

THE EFFECT OF THE ADDITION OF ORGANIC MATERIALS ON THE  
DECOMPOSITION OF AN ORGANIC SOIL

DISSERTATION

Presented in Partial Fulfillment of the Requirements for the  
Degree Doctor of Philosophy in the Graduate School of  
The Ohio State University

By

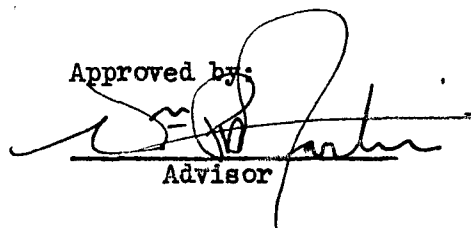
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## TABLE OF CONTENTS

	Page
Introduction . . . . .	1
Review of Literature . . . . .	3
Purpose of the Study . . . . .	8
Methods and Materials . . . . .	9
General . . . . .	9
Materials . . . . .	10
Collection of CO <sub>2</sub> Evolved on Incubation of the Soil . . . . .	11
Biosynthesis of Carbon-14 Labeled Glucose . . . . .	13
Determination of Organic Carbon . . . . .	14
Determination of Radioactivity . . . . .	15
Determination of Total Nitrogen . . . . .	16
Determination of pH . . . . .	16
Extraction of Soluble Material . . . . .	16
Experimental . . . . .	18
Tabulation of Results . . . . .	23
Discussion . . . . .	37
Summary . . . . .	46
Bibliography . . . . .	49
Appendix . . . . .	53

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INTRODUCTION

Organic soils, mucks and peats, generally arise where conditions are favorable for swamp vegetation and when decomposition of the plant remains takes place anaerobically. Decomposition is, therefore, slow and incomplete. Thus, there accumulates a deposit of partially decomposed, relatively resistant organic matter which may, when drained and cleared, provide a soil suitable for the intensive cultivation of high value crops.

Due to the inherent characteristics of the soils and the nature of the type of agriculture carried out on them, certain problems are universally associated with them. The most common of these is the problem of subsidence sometimes termed shrinkage or merely loss of soil.

The soils, being high in colloid content, can hold a large amount of water. After drying they are usually unable to regain the total amount of water they lost. They are thus, by nature, subject to shrinkage. Their specific gravity is quite low leaving them particularly susceptible to wind erosion. Their high organic content and loose physical structure when drained, the frequent cultivations they receive, make them subject to accelerated biological decomposition.

While considerable effort has been directed towards slowing down this biological decomposition, investigations into the possibility of building up the organic content have been limited. A considerable

amount of time and effort has been spent trying to build up, or maintain, the organic matter of inorganic soils by the addition of barnyard manure, green manures and mulching materials. That this is not true for organic soils is probably due to the intensive type of agriculture practiced on them. Crops produced on these soils are generally harvested without leaving any residue and one crop follows another with no time taken during the growing season for green manuring crops. Whether manuring practices would be beneficial, detrimental or economically feasible has never been established. Considering the high productivity of these soils and the magnitude of the problems arising from loss of the soil it would seem that considerable investigation into this phase would be merited.

## LITERATURE REVIEW

Skertchey as reported by Powers (29) cited evidence of subsidence of organic soils near Ely in England. In sixty-five years, ten feet of soil was lost. In Louisiana, Bowers reported loss of eighteen inches in eight years. The depth of the deposit ranged from a few inches up to eighteen feet.

By taking surface levels it was shown (32, 41) that a tule (Scirpus lacustris) peat on the San Joaquin, Sacramento delta farmed twenty years lost an average of 0.19 feet annually in the next ten years. For soil farmed seven years and one year the annual loss was 0.22 and 0.33 feet respectively.

Neller (21) reported that a high water table reduced subsidence of a Florida peat. Annual loss under a thirty six inch table was 1.80 inches. Under a twelve inch table it was 0.55 inches. Neller and Deane (22) further showed that addition of energy supplying materials to these soils caused an increase in microbial activity and resulted in higher crop yields.

Edelman (11) lists some of the problems arising from subsidence of organic soils in Holland.

Very little research on the effect of manuring practices on loss of organic soils has been reported in the literature. The analogous situation in mineral soils i.e., the effect of manuring on the amount of organic matter in the soil, has received a great deal of attention. It should prove helpful in establishing fundamental

principles to review some of the work done on inorganic soils, though it is not to be expected that organic soils will react the same.

One of the first reports was made by Bousingault (5) who showed that decomposition of organic matter in the soil resulted in oxygen utilized and carbon dioxide evolved in almost equal volumes. This led to considerable research on the subject using carbon dioxide as an index of decomposition, a practice extensively reviewed by Potter and Snyder (28), Gainey (12), and Waksman and Starkey (40).

It has been determined that the organic composition of the material added to a soil has considerable effect on the rate and amount of decomposition taking place. Soluble materials such as glucose quickly decompose with a short, sharp burst of carbon dioxide evolution. Adding nitrogen with sucrose, Broadbent (6) measured 100% loss of added carbon in 67 days. Gray (14), by testing for glucose added to a podsol, was unable to find any left after three days. Lees and Porteous (17), using soil percolators, showed that at equilibrium one third of the carbon added as glucose remained in the soil.

Turk (35), incubating water extracted soybeans in a Gerald silt loam, found that taking out the water soluble constituents resulted in a drop of 50% in the carbon evolved during the initial decomposition period. Later the rate of loss was slightly higher than for the complete plant material.

Peevy and Norman (23) incubated straw preparations in a Thurman loamy fine sand. These preparations high in lignin lost the least carbon during a period of 833 days followed by the complete straw, cellulose and dextrose in that order. The high lignin treated soil retained 22.6% of the added carbon, the glucose 2.8%.

On two sandy soils used for truck gardening, Bear (4), over an eight year period attempted to increase the initially low supply of organic matter by a system of crop residue return, green manuring and compost additions. On the sandy soil almost all treatments resulted in an increase. The best results were obtained from a combination of all three practices. On the more loamy soil only the use of lignin or peat as a compost combined with the other two treatments gave an increase. The amount applied over the eight year period was between 34 and 37 tons of dry matter per acre.

Allison et al. (1, 2) pointed out that as long as equal quantities of carbon were applied succulent plant material supplied almost as much organic matter to the soil as more mature material. In one year on the average of three soils, nitrogen added and not added, straw resulted in only 120 pounds per acre more carbon retained than green oats. Applications were at the rate of 6000 pounds of carbon per acre.

Pinck et al. (26, 27) showed that it was possible to increase organic matter content in the soil by nitrogen additions. The effect of nitrogen was due chiefly to its relation to more abundant production of roots and other residues. They concluded from their work



that the decomposition of added material and the effect on the organic content of the soil are functions of: (a) chemical composition of the material, (b) chemical composition of the soil, (c) the carbon-nitrogen relationship. For a given material, reducing the carbon-nitrogen ratio resulted in speedier decomposition and often slightly more organic matter remaining in the soil. Bringing two different materials to the same carbon-nitrogen ratio, e.g., alfalfa and straw, did not mean that they would decompose at the same rate or reach the same degree of decomposition.

Broadbent and Bartholomew (7) found that increasing the amount of manuring material added to the soil resulted in a decrease in the rate of decomposition. They concluded that large infrequent applications of manures were better than small more numerous applications. Pinck et al. (27) concluded that usually green manures were added in such small amounts that build up of soil organic matter this way is difficult or impossible.

Recently Broadbent (6) using isotopic nitrogen and carbon showed that a 1.2% addition of carbonaceous material to a Clarion silt loam caused an increase in evolution of carbon dioxide and nitrogen from the residual soil organic matter. Additions of 1 and 2% had no effect on the organic matter of a virgin Webster clay already high (13%) in carbonaceous organic matter. Broadbent and Norman (8) from similar results suggested that the whole practice of green manuring ought to be reevaluated since it appeared possible

that it could lead to a net loss of organic matter.

This work was reviewed critically by Pinck and Allison (24) who pointed out that the accelerated oxidation of the soil organic matter was almost complete in the first very few days. Furthermore, even though applications were large, the loss of soil organic matter amounted to not more than 5% of the total within the soil. They presented data to show that if there was a loss of soil organic matter it was too small to be of any significance.

## PURPOSE OF THE STUDY

Scientific literature and the history of agricultural marshes in the State of Ohio (10), and throughout the world show that subsidence and loss of organic soil deposits is a problem of considerable importance. It is evident that biological decomposition is to some extent responsible for this loss. Whether or not green manuring practices enhance the biological decomposition of organic matter in inorganic soils is a matter of controversy at the present time. Whether or not it is true in organic soils has not been determined.

This laboratory study on an organic soil from Northern Ohio was carried out to provide a basis for field experimentation on the problem of conserving the organic soils by slowing down or compensating for biological decomposition.

An attempt was made to answer the following questions:

1. Under controlled conditions in the laboratory, does the addition of organic materials to an organic soil speed up the breakdown of the organic matter of the soil, a process hereafter referred to as a priming action.

2. If a priming action does take place, is it large enough to cause a net loss of dry matter?

3. If a priming action does take place, are mature, fibrous plant materials containing a large proportion of insoluble organic constituents, or succulent materials containing a large soluble fraction, chiefly responsible?

## METHODS AND MATERIALS

### 1. General:

Although there are some objections (18) to the use of CO<sub>2</sub> collection as a criterion of biological activity within a soil it has been shown to correlate with loss of carbon as determined by oxidation methods and with weight loss (15). It was used in this study for this reason and because it fitted in well with the use of isotopic carbon. It is possible, by use of organic materials labeled with the latter, to divide the CO<sub>2</sub> evolved from a treated soil into the fraction arising from the added material and the fraction arising from the soil organic matter. In this way, by comparison to CO<sub>2</sub> arising from an untreated soil, the amount of priming can be evaluated.

To determine the priming effect of soluble, readily decomposable material, glucose tagged with carbon-14 was added to the soil. The soil was incubated and CO<sub>2</sub> collected. Complete alfalfa evenly labeled with carbon-14 was used to obtain an estimate of the priming action of complete plant material, and the alfalfa, with water soluble constituents extracted, was used to estimate the effect of the fibrous fraction.

The CO<sub>2</sub> was absorbed in NaOH and precipitated as BaCO<sub>3</sub>. Activity counts were made by plating a weighed sample of BaCO<sub>3</sub> on an aluminium dish. The readings were made on a Landsverk model L-75 electrometer.

