THE EFFECT OF SOIL IN A MEDIUM ON THE ORGANISMS DEVELOPING IN IT AND ON THE pH OF THE MEDIUM.

by

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INTRODUCTION

Some substances in soil inhibit the growth of micro-organisms. Acids are produced as products of the growth of many kinds of micro-organisms (3, 4, 18, 19). Under certain conditions, these acids produce an unfavourable hydrogen-ion concentration, which according to Webb (23) retards spore-formation and according to Meacham (14) suppresses vegetative development. Toxins are produced by certain fungi and bacteria. These toxins inhibit the development of other organisms. This is considered to be the basis of the antagonism of certain spore-forming bacteria to Rhizoctoni solani (22).

On the other hand, certain substances in soil favour the development of specific groups of micro-organisms. For example, sulphur is essential for sulphur bacteria (22) and cellulose is necessary for cellulose-decomposing fungi (20).

In the light of the above facts, it is probable that small amounts of soil added to a nutrient medium in the process of plating contain some principle that affects the numbers of certain organisms developing in it. The present study was undertaken to obtain information on this probability. It was divided into four parts as follows:-

(1) the effect of soil in a medium on organisms that develop in it;

(2) the effect of soil on the rate of growth and sporulation of Helminthosporium sativum;

(3) the effect of soil in a medium on the pH of the medium;

(4) the effect of cultures of certain organisms on the pH of a medium containing soil.

HISTORICAL

Although a vast literature on microbial antagonism has accumulated, only a few references that appear to bear on the subject will be presented.

As early as 1907, Russell and Hutchinson (16) observed that soil, which had been partially sterilized, showed first a decrease and then an increase in numbers of bacteria. The authors attributed this phenomenon to the presence of something active in untreated soils rather than the lack of some principle.

The search for toxic substances in soil probably began after Potter (15), in 1908, found that Pseudomonas destructans produced a potent toxin which not only caused a disease, turnip-rot, but also destroyed the pathogen.

In a study of fungi isolated from Manitoba, Bisby, James and Timonin (1) found that Trichoderma lignorum played an important part in suppressing the virulence of Fusarium culmorum and Helminthosporium sativum on wheat.

Later, Weindling found the lethal principle of this fungus to be present in a filtrate from the culture (24).

Greaney and Machacek (8) suggested that the antagonistic action of Cephalothecium roseum against Helminthosporium sativum was due to a toxic substance which inhibited spore development.

Vandecaveye and Katznelson (17) demonstrated that different soil types supported different populations. One type supported 19 times as many bacteria and 200 times as many fungi as another type; thereby indicating that certain factors in the soils were responsible for the differences in numbers.

Certain investigators studied the effect of soil extracts on the survival and growth of micro-organisms. Greig Smith (10) introduced Bacillus prodigiosus into sterile saline extracts of different soils. He invariably found that the reduction in numbers of cells was greater than in the check medium, which was merely sterile saline. This was presented as evidence of the existence of bacterio-toxins in soil.

Hutchinson and Thayson (10) proceeded further along this line of study. In addition to B. prodigiosus, they used B. fluorescens liquefaciens, which they considered to be one of the commonest soil forms. These investigators found that extracts from various soils differed in their suitability for bacterial growth. In some instances vigorous growth of B. prodigiosus occurred; in others the numbers of introduced organisms fell to a minimum. Extracts of the two soils that rated poorest for the growth of B. prodigiosus produced

excellent growth of B. fluorescens liquefaciens. They accepted this as failure to indicate the presence of toxins in the soil extracts.

Waksman and Woodruff (21) showed that an ether-soluble fraction of soil had marked inhibition upon Bacillus subtilis and Sarcina lutea but not upon Escherichia coli. These authors pointed out that soil extracts used in many cultural media for growth contained growth-inhibiting substances as well as growth-promoting substances.

Finally, Campbell (2) adding soil to Czapek's medium demonstrated that numbers of a saltant of Helminthosporium sativum were lower than in the control. The suppression was significant in the 12 samples of soil studied.

EXPERIMENTAL

The Effect of Soil in a Medium on Organisms developing in it

A preliminary experiment was carried out to ascertain the amount of diluting necessary to give a countable number of colonies on a plate with a given culture. A spore suspension was prepared by introducing sterile water into the culture bottle and scraping the spores from the culture gently with a loop. The number of spores in the suspension was determined by the Neubauer haemocytometer method. Then, the suspension was diluted serially and the 0.01, 0.001, 0.0002 and 0.0001 concentrations were plated in Czapek's agar. By the above procedure it was determined that a spore suspension, containing from 600,000 to 1,000,000 spores, when diluted to 1:5000, would yield the desired number of colonies per plate. For example, a spore suspension of 700,000 Fusarium graminearum Corda produced 7 to 10 colonies per plate. Evidently, not all spores counted by the direct count grew on the agar medium.

The Neubauer haemocytometer method was not applicable to the estimation of numbers of Serratia marcescens Bizio, a species of bacteria. Accordingly, the following smear technique was used. A 4-day nutrient broth culture was diluted in sterile water to 1:100 and shaken vigorously. One ml. of this suspension was pipetted into 9 ml. gelatin (0.05

per cent) in water. After vigorous shaking, 0.01 ml. was spread evenly over a square centimetre of a clean slide. The smear was dried quickly by heating in an oven at 131°C. for about half a minute. It was stained with a 1 per cent aqueous solution of rose bengal, which was allowed to act from 2 to 3 minutes.

Counts of the bacteria were made on 25 random fields and the average count was multiplied by the appropriate factor. A procedure similar to that followed with the fungi was used to ascertain the relation between the microscopic count thus obtained and the plate count and to determine the degree of diluting necessary for a given suspension to provide countable numbers of colonies per plate on nutrient agar. For example, a bacterial suspension of 1,262 million cells diluted to 1:1,000,000 produced plates with about 50 surface colonies per plate. Surface colonies only were counted because of the difficulty in distinguishing between particles of soil in medium and subsurface colonies.

Several species of fungi and one of bacteria were used in this portion of the study to determine whether soil in a medium has the same effect on their development as has been observed with a saltant of Helminthosporium sativum P.K. and B. (12). Three of these fungi were isolated from wheat seeds according to the method of James, Wilson, and Stark (11). They were identified as Penicillium viride-dorsum Biourge, Penicillium Melinii Thom and Aspergillus

fumigatus Fresenius. The remaining cultures were procured from the Dominion Laboratory of Plant Pathology located on this campus. These follow:-

Fusarium avenaceum (Fries) Saccardo,

Fusarium graminearum,

Helminthosporium sativum, and a

saltant of Helminthosporium sativum (7).

These fungi were cultured at 25°C in 6 oz. medicine bottles on Czapek's agar until sufficient spores developed. Serratia marcescens was the only type of bacteria used. It was cultured in nutrient broth at 25°C. for 4 days prior to use. Each culture was plated as in the preliminary experiments in 4 replicate plates without soil, and in addition in 4 replicates with soil. Nine soils ranging in texture from clay to fine sand were used. The equivalent of 0.1 gm. soil was added to each plate. This amount was obtained by transferring 1 ml. of a sterilized, predetermined concentration of soil in water. Ten ml. of Czapek's agar for fungi, or nutrient agar for bacteria, were pipetted into each plate to ensure uniformity in concentration of soil in medium. All plates were incubated at 25°C. until they were countable. The results are presented in the appendix (pg.30) and in summarized form in Table 1. Five of the species grew better in plates containing soil than in control plates. The saltant of Helminthosporium saltivum was depressed by soil in every case. The results on Aspergillus fumigatus were not consistent; as was the case

