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BELOWGROUND BIODIVERSITY IN RELATION TO CROPPING SYSTEMS



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PREAMBLE

Soil, rhetorically described as the 'final frontier', harbours a largely unknown microbial universe whose diversity in quantitative terms is indeed staggering. There are more than 10¹⁶ prokaryotes alone in soil. Recent estimates show that the soil microbial diversity would be at least three times greater than thought earlier.

The belowground biodiversity is not a passive entity; it is a continuum of the life in soil. There is an intimate link between the belowground biodiversity (BGBD) and the processes and sequences of the biodiverse life aboveground (AGBD). Although laser-based technology and the tools of new biology have now made it possible to see through the largely opaque soil and to study the belowground communities *in situ*, it is well nigh impossible to make a direct estimate of the structural and functional diversity of the belowground communities, and to relate these with the biodiversity aboveground. Although the 'last frontier' has not been breached, the few installations made here and there have shown that BGBD as provider of ecosystem goods and services is vitally linked to ecosystem functions, and hence, the welfare of the Universe.

The agro-ecosystems consisting of the temporally variable and spatially heterogeneous cropping systems are candidates for BGBD search in relation to the contributions made by the belowground communities towards sustainability of land and soil productivity under the different cropping systems. *The search has led to more questions than answers and the researchers are now concerned more than ever to make BGBD search relevant to the systems of agricultural production.*

Belowground biodiversity the provider of ecosystem goods and services

A provider of ecosystem goods and services, the belowground biota, as drivers and managers of the natural soil processes, are irrevocably linked to land and soil productivity, that forms the basis of sustainable crop production. The more important aspects of these processes include:

- Nutrient cycling
- Regulation of soil organic matter dynamics
- Soil carbon sequestration and its impact on greenhouse gas (GHG) emission
- Modification of soil physical structure
- Assistance to plant nutrient acquisition mineralization, fixation and mobilization of nutrients
- Enhancement of plant health, and biotic and abiotic stress tolerance

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(1)

Shape and size of the BGBD

The shape and size of the belowground biodiversity is as complex as that of the aboveground. The former consists of the prokaryotes and eukaryotes: the *microflora* bacteria, fungi, algae etc., *micro-* and *mesofauna* protozoa, nematodes, arthropods, mites etc. and the *macrofauna* insects, earthworms, small vertebrates etc. These diverse biotic groups and the roots of plants that grow in the soil, interact with other species, and constitute the life in soil. Apparently, the extent of diversity in numbers of various biotic groups varies directly with their relative dimensions. For every species of vertebrates and earthworms in a soil, there may be hundreds of species of fungi and thousands of species of bacteria. Mathematical interpretation of the diversity abundance distribution relations of the microorganisms in soil, have allowed prediction that there could be 10⁷ prokaryotic taxa in a gram of pristine soil.

The genomic equivalent of prokaryotic diversity of a pasture or a forest soil may be anywhere between x 20-30 larger than that of an average arable soil, although the relative abundance of culturable microorganism populations in soils under such contrasting land utilization systems may be nearly the same. Considering the small number of environmental DNA clones generally analyzed under ecogenomic studies, one is possibly looking at the tip of the iceberg even with the state-of-the-art molecular techniques, such as DGGE, SSCP and tRFLP.

Rhizosphere - a hot spot of belowground biodiversity for the benefit of cropping systems

Root exudates, sloughed off mucilage, dead cells, and other metabolites and by-products are responsible for changed composition and dynamics of the soil biota in the rootsoil interface, the rhizosphere. The hallmark of the rhizosphere lies in the physical and chemical interactions between the roots and the biotic communities of the soil that govern many soil processes influencing the growth of plants. In view of such unique interactions, the 'rhizosphere' is considered as a 'hot spot' for the complimentary relations between the aboveground plant life and the belowground biota.

Molecular tools of ecogenomics have allowed preparation of fairly accurate rhizosphere microbial fingerprints of several crops under varied systems of production, particularly with reference to the biogeochemical cycling and turnover of nutrients nitrogen, phosphorus, iron, sulfur etc., plant growth promotion by non-nutritional mechanisms, and suppression of deleterious microorganisms in the soil-root interface. Use of species-specific molecular probes coupled to gene sequencing, and now the microarrays, permits functional assessment of the rhizosphere biodiversity *in situ*.