## 2. Materials:

### (a) Muck Soil:

The soil samples were taken from the State Experimental Muck Farm in Huron County, Ohio. Two samples were taken, both from the top 6 inches of relatively undecomposed muck which had been farmed five years. The first sample was collected in April after the soil had warmed up and received some spring cultivation. The second sample was collected in January after a prolonged period of rainfall and was frozen to a depth of 6 inches. This sample, used for the alfalfa incubation experiments, evolved less CO<sub>2</sub> per unit time on incubation than the first sample which was used for the glucose and straw vs. rye grass incubations. It, obviously, had been leached of some of its soluble organic material and soluble nitrogen.

The soil had been classified as a Rifle peat (34). The deposit sampled is approximately 6 feet deep, has a pH of about 4.9, exchange capacity of 122 m.e. and contains 2.5% total nitrogen.\*

### (b) Glucose Labeled with Carbon-14:

Labeled glucose was biosynthesized as described in the section on methods.

### (c) Ranger Alfalfa Evenly Labeled with Carbon-14:\*\*

This alfalfa was cultured throughout its growth period in a controlled atmosphere containing C<sup>14</sup>O<sub>2</sub>. It was harvested just before

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\* Analysis by E. E. Barnes, Ohio Agricultural Experiment Station.

\*\* The carbon-14 labeled alfalfa used in this investigation was supplied by The Argonne National Laboratory of the United States Atomic Energy Commission.

blossoming, dried and ground to pass a 60 mesh screen. The amount obtained was 5.9 g. containing 0.6 m/c.

(d) Unlabeled Ranger Alfalfa:

This material, donated by the Argonne Laboratories, was used to dilute the labeled alfalfa. It was the same variety as the labeled material, cultured on the same solution but was not grown in the same rigidly controlled atmosphere.

3. Methods:

(a) Collection of CO<sub>2</sub> Evolved on Incubation of the Soil:

The apparatus used in the incubation experiments was similar to that described by Bartholomew and Broadbent (3) with a glass bead absorber patterned on Heck's (16). Air from an air pressure line was run through a series of scrubbers, (soda lime and saturated Ba(OH)<sub>2</sub>) bubbled through water to saturate it, and into a 4 ft. by 1.5 in. glass manifold with 20 outlets. To these outlets 1 liter Erlenmeyer flasks, containing the soil, were attached. After passing over the soil the air passed through a glass bead bubbler patterned on Heck's absorber. Rate of bubbling was controlled by a section of capillary tubing placed between the incubation flask and the absorber and by a mercury manometer on the air line. The rate of flow through each flask averaged between 1 and 1.5 liters per hour.

In the glucose experiment, 40 ml. of approximately 0.5 N NaOH was measured roughly into each absorbing tower. The absorbed CO<sub>2</sub>

was estimated gravimetrically as follows. The bubbler was washed down into the collection flask with a jet of CO<sub>2</sub> free water. Four ml. of 4 N NH<sub>4</sub>Cl was added through the tower (9) followed by thorough washing into the flask. Ten ml. of 3 N BaCl<sub>2</sub> was added to precipitate the CO<sub>2</sub> absorbed. The flask was then stoppered and placed on the steam plate for 1 hour for digestion. The precipitate was collected on a tared sintered glass filter funnel and dried at 130° C. and weighed.

The CO<sub>2</sub> in all other incubation experiments was determined volumetrically. In the wheat straw vs. rye grass and the alfalfa insolubles vs. complete alfalfa experiments, the bead absorber was modified slightly in that the collection flask, a 500 ml. extra wide Erlenmeyer flask, was changed to a common table glass tumbler which could be placed on a Beckman, Model K Automatic Titrator for the titration of the excess NaOH. The absorbers received an accurately measured (automatic pipette) amount of standard 1N NaOH. Enough NaOH was used so that the quantity neutralized by CO<sub>2</sub> never exceeded two-thirds of the total amount in any collection period.

The bead towers were washed down into the tumblers with CO<sub>2</sub> free water at the end of each collection period. Excess BaCl<sub>2</sub> was added and the excess NaOH titrated on the automatic titrator with standard HCl to a pH of 8.5. This procedure was run through as quickly as possible to minimize absorption of CO<sub>2</sub> from the atmosphere. The precipitate was then digested as before, filtered on sintered glass,

dried and stored in air tight vials for counting.

Since the labeled vs. unlabeled alfalfa experiment dealt with a small weight of soil (20 g.) and thus quite low CO<sub>2</sub> yields, a micro-apparatus was designed for it similar to the larger one just described.. Instead of a large glass manifold, a 2 pound chemical jar was used with 12 outlets in the stopper. Erlenmeyer flasks were used for the incubation flasks and the absorber consisted of a 1.6 by 20 cm. test tube with the inlet extending to the bottom of the tube. Capillaries between the absorber and incubation flask and an H<sub>2</sub>SO<sub>4</sub> bleeding valve on the airline gave control of the aspiration rate. Standard NaOH and CO<sub>2</sub> free water in varying proportions, depending on the amount of CO<sub>2</sub> given off, were measured into the collecting test tube. At the beginning of the collection period this was 6 ml. of 1.002N NaOH and 10 ml. of water. The total volume was kept constant throughout and the amount of base decreased. At the end of each collection period excess BaCl<sub>2</sub> and 2 drops of phenolphthalein were added and the excess NaOH titrated with standard HCl using a stream of nitrogen bubbles for agitation.

(b) Biosynthesis of Carbon-14 Labeled Glucose:

A detached bush bean leaf (fresh weight 3 g.) was used as the synthesis organ. It was depleted of starch by keeping it in the dark 48 hours before exposure to C<sup>14</sup>O<sub>2</sub> and light.

The exposure was carried out as described by Varner (39) using the same chamber. The amount of BaC<sup>14</sup>O<sub>2</sub> used was 2.6 mg. containing



20 microcuries of  $C^{14}$  per mg. and 200 mg. containing only a very small amount of radioactivity.\*

Exposure was continued for 24 hours. The leaf was then frozen with dry ice, ground in a mortar and extracted with 80% alcohol. The solubles obtained were discarded.

Starch was extracted from the remaining material as outlined by Pucher et al. (30) with hydrolysis according to Putman and co-workers (31).

Just before hydrolysis 1 g. of dextrose was added to the extracted starch to act as a carrier. After hydrolysis the solution was purified by running it through a cation and an anion exchange column (Suolite C-3 and Duolite A-3). The resulting solution (300 ml.) was concentrated by slow evaporation to about 40 ml. 7 g. of dextrose was added as a diluent and the resulting solution quick frozen.

An unsuccessful attempt was made to crystallize the material by freeze drying. Instead of crystals a clear amorphous mass was obtained which crystallized readily when dehydrated with absolute alcohol. After desiccation over  $CaSO_4$  the weight of glucose obtained was 8.006 g.

(c) Determination of Organic Carbon:

Organic carbon was determined by wet oxidation using Van Slyke's oxidizing mixture (38) and a combination of the apparatus used by McCready and Hassid (20) and Gortner (13).

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\*The  $BaC^{14}O_3$  used in this investigation was supplied by The Oak Ridge National Laboratory of the United States Atomic Energy Commission.

The CO<sub>2</sub> evolved from unlabeled material was determined gravimetrically by absorption on ascarite. CO<sub>2</sub> from labeled material was collected in NaOH using a bead tower and determined either gravimetrically or volumetrically as described previously.

(d) Determination of Radioactivity:

In this study radioactivity measurements were made on only one material, BaC<sup>14</sup>O<sub>3</sub>. It was plated on an aluminium dish approximately 3.5 cm. in diameter at the bottom.\* A quantity large enough to give maximum thickness (20 mg./cm.<sup>2</sup>) was ground in a small mortar. Two or three drops of water were added and thoroughly mixed with the powder followed by enough 95% alcohol to give a slurry, readily spread with an eye dropper. The slurry was then transferred to the aluminium dish and spread evenly on the bottom. Reproducibility of area and even thickness of the plate depended on having the proper amount of slurry at the correct consistency.

The plates were slowly dried on a hot plate and stored in air tight containers. The weight of the BaC<sup>14</sup>O<sub>3</sub> plate was recorded and corrections for self absorption made as explained by Varner (39). His BaC<sup>14</sup>O<sub>3</sub> graph was used to obtain the correction. All counts were made on a Landsverk Model L-75 Electrometer.

The glucose experiment material was counted on one instrument\*\* and the alfalfa material on another. The latter was found to contain

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\*Kauffman and Lattimer Cat. No. 10676.

\*\*Loaned by the Agricultural Biochemistry Department.

a considerable error due to nonlinearity of the scale. To correct for this a number of samples of different activities were counted across the scale. Using the method of least squares (33) the best fitting straight line was drawn through these points. Three such lines are shown in figure 6 and the summarized data in table 13. The scale error between 0 and 100, on an average of 13 determinations, was 16.9%. This error was distributed over the scale as illustrated in table 14. Point 80 on the scale was taken as the accepted value and all readings corrected to it.

(e) Determination of Total Nitrogen:

The method adopted was that of Murneck and Heinze (19) except that the determination was adapted to micro-Kjeldahl procedure and the catalyst used was selenium and  $\text{Na}_2\text{SO}_4$ . Distillation was carried out on a Pregl-Barnas-Wagner Micro-Kjeldahl Distilling Apparatus and the ammonia was collected in 2% boric acid using a methyl red-bromecresol green mixed indicator.

(f) pH Determination:

pH was measured with a glass electrode. Measurements were made on a 1 to 3 soil-water suspension.

(g) Extraction of Soluble Materials from Alfalfa:

To obtain the insoluble fraction for the alfalfa incubation experiment enough complete alfalfa, 9 g., 29 parts untagged 1 part tagged, were weighed into a 500 ml. flask and shaken 2 hours on an elbow shaker. The alfalfa was transferred to an extraction thimble and extracted with 80% alcohol in a continuous extraction apparatus.

for 3 hours. The contents of the thimble were dried and weighed to determine the percentage of insolubles. The material was transferred to a flask and mixed again by shaking for 2 hours. Portions of this material (1.6 g.) were weighed into each of three of the incubation flasks.

### EXPERIMENTAL

#### 1. Glucose experiment:

Two glucose incubation experiments were carried out. The first duplicated the second in all respects with the exception that unlabeled dextrose was used in place of the labeled glucose. The preliminary experiment indicated that the plan was sound and methods practical.

Three treatments replicated four times were applied in the tagged glucose experiment. These were: (1) no glucose, (2) 0.6 g. glucose and (3) 1.2 g. glucose per 100 g. of air-dry muck. The amount of activity applied was approximately 0.5 and 1.0 microcuries respectively. These applications of glucose were chosen to simulate the soluble material of 2.4 g. and 4.8 g. of dried rye grass on the assumption that it contained 25% soluble material. These latter treatments simulate field applications of 6 and 12 tons per acre respectively.

