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THE EFFECT OF CARBON DIOXIDE ON NITRATE ACCUMULATION IN CARRINGTON LOAM¹

F. B. SMITH, P. E. BROWN AND H. C. MILLAR

Most microorganisms obtain carbon as well as energy from the oxidation of organic compounds but the nitrifying bacteria assimilate carbon dioxide from the air as a source of carbon. The sulfur bacteria and the hydrogen oxidizing bacteria also assimilate carbon dioxide from the air. Other bacteria that can not assimilate carbon dioxide from the air are greatly stimulated by its presence. Increasing the concentration of carbon dioxide in the air about cultures of *Lactobacillus acidophilus* produces higher plate counts and larger colonies than cultures in carbon dioxide-free air. An increased concentration of carbon dioxide in the soil air stimulates the growth of certain of the fungi, particularly those associated with root-rot of the higher plants. Some of the fleshy fungi, the so-called "lignin-destroying" fungi, are greatly retarded in growth by the removal of carbon dioxide from the atmosphere in which they grow. On the other hand, there are cases on record in which carbon dioxide has acted as a growth depressant or germicide. The spores of *Mucor*, *Aspergillus* and *Penicillium* are prevented from germination by very high concentrations of carbon dioxide. The growth rate of yeast and fermentation of solutions have been retarded by certain concentrations of carbon dioxide.

Carbon dioxide is produced in the soil through the decomposition of organic matter and by root respiration of the higher plants. The concentration of carbon dioxide in the soil air is normally higher than that of the atmosphere above the soil but the concentration of carbon dioxide in the soil rarely exceeds 1 or 2 per cent.

In a study of the effects of carbon dioxide on the availability of phosphorus and potassium in Carrington loam, carbon dioxide was added as a gas and in water saturated with the gas. The accumulation of nitrates in this soil was determined at intervals over a period of 5 months and this paper presents the results of that work.

METHODS OF PROCEDURE

Carbon dioxide was applied to the soil as a gas and dissolved

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in water for comparison with untreated soil and soil aerated with a mixture of 1 part oxygen and 4 parts nitrogen. The soil used was a Carrington loam having a pH of 6.33. The soil was passed through a quarter inch screen and thoroughly mixed. Thirty-nine pounds of soil were placed in each of four 4-gallon earthenware pots. Carbon dioxide gas was added from below to the soil in one pot at the rate of five liters per hour for one hour each day throughout the experiment. The soil of another pot was treated from below with a mixture of oxygen and nitrogen, also added at the rate of five liters per hour for an hour each day throughout the experiment. Carbon dioxide water, referred to as carbonic acid and having a pH of 4.2 was added to the soil of a third pot. A fourth pot of soil was left untreated and served as a check. The gases were regulated and measured by means of flow meters as shown in Fig. 1. The moisture content of the soils was adjusted to 25 per cent and maintained at about this amount by frequent additions of distilled water. Samples were taken at regular intervals for nitrate determinations by the phenoldisulfonic acid method. The results obtained are presented in Table I and an analysis of variance of p.p.m. nitrate-nitrogen is given in Table 2. In the calculations of the analysis of variance, the interaction was regarded as the only valid estimate of experimental error, since the two nitrate readings were made on the same soil.

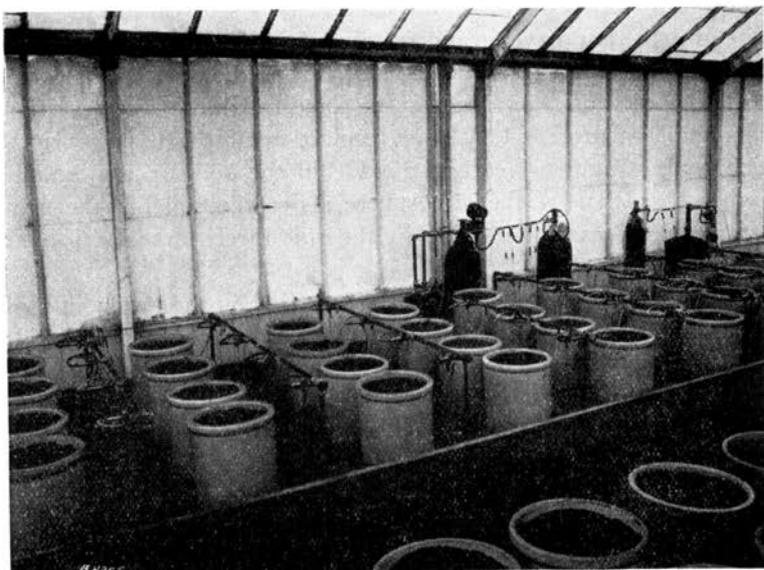


Fig. 1. Apparatus for measuring and regulating gas introduced into the soil.

Table I. Effect of Carbon Dioxide on Nitrate Accumulation in Carrington Loam

Date	P. P. M. Nitrate-Nitrogen				
	Check	O ₂ + N ₂	CO ₂ - gas	H ₂ CO ₃	Mean
November	20.83	20.83	20.83	20.83	20.83
	20.83	20.83	20.83	20.83	
December	28.57	35.08	26.14	39.21	32.22
	28.57	35.08	26.31	38.83	
January	20.08	20.83	20.40	45.45	26.18
	20.08	21.27	17.85	43.47	
February	47.05	71.43	41.66	52.94	53.27
	47.05	71.43	41.66	52.94	
March	47.61	58.82	32.26	50.00	46.62
	47.61	55.25	32.26	49.18	
April	28.16	48.78	29.41	30.30	34.88
	28.16	48.78	27.02	38.46	
Mean	32.05	42.37	28.05	40.20	

Table II. Analysis of Variance of P. P. M. Nitrate-Nitrogen in Carrington Loam

Source of Variation	Degrees of Freedom	Sum of squares	Mean square
Total	23	4677.2595	
Between means of treatments	3	819.3731	273.1244 *
Between means of dates	5	3010.4266	602.0853 **
Interaction (Experimental error)	15	847.4598	56.4973

* Significant.

** Highly significant.

RESULTS

The data in the tables show that there was a significant difference between the means of the nitrate content of the variously treated soils and that all soils varied significantly in the nitrate content for the months December and February. The standard error of difference between the means of treatments is given by

$$\frac{\sqrt{(2)(56.4973)}}{6} = 4.339$$

Corresponding to Fisher's 5 per cent point for 15 degrees of freedom the value of $t = 2.131$; therefore, the least mean difference which can be considered significant is $(2.131)(4.339) = 9.25$ p.p.m. Similarly, the standard error of the difference between any pair of date means is

$$\frac{\sqrt{(2)(56.4973)}}{4} = 5.315$$

and the least mean difference which can be considered significant is $(2.131)(5.315) = 11.33$ p.p.m.

In the light of this analysis it is apparent that the treatment with oxygen and nitrogen gas was effective in stimulating nitrate production but that treatments with carbon dioxide gas or carbonic

acid were without significant effect. Aeration of the soil with oxygen might be expected to stimulate nitrification provided sufficient carbon dioxide were available for the growth of the nitrifying organisms. The failure of the carbon dioxide to affect the nitrate content of this soil indicates that carbon dioxide was not a limiting factor in nitrification.

The apparent decrease in the average nitrate content of the soil treated with carbon dioxide gas under that of the check soil might be caused by an oxygen deficiency and the apparent increase in the nitrate content of the soil treated with the carbonic acid might be caused by an increased solubility of the mineral elements required by the nitrifying organisms. However, these differences, though relatively large, are not greater than might be expected in random sampling and are regarded as insignificant.

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