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ORIGINAL RESEARCH ARTICLE

Soil Mycoprofile of Botanical Garden of Khalsa College, Amritsar, Punjab, India.

Navtej Singh and Ramandeep Kaur*
Department of Agriculture, Khalsa College, Amritsar, Punjab, India.

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Abstract: The samples used for this study were obtained from different rhizosphere garden soil source. The fungi isolates were *Aspergillus spp., Curvularia sp, Cladosporium cladosporioides, Fusarium sp, Fusarium solani, Mucor mucedo, Myrothecium sp., Paecilomyces sp., Phoma sorghina, Rhizopus stolonifer, Sterile mycelium* Yeast sp. and *Verticillum* spp. Some physiological studies show the range of pH from pH 4.92 in CSSS to pH 8.01 in PLRS sample sources. The soil pH values obtained in this study were near neutral ranges, which favour microbial growth. Various species of fungi were encountered in soil sources including the harmful ones. Hence, fungicides can be applied occasionally on these soils to reduce the fungi load in these areas when necessary. Data obtained in this study is valuable to monitor and protect the environment including agriculture products for sustainable economic development.

Key words: Garden soil, Microflora, Physico-chemical, Rhizosphere.

Introduction

A fascinating but little-understood aspect of soil is its biological component. More living organisms occur in soil than in all other ecosystems combined. The living portion of soil is a diverse and dynamic collection of organisms, from types that you can easily see with an unaided eye down to creatures that you can observe only by using a high-powered microscope. Soils contain complex communities of hundreds or thousands of distinct microorganisms. These organisms, which include bacteria, fungi, centipedes, mites, and spiders, feed on the primary consumers. It is a natural culture media for the growth of microorganisms. Maximum microbial growth and activity in the soil is confined around the root systems of plants and this region is called the rhizosphere.

The presence of microbes ensures that nutrients are made available to plants at a steady rate. While the plants are actively growing and requiring more nutrients so do the microbes in the soil. As the weather warms, both the plants and microbes respond at a similar rate. The microbes become increasingly active in their role of breaking down organic materials into forms more readily absorbed by the growing plants that need extra nutrition. As the weather cools and plants require less nutrition so do the microbes. The reduction in their activity means fewer nutrients in the soil are being released to the plants. In this way, the soil can rebuild food reserves. This self-regulating cycle has occurred for millions of years as part of the wisdom of nature. Microbes also help to stabilize the soil by physically binding soil particles together; they release a by-product called glomalin that acts as "glue," binding mineral particles and organisms to each other. This contributes greatly to soil aggregation. All of these processes happen naturally in a healthy, productive soil.

When we feed our plants instead of our soil, we lose all the benefits that microbes contribute. When we say 'feed the soil' it means feed the microbes in the soil, because it is the microbes that make nutrients available for the plants. The way you feed microbes is through the addition of organic material. If you feed with a synthetic chemical fertilizer, you are feeding the plant, not the soil, or the microbes. Adding petrochemical synthetic fertilizer also drives up the salt index in the soil and changes the pH, which can have adverse effects on plants.

More importantly, chemical fertilizers only feed for a short period of time; organic fertilizers offer continual feeding because the microbes cannot digest all of the organic fertilizer at once. With chemical fertilizers, we also lose the microbes' contribution to soil aggregation. Good soil aggregation leads to improvements in tilth, water retention, the rates at which water penetrates the soil, the amount of oxygen in the soil, and the reduction of runoff. All of these desirable soil conditions can be achieved by adding organic material. As you can see, microbes are immeasurably important and essential to the health of all productive soils.

Since the beginning of the soil micro-biological studies (Adametz, 1886) an array of literature has been accumulated from different corners of the world but the workers have restricted their studies on limited soil types specific to the localities. The mycoflora in relation to physico-chemical characters (Chen and Griffin, 1966 b; Kamal, 1968a) and cover vegetation (Christensen, 1969; Deyl, 1938; England and Rice, 1957; Leclerg, 1931, Mallik and Rice, 1966 and Witkamp, 1966) have extensively been studied.The different habitats undertaken for study of soil mycoflora are cultivated fields (Dixon, 1928; Gujati, 1968),

Corresponding Author
Ramandeep Kaur
Nirgunn Overseas Consultants Pvt. Ltd
Level 3, SRK Mall, Mall Road (Above Passport Office),
Amritsar-143001, India.

grasslands (Dwivedi, 1966; Mishra, 1965), sand dunes (Brown, 1958; Moreau and Moreau, 1941; Nicot, 1958; Welby *et al.,* 1952), salt marshes (Baliss-elliot, 1930; Siepmann, 1959 and Turner and Pugh, 1961), coastal soils (Pugh, 1960). Mishra and Kanaujia (1972, 1973 a and 1973 b) laid down the comparative investigation of soil fungi of different localities having diversity in environment and physico-chemical characters. Such studies, however, need further exploration for determination of the soil microflora. In the present investigation an attempt has been made to study the determination of the soil mycoflora of Botanical garden.