All flasks received 3.4 mg. of  $\text{NH}_4\text{Cl}$ , 150 mg. of  $\text{K}_2\text{CO}_3$  and 140 mg. of  $\text{KH}_2\text{PO}_4$ . These salts approximate a field application of 2 tons per acre of a 3-9-18 fertilizer.

The fertilizer salts were ground in a mortar and kept in stoppered containers. The glucose was also ground but contained a number of small lumps. As this material was very hygroscopic the weighing time was set at 3 minutes and weights of 0.6 or 1.2 g.  $\pm$  10 mg. were accepted.

The fertilizer, about one half the soil and glucose were weighed into the incubation flask, stoppered and mixed by rotating the flask. The remaining soil and glucose was then added and mixed in the same way.

The soil was brought to 180% moisture, basis air-dry soil, by weighing 180 g. of water into each flask. The flasks were immediately placed on the aspiration manifold in a constant temperature room and collection of CO<sub>2</sub> begun. Temperature was maintained at 28° C.

Determination of CO<sub>2</sub> evolved was made every half day for 3 days, then every day for 6 days and then at longer intervals as the rate of evolution decreased. The BaCl<sup>4</sup>O<sub>3</sub> collected was stored in vials and counted as time permitted. Total nitrogen and pH were determined before and after incubation. Collection of CO<sub>2</sub> was carried out continuously for 46 days. The flasks were aerated continuously for another 46 days without determination of the CO<sub>2</sub>. Collection was then carried out again for a two week period after which incubation was discontinued.

## 2. Wheat straw vs. rye grass experiment:

Portions of the same soil sample used in the glucose incubation experiment were used in this one. A similar fertilizer application was made except that the following salts were used, 100 mg. KNO<sub>3</sub> and 72 mg. KH<sub>2</sub>PO<sub>4</sub>.

Three treatments replicated three times were applied: a check treatment, wheat straw, and rye grass. These materials were added to give a 1% application of carbon i.e., 2.22 g. wheat straw and 2.44 g. rye grass per 100 g. air-dry soil. They were first ground in a small Wiley Mill using a number 20 screen and then mixed similarly to the glucose in the preceding experiment. Before adding water to bring the moisture content to 180%, 10 g. of the material from each flask was taken out for pH and total nitrogen determinations.

CO<sub>2</sub> was collected at intervals as required for 98 days. Total nitrogen was determined before and after incubation.

### 3. Comparison of tagged and untagged alfalfa:

In the experiment that follows the labeled alfalfa was diluted with unlabeled alfalfa. This dilution served two purposes. There was not enough labeled material to make an application of 2.4 g. per 100 g. of air dry muck. This rate of application had already been chosen and applied in the glucose experiment. To apply it in this experiment would have required the reduction of the amount of soil per flask to less than 40 g. The dilution made it possible to keep the weight of the incubating sample at 100 g. and at the same time bring the activity of the evolved CO<sub>2</sub> into a convenient reading range. This tagged vs. untagged experiment was carried out to determine if there was any difference in the biological breakdown of the two materials as measured by CO<sub>2</sub> evolution.

The soil sample was the same as that used in the alfalfa incubation experiment. Fertilizer salts and water were added in the same proportions. Twenty g. of air dry muck were weighed into each of ten flasks. Five flasks received 480 mg. tagged alfalfa (approximately 50 microcuries of C-14) and five received 480 mg. untagged alfalfa. CO<sub>2</sub> was collected daily for five days. Two flasks were taken off at that time for another experiment and the remaining eight flasks incubated another eight days with CO<sub>2</sub> collected at varying intervals.

#### 4. Alfalfa insolubles vs. complete alfalfa experiment:

This experiment involved three replications of three treatments: a check treatment, 2.4 g. of complete alfalfa (approximately 8 microcuries of C-14), and the insoluble material (1.6 g.) from 2.4 g. of alfalfa (approximately 5.4 microcuries of C-14). The soluble material was extracted as outlined under Methods and Materials. Fertilizer salts were added as in the straw vs. rye grass experiment.

For reasons given above, the labeled alfalfa was diluted. The diluent used was the untagged alfalfa described previously under Methods and Materials. By incubation of 2 or 3 different dilutions and counts of the evolved CO<sub>2</sub> it was found that a dilution of 29 parts untagged to 1 part tagged gave CO<sub>2</sub> that could be accurately counted.

The proper proportions of the complete alfalfa were weighed into three of the flasks and shaken on an elbow shaker for two hours



with frequent tapping and turning of the flask to insure a good mix. The fertilizer salts, the same as in the straw-rye grass incubation, and 100 g. of air-dry soil were weighed into the flasks and shaken for 1 hour on the elbow shaker. Since the alfalfa contained 67% insolubles, 1.6 g. of the extracted alfalfa was added to each of three flasks. Fertilizer and soil were added as with complete alfalfa and the flasks shaken for an hour. Water was added to the flasks as in the previous experiments and they were then connected to the aspiration apparatus. CO<sub>2</sub> was collected at convenient intervals, measured volumetrically and the activity counts made on BaC<sup>14</sup>O<sub>3</sub> as in the glucose experiment. Incubation was carried out for 70 days. Total nitrogen and pH were measured before and after incubation.

## TABULATION OF RESULTS

Note: In tables 1, 2, and 3 the calculated check column is obtained from activity calculations. These calculations are summarized in appendix tables 18, 19, and 20. The measured check is the average CO<sub>2</sub> evolved from the check treatment for the period indicated.

Table 1. Priming Effect 0.6 g. Glucose/100 g. Air Dry Muck

Day	Calculated Check Mg. CO <sub>2</sub>	Measured Check Mg. CO <sub>2</sub>	Priming Mg. CO <sub>2</sub>	Rate of Priming Mg. Carbon per Day
1	150	197	-47	-12.8
2	119	123	- 4	- 1.1
3	111	106	5	1.4
4	94	89	5	1.4
5	61	55	6	1.6
6	54	52	2	0.5
7	51	43	8	2.2
8	53	50	3	0.8
9	49	43	6	1.6
11	82	74	8	1.1
13	63	67	- 4	- 0.6
16	113	109	4	0.4
19	133	122	11	0.6
25	218	196	22	0.1
32	201	177	24	0.9
46	<u>258</u>	<u>239</u>	<u>19</u>	0.3
Total	1810	1742	68 = 19 mg. carbon	

Table 2. Priming Effect 1.2 g. Glucose/100 g. Air Dry Soil

Day	Calculated Check Mg. CO <sub>2</sub>	Measured Check Mg. CO <sub>2</sub>	Priming Mg. CO <sub>2</sub>	Rate of Priming Mg. Carbon per Day
1	140	197	-57	-15.5
2	104	123	-19	- 5.2
3	109	106	3	0.8
4	89	89	0	0.0
5	64	55	9	2.5
6	62	52	10	2.7
7	57	43	14	3.8
8	61	50	11	3.0
9	50	43	7	1.9
11	81	74	7	1.0
13	76	67	9	1.2
16	129	109	20	1.8
19	134	122	12	1.1
25	225	196	29	1.3
32	201	177	24	0.9
46	<u>260</u>	<u>239</u>	<u>21</u>	0.4
Total	1842	1742	100 = 27.3 mg. carbon	

Table 3. Priming Effect

Day	1.6 g. Alfalfa Insolubles				2.4 g. Complete Alfalfa			
	Calcu- lated Check Mg.CO <sub>2</sub>	Measured Check Mg. CO <sub>2</sub>	Priming Mg. CO <sub>2</sub>	Rate of priming Mg. Car- bon/Day	Calcu- lated Check Mg.CO <sub>2</sub>	Measured Check Mg. CO <sub>2</sub>	Priming Mg. CO <sub>2</sub>	Rate of priming Mg. Car- bon/Day
1	55	55	0	0	81	55	26	7.1
2	56	39	17	4.6	118	39	79	21.5
3	51	24	27	7.4	74	24	50	13.6
4	59	40	19	5.2	62	40	22	6.0
6	91	56	35	4.8	106	56	50	6.8
8	72	51	21	2.9	123	51	72	9.8
10	97	54	43	5.9	132	54	78	10.6
12	149	60	89	12.1	108	60	48	6.5
14	148	54	94	12.8	62	54	8	1.1
16	81	41	40	5.5	45	41	4	0.5
20	102	86	16	1.4	81	86	-5	-0.3
25	125	118	7	0.4	110	118	-8	-0.4
32	167	141	26	1.2	143	141	2	0.1
40	201	174	27	0.9	170	174	-4	-0.1
48	171	186	-15	-0.5	165	186	-21	-0.7
60	266	292	-26	-0.6	259	292	-33	-0.8
70	<u>179</u>	<u>215</u>	<u>-36</u>	<u>-1.0</u>	<u>135</u>	<u>215</u>	<u>-80</u>	<u>-2.2</u>
Total	2070	1686	384		1974	1686	288	

384 Mg. CO<sub>2</sub> = 105 Mg. C.288 Mg. CO<sub>2</sub> = 79 Mg. C.

Table 4. Summary of Priming Effects

Treatment	Mg. Carbon Lost	Incubation Period
		Days
0.6 g. Glucose	19	46
1.2 g. Glucose	27	46
1.6 g. Alfalfa insolubles	105	70
2.4 g. Complete alfalfa	79	70

Table 5. Percentage of Added Material Retained

Treatment	Carbon added Mg.	Calculated loss of added carbon	% Carbon retained	Days Incuba- tion
0.6 g. Glucose	240	185	22.9	46
1.2 g. Glucose	480	391	18.5	46
1.6 g. Alfalfa insolubles	661	314	52.5	70
2.4 g. Complete alfalfa	953	573	39.9	70

Table 6. Total Carbon Evolution Straw-Rye Grass Incubation  
Mg. Carbon/100 g. Air Dry Muck

Treatment	Replication			Mean
	1	2	3	
Check	881	880	872	878
Wheat straw	1508	1514	1547	1523
Rye grass	1494	1515	1463	1491
L.S.D. (P = 0.05) = 39				

Table 7. Total Carbon Evolution Alfalfa Incubation  
Air Dry Muck

Treatment	Replication			Mean
	1	2	3	
Check	470	473	451	465
1.6 g. Insolubles	886	886	885	886
2.4 g. Alfalfa	1122	1130	1104	1119
(Standard error of the mean = $\pm$ 10)				
(Incubation period = 70 days)				

Table 8. Total Carbon Evolution Glucose Incubation  
Mg. Carbon/100 g. Air Dry Muck

Treatment	Replication				Mean
	1	2	3	4	
Check	480	482	472	465	475
0.6 g. Glucose	679	673	680	694	682
1.2 g. Glucose	879	839	882	876	869

