Studies on variations in the fungal population of Indian alkaline soils*)

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Introduction

In studies on soil microbiology variations in the mycoflora accompanied by changes in pH, moisture content, salinity, organic matter and certain other factors within short distances pose a problem in obtaining a real picture of the mycoflora of a particular soil. This aspect of study in case of the fertile soils was taken up by Waksman (1931), Rose and Miller (1954) and Mishra (1965), other soil types however, remain largely neglected in this matter. The present study was undertaken mainly to investigate as to whether or not there exists any statistically significant variation in relation to pH, moisture content and number of fungal colonies in a randomly selected patch of **'Usar'** (alkaline) soil which cover vast tracts of barren areas in this country. In addition, results of qualitative analysis of the fungal flora are also presented.

Materials and Methods

Design of Experiment:

The sample plot was so selected as to assure complete representation of the population in terms of specified variables i. e., pH of the soil, moisture content (%) and number of fungal colonies per gm. of dry soil. Hence, the sample was drawn out by delimiting the total population within an area of $60' \times 60'$. The $60' \times 60'$ square was conceived to be composed of 8 equilateral triangles (Fig. 1) and alternate triangles were taken as samples. Each of these four triangles was subdivided into 8 equilateral triangles of which six were subjected to detailed analysis in terms of degree to which three variables were present in each case so that the sample size consisted of $4 \times 6 = 24$ subsamples out of a large area of $60' \times 60'$. This scheme has the adavantage in so far as it ensures 4 samples from the middle, 4 each from the corner and 4 from centre of the plot.

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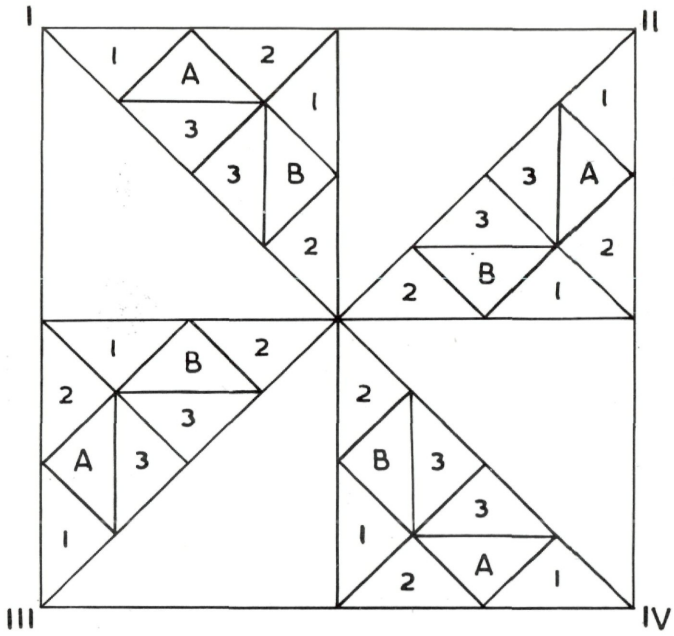
Microbial analysis of soil samples:

Soil samples, from the spot selected for this study, were collected from 2–6″ depth and analysed both by the Dilution-plate and Soil-plate methods (Waksman and Fred, 1922; Warcup, 1950; 1955; Johnson et al., 1960). The dilution-plate method was the one followed for quantitative determination of the mycoflora. pH and moisture content of the soil



were noted in each case. Frequency percentage of different fungal species was calculated using the formula of Tresner's et al. (1954).

Observations

Fungal flora of the investigated soil:

Results of qualitative analysis of the fungal status of 24 sub-samples are presented in Tables I and II.

As is evident from Table I, the minimum-maximum number of species present in various sub-samples show a ratio of 1:2.4.