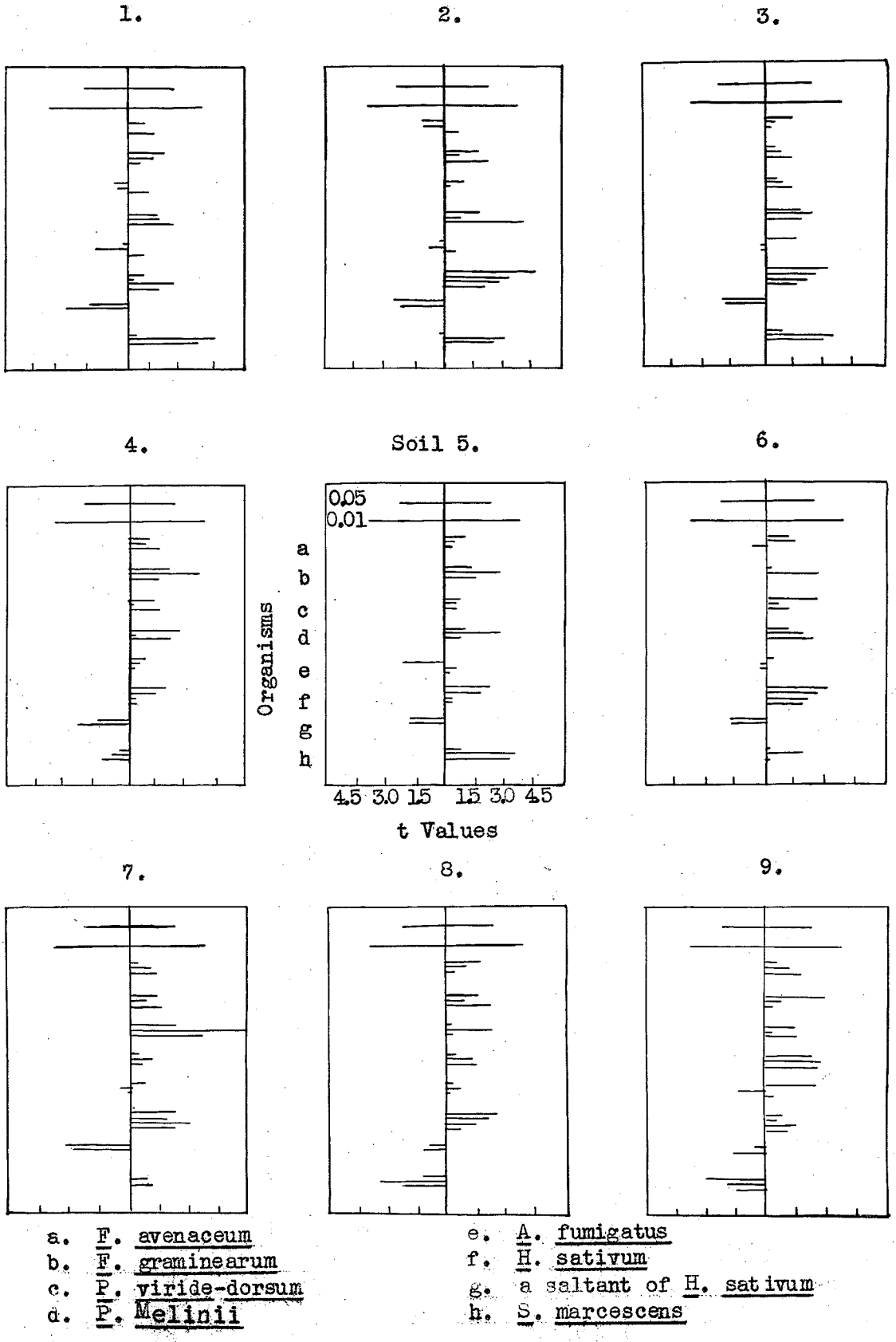
also with Serratia marcescens, except for soils 8 and 9. However, the effect of a given soil on numbers was not the same with different species. For example, numbers of Fusarium graminearum were larger in plates containing soil 9 than in the control plates; whereas the opposite was true in the case of Serratia marcescens in the same soil. Likewise Fusarium avenaceum was stimulated by soil 8 while Serratia marcescens was depressed by this soil.

The differences between soils with a given species and the differences between species with a given soil did not appear to be marked and in some cases lacked consistency from replication to replication. In order to present the findings in another way, the data were submitted to the t test (6). The t values are presented graphically in Fig. 1. The computed replicate t values for each organism are represented by a group of lines of certain lengths. In the majority of cases, the lengths of these lines are less than the lines representing significance. The lines representing t values for each species do not indicate any consistent relationship between species and soils.

Table 1. Change in numbers of organisms developing on a nutrient medium produced by the addition of 0.1 gm. sterile soil to 10 ml. of the medium. (Average of 3 trials for 4 counts per trial).

	Fungi							
Soil	1	2	3	4	5	6	7	8
Control	10.25	14.50	13.42	36.33	21.42	7.00	10.25	29.75
1	11.08	16.58	13.08	43.17	20.92	8.50	7.00	35.42
2	9.50	17.08	14.25	43.58	21.00	11.00	7.50	33.83
3	11.00	16.08	14.92	44.33	21.50	10.75	6.50	38.33
4	11.42	19.08	15.33	42.50	22.75	10.38	8.25	28.25
5	11.08	18.33	14.58	42.00	20.83	9.25	8.00	38.17
6	11.08	19.13	16.58	39.67	20.33	9.63	8.00	31.08
7	11.25	16.50	20.83	39.17	21.42	11.00	6.75	31.12
8	11.58	16.58	16.42	39.25	22.50	10.25	8.50	22.83
9	11.75	16.42	15.25	44.67	21.92	9.00	8.25	20.42

Culture	Medium
1. *Fusarium avenaceum*	-Czapek's agar
2. *Fusarium graminearum*	-Czapek's agar
3. *Penicillium viride-dorsum*	-Czapek's agar
4. *Penicillium Melinii*	-Czapek's agar
5. *Aspergillus fumigatus*	-Czapek's agar
6. *Helminthosporium sativum*	-Czapek's agar
7. a saltant of *Helminthosporium sativum*	-Czapek's agar
8. *Serratia marcescens*	-nutrient agar



a. F. avenaceum
b. F. graminearum
c. P. viride-dorsum
d. P. Melinii
e. A. fumigatus
f. H. sativum
g. a saltant of H. sativum
h. S. marcescens

FIG. 1.- Significance of effect of soil in a medium on growth of various species as indicated by t values - negative on left.

Effect of soil on Rate of Growth and Sporulation of Helminthosporium sativum

While making counts of Helminthosporium sativum, it was noted that the rate of growth and sporulation of this fungus was enhanced by the soil in Czapek's medium. In order to study this phenomenon a new set of plates was prepared and examined daily for 13 days. The experiment was carried out on each of the 9 soils used in the study. At 4 days, colonies were observed in nearly all plates containing soil, whereas very few colonies were present in the control plates. Likewise in the same incubation period, there was a greater number of colonies in plates containing soils 5 to 9 than 1 to 4. Later, a black-green pigmentation, characteristic of spore production, was noted in the plates containing soil. This was quite marked at 6 days. The mycelia of these colonies intertwined to produce a mat. In contrast, the colonies in the control plates were lacking in pigmentation and were very small. However, after 9 day's incubation, many of the colonies in the control plates were also sporulating, as is shown in Fig. 2.

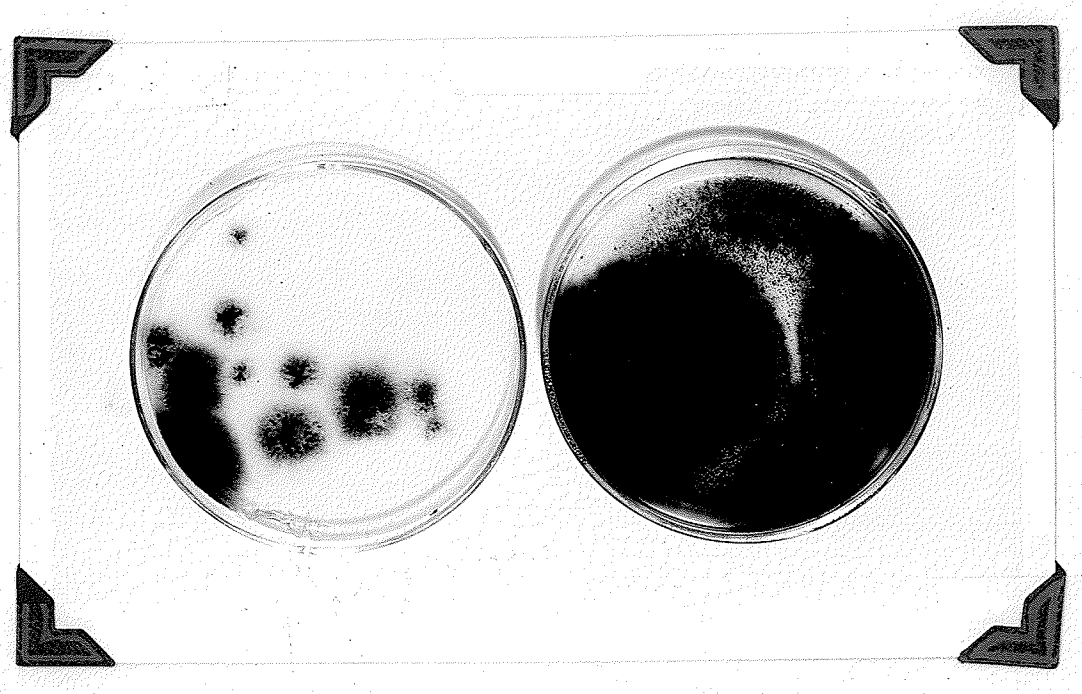


Fig. 2. The stimulating effect of soil on the growth and sporulation of Helminthosporium sativum.