(Standard error of the mean =  $\pm$  6.3)

(Incubation period = 46 days)

Table 9. Net Loss of Carbon

Treatment	Total carbon lost. Mg.	Carbon added Mg.	Net loss	Days Incubated
Check	475	-	475	46
0.6 g. Glucose	682	240	442	46
1.2 g. Glucose	869	480	389	46
Check	878	-	878	98
Wheat straw	1523	1000	523	98
Rye grass	1491	1000	491	98
Check	465	-	465	70
1.6 g. Alfalfa insolubles	886	661	225	70
2.4 g. Complete alfalfa	1119	953	166	70



Table 10. Labeled vs. Unlabeled Alfalfa Incubation  
Carbon Lost Mg./20 g. Air Dry Muck

Day	Tagged				Untagged					
	A	B	C	D	G	H	I	J		
1	26.0	27.8	26.9	22.6	103.3	24.7	23.9	24.1	24.5	97.2
2	18.5	18.2	18.1	18.1	72.9	22.9	22.3	23.3	22.6	91.1
3	11.8	11.6	10.0	11.1*	44.5	13.6	14.3	14.1	13.6	55.6
4	8.9	9.1	9.1	9.2	36.3	10.1	10.3	9.8	10.0	40.2
5	9.6	9.7	9.7	9.7	38.7	11.2	11.3	10.5	10.7	43.7
6	5.7	5.7	5.6	5.8	22.8	7.3	7.3	7.0	7.2	28.8
8	13.5	14.1	13.0	13.6	54.2	18.5	18.4	17.2*	18.3	72.4
10	13.2	13.1	12.9	12.6	51.8	13.2	13.6	13.6	13.5	53.9
13	10.6	9.9	10.3	10.0	40.8	11.1*	9.6	9.8	9.7	40.2
	117.8	119.2	115.6	112.7	465.3	132.6	131.0	129.4	130.1	523.1

Variance Due to	Degree of Freedom	Sum of Squares	Variance	F
Treatment	1	46.40	46.40	105.4**
Days	8	2204.20	275.53	626.2
Treatment - Days	8	16.58	8.32	18.9**
Error	51	22.40	0.44	
Total	68			

\* Missing values

\*\* Significant at 0.01% level

Table 11. Total Nitrogen  
Mg. N/100 g. Oven Dry Muck

Treatment	Before Incubation	After Incubation
Check	3.11	3.14
0.6 g. Glucose	3.11	3.17
1.2 g. Glucose	3.11	3.23
(L.S.D. (P = 0.05) = 0.11 mg.)		
Check	3.06	3.12
Wheat straw	3.00	3.11
Rye grass	3.12	3.18
(L.S.D. (P = 0.05) = 0.10 mg.)		
Check	3.34	3.42
Alfalfa insolubles	3.40	3.08
Complete alfalfa	3.38	3.40
(L.S.D. (P = 0.05) = 0.57 mg.)		

Table 12. pH Determinations

Treatment	Before Incubation	After Incubation
Check	4.9	4.6
0.6 g. Glucose	4.9	4.6
1.2 g. Glucose	4.9	4.6
Check	4.5	4.3
Alfalfa insolubles	4.5	4.2
Complete alfalfa	4.5	4.2

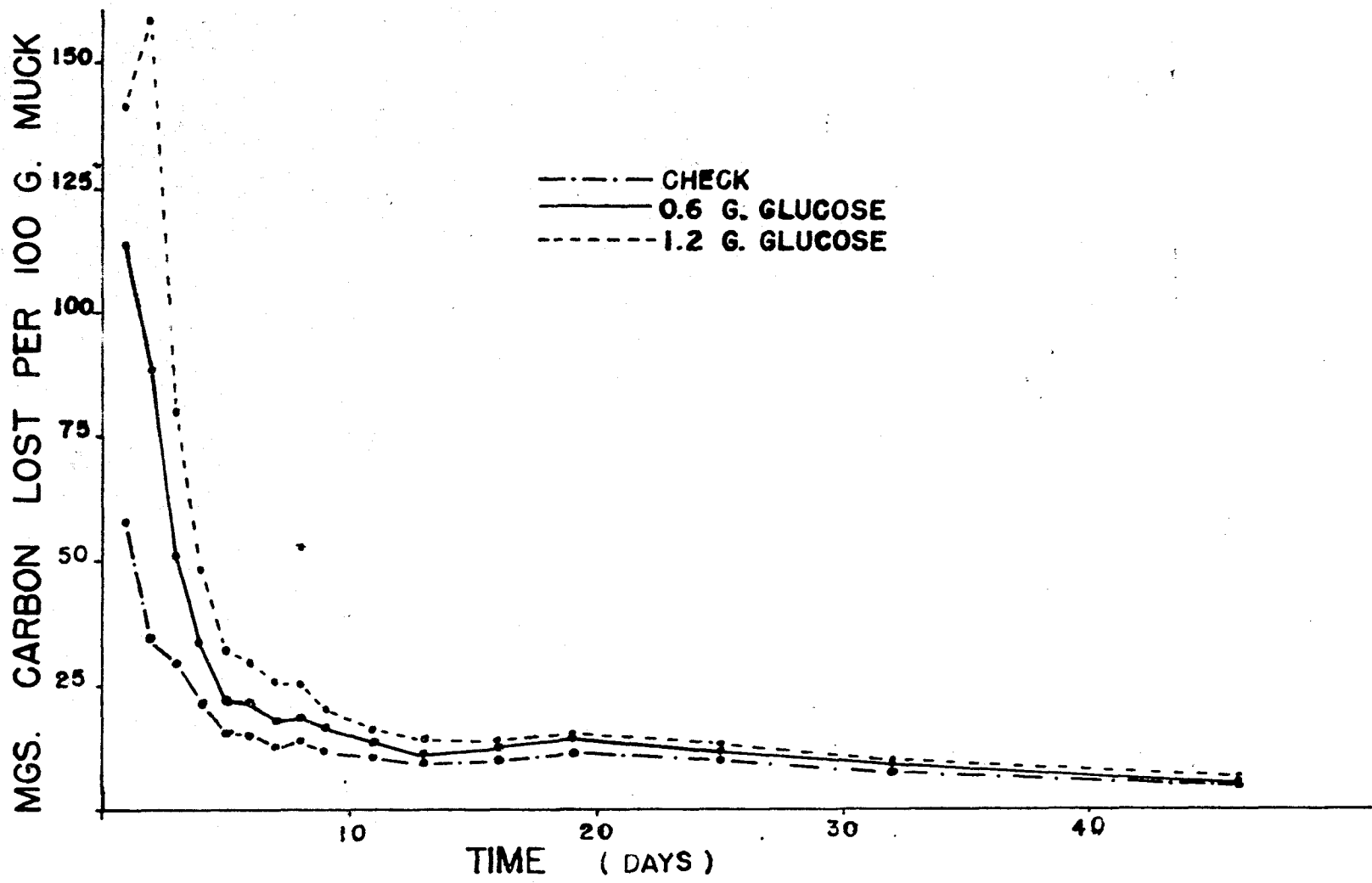


FIGURE 1. RATE OF LOSS OF CARBON AS CO<sub>2</sub> FROM GLUCOSE TREATMENTS.

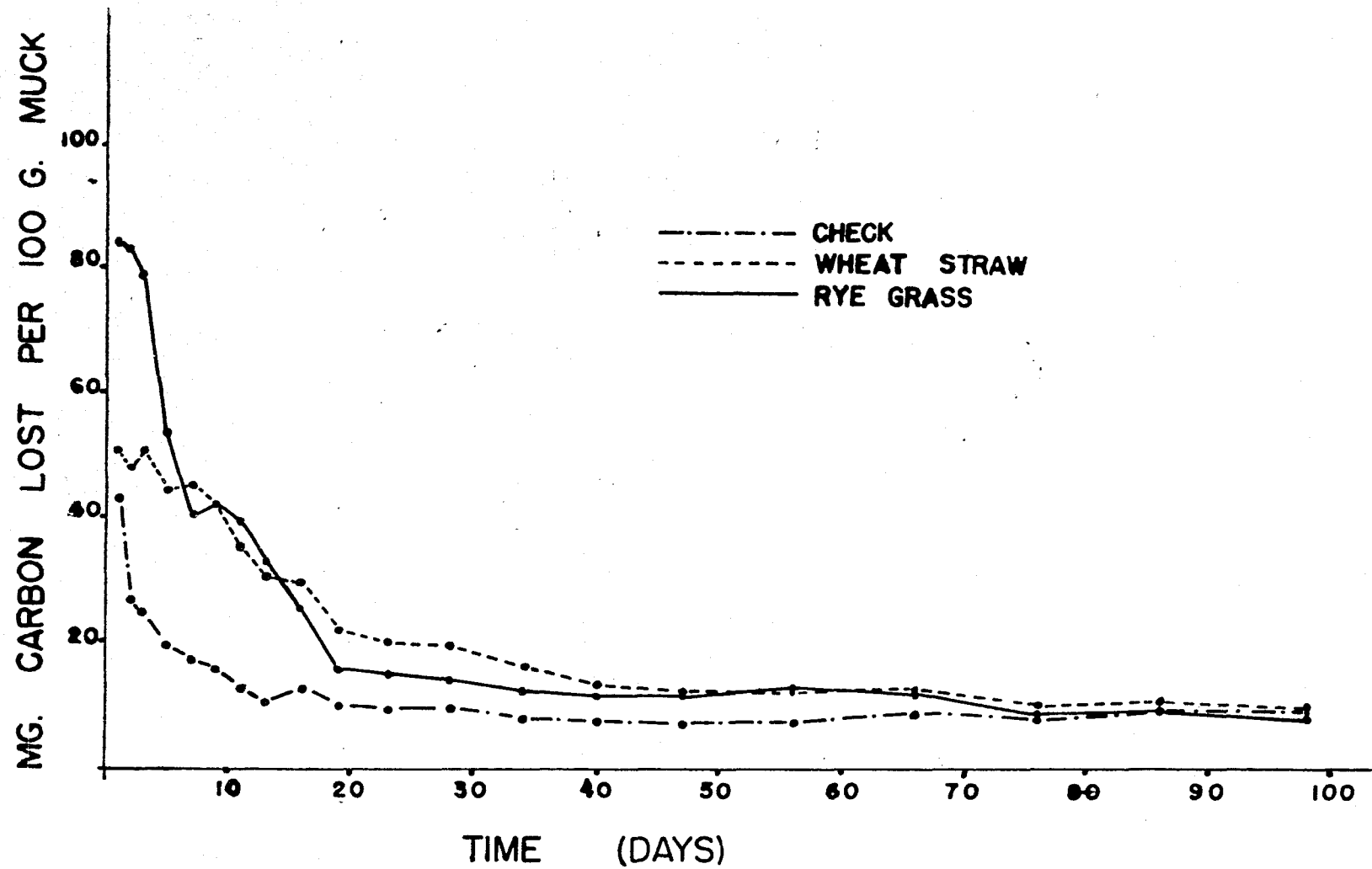


FIGURE 2. RATE OF LOSS OF CARBON AS CO<sub>2</sub> FROM WHEAT STRAW VS. RYE GRASS TREATMENTS.

