# DEVELOPMENT OF FUNGISTASIS IN AUTOCLAVED SOIL BY SOIL FUNGAL INOCULA*

*by* RAJPATI DWIVEDI, *Department of Botany, Banaras Hindu University, Varanasi-5*

(Communicated by R. Misra, F.N.A.)

(*Received* 27 *December* 1971; *after revision* 14 *September* 1972)

Sterilized soil inoculated individually with 35 species of fungi belonging to different groups, viz., phycomycetes—7, ascomycetes—5, deuteromycetes—22 and mycelia sterilia, developed fungistatic activity of different grades after two months of incubation. It was noted that some fungi developed high fungistatic activity while others developed the least. The fungistatic property was apparently more prevalent among deuteromycetous fungi. The higher fungistatic activity in sterile soil was recorded in case of *Cunninghamella bertholletiae, Aspergillusnidulans, A. flavus, Spegazzinia parvum* sp. nov., *Trichoderma album, T. viride, T. koningii, T. lignorum, Alternaria tenuis, Cladosporium herbarum* and *Penicillium nigricans.*

## INTRODUCTION

Sterile soil normally not fungistatic becomes fungistatic following the addition of natural soil or inoculation of specific micro-organisms (Lockwood 1964). Autoclaved soil was made fungistatic by inoculating non-antibiotic as well as antibiotic producing micro-organisms (Griffin 1962; Lockwood and Lingappa 1963). The main objective of the present investigation is to study the capacity of some of the individual soil-inhabiting microfungi to develop fungistatic activity in the autoclaved soil.

## MATERIALS AND METHODS

Thirty-five fungi listed in Table I were inoculated individually in known quantity of sterile grassland soil. Individual fungus was grown on Czapek-agar and after a week of incubation at 25°C, small agar blocks of uniform size were cut with a sterile cork borer from the fungal colony and equal number of them were added to glass jars, each containing 100 g sterile, sieved and moist (60 per cent) soil, and incubated for two months at 25°C to allow the growth of fungi. The fungistatic property was assessed by agar-disc technique of Jackson (1958) with two test fungi viz., *Aspergillus niger* and *Fusarium oxysporum* f. *udum.*

## RESULTS AND DISCUSSION

The antifungal property almost similar to that of natural soil was restored in sterilized soil by inoculating fungi of different classes in the present study. Spore

*A part of thesis approved for Ph.D. degree of the Banaras Hindu University.

TABLE I

*The average % germination of the test fungi on sterile soil pre-inoculated with various soil fungi for development of fungistasis*

| Name of fungi pre-inoculated in sterile soil | Test Fungi | | Average % germination (average of 8 replicates) |
|---|---|---|---|
| | *Aspergillus niger* | *Fusarium oxysporum* f. sp. *udum* | |
| Phycomycetes | | | |
| *Cunninghamella bertholletiae* | 47.24 | 48.25 | 47.74 |
| Sterile phycomycete | 52.19 | 51.63 | 51.91 |
| *Zygorhynchus* sp. | 60.70 | 61.61 | 61.11 |
| *Mucor luteus* | 61.51 | 61.51 | 61.51 |
| *Helicostylum* sp. | 60.56 | 68.46 | 64.51 |
| *Actinomucor* sp. | 70.30 | 70.17 | 70.22 |
| *Absidia spinosa* | 70.03 | 74.83 | 72.43 |
| Ascomycetes | | | |
| *Aspergillus nidulans* | 38.87 | 40.29 | 39.58 |
| *Penicillium javanicum* | 41.81 | 44.12 | 42.96 |
| *P. rubrum* | 44.33 | 44.11 | 44.22 |
| *Chaetomium globosum* | 48.37 | 54.40 | 51.38 |
| *Thielavia terricola* | 53.34 | 58.09 | 55.71 |
| Deuteromycetes | | | |
| *Spegazzinia parvum* sp. nov. | 34.32 | 37.46 | 35.88 |
| *Trichoderma album* | 34.91 | 37.07 | 35.99 |
| *Trichophyton* sp. | 36.29 | 36.23 | 36.23 |
| *Trichoderma koningii* | 36.66 | 37.88 | 36.49 |
| *Alternaria tenuis* | 34.12 | 40.25 | 37.18 |
| *Trichoderma lignorum* | 34.98 | 39.95 | 37.46 |
| *T. viride* | 38.50 | 39.29 | 38.84 |
| *Cladosporium herbarum* | 38.65 | 39.82 | 39.23 |
| *Penicillium nigricans* | 38.53 | 41.87 | 40.20 |
| *Aspergillus flavus* | 38.88 | 42.19 | 40.55 |
| *Fusarium culmorum* | 40.87 | 42.62 | 41.75 |
| *Stachybotrys* sp. | 40.36 | 43.63 | 42 01 |
| *Aspergillus candidus* | 41.16 | 43.63 | 42.13 |
| *Scopulariopsis* sp. | 37.31 | 48.85 | 43.01 |
| *Penicillium citrinum* | 42.38 | 44 91 | 43.64 |
| *Fusarium* sp. | 42.47 | 47.18 | 44.82 |
| *Paecilomyces fusisporus* | 41.44 | 51.99 | 46.71 |
| *Myrothecium verrucaria* | 51.45 | 51.17 | 51.31 |
| *Helminthosporium sativum* | 50.23 | 55.87 | 52.55 |
| *Aspergillus sulphureus* | 50.68 | 55.56 | 53.12 |
| *Penicillium* sp. | 53.71 | 55.97 | 54.84 |
| *Curvularia tetramera* | 59.14 | 66.91 | 63.02 |
| Mycelia Sterilia | | | |
| White sterile mycelium | 39.84 | 44.92 | 42.38 |
| Control (with sterile soil without fungal inoculum) | 89.36 | 98.00 | 93.68 |
| Control (On sterile moist glass wool) | 98.00 | 98.00 | 98.00 |