Left - Colonies of Helminthosporium sativum after 9 day's incubation in Czapek's medium.

Right - Colonies of Helminthosporium sativum after 9 day's incubation in Czapek's medium containing soil.

The Effect of Soil in a Medium on the pH of the Medium.

The previous portion of the study revealed that some factor associated with soil in nutrient agar or in Czapek's medium was affecting the growth of certain organisms. It is conceivable that this factor was pH. In order to study this point, soil was added to a variety of media of different buffer capacities and pH determinations were carried out. All media except soil-extract agar (13) were prepared according to the directions in Fred and Waksman (5). The pH of each medium was tested in duplicate by a Coleman pH meter. Reddings were made on the same medium containing 0.02, 0.01, 0.001, and 0.0001 gms., respectively, of each of the 9 soils studied. The detailed results are presented in the appendix,(pg.51)and in addition those that pertain to the largest amount of soil used are shown in Table 2. The pH of two of the test media was not changed by any of the concentrations of soil used. These were nutrient agar and asparagin mannitol agar. In no case did the amount of soil added change the pH sufficiently to have much effect on the growth of organisms. For example, the maximum change in pH was 0.41 in Czapek's agar, while in media containing 0.001 gm. soil or less, the pH was not appreciably different from that of the medium without the soil (See appendix, page 51).

Table 2. Change in pH of medium produced by the addition of sterile soil to the medium (0.02 gm. soil per 10 ml.).

	Media						
Soil	1	2	3	4	5	6	7
Control	6.80	5.60	5.60	7.29	7.50	7.25	7.50
1	6.80	5.72	5.60	7.30	7.50		
2	6.78	5.61	5.61	7.29	7.50		
3	6.80	5.61	5.60	7.29	7.50		
4	6.77	5.64	5.61	7.29	7.50		
5	6.77	5.60	5.61	7.29	7.50	7.18	7.31
6	6.80	5.62	5.60	7.30	7.50		
7	6.80	5.63	5.57	7.29	7.53	7.20	7.51
8	6.81	5.85	5.70	7.29	7.50		
9	6.81	6.01	5.97	7.30	7.71	7.45	7.53

Medium
1. nutrient agar
2. Czapek's agar
3. potato dextrose agar
4. asparagin mannitol agar
5. sodium albuminate agar
6. soil-extract agar (soil 7)
7. soil-extract agar (soil 9)

The Effect of Cultures of Certain Organisms on the pH of a Medium Containing Soil.

As was observed in the previous section, the change in the pH of a medium was only slight when soil was added to it. This study was carried out to ascertain to what extent the pH of a medium was changed by cultures of certain fungi or bacteria. The experiment was, therefore, carried out on the same suspensions of organisms as were used in the plate counts reported in the previous section. Duplicate control plates containing soil but no organisms were prepared. The experiment was replicated three times on each of the 9 soil samples. In order to have a uniform amount of medium and thereby a uniform concentration of soil in the medium a pipette was used to transfer 10 ml. medium to each plate. After incubation at 25° C. for the periods shown in Tables 3 to 9, the contents of each plate were transferred to a small dish (similar to a crucible) and the pH determined.

The individual results from each organism are presented in the Appendix (page 57). They are presented in summarized form in Tables 3 to 9 and graphically in Fig. 3. It may be observed that all cultures did not change the pH of the medium containing soil to the same extent. For example, Penicillium Melinii produced a smaller change in pH in media containing soils A to I (same as 1 to 9 in Table 1 and 2) than did any other test organisms, while on the other hand

the change in any of the other soils was smaller in not more than 2 other species. In contrast, the change in pH was greatest with Helminthosporium sativum in 8 of 9 soils.

Table 3. The effect of a culture of Fusarium avenaceum on the pH of Czapek's medium containing soil - 25°C. for 7 days.

Soil	pH		
	Without Fungi*	With Fungi**	pH Difference
A	6.01	7.06	1.05
B	6.00	6.93	0.93
C	5.99	6.98	0.99
D	5.98	7.04	1.06
E	5.79	7.06	1.27
F	5.97	7.04	1.07
G	5.71	7.14	1.43
H	6.00	7.08	1.08
I	6.22	7.61	1.39

* Average of 6 pH readings

** Average of 12 pH readings

Table 4. The effect of a culture of Fusarium graminearum on the pH of Czapek's medium containing soil - 25°C. for 7 days.

Soil	pH Without fungi *	pH With fungi **	pH Difference
A	5.99	7.68	1.69
B	5.96	7.75	1.79
C	5.95	7.67	1.72
D	5.94	7.74	1.80
E	5.78	7.77	1.99
F	5.94	7.76	1.82
G	5.75	7.79	2.05
H	6.04	7.83	1.79
I	6.23	7.88	1.65

* Average of 6 pH readings
** Average of 12 pH readings

Table 5. The effect of a culture of Penicillium Melinii on the pH of Czapek's medium containing soil - 25°C. for 8 days.

Soil	pH Without fungi *	pH With fungi **	pH Difference
A	5.97	6.59	0.62
B	5.95	6.46	0.51
C	5.96	6.60	0.64
D	5.97	7.26	1.29
E	5.76	6.56	0.80
F	5.99	6.42	0.43
G	5.69	7.05	1.36
H	6.01	7.25	1.24
I	6.23	6.61	0.38

* Average of 6 pH readings
** Average of 12 pH readings

Table 6. The effect of a culture of Penicillium viride-dorsum on the pH of Czapek's medium containing soil - 25° C. for 8 days.

Soil	pH Without fungi *	pH With fungi **	pH Difference
A	5.96	5.28	0.68
B	5.95	5.09	0.86
C	5.94	5.08	0.86
D	5.94	4.95	0.99
E	5.72	5.12	0.60
F	5.92	4.94	0.98
G	5.72	4.68	1.04
H	6.01	4.64	1.37
I	6.25	5.00	1.25

* Average of 6 pH readings
** Average of 12 pH readings

Table 7. The effect of a culture of Aspergillus Fumigatus on the pH of Czapek's medium containing soil - 25° C. for 8 days.

Soil	pH Without fungi*	pH With fungi**	pH Difference
A	5.97	7.38	1.41
B	6.00	7.49	1.49
C	5.95	7.43	1.49
D	5.95	7.43	1.48
E	5.69	7.46	1.77
F	5.98	7.48	1.50
G	5.67	7.42	1.75
H	6.07	7.39	1.32
I	6.21	7.50	1.29

* Average of 6 pH readings
** Average of 12 pH readings

Table 8. The effect of a culture of Helminthosporium sativum on the pH of Czapek's medium containing soil - 25° C. for 13 days.