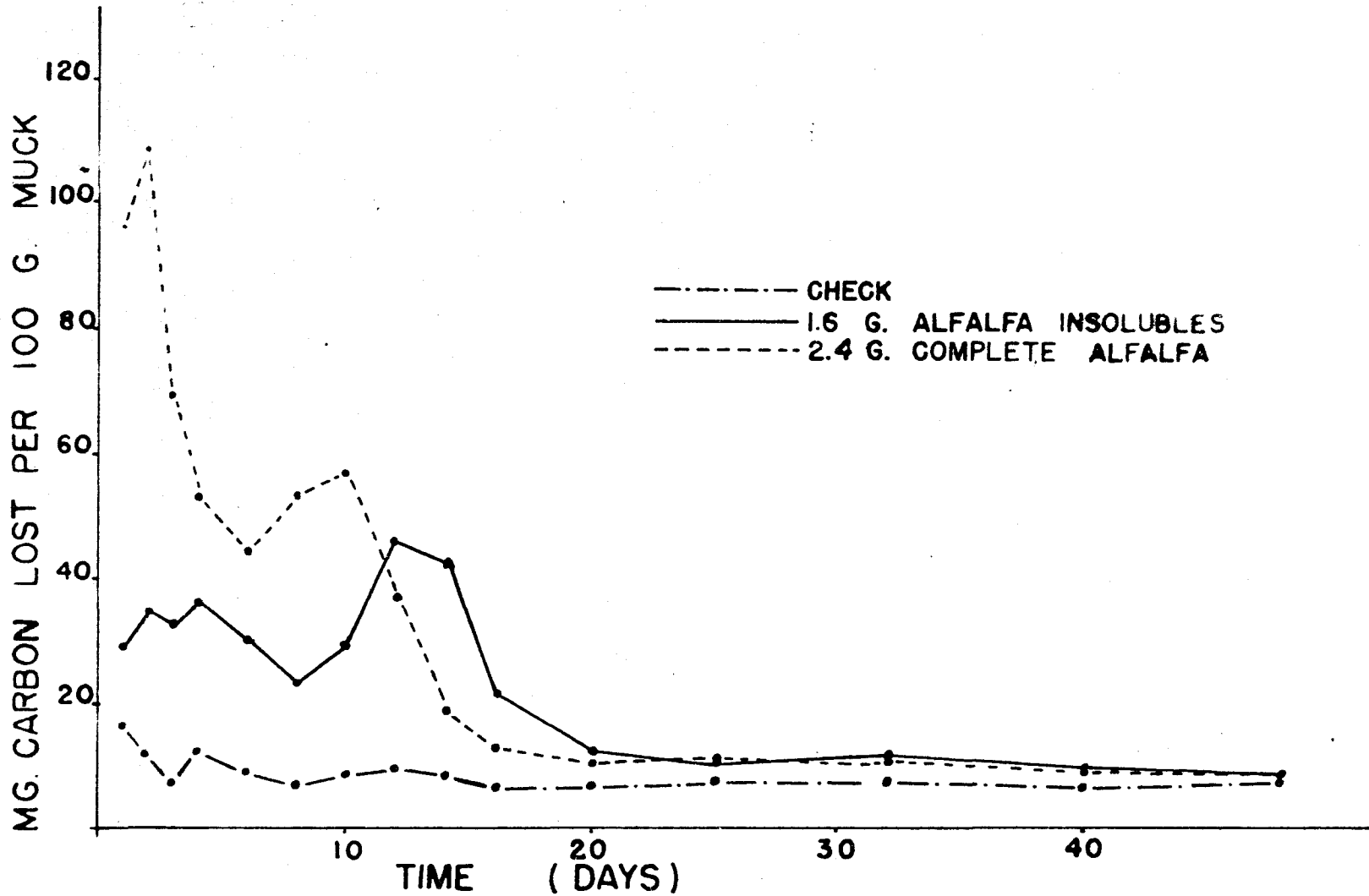


FIGURE 3. RATE OF LOSS OF CARBON AS CO<sub>2</sub> FROM ALFALFA TREATMENTS.

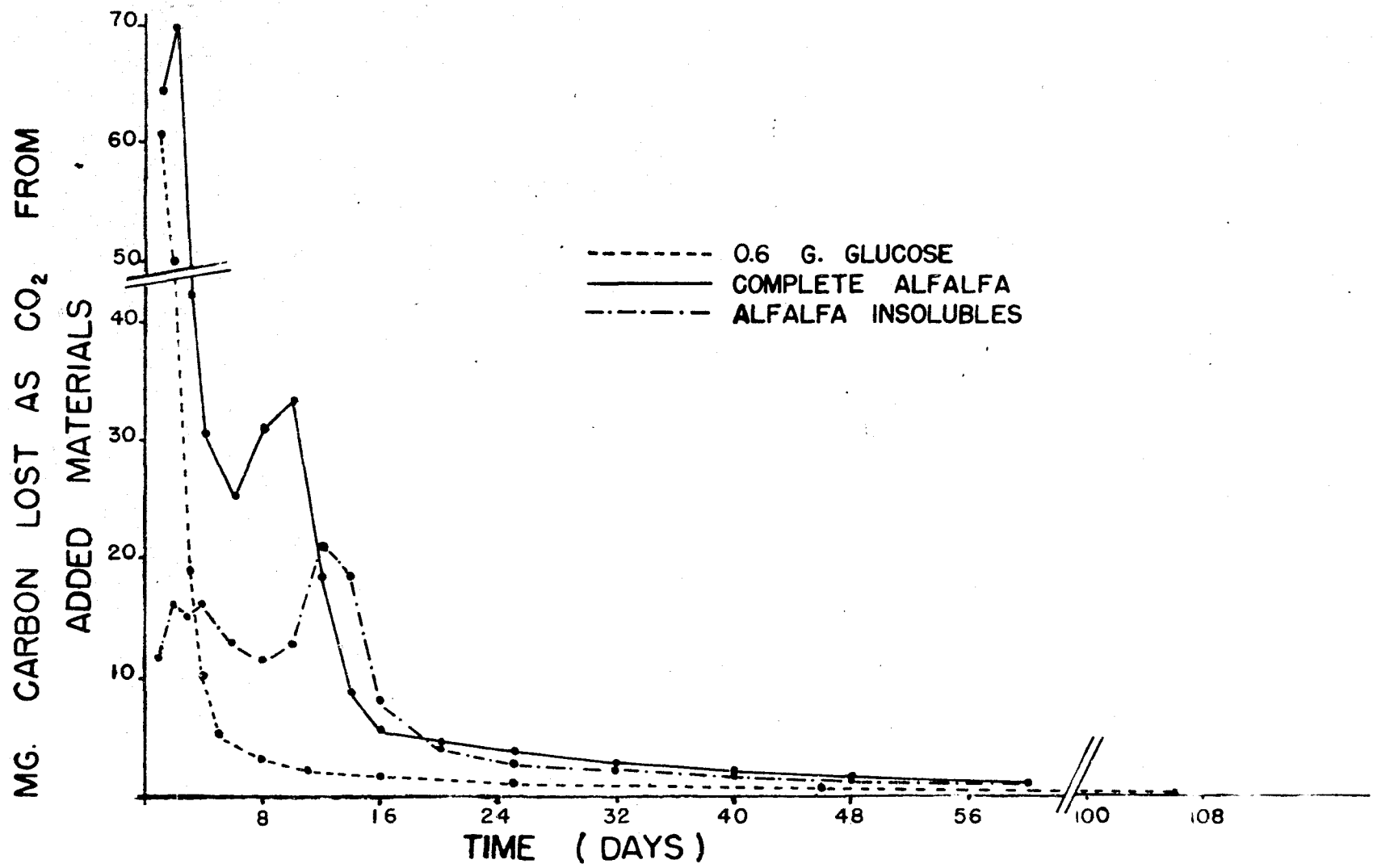


FIGURE 4. RATE OF LOSS OF CARBON FROM ADDED MATERIALS.