Threats and likely causes of BGBD loss in ecosystems

Belowground biodiversity loss is a complex problem involving multiple causative factors. To ascertain the causes of the loss, it is imperative first to know *what determines the biodiversity or the species richness of communities belowground*. In spite of the advances made in BGBD search, it is yet difficult to disentangle the roles of the soil, environment and aboveground biota in determining the BGBD. However, the evidence, like that of the neutral North American deserts having a higher bacterial diversity than the biodiversity rich South American acidic rain forests,

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suggests that the life sustaining (or threatening) abiotic soil environmental factors (for example, pH) may be more important than the aboveground biotic diversity in determining the BGBD. Examples of the kind now available with other belowground biotic communities embolden this argument.

Available evidence suggests that soil biotic communities are more resilient to anthropogenic perturbations than the aboveground diversity, but disturbances in aboveground biota (such as that are caused by land clearing, resettlement, monoculture of crops and trees, use of agrochemicals, intensive tillage and such other practices which negatively affect aboveground biodiversity) may have large impact on the BGBD, and might negatively affect the self-regulatory capacity of the ecosystems. It seems that the high-energy driven intensive agricultural production systems have introduced a kind of vicious cycle, whereby leading the loss in BGBD. As a consequence, the soil biological function, are provided more and more with energy intensive external inputs, bringing further peril for the BGBD, and hence for the sustainability of the production systems.

Belowground biodiversity - methodology base for study

While the traditional methods of sample collection and direct estimation of the faunal component have remained largely valid, the techniques of assessing microbial communities have now undergone a tremendous change. Assessment and identification of the diversity can now be accelerated by using automated methods (e.g. Biolog), and broad scale measures of diversity, e.g., phospholipid fatty acid (PLFA) profiling, fatty acid methyl ester (FAME) profiling, DNA hybridization and reassociation kinetics, etc. These techniques have largely overcome the limitations of the cultivation-based techniques.

Molecular techniques of analysis of the community or species diversity at genetic level based on polymorphism of indicator genes extracted from environmental samples have emerged as alternatives that allow a higher resolution and avoid many of the limitations of the traditional methods. The indicator genes may be functional genes, e.g. genes encoding proteins that perform particular metabolic functions relevant to the ecosystem.

In view of the above background, NAAS organized a Brainstorming Session on "Belowground Biodiversity with Special Reference to Cropping Systems" on March 10, 2006 at Barkatullah University, Bhopal (M.P.) with Dr. B.N. Johri, Emeritus Scientist and Editor NAAS as the Convener. Twenty Eight experts deliberated at length on five major themes: (i) Sustaining Biological Basis of Agriculture, (ii) Development of Tools of BGBD Research, (iii) Climate-Land-Soil and BGBD, (iv) Belowground and Aboveground Biodiversity Interface and Linkages, and (v) Priorities and Researchable Issues. Each theme was introduced by a lead speaker followed by panelists' interventions and open-house discussion of the issues raised. A short plenary session summarized the main points and recommendations.

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BGBD - CONCEPTS AND QUESTIONS

The thematic deliberations addressed the following concepts and questions:

- *"Microbial diversity in soil is beyond practical calculation".* Whatever we talk about diversity is based on our 'observations' made from seemingly, the observed facts. Considering the vastness of the microbial diversity and the variable entity of the soil, it is incomprehensible that we can reach to the bottom of the microbial diversity in soil by empirical observations alone. Mathematics greatly helps us in making observations and drawing conclusions from the observed facts. We possibly need simple mathematical theories to guide us in exploring the BGBD and making predictions.
- Soil biodiversity is an abstract aggregated property of species in the context of communities or ecosystems". Can differences in community or ecosystem function be assigned to the community diversity or species richness of communities belowground? Functioning of terrestrial ecosystems plant biodiversity, productivity, variability and stability seems to depend heavily on community diversity of soil biota. Functional diversity rather than taxonomic diversity (community structure) or species richness per se is the major determinant of ecosystem functioning. It may be thus more important to understand the linkages between the actions of a key species or the functional groups and ecological functions of different ecosystems than to search for the diversity index or the species richness and try to relate the same to ecosystem or community functioning.
- What determines the belowground biodiversity and species richness of communities? How important are soil abiotic factors vs. aboveground biotic diversity and interactions in determining the resulting belowground community? The tenets of species-area relationship, as applied to microorganisms, prokaryotes and eukaroytes, reveal that community diversity and species richness increase with the area owing to the spatial complexity of habitat structure, even within a constant biotic frame aboveground. Environment affects species, and the outcome of their interactions, but not the diversity or ecosystem processes directly. The commonly observed BGBD loss in intensively managed arable soils compared to the native biodiversity of the ecosystems may be a reflection of the intensity and duration of exposure to the changed environment (e.g. pH) under the systems and practices of intensive arable soil and crop management.
- The ecological tenets of structure-function relationship of ecosystems as empirically applied to soil microorganisms would show that, (i) maintenance of a diverse and functional microbial community i.e., functional biodiversity is essential for soil sustainability, and (ii) loss of biodiversity inherently makes the biological systems less resilient to environmental stresses, such as those introduced by intensive soil and crop management practices. This sets the agenda for BGBD search in relation to sustainable productivity management of land and soils for the crops. The key issue here is whether it is

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possible to maintain a tight fit BGBD at threshold level within the ambit of sustainable production system.

