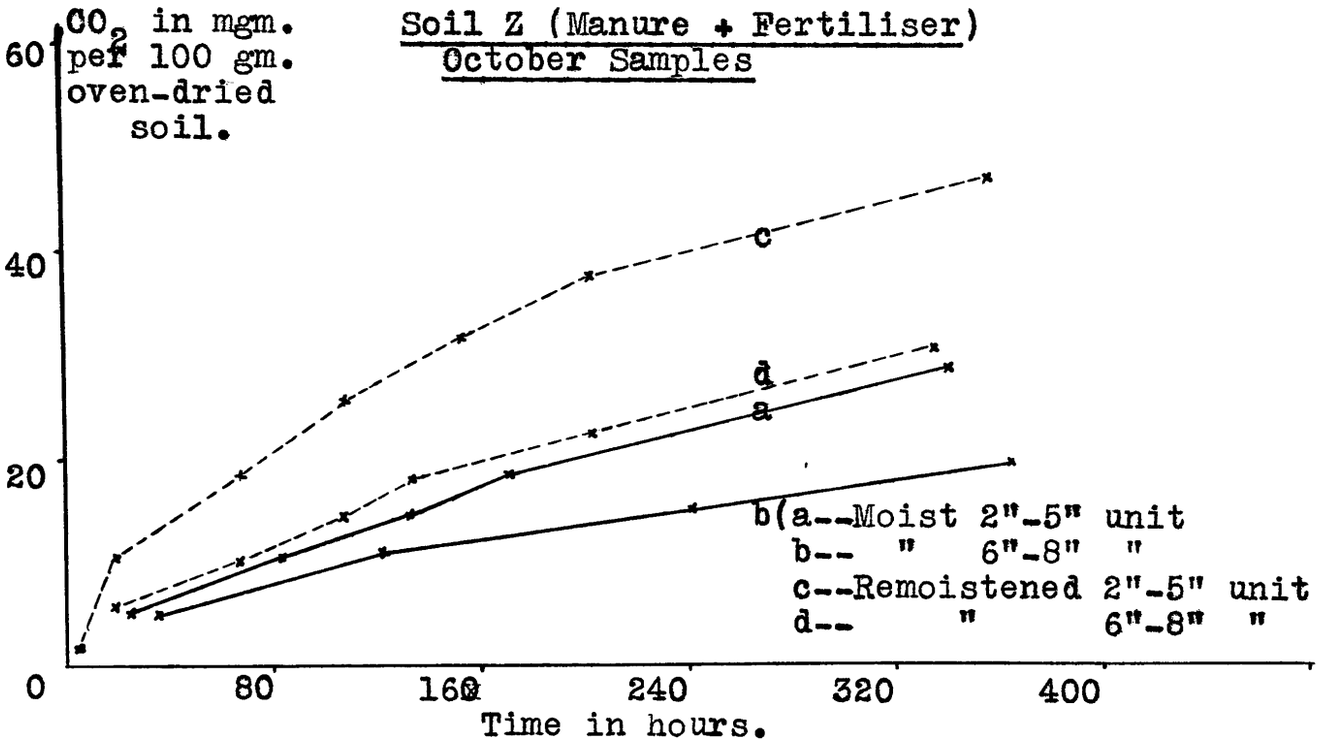


Figure 16.

