## FUNGI AT VARIOUS DEPTHS IN TYPICAL CLEVELAND COUNTY, OKLAHOMA SOILS\*

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In 1902, the subject of soil fungi began to be treated in a systematic and an exact manner when Oudemans and Koning published a paper which represented the first real attempt at a proper study of the occurrence of fungi in the soil.

The very recent work on soil fungi in the United States has been done in New England, in the northwest, and elsewhere. During the first part of 1932 research was done in Texas on the species of fungi found in the soil of a pine forest (1). No general counts have hitherto been reported from Oklahoma, and few indeed from the southern states as a whole.

The work reported here included the fungi in typical Cleveland county, Oklahoma, soils at various depths, extended over a years time; and accurate counts as to the numbers of organisms found were made.

It may be noted that one of the serious problems in micro-organic work has been that of the preparation of a medium on which will grow the greatest proportion of the total number of organisms under consideration. This problem represents one of the first difficulties in the study of the population of the soil. After experimenting with potato dextrose agar, prune agar, and several synthetic preparations, it was found that a synthetic medium, the recipe for which was proposed by Lipman (2) and later by Brown (3), yielded greater and more varied counts of the fungithan any other, and was, therefore, adopted for this work. The Phycomycetes, Ascomycetes, and Fungi Imperfecti which appeared on the agar were counted. The contents of this medium in the proportions given are: 10 grams dextrose, 50 gram potassium phosphate, 20 gram magnesium sulfate, .05 gram peptone, 20 grams agar, and 1,000 cc water.

All materials were prepared before the samples of soil were secured. Glassware and flasks containing distilled water for dilution were sterilized. The pipettes, Petri dishes, etc., were placed in the dry ovens for four hours at 145° C. The flasks were sterilized in the autoclave at fifteen pounds pressure for thirty minutes.

While the sterilized material was cooling, the samples of soil were taken at successive depths of two, eight, twenty, and forty centimeters below the surface. A sterile trowel, which was prepared in the field by being dipped in alcohol and flamed just before the soil was taken, was used for the samples. The soil was placed in squares of parchment paper, wrapped, and tied to exclude atmospheric contamination. The samples were then brought to the laboratory immediately and the dilution work started.

A fresh soil sample was taken and 12.5 grams of it were placed in 25 cc of sterile water. The mixture was shaken thoroughly, and one cc was transferred to 90 cc sterile water. This portion was shaken well and 10 cc was transferred to 90 cc sterile water. This was continued with sterile pipettes until the ultimate dilutions were 1-200,000 and 1-2,000,000 Plates were poured for both dilutions, and three of each, six for each sample, were placed in the incubator for 30 hours at 37° C. At the end of this period the total number of colonies of fungi were counted, the numbers of Aspergillus niger colonies were counted separately, and the per cent of the total number of colonies represented by A. niger was calculated.

<sup>\*</sup>Contribution from the Botanical Laboratories, University of Oklahoma, N. S. No. 21.

It is definitely known that the error introduced in quantitative fungus work is greater than in bacteriological counts. Laboratory air is more greatly contaminated with fungus spores than with bacteria. Then, the total number of fungi in the soil, in general, is less than the total number of bacteria, thus, the per centage of error would be greater in the fungus counts. Statistical methods may be used for determining the error of the counts made. With this explanation the forthcoming data are presented.

The four different types of soils appearing in Cleveland County were studied (Fig. 1). This made possible, between the different types as well as bethe varying tween plant communities. numerical comparisons of the fungi found.

One hundred samtwenty-five locations. or stations. Four different depths in each location were sampled. The total numbers of molds occurring in each sample were counted.

