Impacts of agro-climates and land use systems on culturable microbial population in soils of the Indo-Gangetic Plains, India

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Comprehensive reports on land-use changes and their impact on soil biological properties, specifically microbial population in the Indo-Gangetic Plains (IGP) of India, are lacking. Since IGP is the most fertile land, data on microbial population of IGP may contribute towards the evaluation of various soil quality parameters, disease suppression, organic matter decomposition, plant growth promotion and soil management pattern. To enhance our knowledge on culturable microbial populations in different soil horizons of the agro-ecological sub-regions (AESRs) in the IGP, a study has been undertaken to collect soil samples from the established benchmark (BM) spots of these plains with an objective to investigate the impacts of bioclimates, soil depth, cropping systems, land use systems and management practices on the distribution of culturable microbial population. Bacterial : fungal ratios are significantly different across the land use types. The bacterial and fungal populations are strongly and negatively correlated with soil depth and maximum microbial population (40%) exists in the surface horizon (0–30 cm) than in the subsurface horizon (121–150 cm). Generally, bacterial populations are higher than actinomycetes and fungal populations in all soil profiles of the IGP. Approximately 10% decrease in Shannon diversity index has been observed with increase of 30 cm depth and 89% fall between surface and subsurface profiles. Non-significant difference in microbial population ($P < 0.05$) is noticed across the management and land use systems. Sub-humid (moist) bioclimatic system recorded higher microbial population than sub-humid (dry) and semi-arid bioclimatic systems. Legume-based cropping system has higher microbial population than cereal or vegetable-based cropping.

Keywords: Agro-ecosystems, microbial population, land use type, soil depth.

Introduction

THE Indo-Gangetic Plains (IGP) is one of the largest fertile plains in the world. It is spread over various agro bioclimatic regimes and is one of the most populous areas. The IGP soils have different pedogenetically developed layers

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that may sustain a large number of microorganisms. Pertinent portion of the microbial population inhabitants in the soil is located in the subsurface[1] and these subsurface microbes may have a great influence on bio-transformation processes. Microbial populations that exist in the deeper soil horizons are not well known and the spatial variability exhibited by these populations still remains poorly understood.

Our knowledge of the IGP regarding structural composition and diversity of soil microbial population is mainly limited to surface horizons, since majority of studies emphasize solely on the rhizospheric soil. Likewise, there is a lack of inclusive information on microbial population composition within predefined benchmark (BM) soils. The microbial composition and its diversity are affected by soil edaphic factors, which are non-homogeneous across the landscape. The changes in environmental conditions with soil depth generate differences between surface and subsurface microbial populations, which is also poorly understood in the IGP soils. Hence a study was conducted in 11 predefined BM soil series of the IGP to assess the microbial population cultures across different soil profiles. Simultaneously, the effects of different bioclimates, cropping systems, management and land use systems on microbial population and their Shannon's diversity have also been documented in the IGP soils of India. The data so generated on microbial populations can be considered as part of soil information system for monitoring the soil quality and its evaluation and assessment for soil pollution in the IGP.

Material and methods

Study site and soil sampling

Soil samples were collected from the 11 representative BM sites in the IGP of India (Table 1) covering specific bioclimatic systems. Based on the mean annual rainfall, the IGP is grouped[2] as arid (< 550 mm), semi-arid (dry; SAd) (< 550–850 mm), semi-arid (moist; SAm) (1000–850 mm), sub-humid (dry; SHd) (1100–1000 mm), sub-humid (moist; SHm) (> 1100 mm) and humid (> 1650 mm) in 8 AERs (agro-ecological regions) and 11 AESRs (agro-ecological sub-regions; ref. 3) accounting for 13% (52.01 m ha) of the total geographical area of the country[4]. The soil sampling was performed in such a way that each BM site has two contrasting land use features representing pedons, viz. low management (LM) and high management (HM). The LM areas use low NPK, no manure and take out agricultural residues and biomass during harvesting. By contrast, HM zones use recommended doses of NPK, regular application of manures, incorporate agricultural wastes in the land and adopt soil moisture conservation practices. Horizon-wise soil samples were collected in a sterile polythene bag, labelled and brought to the laboratory for analysis. A questionnaire was used to collect information on land-use practices and physiographic attributes of the BM spots.