RATE OF PRIMING MG. CARBON PER  
100 G. MUCK

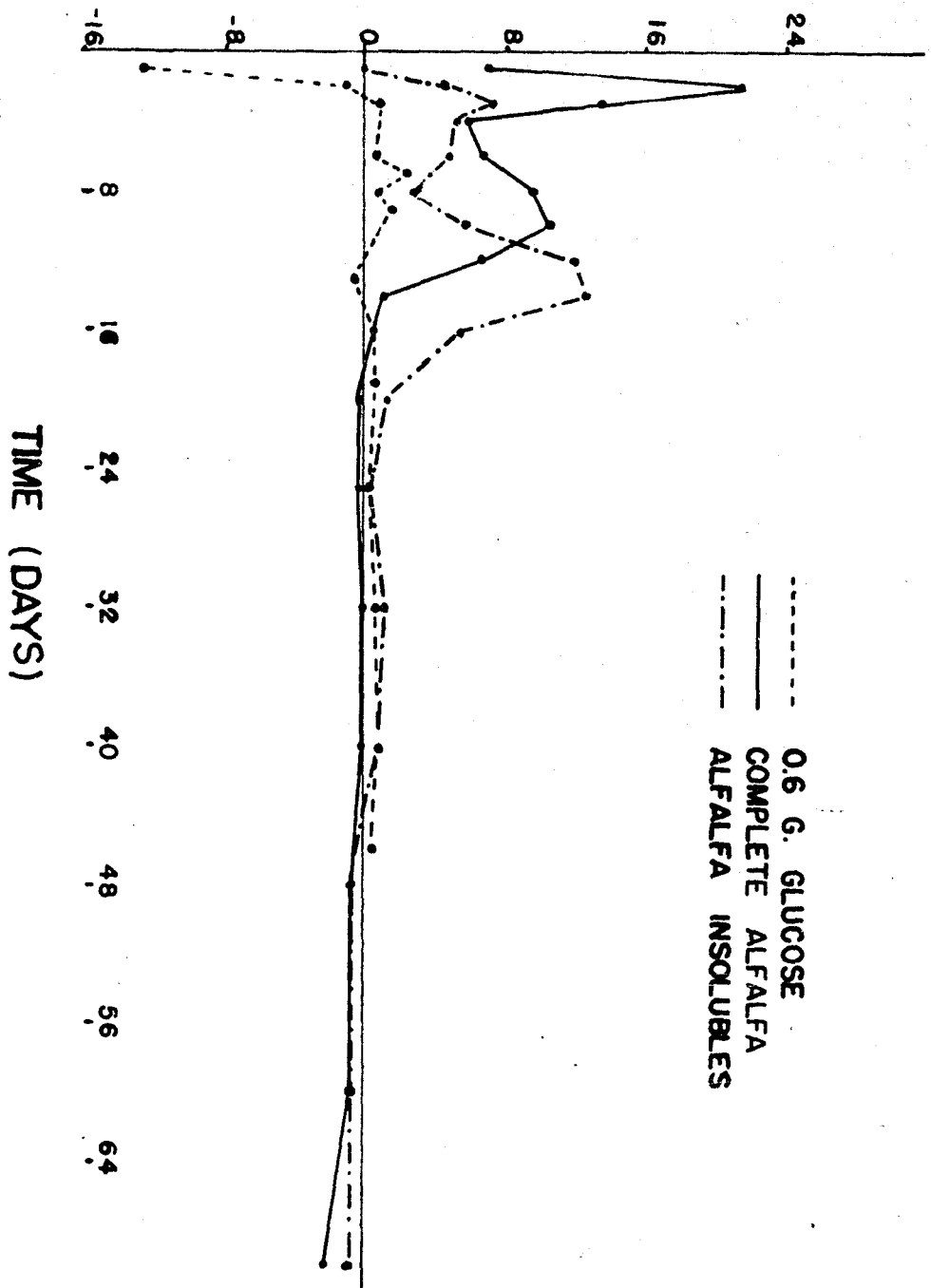


FIGURE 5. PRIMING ACTION - RATE OF LOSS OF NATIVE ORGANIC MATTER.

## DISCUSSION

With the exception of the graphs illustrating rate of total carbon loss, which are presented on the basis of 100 g. of oven dry soil, all treatments and all results are based on air dry soil. In all experiments 100 g. of air dry soil contained  $90 \pm 1$  g. oven dry soil.

All  $\text{BaC}^{14}\text{O}_3$  counts from  $\text{C}^{14}\text{O}_2$  collection in the glucose incubation experiment were compared to the  $\text{BaC}^{14}\text{C}_3$  obtained from chemical oxidation of the glucose. Three plates were made from the  $\text{BaC}^{14}\text{O}_3$  of one glucose oxidation. Eleven separate counts of these plates were made and the results averaged. Using the standard sample mentioned in table 13 this count was converted from divisions per hour to counts per minute.

Maximum activity of  $\text{BaC}^{14}\text{O}_3$  from glucose oxidation -  
154.4 Div./Hr.

Calibration of instrument - 1 Div./Hr. - 46.4 c./min.

Maximum activity of  $\text{BaC}^{14}\text{O}_3$  - 7160 c./min.

$\text{BaC}^{14}\text{O}_3$  arising from  $\text{CO}_2$  collection in the alfalfa incubation experiment was compared to the  $\text{BaC}^{14}\text{O}_3$  from the chemical oxidation of either the insoluble fraction or the complete alfalfa depending on which treatment it arose from. Table 13 appendix shows that at 80 on the scale the standard sample read 38.5 Div./hr. Therefore 1 Div./hr. is equivalent to 57 c./min. The average of the five readings for insolubles oxidation is 426.4 Div./hr. or 24,310 c./min. Likewise the count of the complete alfalfa is 22,610 c./min.



Figures 1 and 5 show that the rate of carbon lost from glucose treated soils quickly reached a peak and dropped down again in about 4 days. The rate of loss of carbon added as glucose follows the same pattern. However, not all the added carbon was lost at the end of 46 days or even at the end of 106 days. At the end of 46 days only about 80% of the added carbon had been lost. The remaining 20% was probably entirely incorporated in synthesized biological tissue. Gray (14), with a much smaller application of glucose to an inorganic soil, found no glucose as such remained after the third day. At this time only 41% of the theoretical amount of CO<sub>2</sub>, priming action not considered, had been evolved. It is possible that all the glucose added to the muck was decomposed in the first week.

Tables 1 and 2 show that instead of a priming action during the first two days there was actually less soil organic matter lost than in the check. The effect was greater with the larger glucose application. This would indicate that the added soluble material was utilized as an energy source in preference to the soil organic matter. This initial effect was so large that it almost counteracted the small priming effect that followed reaching a peak at about the eighth day.

Since this peak occurred about the estimated time of the glucose disappearance, it would follow that any priming effect of the soluble material is brought about by lack of a suitable substrate other than the soil organic matter itself. The reason the

priming action was so small may be due to the fact that the population built up when soluble material is added is highly selective as to substrate.

The percentage of total nitrogen rose slightly on incubation indicating that the carbon loss was greater than the nitrogen loss. This apparent nitrogen increase was statistically significant at the 0.05% level only in the high glucose treatment. The pH of the soil fell slightly.

In the wheat straw vs. rye grass experiment, table 6 shows that there is no significant difference in the amount of carbon lost due to the two treatments. Pinck et al. (27) found no consistent significant difference in carbon lost when young and mature millet were added to two inorganic soils. Those that were significantly different gave in all cases a greater carbon loss for the younger materials.

Figure 2 shows the effect of the higher content of soluble material and nitrogen in the rye grass in that it lost considerably more carbon in the first few days than the straw did. From the fifteenth to the fiftieth day, however, the straw treated soil lost more.

It is to be expected that the supply of nitrogen was not a factor here since there was sufficient added to bring the C/N ratio of the straw to 21:1 and also there would be nitrogen mineralized from the soil organic matter (carbon nitrogen ratio 16:1) as shown by Broadbent and Norman (8). As in the glucose experiment there was

a tendency for the percentage of total nitrogen to increase on incubation. This increase was significant at the 5% level only in the straw treatment.

The net effect after 98 days of incubation under these conditions was an increase in organic matter over the untreated soil. It should be pointed out that neither of these treatments resulted in an increase in the amount of organic matter of the muck but rather a smaller net loss than from the untreated soil. This was true of all experiments including the glucose though the effect was much smaller there.

Analysis of the complete data in the tagged vs. untagged alfalfa incubation, table 10, gives a highly significant treatment difference. It may be concluded, then, that the labeled alfalfa does not lose carbon as  $\text{CO}_2$  at the same rate as the unlabeled material. This difference could be caused by the radioactivity or by a difference in organic composition of the alfalfa brought about by the slightly different environment under which the two materials were cultured. Since the treatment x day interaction was also highly significant, it is possible that the difference eventually disappeared as indicated by the data. From the second to the eighth day there is a consistently greater loss of the untagged material averaging about 18%. It is then quite possible that the calculated priming effect in the alfalfa vs. insoluble experiment is too large by that amount during that period.

Figure 3 shows a high initial peak of carbon loss from the complete alfalfa treatment similar to the rye grass treatment mentioned previously. These peaks are similar to the one produced by treatment with glucose. They must, then, represent the rapid initial loss of the soluble fraction of the added material. Neither the wheat straw or the alfalfa insolubles give rise to this high peak. This is to be expected as they do not contain an appreciable soluble fraction. Both straw and the alfalfa insolubles have a small peak in this period. This could be due to the presence of some readily available part of the insoluble material and to the presence of a small amount of solubles in the soil as indicated by the check treatment.

The significant points illustrated by the curves are the presence of secondary peaks and the fact that, beginning about the twelfth day, the rate of carbon loss from the insoluble treated soil exceeds that of the complete alfalfa treatment. This increase is quite marked for about a week and then the rates of loss become almost equal. The change in rank takes place at the same time the rate of carbon loss for insolubles reaches a peak. This coincides also with the maximum rate of priming action due to the insolubles, table 3, figure 5. The complete alfalfa priming action also reaches a peak at the same time the total carbon lost for alfalfa reaches its secondary peak. It is possible that these secondary peaks are at least partially due to the increase in priming action. That other factors are involved is indicated in that the complete alfalfa peak is larger than the one for insolubles whereas its priming action

peak is smaller.

The maximum number of microorganisms does not coincide with the initial CO<sub>2</sub> peak but comes one or two weeks later (15, 36, 37). In a peat soil Vandecaveye (36), and in a podsol, Gray and Taylor (14), showed that when organic matter is added to the soil there is a large decrease in the number of bacteria in the first 3 or 4 days accompanied by a steady continuing increase in the number of fungi. It is possible that the secondary peaks are due to this increased fungal population. It is also probable that the priming peaks develop at this time for the same reason. Fungi are better able to utilize complex material than bacteria and as their number increases they come in contact with a larger amount of soil organic matter.

Table 3 and figure 5 show that both treatments eventually caused a smaller carbon loss from the soil organic matter than in the untreated check. This is much more evident in the complete alfalfa treatment and is the reason for the greater total priming action of the insoluble alfalfa. It will be noted that the complete alfalfa treatment caused a greater total priming action up to the thirty second day. Thereafter it was largely negative. The probable explanation of this greater carbon loss from the soil lies, as postulated in the glucose treatment, in availability of substrate. Pinck (24) suggested that this might take place. It would then follow that the reason it is greater in the complete alfalfa treatment is due to a different population - one less able to utilize the

complex material of the soil organic matter.

Figure 5 shows that there was a difference in the priming effect of the solubles when added as glucose and when added as a fraction of plant material such as the complete alfalfa. In the light of the other data and the foregoing explanation it would be expected that the rate of priming for complete alfalfa would be quite low in the beginning. Instead there is a pronounced peak for the second and third days. This peak would indicate that the breakdown of the solubles in the alfalfa caused considerable priming effect, a conclusion not in keeping with the glucose data.

It should be pointed out that, as mentioned previously, the calculated rate of priming for this period is approximately 18% too high due to the differential rate of decomposition of the tagged and untagged material.

Table 3 shows that this peak is due to both an increase in the calculated check and a decrease in the measured check. This two day decrease is unusual. If it was not present the rate of priming at this point would be reduced another 6% on the second day and about 50% on the third day.

It should be pointed out again that the soil for the alfalfa incubation and the soil for the glucose incubation were collected under entirely different circumstances. Also, through an error, the glucose treated soil received its nitrogen fertilizer as  $\text{NH}_4\text{Cl}$ , the alfalfa treated soil as  $\text{KNO}_3$ . It does not seem logical that these differences should cause such a marked difference in priming action.