Materials and Methods

Sample Sources

The soil samples used for this study were obtained from different rhizosphere places of Botanical garden of Khalsa College, Amritsar in the September, 2014. The soil samples were collected using a sterile spoon for removal of the sub-surface soil into sterile sample bottles which were then transferred to the laboratory for analysis. The samples were preserved at 4^0 in refrigerator to slow down biological activities and reduce chemical reactions in nature.

Inoculums Preparation and Microbiological Analysis of Soil Samples

The stock cultures for this study were prepared by weighing 1g of the soil samples into test tubes containing 10ml of sterile distilled water and shaken properly for homogeneity. A pour plate technique was used to estimate the soil microbial population. The rhizosphere fungi were enumerated by Serial dilution method (Waksman, 1992). Appropriate diluents were plated for microbial enumeration. Potato dextrose agar (PDA) medium was used for fungi cultivation. The plates were allowed to solidify, inverted and incubated at 25^0 for 5 days for fungi [Prescott *et al.,* 2008]. Total microbial counts were estimated for the sample sources.

Identification of Fungi

Each fungal colony was picked from the plates containing massive growth of organisms with sterile inoculating needle and stabbed on sterile plates were prepared containing potato dextrose agar using an aseptic technique. After inoculation, the prepared plates containing each colony were incubated invertedly at 25^0 for 5 days for further identification purposes. A small portion of the mycelium was removed from the fungal culture and teased in the drop of lactophenol cotton blue stain earlier introduced on a clean microscopic slide. The organisms were teased very well and covered with a cover slip, then examined with the aid of a light microscope under objective x40 [Domsch *et al.,* 1995]. The compendium of fungi was used for the identification of fungal isolates.

Physiochemical Analysis

Some physicochemical parameters of the sample sources, such as colour, size, odour, and texture were physically determined amongst other parameters such as moisture contents, pH values, as described below.

Moisture Contents Determination

Clean Petri-dish used for this study was dried in an oven at 80^0 for about 30 minutes. It was then cooled and weighed (W_1). A known weight of the prepared sample sources was placed into the Petri-dish, the weight was also noted (W_2) and kept in an oven maintained at 105^0 for about 3hrs. The Petri-dish with the sample was then removed at hourly intervals for the purpose of getting a constant weight. It was cooled and weighed until a constant weight (W_3) was obtained. The loss in weight during drying is equal to the moisture content of the sample as calculated below:

5g of the soil sample sources were used for the experiments:

Weight of the Petri dish = (W_1)

Weight of the petri dish + soil sample = (W_2)

Petri dish+ dried soil sample ---------- = (W_3)

% moisture = loss of weight due to drying × 100 = W_2-W_3/ W_2-W_1

pH: The pH of each soil sample was measured by using pH meter in the laboratory. The meter was standardized with buffer at pH 4.7 and 9 before use. The pH 7 was equally determined. The sensitive bulb was then immersed into the sample to get appropriate reading for this study (Table 2).

Results

Sample obtained from rhizosphere soil sources for the purpose of this study were determined for some microbiological and physico-chemical parameters. Total of 22 fungal species, comprising were isolated (Tables 1) *viz. Aspergillus spp., Curvularia sp, Cladosporium cladosporioides, Fusarium sp, Fusarium solani, Mucor mucedo, Myrothecium sp., Paecilomyces sp., Phoma sorghina, Rhizopus stolonifer, Sterile mycelium,* Yeast sp. *Verticillum* spp. The *Aspergillus* species were the predominant organism constituting 38.0% of the total isolates while the rest were in least category of the total. The distribution patterns of the fungal species in different rhizoshpere sources were also determined. Here the *Aspergillus* spp. were encountered in all the rhizosphere soil sources.

Table 1: Analysis of soil mycoprofile of Botanical Garden of Khalsa College, Amritsar in 2014.