Table II shows that out of 72 fungal forms isolated during the course of this study, *Absidia corymbifera, Aspergillus carneus, A. fischeri, A. fumigatus, A. niger, Tripterospora tetraspora* and hyaline strains of Mycelia

Table I. Showing number of fungal species in various subsamples of all the four corners

Corner and sample numbers			Number of fungal species
Corners	Samples	sub samples	
CORNER I	A	1	21
		2	13
		3	14
	B	1	19
		2	16
		2	9
CORNER II	A	1	21
		2	16
		3	17
	B	1	21
		2	17
		3	17
CORNER III	A	1	22
		2	19
		3	16
	B	1	16
		2	13
		3	16
CORNER IV	A	1	17
		2	18
		3	16
	B	1	20
		2	17
		3	10

Sterilia were of common occurrence and were isolated from all the four corners of the plot whereas, forms like *Acrophialophora nainiana, Alternaria tenuis, Aspergillus fumigatus* var. *albus, A. giganteus, A. terreus* var. *globosus, Cunninghamella blakesleeana, Curvularia tuberculata, Fusarium semitectum, Humicola fuscoatra, Penicillium cyclopium, P. funiculosum, Periconia saraswathipurensis, Phoma hibernica, Sordaria humana, Thielavia sepedonium, Trichoderma lignorum* and *Trichothecium roseum*

Table II. Showing frequency % of different fungi in soil samples of each of the four corners

Name of fungal species	Frequency % of different fungal species			
	CORNER I	CORNER II	CORNER III	CORNER IV
Absidia corymbifera	83	50	33	50
Achaetomium strumarium		50		16
Acrophialophora nainiana	33	83		33
Acrostalagmus cinnabarinus		33	50	
Alternaria tenuis	50		100	33
Aspergillus amstelodami	66	50		
A. carneus	33	33	33	50
A. fischeri	16	33	16	16
A. flavipes			16	33
A. flavus	50	50		
A. fumigatus	50	33	50	83
A. fumigatus var. *albus*	16		33	33
A. fumigatus var. *griseo-brunneus*	50	33		
A. giganteus	16	33	33	
A. nidulans	50	83		
A. niger	100	50	33	50
A. ochraceus				33
A. penicilliformis	33		50	
Aspergillus sp.	33	33		
A. sclerotiorum	33		66	
A. sulphureus	50		100	
A. terreus	33	83		
A. terreus var. *globosus*	50		33	16
A. ustus		50		100
A. versicolor	83		16	
Cephalosporium sp.		50		
Chaetomium arcuatum	66			
C. globosum		33		50
C. indicum				33
C. lucknowense		16		16
Circinella muscae	50	16		
Cladosporium cladosporioides		50	33	
Cunninghamella blakesleeana	33	33	33	
C. echinulata		16	33	
Curvularia lunata var. *aeria*			66	
C. tuberculata	16	50	50	
C. verruculosa			16	50
Dactylium fusarioides			33	33
Fusarium concolor		16	33	
F. moniliforme		66		33
F. semitectum	33		33	33
F. solani		16		66
Graphium sp.		16		16

Name of fungal species	Frequency % of different fungal species CORNER I	CORNER II	CORNER III	CORNER IV
Helminthosporium sp.			16	33
H. hawaiiense		33		83
Humicola fuscoatra	50	16		66
Memnoniella echinata		16	16	
Mucor sp.		83		
Monilia sitophila		33		
Myrothecium striatisporum			16	16
Mycelia sterilia (Hyaline)	16	16	50	16
Mycelia sterilia (Dark coloured)		16	83	
Neocosmospora vasinfecta	33		83	
Paecilomyces persicinus		50		
P. varioti		50		83
Penicillium brefeldianum	16		33	
P. citrinum				66
P. cyclopium	33	16	50	
P. funiculosum	33	16		66
P. steckii	16		50	
Periconia saraswathipurensis		16	16	16
Phoma glomerata			16	33
P. hibernica		33	16	16
Pullularia pullulans			33	
Rhizoctonia sp.		50		50
Sordaria humana		16	16	16
Starkeomyces koorchalomoides	16		16	
Syncephalastrum racemosum		33		33
Thielavia sepedonium	33	16	33	
Trichoderma lignorum	50	16	16	
Trichothecium roseum	16	16	16	
Tripterospora tetraspora	33	33	50	33

were found to occur in three corners only. The other fungi were isolated either from two or only one corner of the investigated **'Usar'** soil plot.