- Can the positive relationships between soil (biological) quality, soil functions, and soil productivity as clearly discernible in the undisturbed natural ecosystems be extended to agricultural production systems and exploited for sustainable production management of crops? The biologically driven alternative agricultural production systems and practices have provided empirical information about the changes in soil biological processes and the quality traits. One important question to answer in this context is whether the observed changes and benefits are in integration or in segregation of the different land use systems?
- Can we delineate keystone BGBD markers across the functionality of different production systems? The state-of-the-art of the BGBD search in relation to the status of soil health and productive functions of different land use systems relies heavily on soil biological quality parameters (soil organic matter (SOM), microbial biomass, soil enzymes, etc.). For linking BGBD functions with the functionality and stability of different production systems and generating principles and practices of their sustainable management, we need broad based biotic indicators. Against the current state of our knowledge, it is different production systems and their functions. Instead, system and function specific biotic indicators, which satisfy the stringent requirements of functionality, reliability and reproducibility will help in advancing the cause of BGBD search for cropping system management. Could new molecular tools help us to reach such indicators?
- The nexus between system productivity and soil organic matter (SOM), especially the more labile part of it, through aggradations of fertility and physical structure underwrites the SOM-BGBD-sustainability linkage. Organic matter management being central to soil sustainability management, can a working model be developed based on SOM or any relevant part of it including its role in buffering the soil reaction, to provide for the indicators of SOM BGBD sustainability linkage?
- Carbon sequestered in soil is three times more compared to that in aboveground biomass. In view of the mycorrhizal fungi functioning as a main carbon sink in soil with direct links to the atmosphere, can the arbuscular and ectomycorrhizal fungi as distributed in the ecological zones be considered as indicators of carbon cycling and the interconnected ecosystem processes vis á vis plant production and GHG management?
- Nematodes, especially the bacteria feeding ones, are good indicators of soil disturbance.
 Can nematodes be developed as indicators of soil stress in disturbed production systems?
 Could ants be used as keystone species in cropping systems? Can the role of collembolans as ecosystem engineers at micro-level be exploited to relate to ecosystem health?

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 Can any indicator ratio or quotient, such as that of the proportion of r and k strategists or that of termites vs. earthworms be used to monitor production system sustainability?

RECOMMENDATIONS

- Considering the enormous diversity and heterogeneity of the microbial forms and spatial variability of habitat structure even within the similar production systems, research effort is needed to develop mathematical models for assessment of the BGBD-cropping system interaction. The microbial ecologists, crop production specialists, molecular biologists and statisticians need to collaborate in this endeavour.
- In view of the alarming scenario of global warming vis á vis GHG emission, BGBD contributions towards carbon sequestration in soil should receive active consideration. The impact of GHG atmospheric warming nexus on BGBD ecosystem functionality links needs precise understanding. The agricultural production systems, such as wetland rice should receive adequate attention from the point of view of reducing the risk of GHG emissions.
- In view of the ongoing degradation of the biological base of soil productivity in the prime agro-ecosystems (such as the rice wheat system of the IGP), efforts should be directed to precisely assess, evaluate and monitor the BGBD structure and functioning in select agro-ecosystems. Inventorization, identification, characterization and conservation of the soil microorganisms using molecular and traditional tools should be a priority objective. Agro-ecosystems such as dryland agriculture, hill agriculture, coastal zone agriculture and mixed farming should be the immediate targets.
- Intensive agriculture such as that practiced in rice-rice or rice-wheat cropping systems over a large area of the country, utilizes heavy doses of chemical fertilizers and pesticides, whereas these crops show low belowground : aboveground biomass ratio on account of considerable ground and surface water utilization. This has brought about considerable changes in the belowground habitat, and introduced stresses and strains that are likely to result in at least short-term disturbances in microbial community structure and functioning in the soil and the rhizosphere. Forms that degrade xenobiotics are likely to be abundant under such situations. These production systems offer scope for studying the extent of soil degradation that allow microbial functionality at an acceptable level under the broad principles of BGBD conservation and management in intensive cropping systems. Hence, long-term restoration studies of the degraded systems shall help arrive at quantifiable indices of soil microbial diversity-functionality relationships of the agroecosystems.