S.No	Identification of Fungus	Number of Colonies per Serial Dilution					
		Stock 1g/100ml	1:10	1:100	1:1000	1:10000	1:100000
1.	*Aspergillus niger-I*	17	8	1	2	-	-
2.	*Aspergillus niger-I*	14	10	16	5	2	3
3.	*Aspergillus fumigates*	3	1	-	1	-	-
4.	*Aspergillus tamarii*	4	-	1	-	-	-
5.	*Aspergillus sydowii*	1	-	3	-	3	1
6.	*Aspergillus niger-II*	2	3	-	1	-	-
7.	*Aspergillus flavus-II*	-	3	-	-	1	-
8.	*Aspergillus japonicas*	-	2	3	1	-	-
9.	*Curvularia sp.*	-	2	3	1	3	2
10.	*Cladosporium cladosporioides*	3	1	-	1	-	-
11.	*Fusarium sp.*	4	-	2	-	-	1
12.	*Fusarium solani*	4	8	3	5	2	-
13.	*Mucor mucedo*	-	-	1	-	-	-
14.	*Myrothecium sp.*	-	3	-	-	-	-
15.	*Paecilomyces sp.*	2	-	1	2	-	-
16.	*Penicillium chrysogenum*	-	4	5	4	1	1
17.	*Paecilomyces variotii*	2	-	-	-	-	-
18.	*Phoma sorghina*	-	-	1	-	-	-
19.	*Rhizopus stolonifer*	4	3	1	-	-	-
20.	*Sterile mycelium*	-	1	1	-	-	-
21.	Yeast sp.	-	3	3	1	2	1
22.	*Verticillum* spp.	-	-	1	-	1	-
Total number of isolated fungal colonies.		**60**	**52**	**46**	**24**	**15**	**9**

Table 2: Physico-Chemical parameters of the soil samples.

Physico-Chemical Parameters	Sample
Colour	Dark Brown
Odour	Odourless
Size	Small
Moisture Content %	6.67
pH	6.09
Texture	Smooth

Discussion

Various types of microorganism were isolated from the different rhizosphere garden soil samples collected. In the fungal group *Aspergillus* spp is one of the organisms recovered. An immunocompromised individual exposed to high doses of fungi spores such as *Aspergillus* spp, can come down with respiratory problems e.g. asthma. Dahlgren and Driscoll [Dahlgren *et al.,* 1994] showed that some genera of fungi causes diseases of hair, skin, and the nail e.g. *Trichophyton* spp and *Epidermophyton* spp which were also isolated in this study. The nature of isolates obtained in this study also correlates with the report of Thorn [Thorn.,1997] which showed common genera of fungi such as *Chaetomium, Fusarium, Penicillium, Aspergillus, Mucor* and *Alternaria* isolated in the soil. According to Dighton *et al.,* 2005, the development of fungi is especially favoured by the soil having an acidic reaction and where an aerobic condition is likely to be present near the surface since they exist both in mycelia and spore stages. The benefits derived from these fungi include active decomposition of cellulose and lignin of plant tissues and formation of water stable aggregates. Yeasts were not isolated much in this study in corroboration with report of Dighton *et al.,* 2005 that yeasts are generally not found in large number except in soils of vineyards and orchards. Table 2 shows some physico-chemical parameters of the soil sample sources including the colour which was dark brown and the size which were observed as small, smooth texture respectively among other values. This study shows that there are both beneficial and harmful groups of fungi present in the soil. Similarly, some forms of bacterial species encountered apart from coliforms which signify contamination may be indicative of some crop microflora in the planting zone. Some precautionary measures should be taken to protect our environment and assess the applicability of fertilizer based on the microbial population or soil structure of the sampled site. Nevertheless, modern technology (nuclei acid probes) to obtain such detailed overview of microbial diversity can be intensified in extension of this investigation in future. Fungicides can be applied occasionally on this soil sources especially when there are lots of people inhabiting the areas to reduce the fungi load for epidemiological reasons. According to Inderjit *et al.,* 2005 root pathogenic fungi such as Verticillum causes major economic losses in agriculture, so care should be taken to monitor soil sources where these group of organisms are encountered in order to protect our agricultural systems. Hence, the data obtained in this study can serve as a guide to monitor and protect human health including the agriculture products for sustainable economic development.

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Refrences

1. Adametz, L. "Untersuchungen uber die niederen Pilze der Acker–krume". *Inaugural-Dissertation.* Leipzig (1886). 78.

2. Bayliss-Elliot, J. S. "The soil fungi of Dovey salt marshes". *Ann.Appl. Biol.,* 17 (1930): 284.

3. Brown, J. C. "Soil fungi of some British dunes in relation to soil type and succession". *Jour. Ecol.,* 46 (1958): 641 - 664.

4. Chen, A. W. C. and Griffin, D. M. "Soil physical factors and ecology of soil fungi. VI. Interactions between temperature and soil moisture." *Ibid.* 49 (1966b): 551-561.