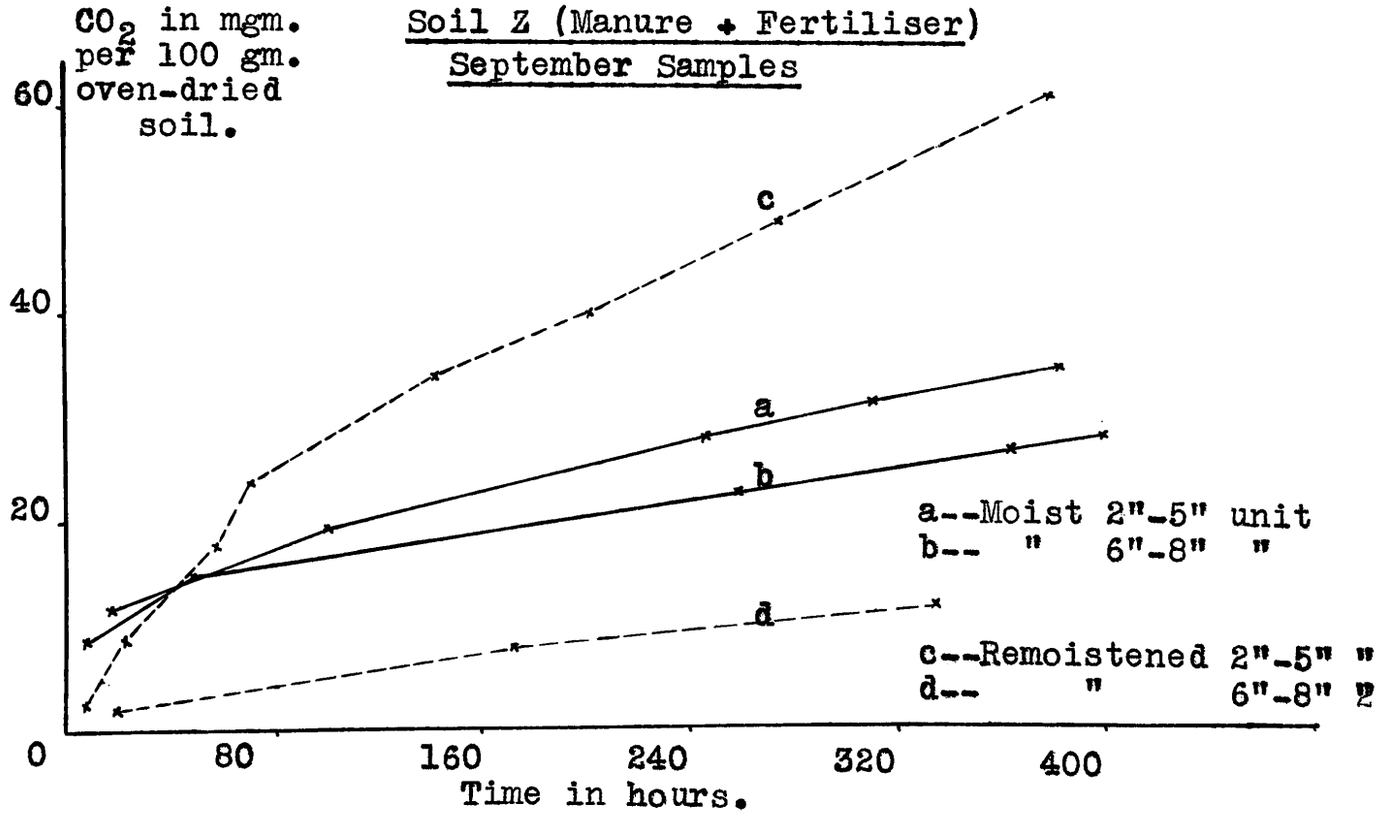
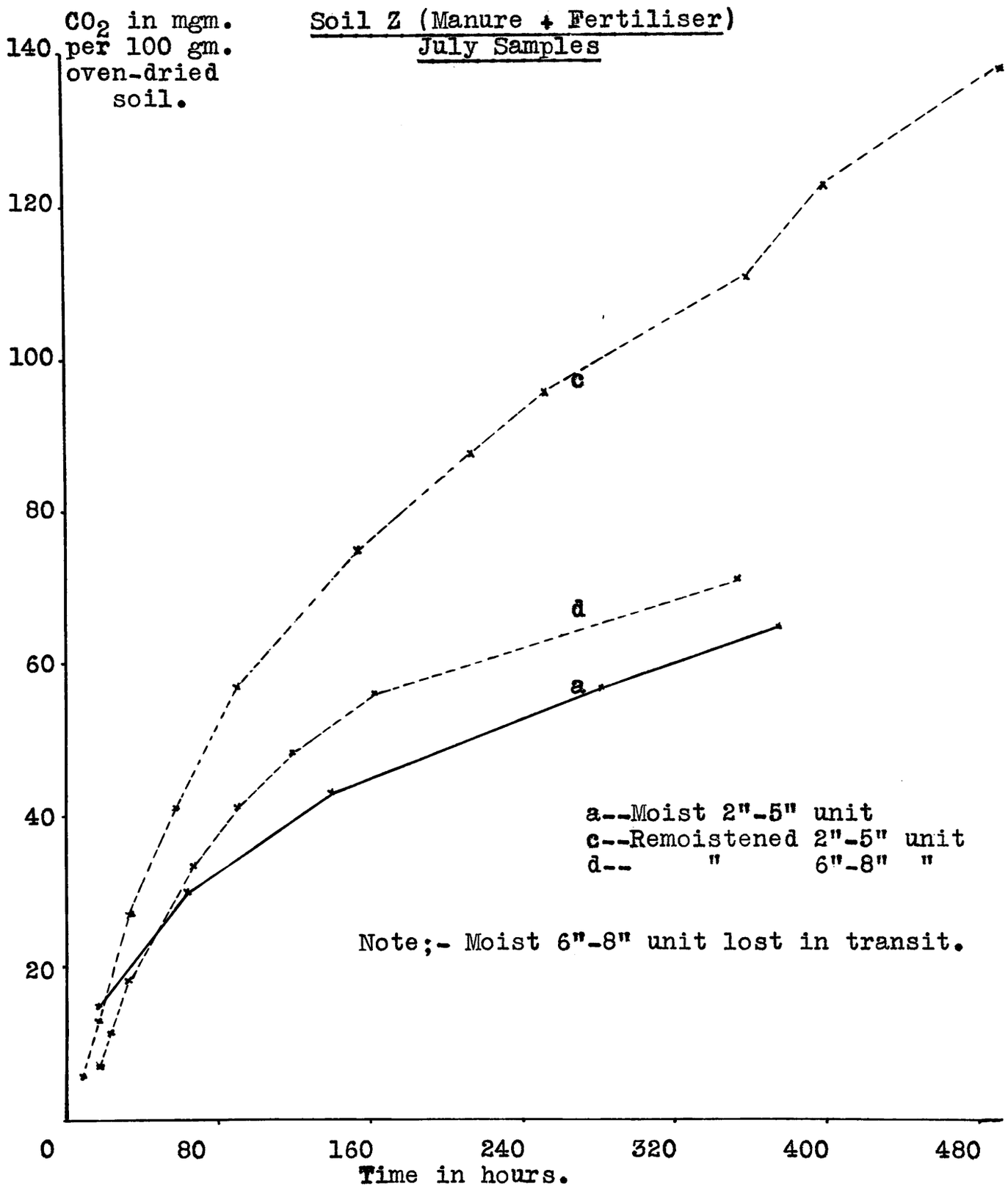


Figure 17.


CO_2 in mgm. per 100 gm. oven-dried soil.
Soil Z (Manure + Fertiliser)
July Samples
140
120
100
80
60
40
20
0
80
160
240
320
400
480
Time in hours.
c
d
a
a--Moist 2"-5" unit
c--Remoistened 2"-5" unit
d-- " 6"-8" "
Note;- Moist 6"-8" unit lost in transit.