Analysis of variance:

Data on pH, moisture content (%) and microbial level of the selected patch of **'Usar'** soil is given in Table III which indicates that the pH ranged from 8.0–10.5, moisture content from 3.0–5.2% and, number of colonies per gm. of dry soil from 4790–8620. Thus minimum – maximum ratio of the three variables between all the 24 subsamples was 1 : 1.3, 1 : 1.4 and 1 : 1.8 respectively. However, as is shown below statistically these variations are not significant.

The estimated inter- and intra-subsample variations indicated in Table IV reveals that the estimated variance and the value of variance ratio 'F' is not significant. The estimated value of 'F' for pH is 0.26; for moisture content 2.63 and for number of colonies 1.33 as against the tabula-

Table III. Showing variations in pH, moisture content (%) and number of fungal colonies per gm. of dry soil

Corner and sample numbers			Name of the variables		
Corners	Samples	Sub-samples	pH	Moisture content %	No. of colonies per gm. of dry soil
		1	9.0	4.3	5850
	A	2	8.0	5.0	6310
		3	9.5	5.1	6480
CORNER I					
		1	8.5	3.8	5280
	B	2	9.0	4.1	6160
		3	10.0	4.8	6930
		1	10.0	3.0	6800
	A	2	9.0	4.8	6720
		3	10.5	5.0	7150
CORNER II					
		1	8.5	3.5	7740
	B	2	9.5	3.5	6840
		3	10.0	3.0	7010
		1	8.5	5.1	6530
	A	2	9.0	4.8	7350
		3	10.5	4.0	5830
CORNER III					
		1	9.5	3.8	5610
	B	2	9.0	3.7	6430
		3	8.0	5.2	6110
		1	8.0	3.7	4790
	A	2	8.5	4.0	5620
		3	9.5	4.8	6930
CORNER IV					
		1	8.5	5.0	8620
	B	2	9.0	3.6	7050
		3	10.5	4.7	5990

ted value of 3.10. This means that there are no significant differences between various patches of a piece of randomly selected **'Usar'** land. Hence, it can be concluded that variations in respect of these three variables are not likely to be of any great significance or that each of our sub-sample has been drawn from the same population and is representative of the same.

Table IV. Showing analysis of variance for pH, moisture content (%) and number of fungal colonies per gm. of dry soil

Name of variables	Source of variation	Analysis of Variance: Sum of squares	Degree of freedom	Variance estimate	Variance ratio 'F'
pH of soil samples	Between corners	1.62	3	0.54	0.26
	Within corners	41.94	20	2.10	
Moisture content (%)	Between corners	3.24	3	1.08	2.63
	Within corners	8.09	20	0.41	
Number of fungal colonies per gm. of dry soil	Between corners	2646378	3	882126	1.33
	Within corners	13266922	20	663346	

Discussion

Soil being a dynamic system constantly keeps on changing. Hence, large variations in the mineral and organic matter content between short distances of the soil are expected. Such variations should be greatly magnified in fertile soils due to the presence of prolific organic decay processes resulting in micro or macro-pockets of humus. Indeed, the same trend is reflected through variations in the fungal population of fertile soils. Waksman (1931) reported variations to the tune of 1:3.25 and Rose and Miller (1954) found it to be of the order of 1:13. The present study on **'Usar'** soils revealed variations in the fungal numbers only of 1:1.8 which is much less than that observed in the fertile soils. More so, these variations are statistically not significant.