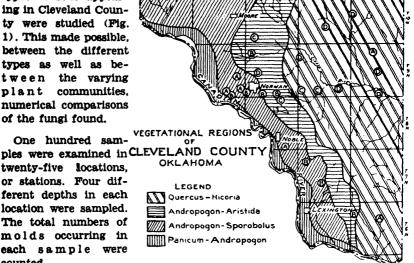
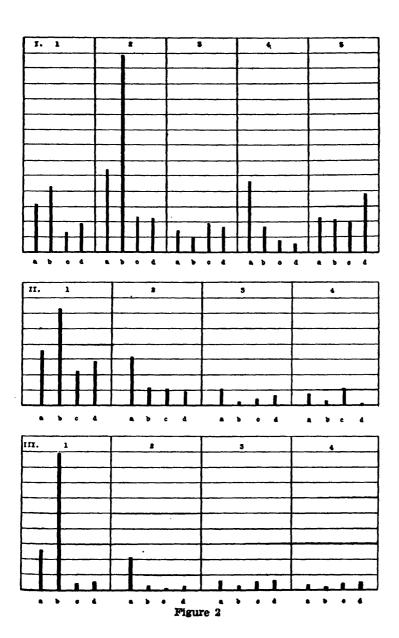


Figure 1. A-stations where soil was sandy. B-stations of silt soil. C-stations of loam. D-stations of clay.

Results for this study show that at a depth of 8 cm below the surface there were many more fungi per gram of soil than at the other depths (Fig. 2, Part I, Section 1, Line b). Upon examination there appeared to be more humus deposited at this depth than on the surface or at the other depths studied. Since humus harbors more micro-organisms than soil devoid of organic material this may explain the greater numbers at 8 cm. The very loose sandy soils had abundant populations of Phycomycetes. Ascomycetes, and Fungi Imperfecti. The numbers of these organisms diminish as the porosity of the soil becomes less, with the exception of the soils containing large percentages of humus (Fig. 2, Part I, Section 2).

Grasslands showed more molds per gram than soil on which there were other types of plant growth. The woods showed the next greatest numbers. Shrub communities had few fungi, while those samples taken where only annuals and ruderals were growing showed the least numbers present (Fig. 2. Part II).

A surprising fact is the large percentages of Aspergillus niger present (Fig. 2, Part III.) Of all the fungi counted 36.3 percent belonged to the A niger group. The percentages in the individual samples ranged from 0 to 100



percent. This phenomenon is very difficult to explain. According to Thom (4) A. niger is strictly aerobic and also is able to grow quite high temperatures. The following facts may offer some explanation. Soils in southern climates are warmer the year around than those in the north. This would make the temperature of the soil in the south more nearly constant. Frozen soil is more tightly packed, and thus would be less aerated than warm soils. Lack of sufficient data on A. niger in northern soils makes further comparison impossible. Soils in the southern states are much higher in fungi content, in general, than are the soils in the northern United States (5).

- Figure 2. I. Fungi in different types of soil. Section I shows the average numbers of fungi per gram of soil for all soils studied, Section 2, the average for the loose, sandy soils. Section 3, the average for silt. Section 4, for loam. Section 5, clay. For all sections: a represents the depth of 2 cm below the surface; b represents the depth of 8 cm; c, the depth of 20 cm; and d, 40 cm.
- II. Plant communities and numbers of fungi. Section 1 shows the average for prairie or grasslands; section 2, for woods; section 3, for shrub communities; and section 4, for collections of soil made where only annuals and ruderals were growing. (Lines a, b, c, and d—same as in I.)
- III. Aspergillus niger group occurrence in different types of soil. Section 1 represents the average numbers in loose, sandy soils; section 2, for loam; section 3, silt; and section 4, clay. (Lines a, b, c, and d—same as in I.)

Scale for Fig. 1. Horizontal space divisions represent 100,000 each.

## CONCLUSIONS

- 1. More fungi were found at a depth of 8 cm below the surface than at any other depth studied.
- 2. Sandy soils showed a larger number of fungi than any other type of soil.
- 3. Aspergillus niger inhabits the soil in large numbers. This group of organisms shows a preference for sandy soils, as the percentages of A. niger in loose, porous soils were very high.
- 4. Striking variations in numbers of fungi in the same types of soil may occur as a result of cultivation, climatic conditions, plant growth, and other factors.

## LITERATURE CITED

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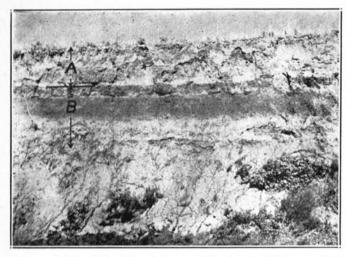


Figure 1. A buried soil located east of Waukomis, Oklahoma along Skeleton Creek.