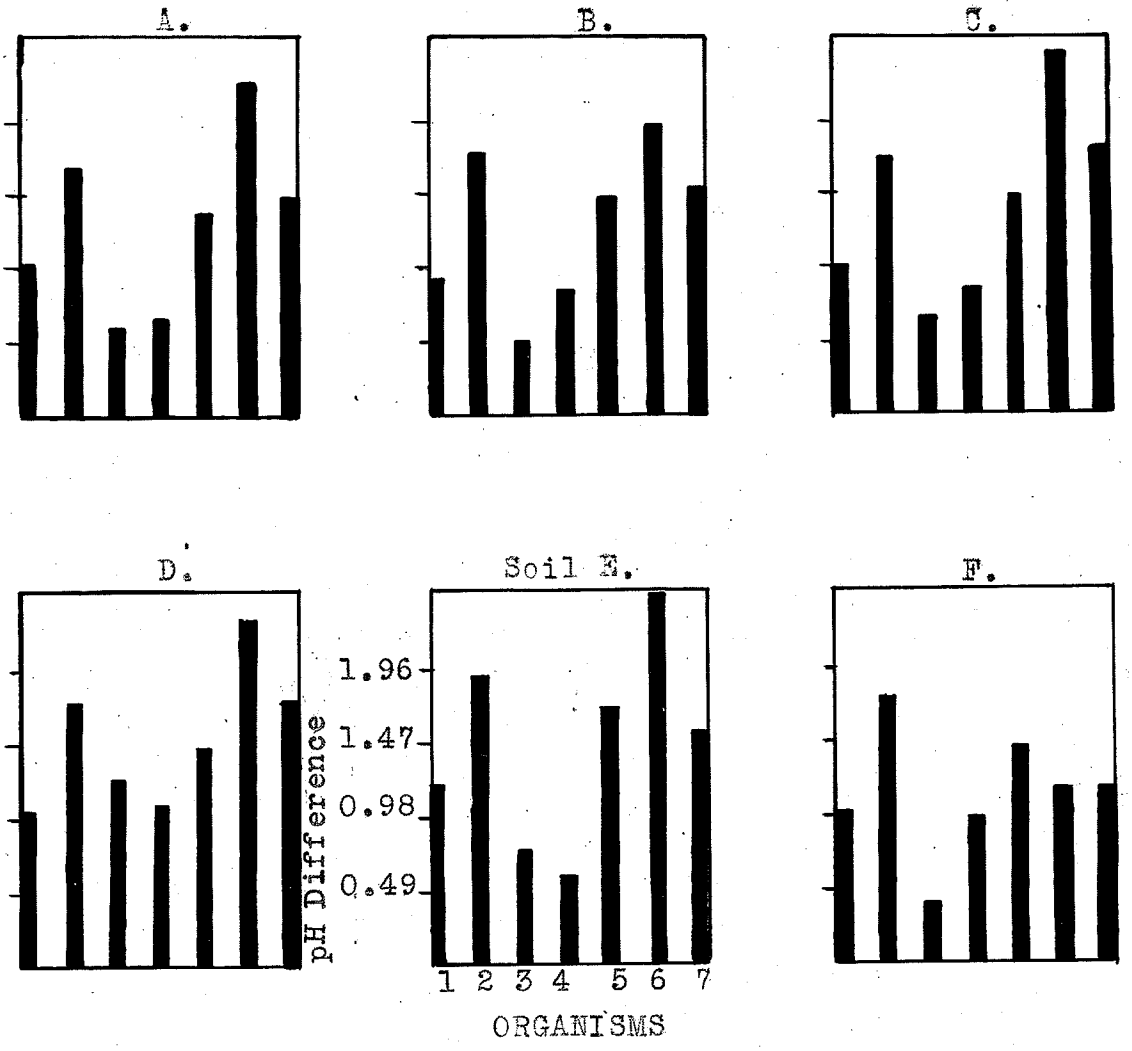
Soil	pH Without fungi *	pH With fungi **	pH Difference
A	6.01	8.30	2.29
B	6.00	7.96	1.96
C	5.96	8.43	2.47
D	5.96	8.34	2.38
E	5.75	8.32	2.57
F	6.01	7.21	1.20
G	5.74	8.32	2.58
H	6.11	8.24	2.13
I	6.29	8.30	2.01

* Average of 4 pH readings
** Average of 8 pH readings

Table 9. The effect of a culture of Serratia marcescens on the pH of nutrient agar - 25° C. for 8 days.

Soil	pH Without bacteria *	pH With bacteria **	pH Difference
A	6.61	8.10	1.49
B	6.43	7.95	1.52
C	6.32	8.12	1.80
D	6.32	8.09	1.77
E	6.51	8.08	1.57
F	6.84	8.14	1.30
G	6.52	8.09	1.57
H	6.68	7.97	1.29
I	6.82	8.04	1.22

* Average of 6 pH readings
** Average of 12 pH readings



1. F. avenaceum
2. F. graminearum
3. P. Melinii
4. P. viride-dorsum
5. A. fumigatus
6. H. sativum
7. S. marcescens

Fig. 3. Effect of cultures of organisms on the pH of media containing soil.

DISCUSSION

The reason for the different effects produced by different soils on the various species studied is not apparent. It is known that some soil organisms produce certain metabolic products which are toxic to themselves or to other organisms. These same substances may not be inhibitory to other organisms and may even serve as food for them. Weindling (25) pointed out that simple products are produced from the breakdown of carbohydrates by one or more associated organisms in cultural media. These metabolic products of one of the associated organisms may serve as food for the other or may accumulate in a medium to a poisonous concentration for the associate.

Sterilization may change the chemical nature of the soil, making certain substances soluble and causing disintegration of microbial cells and thereby releasing nutrients for organisms. Undoubtedly some of these substances could bring about better growth of a given species or inhibit it. The work of Greaves (9) supports this contention. This author found that certain salts known to be necessary for higher life were essential for lower life as well. However, if these salts were in too great concentration, they became toxic. He further pointed out that salts of nitrates were the most toxic and that salts such as calcium chloride were stimulating for ammonifying bacteria. The toxic concentration of these salts differed with each salt and was not the same for all species.

The breakdown of microbial cell substance may release into soil certain trace elements and vitamins not normally present in the soil in appreciable quantities. These substances may enhance the growth of certain species. It has been suggested by Weindling (25) that stimulation of sporulation of fungi arises in culture from proportionately a greater ratio of accessory substances to food materials in the early stage of growth. The presence of these substances in a medium may account, in part, for the rapid growth and sporulation of Helminthosporium sativum in plates containing soil.

Undoubtedly one of the three phenomena referred to above or a combination of two or more of them may be responsible for the finding reported in Table 1 that the addition of some soils to a medium increased numbers of certain species of organisms growing in it, whilst the numbers of other species were reduced.

The small change in pH resulting from the presence of soil in media observed in the second study could scarcely be responsible for the different numbers of the various species found in the media containing soil. In the actual plating of soil from a field, this effect would be much less, since a plate at 0.001 dilution would contain only 0.01 times as much soil as was used in these tests.

Finally, from the last section, it may be noted that a culture of one species in a medium containing one soil produced a greater change in the pH in the medium than did

a culture of another species in the medium with the same soil. The change in pH must be accepted as merely the final result of metabolic processes of the culture. It may result from the acid or base produced from some ingredient in the medium. It is conceivable that an ingredient from one soil in a medium might affect the final pH produced by a culture, whereas that ingredient might be lacking in another soil.

SUMMARY

1. Eight species, 7 of fungi and 1 of bacteria were tested for the effect of soil in a medium on numbers developing in it. For this purpose, 9 soils were used.

2. With five of the eight species, a larger number of organisms occurred in plates containing soil than in control plates. Two of the species gave inconsistent results, while with another all soils depressed growth.

3. In addition, one of the species, Helminthosporium sativum, appeared to grow more rapidly and sporulated earlier in a medium containing soil than in the control. This was found to be the case with all soils tested, although the effect was more marked with certain soils.

4. Soil in a medium was found to change the pH to such a small extent that the change in pH scarcely could account for the lower number of fungi found in media containing soil.

5. The pH of a medium was found to change with development of a culture. This change was not the same for all the cultures tested.

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APPENDIX

Trial 1. The effect of sterile soil in Czapek's medium on numbers of *Fusarium avenaceum*.

Soil	Replicate Counts				Average Count	t Value
	A.	B.	C.	D.		
Control	14	11	13	11	12.25	
1	11	14	13	15	13.25	0.88
2	8	12	10	13	10.75	-1.12
3	15	16	13	11	13.75	1.12
4	15	13	12	14	13.50	1.26
5	16	15	13	11	13.75	1.12
6	14	12	12	17	13.75	1.07
7	11	11	15	13	12.50	0.21
8	16	13	14	13	14.00	1.70
9	12	11	13	15	12.75	0.44

t.05 = 2.45
t.01 = 3.71

Trial 2.

Soil	Replicate Counts				Average Count	t Value
	A.	B.	C.	D.		
Control	12	8	7	9	9.00	
1	9	9	8	10	9.00	0.00
2	6	7	7	10	7.50	-1.09
3	9	9	10	10	9.50	0.45
4	9	11	8	12	10.00	0.66
5	10	9	12	8	9.75	0.54
6	11	12	10	9	10.50	1.19
7	10	11	8	12	10.25	0.90
8	9	10	14	9	10.50	0.93
9	12	10	9	12	10.75	1.33

$t_{.05} = 2.45$
$t_{.01} = 3.71$

Trial 3.