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- Attempts should be made to work out the threshold level of the functional microorganism groups and their phenological dynamics in the rhizosphere in relation to the turnover and cycling of nutrients for better management of sustainability of the production systems. The holistic approach should take care of the metabiosis in the rhizosphere that determines the successional colonization of niche by individuals and population groups.
- Plant breeding has generally tended to ignore the functional contributions of microorganisms in soil and rhizhosphere (e.g. AMF and *Rhizobium* symbioses), and the plant genetic traits like enhanced association with and response to the beneficial microbial community groups in soil. Crop-genotype-specific differences in supporting soil biological processes can open a new area of plant breeding and biotechnology for exploiting soil biology for better production management of crops and cropping systems in a sustainable manner. The conventional and modern tools of plant breeding can be taken advantage of in breeding crop cultivars with enhanced response to the specific belowground communities. Such varieties shall be of greater advantage in case of alternative, low input systems of production.
- The root exudates microorganisms interaction in the rhizosphere can be a potential area of soil biological management for sustainability. The genetic control of root exudation remains largely unknown, and if known, shall offer scope for modification by genetic engineering to exploit rhizosphere build up of the beneficial microbiota and to facilitate suppression of the deleterious indigenous ones. Also, there is scope to exploit allelo-chemicals in this context to suppress unwanted weeds, pathogens and herbivores. Bioinformatics can be a useful tool to advance the concept.
- Fine roots, < 30 um are considered, in a broad sense, a part of the soil biota and functionally interact with the microbial community groups in many diverse ways. The interactions between fine root architecture and soil microorganisms in relation to dynamics and turnover of nutrients, herbivory, escape and tolerance to deleterious microorganisms, etc., should be studied employing modern tools, like radiotracer techniques to generate data for genetic manipulation and modeling.
- The major representative crops and cropping systems should be utilized to generate data on interventions directed towards the re-establishment of soil biology using biomarkers, such as the AMF, nematodes, earthworms, etc., so that the interventions with organics and biologicals can be based on sound scientific knowledge. Data generation is particularly important where new crop types inbreds, hybrids and transgenics are involved.
- Considering the steady increase in the exploitation of transgenic crops, it is important to take up, on priority, assessment of the ecological impact of their

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residues and rhizodeposits on BGBD structure and function; as for example, the overall impact of transgenes directed to suppress the insects or the nematodes, or the impact of crop genotypes modified in the pathway of recognition and defense to fungal pathogens or AMF colonization. This should also take care of the horizontal gene transfer from the transgenics to the microorganisms. This information, apart from putting transgenics use on sound ecological understanding, shall help in understanding the microbial gene expressions in soil.

- The IPM practices provide a platform for studying the relations of the ecologically driven IPM interventions with other soil processes as also the signal transduction mechanisms against pathogens and pests. This may be applicable in a reverse way on the likely impact of biofertilizers and PGPRs (AMF, Azospirillum, Rhizobium, fluorescent Pseudomonas etc.) on signal transductions for defense against the pathogens. Such information is likely to go a long way to design appropriate plant disease and pest management practices based on sound ecological principles.
- A more coherent and directed effort is necessary to assess the indigenous diversity, especially of the growth promoting soil microorganisms. This will help in extending the advantages of the natural bio-resource base (that the BGBD is) to intensive agriculture and maintenance of soil health.
- To meet the goals stated above, it is essential (a) to put in place a research initiative on Long Term Ecological Research (LTER) for varied ecosystems to investigate the BGBD cropping or agro-ecosystem functional linkages, and to utilize the information for more balanced and productive cropping systems, and (b) to upgrade soil microbiology, including molecular biology research tools and methods. These efforts will not be successful without the well trained and dedicated manpower in the fields of molecular and morphometric taxonomy, biosystematics, molecular ecology, and bioinformatics - BGBD-cropping system interaction research. This will require considerable investment for laboratory upgradation, introduction of teaching and training programmes, and adequate research support to the institutes and universities. Creation of Centres of Excellence around outstanding microbial ecologists will be a desirable step in this direction.
- The recently established National Biodiversity Authority for implementing the Biodiversity Act (2002) should set up an Expert Group to develop guidelines for the conservation, and sustainable and equitable use of soil micro-flora and meso-fauna.

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