The alfalfa soil was probably low in soluble nitrogen and soluble organic matter but sufficient nitrogen was added to supply the immediate needs of the microorganisms. The lower content of soluble substrate in the alfalfa incubated soil would not be a factor since there is plenty of available added substrate. A low soluble substrate in the soil at this stage of incubation would be expected to decrease rather than increase priming.

This leaves the added substrate or an unaccountable activity dilution effect to explain the discrepancy. The dilution has been shown to be somewhat responsible. Perhaps its magnitude was underestimated. Unfortunately, no breakdown of the nitrogen fraction of the alfalfa or of its organic composition was made so it is impossible to determine whether these may have been responsible.

In order to make a sound explanation of these priming effects more data are required. Biological counts should be made so that changes within the population could be correlated with changes in the loss of soil organic materials. The nitrogen fraction of both soil and added materials should be thoroughly defined and a knowledge of the organic composition would be helpful.

Not considering priming, table 9 , the amount of carbon lost in excess of the check, expressed as a percentage of added carbon, is 64% for the insoluble and 69% for the complete alfalfa. If they had been added in equal amounts, assuming that quantity has little effect on the percentage loss, the insolubles would result in essentially the same carbon loss as the complete alfalfa. This is borne out by

the rye grass vs. wheat straw experiment and by Pinck (27) using mature vs. immature manuring materials. This takes place despite the fact that more of the carbon of the added insolubles than of the complete alfalfa remains in the soil, table 5, and can only be explained in the greater priming effect of the insolubles.

There was no tendency toward a percentage increase in total nitrogen as in the preceding experiments. There was a slight decrease in pH.



## SUMMARY

1. A study was made of the effect of certain added organic materials on the breakdown of the organic matter of a Rifle Peat. This breakdown has been referred to as a priming action.
2. Methods are outlined for the use of carbon-14 in a study of this type.
3. Glucose labeled with carbon-14 was incubated with the soil to determine the priming effect of soluble material.
4. Alfalfa evenly labeled with carbon-14 and the insoluble fraction of the alfalfa were incubated with the soil to determine the priming effect of this plant material and of fibrous, insoluble material.
5. Rye grass and wheat straw were added to the soil. Carbon lost as CO<sub>2</sub> was measured over a period of 98 days to determine which would have the greatest effect, if any, in decreasing subsidence of an organic soil.
6. It was found that glucose alone caused a very small loss of soil organic matter. Instead of a priming effect there was a depression in the amount of soil organic matter lost during the first 2 days.
7. Rate of priming with both complete alfalfa and alfalfa insolubles reached two peaks that corresponded with peaks of total CO<sub>2</sub> loss. The priming action eventually became negative as a result of both treatments but this effect developed sooner and became more marked

in the complete alfalfa treatment. The total loss due to priming was greater from the insoluble fraction than from the complete alfalfa treatment.

8. It was suggested that priming action was a function of availability of substrate. A microbial population built up due to added substrate would, as that added substrate decreased, utilize the soil organic matter as an energy source. If only readily available material is added a population best adapted to utilize such material quickly develops and uses it in preference to similar material in the soil. This is probably due largely because the concentration of the added material in the soil solution is greater than the concentration of soil organic matter solubles and possibly, in part, because the added material is more chemically available. As the added material disappears, more of the more soluble soil organic matter is utilized but since this fraction is usually limited no large loss of soil organic matter takes place.

If the added material is largely insoluble the population that develops is better adapted to utilize complex substances. Following reasoning similar to that above, it would be concluded that considerable soil organic matter may be lost.

9. Under controlled conditions in the laboratory it was found that, from the Rifle Peat, the loss of soil organic matter due to the priming effect of added organic materials was not sufficient to

offset the gain due to the residue of these materials.

10. Under these conditions it was found that added material high in solubles had about the same net effect on loss of organic matter as more mature materials with a higher content of insolubles. The latter materials cause a greater loss of the organic matter of the soil but leave a larger residue.
11. It was not possible, under laboratory conditions and with the amounts added, to effect an increase in the weight of the soil over a period of time by adding organic materials. In all cases more carbon was lost than was added. However, the net loss when these organic materials were added was always less than the total loss from the untreated soil.

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APPENDIX



Table 13. Error Due to Nonlinearity of Electrometer Scale

Sample	Sample Counts in Division/ Hour From Graph - Fig. 6				Scale error Div./ Hr.	% error
	Scale	Scale	Scale	Scale		
	point 100	point 0	point 50	point 80		
Chemical Oxidation #1						
Alfalfa Insolubles						
Plate 1	423.0	361.6	392.5	411.0	61.4	15.6
Plate 2	431.0	367.3	403.5	426.0	63.7	15.8
Plate 3**	436.0	368.5	402.5	423.0	67.5	16.8
Chemical Oxidation #2						
Alfalfa Insolubles						
Plate 1	443.0	365.3	409.0	435.5	77.7	19.0
Plate 2**	450.5	380.4	415.0	436.5	70.1	16.9
Chemical Oxidation #1						
Complete Alfalfa						
Plate 1	409.8	350.0	379.9	397.8	59.8	15.7
Plate 2	400.0	332.0	366.2	386.5	68.0	18.5
Plate 3	411.0	345.0	378.0	397.8	66.0	17.4
Chemical Oxidation #2						
Complete Alfalfa						
Plate 1	421.7	352.0	386.9	403.8	69.7	18.0
Plate 2	408.7	354.0	381.4	397.8	54.7	14.3
Chemical Oxidation of Glucose						
Plate 1	137.1	115.8	126.4	132.8	21.3	16.9
Plate 2	134.3	112.0	123.3	129.8	22.3	18.1
*Standard Sample						
2200 Counts per minute**	39.8	33.3	36.5	38.5	6.5	17.8
Average percent correction						16.9

\*Supplied by J. E. Varner, Agricultural Biochemistry Department.

\*\*Plate counts illustrated in Figure 6.

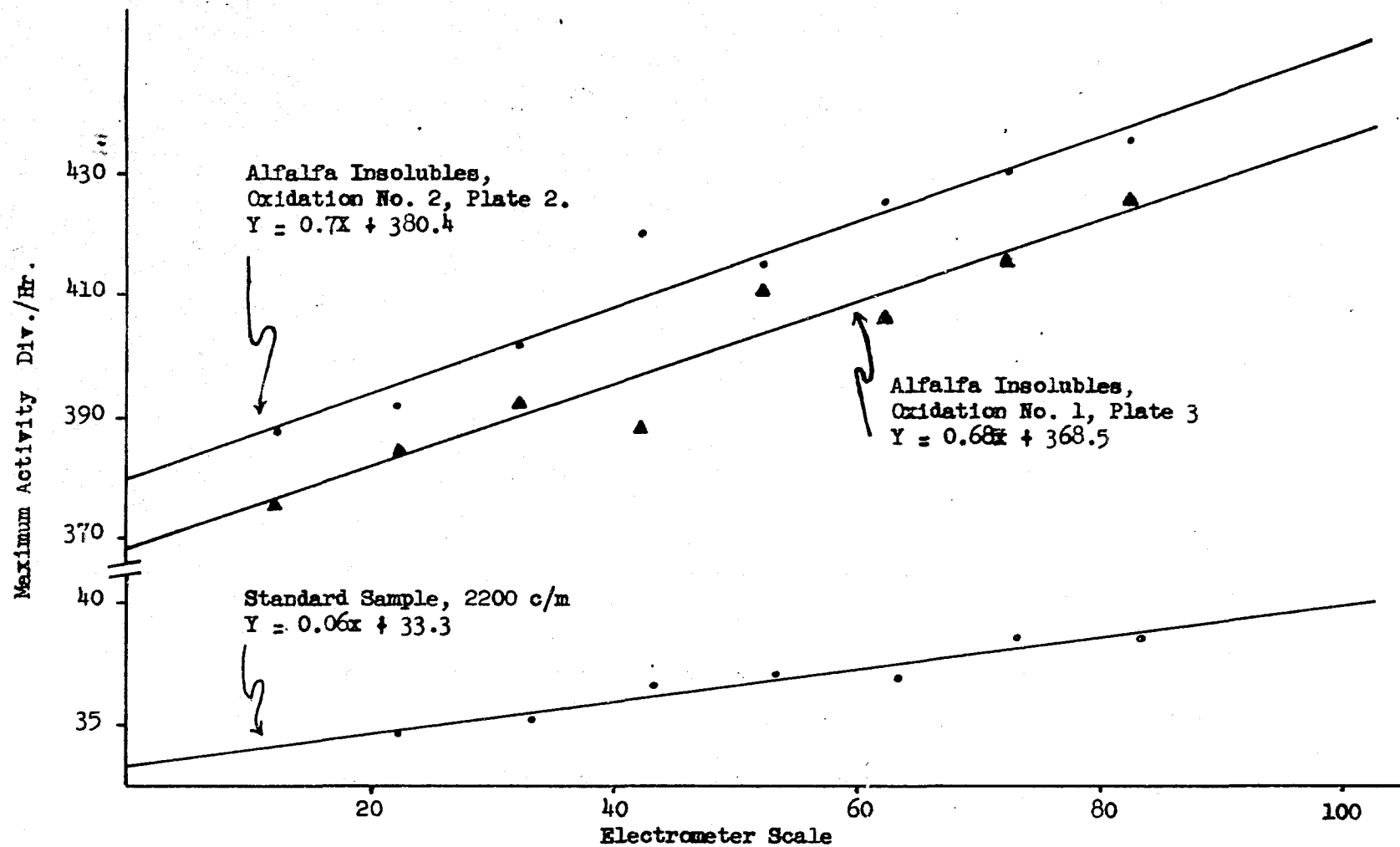


FIGURE 6. SCALE ERROR LANDSVERK ELECTROMETER NO. 1136

Table 14. Correction for Nonlinearity of Scale.  
Landsverk Electrometer Model L-75, Serial 1136

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<u>Scale</u>	<u>% Correction</u>
86-80	-1
80-74	+1
74-68	+2
68-62	+3
62-56	+4
56-50	+5
50-44	+6
44-38	+7
38-32	+8
32-26	+9
26-20	+10
20-14	+11
14-8	+12

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Table 15.  $\text{BaC}^{14}\text{O}_3$  Counts 0.6 g. Glucose Treatment

Day	Flask No.	Maximum activity Div./Hr.	Average activity Div./Hr.	Average activity C/M	Activity ratio
0.5	5	51.1	45.6	2115	0.295
	6	45.8			
	7	40.8			
	8	44.5			
1.0	5	105.3	109.3	5070	0.708
	6	113.3			
1.5	6	102.9	105.0	4870	0.680
	8	107.0			
2.0	5	79.0	78.9	3660	0.571
	6	78.8			
2.5	6	62.1	64.7	3000	0.419
	7	67.3			
3.0	5	53.2	52.0	2410	0.336
	6	50.8			
4	5	41.2	43.1	2000	0.279
	8	44.9			
5	5	35.9	36.6	1700	0.237
	6	37.3			
6	7	31.1	33.4		
	8	35.6			
7	5	33.3	31.4	1460	0.204
	6	29.4			
8	5	25.9	26.6	1230	0.172
	7	27.2			
9	5	22.6	22.2	1030	0.144
	6	21.8			
11	6	22.1	23.1	1070	0.149
	8	24.1			

Table 15. Continued

Day	Flask No.	Maximum activity Div./Hr.	Average activity Div./Hr.	Average activity C/M	Activity ratio
13	5	16.3	16.5	770	0.108
	6	16.7			
16	6	17.0	17.2	800	0.112
	7	17.4			
19	5	14.7	13.8	640	0.089
	6	12.8			
25	6	11.4	11.9	550	0.077
	7	12.3			
32	5	12.9	12.7	590	0.082
	7	12.5			
46	5	7.9	7.6	350	0.049
	7	7.4			
*106	6	2.6	2.9	130	0.018
	8	3.1			

\*Represents activity in BaCO<sub>3</sub> collected from 92-106 days, i.e., no counts made on BaCO<sub>3</sub> from 46th-92nd day.