Microbial enumeration and diversity analysis

The soils from different BM sites were passed through 2 mm sieve and used for enumeration of microbial population following standard serial dilution method (10^{-3}) (ref. 5). General microbiological media such as nutrient agar, actinomycetes isolation agar with 0.025% (w/v) nystatin and potato dextrose agar with 0.10% (w/v) streptomycin sulphate have been used for enumeration of bacteria, actinomycetes and fungi respectively. The plates in triplicate were incubated at optimum temperature (25 ± 1°C for bacteria; 30 ± 2°C for fungi and actinomycetes). The microbial colonies appearing after the incubation (3 days for bacteria, 5 days for fungi and 7 days for actinomycetes) were counted as total cultured colony forming units (cfu) and expressed as $\log_{10}$ cfu/g of dry soil. The microbial diversity index (Shannon diversity index (H')) was determined using the following equation[6]

$$H' = \sum pi * \ln pi,$$

where ln is the natural logarithm and pi the proportion of individual microbial colony found in the ith BM spot.

Statistical analysis

To study the impact of different factors (bioclimates, cropping systems, land use and management practices) on cultured microbial population and diversity index, the data from different BM sites of the IGP under HM and LM were grouped together and analysed by one-way ANOVA. All analyses were performed using statistical software SPSS 16.0.

Results and discussion

Microbial population in the IGP soils

The soil culture microbial population shows a decline in all the BM spots with increase in soil depth (Tables 2–4). The maximum microbial populations are restricted to surface profiles and 40% of total population is confined to surface soil. The mean counts of bacteria, actinomycetes and fungi are in the order of 5.35, 4.97 and 4.36 $\log_{10}$ cfu g^{-1} in the surface soil. In the subsurface (121–150 cm) soil, the mean population is 4.87, 4.53 and 3.71 $\log_{10}$ cfu g^{-1} respectively. The surface soils recorded higher bacteria (41.3%), actinomycetes (38.4%) and fungi (26.1%) population compared to the subsurface horizons. Bacteria are the dominant group followed by actinomycetes and fungi are the least dominant among the three

Table 1. Characteristics of selected benchmark spots in IGP of India

AESR	Bioclimate	MAR (mm)	Soil series	MSL (m)	District	State	Textural class	Cropping systems
16.3	Humid	2500	Seoraguri	42	Coochbehar	West Bengal	Loamy	Rice/jute–potato
15.3	Humid	1800	Nayanpur	120	WestTripura	Tripura	Very fine	Rice/vegetables
18.5	Humid	1783	Sagar	5	24 Parganas (South)	West Bengal	Fine	Rice–Chilli/Green gram
13.2	Sub-humid (M)	1154	Haldi	238	Udhamsingh Nagar	Uttarakhand	Coarse loamy	Wheat/maize–soybean
15.1	Sub-humid (M)	1150	Madhpur	18	Burdhman	West Bengal	Fine	Rice–mustard/potato
12.3	Sub-humid (M)	1130	Gopalpur	38	Bhirbhoom	West Bengal	Fine	Rice–potato/Green gram
9.2	Sub-humid (M)	1110	Itwa	57	Chandauli	Uttar Pradesh	Fine	Rice–wheat
13.1	Sub-humid (M)	1105	Ekchari	30	Bhagalpur	Bihar	Fine	Rice–wheat/maize
9.1	Sub-humid (M)	950	Fatehpur	230	Ludhiana	Punjab	Coarse loamy	Wheat + mustard–pigeon pea
4.1	Semi-arid (D)	800	Zarifaviran	285	Karnal	Haryana	Fine silty	Rice–wheat
4.3	Semi-arid (D)	790	Sakit	149	Etah	Uttar Pradesh	Fine loamy	Rice–wheat

AESR, Agro-ecological sub-regions; MAR, Mean annual rainfall; MSL, Elevation above mean sea level; M, Moist; D, Dry; '/', or '+', Intercropping; '–', Followed by.

Table 2. Bacterial population in benchmark spots of IGP

	Bacteria population ($\log_{10}$ cfu/g soil)				
	Soil depth (cm)				
BM spot	0–30	30–60	60–90	90–120	120–150
Zarifa Viran	5.35	5.24	5.24	5.14	4.90
Sakit	5.32	5.24	5.21	5.13	4.91
Fatehpur	5.33	5.22	5.19	5.14	4.93
Itwa	5.34	5.28	5.17	5.13	4.79
Gopalpur	5.40	5.31	5.21	5.15	4.84
Ekchari	5.38	5.14	5.23	5.19	4.94
Haldi	5.41	5.32	5.24	5.12	4.91
Madhpur	5.37	5.33	5.29	5.18	4.91
Seoraguri	5.31	5.22	5.27	5.12	4.83
Nayanpur	5.29	5.20	5.00	4.84	4.69
Sagar	5.34	5.25	5.18	5.12	4.91
Mean	5.35	5.25	5.20	5.12	4.87
CD (0.05)	0.029	0.020	0.020	0.062	0.061
CV (%)	0.280	0.205	0.205	0.630	0.660