Soil	Replicate Counts				Average Count	t Value
	A.	B.	C.	D.		
Control	11	9	10	8	9.50	
1	10	12	13	9	11.00	1.34
2	9	10	12	10	10.25	0.83
3	9	10	9	11	9.75	0.31
4	9	11	11	12	10.75	1.39
5	10	9	9	11	9.75	0.31
6	9	10	9	8	9.00	-0.65
7	10	12	13	9	11.00	1.34
8	9	11	8	13	10.25	0.59
9	14	10	10	13	11.75	1.85

$t_{.05} = 2.45$
$t_{.01} = 3.71$

Trial 1. The effect of sterile soil in Czapek's medium on numbers of *Fusarium graminearum*.

Soil	Replicate Counts				Average Count	t Value
	A.	B.	C.	D.		
Control	11	16	15	15	14.25	
1	14	21	20	18	18.25	1.81
2	15	17	17	23	18.00	1.82
3	13	18	16	13	15.00	0.45
4	20	19	17	14	17.50	1.88
5	16	22	18	13	17.25	1.37
6	21	19	15	12	16.75	0.11
7	17	11	26	18	18.00	1.15
8	13	19	20	14	16.50	1.80
9	19	20	16	20	18.75	3.08*

t.05 = 2.45
t.01 = 3.71

Trial 2.

Soil	Replicate Counts				Average Count	t Value
	A.	B.	C.	D.		
Control	14	14	16	17	15.25	
1	18	14	18	18	17.00	1.40
2	13	14	18	21	16.50	0.63
3	16	15	15	19	16.25	0.83
4	20	18	25	26	22.25	3.38 *
5	21	22	16	19	19.50	2.78 *
6	21	17	24	24	21.50	3.44 *
7	15	14	17	18	16.00	0.64
8	17	18	19	13	16.75	0.99
9	17	14	15	19	16.25	0.75

t.05 = 2.45
t.01 = 3.71

Trial 3.

Soil	A.	B.	C.	D.	Average Count	t Value
Control	13	13	16	14	14.00	
1	15	13	14	16	14.50	0.52
2	17	17	19	14	16.75	2.20
3	15	19	22	12	17.00	1.30
4	19	23	16	12	17.50	1.50
5	18	20	15	20	18.25	1.59
6	23					0
7	16	17	14	15	15.50	1.57
8	18	14	16	18	16.50	2.10
9	13	16	14	14	14.25	0.26

$t_{.05} = 2.45$
$t_{.01} = 3.71$

Trial 1. The effect of sterile soil in Czapek's medium on numbers of *Penicillium viride-dorsum*.

Soil	Replicate Counts				Average Count	t Value
	A.	B.	C.	D.		
Control	12	16	21	15	16.00	
1	14	18	11	15	14.50	-0.64
2	18	16	20	18	18.00	0.98
3	11	21	17	20	17.25	0.43
4	26	19	12	18	18.75	1.13
5	18	24	15	15	18.00	0.71
6	23	16	26	22	21.75	2.40
7	22	19			20.50	2.29
8	19	18	18	12	16.75	0.32
9	20	18	19	18	18.75	1.42

t.05 = 2.45
t.01 = 3.71

Trial 2.

Soil	Replicate Counts				Average Count	t Value
	A.	B.	C.	D.		
Control	18	13	19	14	16.00	
1	18	12	15	15	15.00	0.52
2	13	19	21	13	16.50	0.20
3	14	20	15	21	17.50	0.66
4	23	19	14	10	16.50	0.16
5	12	18	16	14	15.00	0.51
6	18	13	23	15	17.25	0.48
7	28	27	33	36	31.00	5.81**
8	26	14	27	27	23.50	2.14
9	15	17	16	14	15.50	0.31

t.05 = 2.45
t.01 = 3.71

Trial 3.

Soil	Replicate Counts				Average Count	t Value
	A.	B.	C.	D.		
Control	11	8	9	5	8.25	
1	9	13	9	8	9.75	0.90
2	8	8	7	10	8.25	0.00
3	12	8	9	11	10.00	1.13
4	10	8	14	11	10.75	1.41
5	14	12	8	9	10.75	0.51
6	11	7	14	11	10.75	1.05
7	8	11	12	13	11.00	3.48*
8	7	5	13	11	9.00	0.34
9	17	8	10	11	11.50	1.41

t.05 = 2.45
t.01 = 3.71

Trial 1. The effect of sterile soil in Czapek's medium on numbers of Penicillium Melinii.

Soil	Replicate Counts A.	B.	C.	D.	Average Count	t Value
Control	41	31	33	26	32.75	
1	40	49	30	39	39.50	1.36
2	28	44	50	49	42.75	1.68
3	40	38	34	53	41.25	1.65
4	49	42	43	36	42.50	2.38
5	29	44	32	49	38.50	1.01
6	44	29	32	49	38.50	1.01
7	38	31	33	36	34.50	0.50
8	29	30	34	47	35.00	0.43
9	35	45	42	41	40.75	2.13

t.05 = 2.45
t.01 = 3.71

Trial 2.

Soil	Replicate Counts				Average Counts	t Value
	A.	B.	C.	D.		
Control	46	38	36	41	40.25	
1	44	52	39	49	46.00	1.60
2	44	38	42	45	42.25	0.75
3	41	54	46	50	47.75	2.13
4	31	43	45	48	41.75	0.35
5	48	47	50	44	47.25	2.83*
6	51	45	40	49	36.25	1.84
7	58	46	33	49	46.50	1.11
8	45	44	39	46	43.50	1.22
9	43	49	48	51	47.75	2.72 *

t.05 = 2.45
t.01 = 3.71

Trial 3.

Soil	Replicate Counts				Average Count	t Value
	A.	B.	C.	D.		
Control	32	42	35	35	36.00	
1	51	38	44	43	44.00	2.34
2	49	46	43	45	45.75	3.96**
3	43	35	54	44	44.00	1.80
4	44	47	34	48	43.25	1.89
5	31	49	46	35	40.25	0.86
6	41	38	49	49	44.25	2.34
7	28	36	48	43	38.75	0.57
8	41	37	38	41	39.25	1.38
9	45	42	54	41	45.50	2.61*

t.05 = 2.45
t.01 = 3.71

Trial 1. The effect of sterile soil in Czapek's medium on numbers of Aspergillus fumigatus.

Soil	Replicate Counts				Average Count	t Value
	A.	B.	C.	D.		
Control	25	23	20	22	22.50	
1	23	26	20	19	22.00	-0.26
2	20	28	17	24	22.25	-0.10
3	26	21	29	26	25.50	1.53
4	21	24	29	22	24.00	0.73
5	25	23	22	19	22.25	-0.16
6	22	25	21	25	23.25	0.51
7	19	29	22	28	24.50	0.77
8	26	27	25	15	23.25	0.25
9	23	31	27	29	27.50	2.50 *

t.05 = 2.45
t.01 = 3.71

Trial 2.

Soil	Replicate Counts				Average Count	t Value
	A.	B.	C.	D.		
Control	26	18	25	22	22.75	
1	19	11	21	20	17.75	-1.72
2	15	11	28	25	19.75	-0.68
3	27	13	27	16	20.75	-0.49
4	21	24	29	22	24.00	0.49
5	20	21	16	17	18.50	-1.97
6	17	13	25	27	20.50	-0.60
7	27	18	19	21	21.25	-0.56
8	24	26	22	26	24.50	0.86
9	22	21	14	14	17.75	-1.35

t.05 = 2.45
t.01 = 3.71

Trial 3.

Soil	Replicate Counts				Average Count	t Value
	A.	B.	C.	D.		
Control	19	11	29	17	19.00	
1	18	25	27	22	23.00	0.95
2	13	23	23	25	21.00	0.43
3	11	22	18	22	18.25	-0.16
4	20	16	23	22	20.25	0.31
5	19	27	21	20	21.75	0.56
6	17	21	15	16	17.25	0.44
7	16	17	22	19	18.50	-0.13
8	13	19	29	18	19.75	0.15
9	20	22	24	16	20.50	9.36

t.05 = 2.45
t.01 = 3.71

Trial 1. The effect of sterile soil in Czapek's agar on numbers of Helminthosporum sativum.