5. Christensen, M. "The soil microfungi of dry to mesic hardwood conifer forest in Northern Wisconsin". *Ecology,* 50 (1969): 9 - 27.

6. Deyl, M. "Uber die Mikrobiologie der alpinen Boden in den Ostkarpathen". *Stud. Bot. Cechoslovacia,* 1(1938): 10-19.

7. Dighton, J., White, J.F. and Qudeman, S.P. "The fungal Community; Taylor and Francis". *3rd Edition.* (2005): 17-21.

8. Dahlgren, R.A. and Driscoll, C.T. "The effects of whole-tree clear-cutting on soil processes at the Hubbard Brook Experimental Forest, New Hamphire". *USA. Plant Soil.* 158(2) (1994): 239-262.

9. Domsch, K.H., Gams, W. and Anderson, T.H. Compendium of Soil Fungi, Second Edition. *Lubrecht & Cramer Ltd*, (1995). USA.

10. Dixon, D. "The microorganisms of cultivated and bush soils in Victoria". *Aust. J. Exp. Biol. Med. Sci.,* 5(1928).: 223-232.

11. Dwivedi, R. S. "Ecology of the soil fungi of some grasslands of Varanasi. II." *Distribution of soil mycoflora.* Trop. Ecol., 7 (1966): 84 - 99.

12. England, C. M. and Rice, E. L. "A comparison of the soil fungi of a tall grass prairie and of an abundaned field in Central Oklahoma." *Bot.Gaz.,* (1957). 188: 180-190.

13. Gtjjrati, S. "Ecology of soil fungi of cultivated fields". *Proc. Natl.Acad. Sci.* (India), 38 (1968). B: 66-80.

14. Inderjit, P.O., Jenkins, L.A. and Sumbali, O. "History of systematic botany and ecology." *U.S.A. 6th Edition.* (2005). 20.

15. Kanaujia, R. S. "Studies on certain ecological aspects of soil fungi". *Trop. Ecol.,* 13(1972): 5- 11.

16. Kanaujia, R. S. "Studies on certain ecological aspects of soil fungi. II.Distribution of soil fungi in relation to cover vegetation and physicochemical characters of the soils". *Anles Edafol. Agrobiol.,* 32(1973a): 21 - 34.

17. Kanaujia, R. S. "Studies on certain ecological aspects of soil fungi. III. Soil fungi in relation to varying cover vegetation, climatic conditions, soil depths and physico-chemical characters of the soil". *Proc. Natl. Acad. Sci.,* India, 1973 (in Press).

18. Leclerg, E. L. "Distribution of certain fungi in Colorado soils". *Ibid,* 21(1931): 1073-1081.

19. Mallik, M. A. B. and Rice, E. L. "Relation between soil fungi and seed plants in three successional forest communities in Oklahoma". *Bot.Gaz.,* 127(1966): 120-127.

20. Mishra, R. R. "Seasonal distribution of fungi in four different grass concociations of Varanasi (India)". *Trop. Ecol.,* 6(1965): 133 -140.

21. Moreatj, M. and Moreau, F. "Premiere contribution a l'etude de la microflore des dunes". *Rev. Mycol.,* 6(1941): 49 - 94.

22. Nicot, M. M. E. J. "Some characteristics to the mycoflora in desert sands". In: The Ecology of Soil Fungi. *Liverpool Univ.* Press, (1960). 94-97.

23. Pugh, G. J. F. "The fungal flora of tidal mud flats. In: The Ecology of Soil Fungi." *Liverpool Univ. Press,* (1960). 202 -208.

24. Siepmann, R. "Ein Beitrag zur saprophytischen Pilzflora des Wattes der Wesermundung. Veroffent. Inst. Meeresforsch". *Bremerhaven*,6(1959): 213-301.

25. Thorn, F.L. "Laboratory technique of isolating fungi". *2nd Edition.* (1997). pp 45.

26. Turner, M. and Pugh, G. J. F. Species of *Mortierella* from a salt marsh. Trans. *Brit, mycol. Soc,* 44(1961): 243-252.

27. Waksman, S. A. "A method for counting the numbers of fungi in the soil". *J. Bot.***7**(1992):339-341.

28. Webley, D. M., Eastwood, D. G. and Gimingham, C. H. "The development of a soil mycroflora in relation to plant succession on sand dunes including rhizosphere flora associated with colonizing species". *Jour.Ecol.,* 40(1952): 168-178.

29. Witkamp, M. "Macroflora, microflora and soil relationships in a pine plantation". *Ecology,* 47(1966).: 238 -244.

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