'Usar' (alkaline) soils present very drastic conditions due to high levels of pH, salinity etc. (Rai et al., 1970a, 1971). They are therefore, largely barren except for some patches of poor grassy-growth, a few herbs specially in the damper areas and a few shrubs and trees. However, during the rains when the salts are partially leached down, a few more herbs make their appearance. Such soils are obviously expected to be poorer in organic matter and hence will show much less variability in the presence of humus in adjacent areas of the soil. Agarwal and Gupta (1968) reported that "saline and alkali soils are generally poor in humus and the latter are particularly deficient in humus, nitrogen and phosphorus; high alkalinity is responsible for the dispersion of humus". It is noteworthy that the soils investigated during this study were of alkali type.

Several recent reports have emphasized the major role of organic matter in governing the fungal population of soil. It is therefore, attrac-

tive to correlate directly the low variability of fungal population with the low humus content of **'Usar'** soils. However, more experimental work is needed to confirm this proposition. The likeness between the behaviour of pigmented bacteria in **'Usar'** and coastal saline soils (Turner and Jervis, 1968; Rai et al., 1970b) and the salinity optima as affected by temperature for **'Usar'** and marine fungi (Rai and Agarwal, 1973) makes this view doubly fascinating. It is noteworthy that in coastal soils also the organic matter is the major single factor governing the fungal population (Pawar and Thirumalachar, 1966; Pugh, 1961).

Summary

The paper deals with the microbial population of a number of samples collected from a standardized **'Usar'** soil plot which was statistically analysed. The total number of colonies per gram of dry soil showed a statistically insignificant minimum – maximum ratio of 1 : 1.8. This low variation in the fungal population of these soils may be attributed to the poor organic matter content of these highly alkaline soils.

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References

Agarwal, R. R. and Gupta, R. N. (1968) – Saline – Alkali Soils in India. Indian Council Agric. Res., New Delhi.

Mishra, R. R. (1965) – Sampling variations in the fungal flora of grass land. J. Sci. Res. (B. H. U.), **15:** 19–21.

Pawar, V. H. and Thirumalachar, M. J. (1966) – Studies on halophilic soil fungi from Bombay. Nova Hedwigia, **12:** 497–508.

Pugh, G. J. F. (1961) – Fungal colonization of a developing salt marsh. Nature (Lond.), **190:** 1032–1033.

Rai, J. N., Sharma, B. B. and Agarwal, S. C. (1970a) – Increased pH-tolerance of some Aspergilli isolated from **'Usar'** (alkaline) soils. A possible indication of ecological specialization. Sydowia, Annales Mycologici, **24:** 336–344.

— Tewari, J. P. and Agarwal, S. C. (1970b) – Relative abundance of pigmented bacteria in Indian 'Usar' (alkaline) soils – A possible indication of ecological specialization. J. Gen. Apll. Microbiol., **16:** 315–319.

— Agarwal, S. C. and Tewari, J. P. (1971) – Fungal microflora of 'Usar' soils of India. Jour. Indian Bot. Soc., **50:** 63–74.

— and Agarwal, S. C. (1973) – Salinity optima as affected by temperature for some 'Usas' soil Aspergilli. Mycopathol. et Mycol. Appl., **50:** 307–312.

Rose, R. E. and Miller, J. G. (1954) – Some sampling variations in soil fungal numbers. J. Gen. Microbiol., **10:** 1–10.

Tresner, H. D., Backus, M. P. and Curtis, J. T. (1954) – Soil microfungi in relation to the hardwood forest continum in Southern Wisconsin. Mycologia, **46:** 314–333.

Turner, M. and Jervis, D. I. (1968) – The distribution of pigmented *Bacillus* species in salt marsh and other saline and non-saline soils. Nova Hedwigia, **16:** 293–298.

Waksman, S. A. and Fred, E. B. (1922) – A tentative outline of the platemethod for determining the number of microorganisms in the soil. Soil. Sci., **14:** 27–28.

— (1931) – Principles of Soil Microbiology, 2nd Ed. London: Bailliere, Tindall and Cox.

Warcup, J. H. (1950) – The soil plate method for the isolation of fungi from soil. Nature, **166:** 117–118.

— (1955) – Isolation of fungi from hyphae present in soil. Nature, **175:** 953–954.