Soil	Replicate Counts				Average Count	t_1 # Value	t_2 ## Value
	A.	B.	C.	D.			
Control 1	9	10	5	8	8.00		
2	8	5	12	9	8.50		
1	12	8	7	9	9.00	0.66	0.28
2	14	13	15	12	13.50	4.38**	3.16*
3	13	12	11	17	13.25	3.09*	2.43
4	14	10	17	7	12.00	1.63	1.33
5	10	11	18	12	12.75	2.27	1.85
6	16	15	13	10	13.50	3.22*	2.56
7	12	8	15	16	12.75	2.27	1.85
8	11	9	16	15	12.75	2.41	1.94
9	18	4	10	9	10.25	0.73	0.54
						t.05 =	2.45
						t.01 =	3.71

\# Control 1 used in calculating t_1

\## Control 2 used in calculating t_2

Trial 2.

Soil	Replicate Counts A.	B.	C.	D.	Average Count	t_1# Value	t_2## Value
Control 1	6	5	4	7	5.50		
2	5	7	4	8	6.00		
1	7	9	6	10	8.00	2.24	1.55
2	8	7	8	11	8.50	2.78*	1.99
3	7	6	8	12	8.25	1.88	1.41
4	6	5	6	6	8.75	0.36	0.26
5	5	6	8	4	5.75	0.21	0.20
6	13	9	10	5	5.75	2.12	1.72
7	7	8	9	11	9.25	3.05*	2.20
8	9	4	8	10	7.75	1.54	1.09
9	9	4	10	8	7.75	1.54	1.09
						t.05 =	2.45
						t.01 =	3.71

\# Control 1 used in calculating t_1.

\## Control 2 used in calculating t_2.

Trial 1. The effect of sterile soil in Czapek's agar on numbers of a saltant of Helminthosporium sativum

Soil	Replicate Counts				Average Count	t_1 # Value	t_2 ## Value
	A.	B.	C.	D.			
Control 1	9	9	9	10	9.25		
2	14	10	11	10	11.25		
1	4	8	7	9	7.00	-2.03	-2.96*
2	8	9	7	6	7.50	-2.53*	-2.28
3	7	3	8	8	6.50	-2.26	-3.12*
4	8	8	10	7	8.25	-1.48	-2.64*
5	9	6	9	8	8.00	-1.67	-1.75
6	8	7	10	7	8.00	-1.67	-1.75
7	8	5	8	6	6.75	-3.16*	-2.73*
8	9	11	7	7	8.50	-0.76	-1.04
9	13	7	7	6	8.25	-0.62	-1.61

$t_{.05} = 2.45$
$t_{.01} = 3.71$

\# Control 1 used in calculating t_1.

\## Control 2 used in calculating t_2.

Trial 1. The effect of sterile soil in nutrient agar on numbers of *Serratia marcescens*

Soil	Replicate Counts				Average Count	t Value
	A.	B.	C.	D.		
Control	36	39	40	56	42.75	
1	45	47	39	45	44.00	0.26
2	42	30	45	48	41.25	-0.25
3	40	48	40	70	49.50	0.80
4	31	40	44	48	40.75	-0.46
5	39	43	45	65	48.00	0.72
6	52	49	39	33	43.25	0.08
7	46	54	37	34	42.75	0.00
8	27	34	49	32	35.50	-1.11
9	27	25	21	34	26.75	-3.03**

t.05 = 2.45
t.01 = 3.71

Trial 2.

Soil	Replicate Counts				Average Count	t Value
	A.	B.	C.	D.		
Control	27	22	23	26	24.50	
1	30	36	34	30	32.50	4.18 **
2	36	33	31	26	31.50	2.90 *
3	38	29	29	37	33.25	3.20 *
4	23	21	22	27	23.25	-0.71
5	41	36	34	28	34.75	3.49 *
6	30	29	28	24	27.75	1.83
7	22	31	29	24	26.50	0.83
8	18	14	22	17	17.75	-3.32 *
9	23	22	18	11	18.50	-2.02

t.05 = 2.45
t.01 = 3.71

Trial 3.

Soil	Replicate Counts				Average Counts	t Value
	A.	B.	C.	D.		
Control	26	23	20	19	22.00	
1	28	31	27	33	29.75	3.58 *
2	33	30	22	30	28.75	2.38
3	40	29	25	35	32.25	2.82 *
4	20	18	21	24	20.75	-1.36
5	38	33	31	25	31.75	3.13 *
6	27	26	25	21	22.25	0.10
7	20	29	26	21	24.10	0.76
8	12	11	9	29	15.25	-2.21
9	21	19	10	14	16.00	-1.62

t.05 = 2.45
t.01 = 3.71

The effect of soil on the pH of nutrient agar.

Soil		Soil concentrations per 10 ml. of agar				
		0.00	0.02	0.01	0.001	0.0001
1.	A	6.80	6.79	6.74	6.80	6.80
	B	6.80	6.80	6.80	6.81	6.79
2.	A	6.80	6.79	6.75	6.79	6.79
	B	6.80	6.76	6.79	6.81	6.80
3.	A	6.80	6.81	6.79	6.79	6.80
	B	6.80	6.79	6.81	6.80	6.80
4.	A	6.80	6.80	6.79	6.74	6.76
	B	6.80	6.74	6.80	6.81	6.80
5.	A	6.80	6.79	6.81	6.80	6.74
	B	6.80	6.74	6.80	6.80	6.79
6.	A	6.80	6.79	6.74	6.80	6.79
	B	6.80	6.80	6.79	6.81	6.80
7.	A	6.80	6.80	6.75	6.81	6.80
	B	6.80	6.80	6.79	6.80	6.80
8.	A	6.80	6.81	6.81	6.80	6.80
	B	6.80	6.80	6.79	6.80	6.80
9.	A	6.80	6.80	6.80	6.81	6.80
	B	6.80	6.81	6.81	6.79	6.80

. The effect of soil on the pH of Czapek's agar

Soil		Soil concentrations per 10 ml. of agar				
		0.00	0.02	0.01	0.001	0.0001
1.	A	5.60	5.75	5.65	5.65	5.60
	B	5.60	5.69	5.70	5.60	5.61
2.	A	5.60	5.61	5.60	5.61	5.60
	B	5.60	5.61	5.59	5.60	5.60
3.	A	5.60	5.61	5.61	5.61	5.60
	B	5.60	5.60	5.60	5.61	5.60
4.	A	5.59	5.64	5.60	5.61	5.60
	B	5.60	5.64	5.60	5.65	5.60
5.	A	5.60	5.60	5.60	5.60	5.59
	B	5.60	5.60	5.60	5.60	5.60
6.	A	5.60	5.60	5.60	5.60	5.59
	B	5.60	5.64	5.64	5.60	5.59
7.	A	5.60	5.65	5.60	5.65	5.60
	B	5.60	5.60	5.61	5.60	5.60
8.	A	5.59	5.85	5.95	5.65	5.61
	B	5.59	5.84	5.84	5.65	5.61
9.	A	5.60	6.00	5.85	5.74	5.74
	B	5.60	6.01	5.84	5.75	5.70

The effect of soil on the pH of potato dextrose agar.

Soil		Soil concentrations per 10 ml. of dextrose agar				
		0.00	0.02	0.01	0.001	0.0001
1.	A	5.60	5.60	5.60	5.59	5.59
	B	5.60	5.59	5.59	5.60	5.60
2.	A	5.60	5.60	5.60	5.60	5.60
	B	5.60	5.61	5.61	5.60	5.60
3.	A	5.60	5.60	5.60	5.60	5.60
	B	5.61	5.60	5.61	5.61	5.60
4.	A	5.60	5.61	5.61	5.60	5.60
	B	5.60	5.60	5.61	5.60	5.60
5.	A	5.60	5.61	5.61	5.60	5.60
	B	5.60	5.60	5.61	5.60	5.60
6.	A	5.60	5.60	5.61	5.60	5.60
	B	5.60	5.60	5.60	5.60	5.60
7.	A	5.60	5.55	5.60	5.60	5.60
	B	5.61	5.59	5.60	5.60	5.59
8.	A	5.60	5.70	5.65	5.60	5.60
	B	5.60	5.71	5.61	5.60	5.61
9.	A	5.60	5.99	5.80	5.61	5.60
	B	5.60	5.94	5.81	5.64	5.65

The effect of soil on the pH of asparagin mannitol agar.