Table 3. Actinomycetes population in benchmark spots of IGP

	Actinomycetes population ($\log_{10}$ cfu/g soil)				
	Soil depth (cm)				
BM spot	0–30	30–60	60–90	90–120	120–150
Zarifaviran	5.09	4.99	4.97	4.94	4.58
Sakit	5.06	4.94	4.91	4.72	4.53
Fatehpur	5.18	5.06	4.98	4.95	4.72
Itwa	4.92	4.86	4.68	4.62	4.49
Gopalpur	5.12	5.06	4.99	4.88	4.70
Ekchari	4.92	4.79	4.66	4.60	4.49
Haldi	5.07	4.98	4.22	4.69	4.56
Madhpur	5.08	5.00	4.95	4.80	4.64
Seoraguri	4.56	4.50	4.49	4.40	4.34
Nayanpur	4.63	4.57	4.50	4.17	3.99
Sagar	5.05	4.99	4.91	4.87	4.78
Mean	4.97	4.89	4.75	4.69	4.53
CD (0.05)	0.077	0.116	0.110	0.106	0.133
CV (%)	0.81	1.25	0.12	1.19	1.53

Table 4. Fungi population in benchmark spots of IGP

	Fungi population ($\log_{10}$ cfu/g soil)				
	Soil depth (cm)				
BM spot	0–30	30–60	60–90	90–120	120–150
ZarifaViran	4.47	4.40	4.30	4.10	3.60
Sakit	4.33	4.28	4.12	3.87	3.48
Fatehpur	4.36	4.27	4.16	4.07	3.48
Itwa	4.41	4.37	4.17	4.05	3.87
Gopalpur	4.50	4.46	4.30	4.05	3.75
Ekchari	4.37	4.25	4.07	3.92	3.75
Haldi	4.36	4.26	4.18	4.04	3.73
Madhpur	4.40	4.36	4.26	4.09	3.82
Seoraguri	4.19	4.08	3.94	3.82	3.67
Nayanpur	4.21	4.12	4.08	3.99	3.82
Sagar	4.38	4.39	4.31	4.04	3.80
Mean	4.36	4.29	4.17	4.00	3.71
CD (0.05)	0.102	0.089	0.162	0.154	0.216
CV (%)	1.23	1.09	2.02	2.01	3.06

groups of microorganisms. Higher populations of heterotrophic bacteria and actinomycetes can be attributed to their higher tolerance and adaptation to wide variations of the soil properties[7]. The low fungal counts in the IGP soils can be attributed to their non-acidic nature. Significant correlation ($P < 0.05$) has been observed between microbial counts and soil depth (bacteria ($r = -0.98$), fungi ($r = -0.98$), actinomycetes ($r = -0.97$)). The present study confirms the findings of Hartmann *et al.*[8] that microbial biomass exponentially decreases with soil depth. The decline in the number of microbes in subsurface soils implies the deficiency of soil nutrients, water and aeration in deeper layers compared with surface horizon. The result of the vertical distribution of fungi and actinomycetes is similar to observations by some others[9,10] as they have also reported the abundance of fungi in the surface soils compared to subsurface soils. Zhou *et al.*[11] reported a decreasing trend of organic matter, nitrogen and

phosphorus down the depth soil. This influences the microbial communities in the subsurface soils.

Shannon's diversity index

Pooled data on Shannon's diversity index (H') show a significant variation ($P < 0.05$) with soil depth (Table 5). With every increase in 30 cm, approximately 10% decrease in diversity index has been observed in the IGP soils. Around 89% decline in microbial diversity index is observed from surface to subsurface soils. Comparatively, farms which are better managed (i.e. high management) are rich in microbial population showing more diversity than in the poorly managed farms. Among the BM spots, higher H' (2.42) has been recorded in surface layers of Gopalpur soils (West Bengal). Microbial population varies in surface and subsurface soils. This supports earlier observations suggesting different environments in deeper soils which require different adaptation strategies of microbes to soil edaphic factors[10,12,13]. Our results corroborate with the findings of Zhou *et al.*[11] that the surface bacterial population has higher diversity index values that are 2–3 orders of magnitude greater than those for the subsurface population. Decrease of microbial diversity following a perpendicular spatial model finds support from the available literature as well as the present study.