Table 16.  $BaC^{14}O_3$  Counts Alfalfa Insolubles Treatment

Day	Flask No.	Maximum activity Div./Hr.	Average activity Div./Hr.	Average activity C/M	Activity ratio
1	4	187.4	187.7	10700	0.440
	5	199.6			
	6	176.2			
2	5	219.1	219.2	12490	0.514
	6	219.3			
3	4	223.0	225.6	12860	0.529
	5	228.2			
4	4	209.2	214.0	12200	0.502
	6	218.8			
6	4	214.9	219.8	12500	0.514
	5	223.6			
8	5	233.2	228.6	13030	0.536
	6	224.2			
10	4	217.4	216.6	12350	0.508
	6	215.8			
12	4	216.3	218.0	12430	0.511
	6	219.7			
14	5	205.1	204.0	11630	0.478
	6	202.9			
16	5	183.4	181.9	10370	0.427
	6	180.4			
20	5	154.8	156.3	8910	0.367
	6	157.7			
25	4	124.3	122.6	6990	0.288
	6	120.9			
32	4	100.3	102.8	5860	0.241
	6	105.3			

Table 16. Continued

Day	Flask No.	Maximum activity Div./Hr.	Average activity Div./Hr.	Average activity C/M	Activity ratio
40	4	78.7	79.1	4510	0.186
	5	79.4			
48	4	61.0	61.4	3500	0.144
	4	59.7			
	5	60.5			
	6	63.5			
60	4	42.6	41.4	2360	0.097
	5	42.4			
70	4	63.0	62.5	3560	0.146
	6	61.9			

Table 17.  $BaC^{14}O_3$  Counts Complete Alfalfa Treatment

Day	Flask No.	Maximum activity Div./Hr.	Average activity Div./Hr.	Average activity C/M	Activity ratio
1	8	299.5	295.7	16850	0.745
	9	291.9			
2	7	273.0	270.9	15440	0.683
	9	268.7			
3	8	269.2	269.3	15350	0.679
	9	269.4			
4	7	255.0	255.7	14570	0.644
	9	256.4			
6	8	252.7	252.4	14390	0.636
	9	252.1			
8	7	256.1	257.4	14670	0.649
	9	258.7			
10	7	250.4	255.0	14540	0.643
	8	259.5			
12	7	216.5	220.4	12560	0.556
	8	224.3			
14	7	200.3	199.9	11390	0.504
	8	203.5			
	9	195.8			
16	7	189.1	188.7	10760	0.476
	8	188.3			
20	8	176.6	177.9	10140	0.449
	9	179.1			
25	7	156.2	153.6	8760	0.387
	9	151.0			
32	7	131.6	131.5	7500	0.332
	8	131.3			



Table 17. Continued

Day	Flask No.	Maximum activity Div./Hr.	Average activity Div./Hr.	Average activity C/M	Activity ratio
40	7	99.6	101.4	5780	0.256
	9	103.2			
48	7	81.1	82.9	4730	0.209
	8	84.2			
	9	83.3			
60	6	63.3	64.0	3650	0.161
	6	64.6			
70	7	77.4	78.0	4450	0.197
	9	78.6			

Table 18. Calculated Check - 0.6 g. Glucose

Day	Activity ratio	Mg. CO <sub>2</sub> soil + treatment	Average	Calculated Mg. CO <sub>2</sub> from glucose	Average	Calculated Check Mg. CO <sub>2</sub>
0.5	0.295	*141	100	-	30	70
		97		29		
		98		29		
		105		31		
1.0	0.708	293	273	208	193	80
		317		225		
		252		178		
		229		162		
1.5	0.680	171	175	116	118	57
		175		119		
		182		124		
		171		116		
2.0	0.511	127	127	65	65	62
		132		68		
		123		63		
		128		65		
2.5	0.419	*90	102	-	43	59
		97		41		
		106		44		
		103		43		
3.0	0.336	78	79	26	27	52
		79		27		
		81		27		
		*85		-		
4	0.279	129	131	36	37	94
		135		38		
		128		36		
		134		37		
5	0.237	*59	80	-	19	61
		77		18		
		80		19		
		83		20		

Table 18. Continued

Day	Activity ratio	Mg. CO <sub>2</sub> soil + treatment	Average	Calculated Mg. CO <sub>2</sub> from glucose	Average	Calculated Check Mg. CO <sub>2</sub>
6	0.217	*85 67 70 72	70	- 15 15 16	16	54
7	0.204	62 *46 62 68	64	13 - 13 14	13	51
8	0.172	63 *80 62 66	64	11 - 11 11	11	53
9	0.144	*51 55 59 58	57	- 8 9 8	8	49
11	0.149	92 *89 96 101	96	14 - 14 15	14	82
13	0.108	69 71 *89 73	71	7 8 - 8	8	63
16	0.112	134 124 123 *147	127	15 14 14 -	14	113
19	0.089	142 *134 146 151	146	13 - 13 14	13	133

Table 18. Continued

Day	Activity ratio	Mg. CO <sub>2</sub> soil + treatment	Average	Calculated Mg. CO <sub>2</sub> from glucose	Average	Calculated Check Mg. CO <sub>2</sub>
25	0.077	223	236	17	18	218
		227		17		
		242		19		
		253		19		
32	0.082	211	219	17	18	201
		*202		-		
		221		18		
		226		19		
46	0.049	272	271	13	13	258
		265		13		
		275		13		
		*293		-		
92- 106	0.018	376	382	7	7	375
		377		7		
		*365		-		
		392		7		

\* These replications exceeded twice the standard deviation of the other three and were omitted on this basis.

Table 19. Calculated Check - 1.6 g. Alfalfa Insolubles

Day	Activity ratio	Mg. CO <sub>2</sub> soil + treatment	Average	Calculated Mg. CO <sub>2</sub> from insolubles	Average	Calculated check Mg. CO <sub>2</sub>
1	0.440	101 112 81	98	45 49 36	43	55
2	0.514	117 120 108	115	60 62 56	59	56
3	0.529	111 110 106	109	59 58 56	58	51
4	0.502	124 109 123	119	62 55 62	60	59
6	0.514	183 177 200	187	94 91 103	96	91
8	0.536	150 160 159	156	80 86 85	84	72
10	0.508	167 215 189	190	85 109 96	93	97
12	0.511	306 319 294	306	157 163 150	157	149
14	0.478	297 263 288	283	142 126 138	135	148
16	0.427	148 124 150	141	63 53 64	60	81

Table 19. Continued

Day	Activity ratio	Mg. CO <sub>2</sub> soil + treatment	Average	Calculated Mg. CO <sub>2</sub> from insolubles	Average	Calculated check Mg. CO <sub>2</sub>
20	0.367	162 154 166	161	59 56 61	59	102
25	0.288	172 178 177	176	50 51 51	51	125
32	0.241	221 226 212	220	53 55 51	53	167
40	0.186	248 242 252	247	46 45 47	46	201
48	0.144	212 205 212	210	39 38 39	39	171
60	0.097	296 298 292	295	29 29 28	29	266
70	0.146	208 - 212	210	30 - 31	31	179

Table 20. Calculated Check - 2.4 g. Complete Alfalfa

Day	Activity ratio	Mg. CO <sub>2</sub> soil † treatment	Average	Calculated Mg. CO <sub>2</sub> from alfalfa	Average	Calculated check Mg. CO <sub>2</sub>
1	0.745	330 309 312	317	246 230 233	236	81
2	0.683	375 378 371	374	256 258 253	256	118
3	0.679	234 222 230	229	159 151 156	155	74
4	0.644	181 164 178	174	117 106 115	112	62
6	0.636	- 290 294	292	- 185 187	186	106
8	0.649	360 344 349	351	234 223 226	228	123
10	0.643	389 381 357	376	252 247 232	244	132
12	0.556	227 282 217	242	126 157 121	134	108
14	0.504	122 129 128	126	62 65 65	64	62
16	0.476	82 87 86	85	39 41 41	40	45

Table 20. Continued

Day	Activity ratio	Mg. CO <sub>2</sub> soil + treatment	Average	Calculated Mg. CO <sub>2</sub> from alfalfa	Average	Calculated check Mg. CO <sub>2</sub>
20	0.449	143 150 149	147	64 67 67	66	81
25	0.387	178 180 178	179	69 70 69	69	110
32	0.332	216 216 211	214	72 72 70	71	143
40	0.356	224 232 228	228	57 59 58	58	170
48	0.209	213 208 205	209	45 44 43	44	165
60	0.161	306 318 304	309	49 51 49	50	259
70	0.197	219 227 220	222	86 88 86	87	135



## AUTOBIOGRAPHY

I, Carl Woodrow Bingeman, was born on July 1, 1919, at Esther, Alberta, Canada. I received my elementary education and part of my secondary schooling in a one roomed rural school near Esther. The remainder of my secondary education was received in high schools in surrounding villages. I attended The Edmonton Normal School for one year and subsequently taught in rural schools in The Province of Alberta for four years. From 1941 to 1946 I served in the Canadian Army with the Royal Canadian Artillery. In 1946 I entered Macdonald College of McGill University receiving a Bachelor of Science degree three years later. In 1949 I received an appointment as a Research Assistant with the Ohio Agricultural Experiment Station. During the tenure of this assistantship I completed requirements for the degree of Doctor of Philosophy at Ohio State University.