Soil		Soil concentrations per 10 ml. of agar				
		0.00	0.02	0.01	0.001	0.0001
1.	A	7.29	7.29	7.30	7.29	7.29
	B	7.29	7.30	7.29	7.29	7.29
2.	A	7.29	7.29	7.29	7.30	7.29
	B	7.29	7.29	7.29	7.29	7.29
3.	A	7.29	7.29	7.30	7.29	7.29
	B	7.29	7.29	7.29	7.29	7.29
4.	A	7.29	7.29	7.29	7.29	7.29
	B	7.30	7.29	7.30	7.29	7.29
5.	A	7.29	7.29	7.30	7.29	7.29
	B	7.30	7.29	7.29	7.30	7.29
6.	A	7.30	7.29	7.29	7.29	7.30
	B	7.29	7.30	7.30	7.29	7.30
7.	A	7.29	7.29	7.29	7.29	7.29
	B	7.29	7.29	7.29	7.29	7.29
8.	A	7.29	7.29	7.30	7.29	7.29
	B	7.29	7.29	7.29	7.30	7.29
9.	A	7.29	7.30	7.29	7.30	7.29
	B	7.29	7.29	7.29	7.29	7.29

. The effect of soil on the pH of sodium albuminate agar

Soil		Soil concentrations per 10 ml. of agar				
		0.00	0.02	0.01	0.001	0.0001
1.	A	7.50	7.51	7.50	7.50	7.50
	B	7.50	7.49	7.50	7.50	7.50
2.	A	7.50	7.50	7.50	7.50	7.49
	B	7.51	7.50	7.51	7.51	7.50
3.	A	7.50	7.50	7.51	7.49	7.50
	B	7.49	7.50	7.50	7.50	7.50
4.	A	7.51	7.50	7.51	7.50	7.49
	B	7.51	7.50	7.50	7.50	7.50
5.	A	7.50	7.50	7.50	7.50	7.50
	B	7.49	7.50	7.50	7.49	7.49
6.	A	7.50	7.50	7.55	7.50	7.51
	B	7.49	7.50	7.50	7.49	7.50
7.	A	7.50	7.55	7.50	7.51	7.50
	B	7.50	7.50	7.50	7.50	7.49
8.	A	7.50	7.50	7.50	7.50	7.45
	B	7.50	7.50	7.49	7.50	7.50
9.	A	7.50	7.71	7.71	7.71	7.64
	B	7.50	7.70	7.71	7.71	7.64

The effect of soil on the pH of soil-extract agar (Soil 7).

Soil		Soil concentrations per 10 ml. agar				
		0.00	0.02	0.01	0.001	0.0001
5.	A	7.25	7.15	7.20	7.25	7.20
	B	7.24	7.20	7.24	7.21	7.20
7.	A	7.25	7.21	7.24	7.21	7.21
	B	7.21	7.19	7.24	7.25	7.21
9.	A	7.25	7.45	7.24	7.29	7.29
	B	7.24	7.44	7.21	7.25	7.24

The effect of soil on the pH of soil-extract agar (Soil 9).

Soil		Soil concentrations per 10 ml. agar				
		0.00	0.02	0.01	0.001	0.0001
5.	A	7.50	7.31	7.30	7.50	7.45
	B	7.54	7.31	7.29	7.50	7.49
6.	A	7.50	7.50	7.49	7.51	7.51
	B	7.51	7.51	7.50	7.50	7.49
9.	A	7.50	7.51	7.49	7.50	7.54
	B	7.54	7.54	7.49	7.49	7.49

pH of Media Containing Different Soils

Fusarium avenaceum

Replication	Soil	Without fungi replicates		With fungi replicates			
		A.	B.	A.	B.	C.	D.
1.	A	5.99	6.00	7.25	7.54	7.47	7.30
	B	5.99	6.00	7.01	7.25	7.40	7.01
	C	6.01	5.99	7.31	6.90	6.85	7.01
	D	6.00	5.96	7.01	7.31	7.29	7.40
	E	7.74	7.75	7.40	7.20	6.99	7.01
	F	6.92	6.95	7.00	6.99	6.95	6.75
	G	5.70	5.74	7.51	7.49	7.44	7.01
	H	5.96	5.99	7.49	7.15	7.20	7.15
	I	6.25	6.24	7.80	7.61	7.40	7.79
2.	A	6.01	5.99	6.89	6.65	6.90	7.01
	B	6.01	6.05	6.70	6.75	6.90	6.75
	C	6.00	6.01	6.75	6.85	6.90	7.14
	D	5.99	6.00	6.85	6.85	7.01	6.71
	E	5.80	5.84	6.81	6.75	6.89	7.04
	F	5.99	5.99	6.95	7.20	7.19	7.30
	G	5.69	5.75	7.01	7.04	7.20	6.99
	H	6.00	5.99	7.01	7.11	6.90	6.99
	I	6.21	6.24	8.20	7.70	7.40	7.70
3.	A	6.00	6.00	6.90	6.85	7.01	7.00
	B	6.00	5.96	6.70	6.90	6.89	6.90
	C	5.94	6.00	6.90	6.95	7.20	7.00
	D	5.95	6.00	6.95	7.01	7.04	7.01
	E	5.80	5.81	6.99	7.20	7.21	7.20
	F	6.00	5.99	6.99	7.00	6.99	7.14
	G	5.71	5.69	7.00	6.99	7.11	6.85
	H	6.00	6.04	6.99	7.20	6.75	6.99
	I	6.00	6.04	7.44	7.70	6.99	7.54

pH of Media Containing Different Soils

Fusarium graminearum

Replication	Soil	Without fungi replicates		With fungi replicates			
		A.	B.	A.	B.	C.	D.
1.	A	5.99	6.00	7.60	7.80	7.75	7.62
	B	5.95	5.94	7.76	7.91	7.75	7.61
	C	5.96	5.95	7.50	7.60	7.75	7.79
	D	5.96	5.94	7.61	7.55	7.89	7.75
	E	5.79	5.75	7.75	7.70	7.66	7.74
	F	5.95	5.93	7.70	7.81	7.65	
	G	5.76	5.79	7.64	7.55	7.80	7.69
	H	5.99	6.00	7.71	7.90	7.90	7.79
	I	6.25	6.21	8.00	8.10	7.81	8.15
2.	A	5.99	5.96	7.70	7.65	7.61	7.69
	B	5.94		7.74	7.79	7.90	7.61
	C	5.91	5.96	7.69	7.70	8.01	7.59
	D	5.95	5.94	7.80	8.01	7.61	7.59
	E	5.75	5.79	7.79	7.90	7.69	7.94
	F	5.94	5.90	7.50			
	G	7.76	5.69	7.79	7.90	7.70	7.81
	H	6.00	6.04	7.91	7.60	7.79	8.00
	I	6.21	6.26	7.72	7.90	7.74	7.79
3.	A	5.99	6.01	7.65	7.60	7.75	7.70
	B	6.00	5.99	7.60	7.59	7.79	7.91
	C	5.94	5.97	7.50	7.50	7.54	7.80
	D	5.91	5.94	7.80	7.71	7.70	7.84
	E	5.81	5.79	7.75	7.91	7.60	7.75
	F	5.94	5.97	7.75	7.80	7.90	7.99
	G	5.70	5.75	7.74	7.80	8.00	8.01
	H	6.00	5.99	7.81	7.84	8.04	7.69
	I	6.26	6.20	7.91	7.70	7.72	7.95