germination of the test fungi was reduced by all the fungi of which 7 belonged to phycomycetes, 5 to ascomycetes, 22 to deuteromycetes and 1 to mycelia sterilia (Table I). Fungi of different groups are arranged according to their capacity to develop fungistatic activity in sterilized soil. The highest fungistatic ones in each group are towards the top and those with the least fungistatic activity are towards the bottom. Among phycomycetes, *Cunninghamella bertholletiae*; among ascomycetes, *Aspergillus nidulans, Penicillium javanicum* and *P. rubrum*; among deuteromycetes *Spegazzinia parvum* sp. nov., *Trichoderma album, T. koningii, Trichophyton* sp., *Alternaria tenuis, T. lignorum, T. viride, Cladosporium herbarum, Penicillium nigricans, Aspergillus flavus, Fusarium culmorum, Stachybotrys* sp. *Aspergillus candidus, Scopulariopsis* sp., *Penicillium citrinum, Fusarium* sp., *Paecilomyces fusisporus* and white sterile mycelium developed high fungistatic activity as the average per cent germination of the test fungi was less than 50 per cent. Sterile phycomycete, *Thielavia terricola, Myrothecium verrucaria, Helminthosporium sativum, Aspergillus sulphureus* and *Penicillium* sp. developed less fungistatic activity as the average per cent germination of the test fungi was above 50 per cent and less than 60 per cent. *Zygorhynchus* sp., *Mucor luteus, Helicostylum* sp. and *Curvularia tetramera* developed still less fungistatic activity, whereas *Actinomucor* sp. and *Absidia spinosa* developed the least fungistatic activity as the average per cent germination of the test fungi was still higher. In control with sterile soil and without soil the average per cent germination of the test fungi was 93.68 and 98.00 respectively. Thus it may be presumed that some of the members of different groups of fungi in soil play an important role in imparting soils the fungistatic property. Lockwood (1959) reported that reinoculation of autoclaved soil with *Streptomyces* spp., known to be lytic in culture, induced lytic and fungistatic properties in soil. Lingappa and Lockwood (1961) and Lockwood (1959, 1960) suggested that a wide variety of soil microflora could contribute to soil mycostasis. Further, Lockwood and Lingappa (1963) reported that each of the 67 randomly selected actinomycetes, bacteria and fungi caused inhibition of spore germination of the test organism in direct assays of fungistasis in inoculated soils. Steiner and Lockwood (1970) stated that subsoil which was reinoculated with *Streptomyces* sp., *Verticillium albo-atrum* or natural soil became fungistatic to *Glomerella cingulata*, although this fungus grew well in aqueous extract of the reinoculated subsoil.

## Acknowledgements

Thanks are due to Dr. R. Y. Roy and Dr. R. S. Dwivedi, for suggestions and criticism; Prof R. Misra, F.N.A, for laboratory facilities; and the Director, P.G.I.I.M., B.H.U., for financial assistance.

## References

Griffin, C. G. (1962). Production of fungistatic effect by soil microflora in autoclaved soil. *Phytopathology*, **52**, 90-91.

Jackson, R. M. (1958) Some aspects of soil fungistasis. *J. gen. Microbiol.*, **19**, 319-341.

Lingappa, B. T., and Lockwood, J. L. (1961). The nature of the widespread soil fungistasis. *J. gen. Microbiol.*, **26**, 473.

Lockwood, J. L. (1959). *Streptomyces* spp. as a cause of natural fungitoxicity in soil. *Phytopathology*, **49**, 327-331.

————(1960). Lysis of mycelium of plant pathogenic fungi by natural soil. *Phytopathology*, **50**, 787-789.

————(1964). Soil fungistasis. *A. Rev. Phytopath.*, **2**, 341-362.

Lockwood, J. L., and Lingappa, B. T. (1963). Fungitoxicity of sterilized soil inoculated with soil microflora. *Phytopathology*, **58**, 917-920.

Steiner, G. W., and Lockwood, J. L. (1970). Soil fungistasis : Mechanism in sterilized reinoculated soil. *Phytopathology*, **60**, 89-91.