Impact of bioclimates on microbial population in the IGP soils

Significant differences are observed in microbial population belonging to different bioclimates of the IGP (Table 6). In surface horizon (0–30 cm), higher bacterial population (5.46 $\log_{10}$ cfu g^{-1}) is recorded in sub-humid moist bioclimate followed by semi-arid (5.34 $\log_{10}$ cfu g^{-1} soil); however, bacterial population is more or less same in humid (5.32 $\log_{10}$ cfu g^{-1} soil) and sub-humid dry bioclimates (5.31 $\log_{10}$ cfu g^{-1} soil). Actinomycetes population is maximum in sub-humid dry (5.13 $\log_{10}$ cfu g^{-1} soil) and the least in humid region (4.84 $\log_{10}$ cfu g^{-1} soil). Fungal population is highest (4.42 $\log_{10}$ cfu g^{-1} soil) in subhumid moist bioclimatic zone and lowest in humid bioclimate (4.32 $\log_{10}$ cfu g^{-1} soil). Rainfall is an important physical phenomenon, which directly influences the microbial population through soil water precipitation and moisture retention. The reduction in microbial population may be explained by a combination of biological and climatic factors. Reduction in labile carbon and increase in recalcitrant substrates in the organic horizon, may have had a negative effect on microbial population. Gram-positive bacteria possess thicker and stronger cell walls[14] and can produce a large number of osmoregulatory solutes than Gram-negative bacteria[15]. Based on these physiological differences, Gram-positive bacteria are believed to be better adapted to stress and large fluctuations in soil moisture[15,16]. Gram-negative bacteria may benefit more from elevated precipitation and soil moisture, because they are more abundant in surface soils[9], which are more likely to wet up during the rains[17]. A previous report indicates that the relative abundance of soil bacteria increases under high soil moisture conditions[1] and is confirmed by the presence of high microbial density in the sub-humid (moist) IGP, whereas fungi dominate carbon and nitrogen cycles in dry soil[18]. These patterns follow logically from the metabolic and physiological requirement of these two major microbial groups. Soil bacteria depend on water for movement and nutrient acquisition[15]. Fungi are aerobic organisms that are more tolerant to dry conditions[19]. For bacteria, dry pore spaces are barriers for movement, diffusion of resources and nutrient uptake and can be a key reason for the low cfu in subsurface profile of IGP. Soil fungi can extend hyphae through air-filled pore spaces to access moisture and nutrients, and can translocate these resources to water and nutrient-limited cells within their mycelia network[20].

Table 5. Shannon's diversity index (H') in different benchmark spots

BM spot	Shannon's index (cm)	
	0–30	120–150
ZarifaViran	2.36	1.81
Sakit	2.34	1.94
Fatehpur	2.38	1.96
Itwa	2.26	1.98
Gopalpur	2.42	1.82
Ekchari	2.15	1.79
Haldi	2.27	1.85
Madhpur	2.30	1.76
Seoraguri	2.19	1.80
Nayanpur	2.00	1.96
Sagar	2.41	1.64
Mean	2.28	1.85
CD (0.01)	0.050	0.062
CV (%)	1.28	1.98

Impact of management regimes on microbial population

The pooled data on microbial population in surface soil (0–30 cm) and subsurface soil of IGP do not indicate a

Table 6. Distribution of microbial population in different bioclimates of IGP

Bioclimate	Bacteria*	Actinomycetes*	Fungi*
Sub-humid (moist)	5.46	5.07	4.42
Sub-humid (dry)	5.31	5.13	4.36
Humid	5.32	4.84	4.32
Semi-arid	5.34	5.08	4.38
CD (0.05)	0.055	0.055	NS
CV (%)	0.54	0.58	0.91

*$\log_{10}$ cfu/g soil.

Table 7. Distribution of microbial population in different management systems

Soil depth	Bacteria#	Actinomycetes#	Fungi#
0–30 cm			
High management	5.38	4.91	4.27
Low management	5.28	4.84	4.17
Mean	5.33	4.88	4.22
CD (0.05)	ns*	ns*	ns*
CV	0.175	2.23	1.68
120–150 cm			
High management	4.86	4.50	3.62
Low management	4.78	4.33	3.56
Mean	4.82	4.42	3.59
CD (0.05)	ns*	ns*	ns*
CV (%)	1.01	2.06	2.04

*Non significant, #$\log_{10}$ cfu/g soil.