pH of Media Containing Different Soils

Penicillium Melinii

Replication	Soil	Without fungi replicates		With fungi replicates			
		A.	B.	A.	B.	C.	D.
1.	A	5.90	5.95	6.50	7.25	6.25	6.01
	B	5.91	5.95	5.65	5.45	5.70	6.00
	C	5.99	6.00	7.61	5.80	5.94	7.04
	D	5.94	5.99	7.21	6.94	7.74	7.05
	E	5.75	5.70	7.01	7.45	5.90	6.41
	F	5.99	6.01	5.01	7.41	5.99	6.21
	G	5.65	5.71	7.40	6.94	7.50	7.55
	H	6.05	6.24	7.40	5.99	6.61	7.41
	I	6.20	6.24	7.40	5.99	6.61	7.41
2.	A	5.99	6.00	5.85	6.80	6.99	7.41
	B	5.94	5.95	6.99	7.44	7.71	7.01
	C	5.90	5.95	7.24	6.30	6.29	6.40
	D	5.99	5.94	7.61	7.14	7.05	7.20
	E	5.80	5.75	6.10	6.44	6.74	7.25
	F	5.99	5.95	7.30	6.55	6.71	6.30
	G	5.75	5.69	6.80	7.01	7.21	6.85
	H	6.05	6.09	6.75	7.00	7.20	7.65
	I	6.21	6.25	6.89	6.60	6.80	6.39
3.	A	5.95	5.99	6.65	6.60	6.81	5.90
	B	5.99	5.95	5.70	7.35	5.45	6.90
	C	5.95	5.99	6.50	7.01	6.41	6.70
	D	5.95	5.99	7.64	7.14	7.06	7.51
	E	5.75	5.79	5.55	7.45		5.70
	F	6.00	6.00	6.50	6.10	6.50	6.41
	G	5.69	5.65	6.90	6.95	6.50	6.95
	H	6.09	6.10	7.01	7.00	7.41	7.50
	I	6.21	6.25	6.71	6.50	5.50	6.51

pH of Media Containing Different Soils

Penicillium viride-dorsum

Replication	Soil	Without fungi replicates		With fungi replicates			
		A.	B.	A.	B.	C.	D.
1.	A	5.94	5.99	5.40	5.31	5.41	5.21
	B	5.94	5.99	5.51	5.71	5.03	5.24
	C	5.91	6.00	5.61	5.01	4.85	5.44
	D	5.95	5.99	4.60	5.40	5.60	5.40
	E	5.75	5.65	5.60	4.95	5.46	5.49
	F	5.90	5.91	4.95	5.40	4.90	5.20
	G	5.70	5.69	4.71	4.70	4.99	5.01
	H	6.00	6.01	4.44	5.04	4.48	5.53
	I	6.24	6.34	5.15	4.81	5.35	5.60
2.	A	5.99	5.96	4.99	5.69	4.99	5.20
	B	5.94	5.99	5.30	5.10	4.90	4.79
	C	5.91	5.94	5.10	5.20	4.79	5.30
	D	5.94	5.95	4.30	4.85	4.65	4.90
	E	5.69	5.70	5.00	5.10	5.50	5.10
	F	5.95	5.91	5.09	4.90	4.60	5.35
	G	5.75	5.75	4.70	4.75	4.89	5.00
	H	6.00	6.05	4.72	4.68	4.23	4.63
	I	6.24	6.20	5.00	5.10	4.85	4.90
3.	A	5.94	5.91	4.83	5.69	4.99	5.69
	B	5.95	5.90	5.25	4.50	5.00	4.79
	C	5.90	5.91	5.20	4.79	4.50	5.13
	D	5.90	5.91	4.30	4.94	5.10	5.31
	E	5.75	5.79	5.25	4.61	4.55	4.81
	F	5.90	5.95	5.01	4.41	4.45	5.05
	G	5.75	5.69	4.45	4.75	4.14	4.01
	H	5.99	6.00	4.67	4.83	4.23	4.18
	I	6.20	6.26	5.01	4.85	4.35	5.01

pH of Media Containing Different Soils

Aspergillus fumigatus

Replication	Soil	Without fungi replicates		With fungi replicates			
		A.	B.	A.	B.	C.	D.
1.	A	5.99	5.94	7.19	7.70	7.75	7.20
	B	6.01	6.00	7.45	7.69	7.59	7.60
	C	5.95	5.90	7.69	7.50	7.45	7.35
	D	5.95	5.94	7.61	7.81	7.45	7.55
	E	5.74	5.79	7.54	7.05	7.61	7.40
	F	5.99	5.95	7.81	7.41	7.29	7.69
	G	5.69	5.70	7.41	7.55	7.54	7.40
	H	6.09	6.00	7.31	7.61	7.51	7.41
	I	6.29	6.20	7.71	7.51	7.55	7.86
2.	A	5.91	6.00	7.50	7.46	7.44	6.91
	B	6.00	6.01	7.59	7.60	7.60	7.01
	C	5.94	5.89	7.65	7.51	7.35	7.36
	D	5.95	5.99	7.34	7.69	6.90	7.14
	E	5.65	5.71	6.89	7.51	7.21	7.55
	F	6.00	5.99	7.45	7.26	7.41	7.35
	G	5.65	5.69		6.91	7.41	7.61
	H	6.01	6.09	7.34	7.41	7.19	7.30
	I	6.10	6.10	7.05	7.29	7.54	7.45
3.	A	6.00	5.95	7.40	7.75	7.20	7.09
	B	6.00	5.94	7.60	7.45	7.10	7.60
	C	5.95	5.90	7.69	7.10	7.30	7.15
	D	5.90	5.95	7.41	7.09	7.40	7.50
	E	5.70	5.59	7.54	7.05	7.20	7.60
	F	5.99	5.95	7.70	7.50	7.40	7.10
	G	5.65	5.65	7.40	7.60	7.30	7.45
	H	6.10	6.15	7.30	7.60	7.50	7.20
	I	6.25	6.29	7.79	7.29	7.56	7.41

pH of Media Containing Different Soils

Helminthosporium sativum

Replication	Soil	Without fungi replicates		With fungi replicates			
		A.	B.	A.	B.	C.	D.
1.	A	6.01	6.10	8.70	8.50	8.21	8.00
	B	6.05	6.00	8.25	7.41	8.10	7.79
	C	5.99	6.00	8.41	8.75	8.45	8.50
	D	5.99	6.00	8.55	8.60	8.71	8.79
	E	5.70	6.05	8.21	8.31	8.71	8.29
	F	6.00	6.05	8.20	8.40	8.61	7.41
	G	7.75	5.81	8.41	8.20	8.41	8.60
	H	6.09	6.10	8.74	8.50	7.49	8.00
	I	6.29	6.30	8.25	8.45	8.00	8.01
2.	A	5.94	5.99	8.40	8.01	8.30	8.31
	B	6.00	5.95	8.00	8.25	7.90	7.94
	C	5.90	5.95	8.41	7.90	8.60	8.40
	D	5.95	5.90	8.41	8.00	7.90	7.74
	E	5.75	5.79	7.79	8.20	8.21	8.30
	F	6.00	5.99	8.34	7.79	7.90	7.99
	G	5.79	5.71	8.01	8.30	7.90	8.71
	H	6.15	6.25	8.41	8.55	8.31	8.41
	I	6.30	6.25	8.41	8.55	8.31	8.41

pH of Media Containing Different Soils

S. marcescens

Replication	Soil	Without bacteria replicates		With bacteria replicates			
		A.	B.	A.	B.	C.	D.
1.	A	6.60	6.55	8.00	8.15	8.30	7.90
	B	6.50	6.46	8.10	8.00	7.56	7.99
	C	6.31	6.35	8.14	8.10	8.15	8.20
	D	6.29	6.35	8.20	8.10	8.25	7.91
	E	6.50	6.45	7.90	7.90	8.01	8.00
	F	6.89	6.91	8.01	8.15	8.20	8.05
	G	6.49	6.55	7.90	7.99	8.01	8.14
	H	6.65	6.70	7.91	8.00	8.10	8.01
	I	6.81	6.85	8.01	8.00	8.15	7.95
2.	A	6.65	6.69	8.01	8.15	8.35	7.95
	B	6.51	6.49	8.15	7.90	7.99	7.95
	C	6.29	6.34	8.20	8.35	7.99	8.00
	D	6.30	6.35	7.90	7.91	7.79	8.21
	E	6.55	6.49	7.94	7.95	7.80	8.31
	F	6.80	6.59	8.15	8.20	8.15	8.21
	G	6.51	6.59	8.15	8.20	8.15	8.21
	H	6.69	6.71	7.91	7.99	8.00	8.15
	I	6.80	6.85	8.04	8.00	8.09	8.31
3.	A	6.61	6.54	8.19	8.29	7.90	8.01
	B	6.55	6.49	8.35	7.99	7.56	7.91
	C	6.30	6.31	8.14	8.10	8.05	8.06
	D	6.35	6.30	8.10	8.25	8.01	8.45
	E	6.50	6.55	7.90	8.31	7.90	8.40
	F	6.85	6.80	7.91	8.00	8.01	8.45
	G	6.45	6.50	8.14	7.79	8.45	8.00
	H	6.61	6.70	7.90	7.70	7.79	8.21
	I	6.80	6.81	8.00	7.90	7.99	8.01