significant difference between management practices HM and LM (Table 7). Higher microbial population was observed in all highly managed BM spots of the IGP. In surface horizon, highest bacterial population (5.41 $\log_{10}$ cfu g^{-1} soil) was recorded in Haldi soils of Uttarakhand and the lowest bacterial population (5.29 $\log_{10}$ cfu g^{-1} soil) was observed in Nayanpur soils of Tripura. Actinomycetes (5.12 $\log_{10}$ cfu g^{-1} soil) was highest in Bhirbhoom district of West Bengal and fungi population was highest (4.50 $\log_{10}$ cfu g^{-1} soil) in Bhirbhoom district of West Bengal and 24 Parganas of West Bengal. Cropping systems and soil management can markedly affect the activity of soil microorganisms and their diversity[21]. Deep tillage may be the possible cause for the higher bacterial population and low fungal population, as deep tillage can damage the fungal hyphae and allow the sedimentation of plant residues into the soil; thereby bacteria in agricultural fields flourish because the contact surface between the substrate and bacteria is increased. Elementary physiological and ecological differences in bacterial and fungal populations would not only be responsible for the biogeography of individual groups of microbial distribution, but are also controlled by separate edaphic factors which may vary among management and land use[22]. The differences in substrate quality and a shift in the proportion of Gram-positive to Gram-negative bacteria in soils may alter nutrient cycling rates. Most Gram-negative bacteria appear to grow quicker on labile substrates[9,23], signifying a higher relative abundance of Gram-negative bacteria associated with faster nutrient cycling rates. Deposition of intractable material at greater depth than the first two surface profiles shows an indirect evidence for commencement of low microbial density and nutrient cycling efficiency at lower depths of soil[9].

Impact of cropping systems on microbial population

The IGP is better known for the cultivation of cereal-based crops such as rice and wheat prominently in rotation due to availability of irrigation source and favourable environmental conditions. The microbial population showed significant difference ($P < 0.05$) among the various crops. The farms with legume-based cropping system (chickpea/potato rotation) recorded higher microbial population, while vegetable-based cropping recorded the least (Table 8). In legume-based cropping system, chickpea field had higher bacterial, actinomycetes and fungi populations (5.43/5.14/4.45 $\log_{10}$ cfu g^{-1} soil) followed by oilseed-based cropping field (mustard and soybean; 5.42/5.13/4.39 $\log_{10}$ cfu g^{-1} soil). In cereal cropping system, maize field had higher microbial population (5.40/5.36/4.41 $\log_{10}$ cfu g^{-1} soil) followed by rice–wheat (5.38/4.99/4.39 $\log_{10}$ cfu g^{-1} soil) and lowest in vegetable-based cropping system (5.41/4.94/4.38 $\log_{10}$ cfu g^{-1} soil). Land management, vegetation and climate are key factors which influence the abundance of microbial communities and their diversity in the top profiles.

Table 8. Distribution of microbial population in different crops of IGP

	Microbial population		
Cropping system	Bacteria*	Actinomycetes*	Fungi*
Cereal (rice)	5.38	4.99	4.39
Legume crops	5.43	5.14	4.45
Vegetable	5.41	4.94	4.38
Oilseed	5.42	5.13	4.39
CD (0.05)	0.099	0.12	0.146
CV (%)	0.97	1.28	1.76

*$\log_{10}$ cfu/g soil.

Crop rotation and residue amendment influence soil microbial density. Various soil and crop management systems can result in different substrate availabilities to affect the establishment of different microbial groups. In a cereal/legume crop rotation system, Alvey *et al.*[24] have reported that different crops affected the number, species and diversity of soil microorganisms. The higher microbial community noticed in legume-based cropping system of the IGP is attributed to the higher soil nutrients and organic carbon along with stimulated microbial activity. Different root exudates and root residues of rotational crops decomposed in the soil may affect nutrient availability to soil microbes.

The present study is in line with the findings of Kathryn *et al.*[25] that surface or organic profiles contain higher microbial density than any profiles of an agricultural field. Soils under crop rotation with high agricultural input have high available micro- and macronutrients, resulting in high microbial population and enzymatic activities than monocropping and poorly managed agricultural fields[26,27].

Conclusions

Soil heterotrophic aerobic microbial communities decrease with increase in soil depth in all the BM spots of the IGP.

The maximum microbial communities are restricted to 30 cm depth (surface horizon). Shannon's microbial diversity index is the highest in the surface soil and decreases by 10% for every 30 cm depth. Further survey of more BM soils of the IGP would certainly provide better interpretation and understanding of microbial diversity and population composition.

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