

**SOIL MICROBIAL DIVERSITY IN THE DISTURBED AND
UNDISTURBED DECIDUOUS FORESTS
OF MEGHALAYA**

**BY
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IN LOVING MEMORY
OF MY
BELOVED GRANDPARENTS

**NORTH EASTERN HILL UNIVERSITY
SHILLONG**

I, **Melboreen Dkhar**, hereby, declare that the subject matter of this thesis entitled "Soil microbial diversity in the disturbed and undisturbed deciduous forests of Meghalaya" is the record of work done by me, that the contents of this thesis did not form basis of the award of any previous degree to me or to the best of my knowledge to anybody else, and that the thesis has not been submitted by me for any research degree in any other University/Institute.

This is being submitted to the North Eastern Hill University, Shillong for the award of the degree of Doctor of Philosophy in Botany.

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INTRODUCTION

Microbial diversity is an unseen national as well as international resource that deserves greater attention than it has been receiving. It encompasses the spectrum of variability among all types of microorganisms in the natural world and as altered by human intervention. Microbial diversity studies are important in order to understand the microbial ecology in soil and other ecosystems (Atlas, 1984 and Reid, 1994). It plays an important role in both natural and agroecosystems. The diversity of plants and animals in forests and agro-ecosystems receives a great deal of scientific attention, whereas the diversity of microorganisms is often ignored. Therefore, much more needs to be done to understand the role of microorganisms and inventory their diversity and to find ways to exploit them beneficially.

Diversity means state of being diverse, difference, unlikeness and variety. The total variability of life on earth means the variability among living organisms from all sources including interalia, terrestrial, marine and other aquatic ecosystems and the ecological complexes of which they are part of; this includes diversity within species and of ecosystems. Strictly speaking, the biodiversity refers to the quality, range or extent of differences between the biological entities in a given set. In total, it would thus be the diversity of all life and is a characterizing property of nature, not in entity or a resource. Biodiversity is the established norm of the nature. The term "diversity", as used today, stands from a molecular to a global level of biological organization. It can be applied to different ecosystems, populations, and even to different individuals. Biological diversity can be defined as "the variety of species in ecosystems as well as the genetic variability within each species" (Conservation, 1987). Diversity is, therefore, the range of

significantly different kinds of organisms and their relative abundance in natural assemblage and habitat. An estimate of microbial diversity is a prerequisite for understanding the functional activities of microorganisms in ecosystems (Garland and Mills, 1994; Zak *et al.*, 1994).

Studies on biodiversity and its relation to ecosystem structure and function have mainly focused on macroorganisms, and little attention has been directed towards microorganisms. Microorganisms, however, are important, especially due to the fact that life is dependent on microbial processes. The last decade has produced an extraordinary new awareness of microbial diversity (Dykhuisen, 1997). Microorganisms have an enormous impact and role in our daily lives, including everything from maintaining the biosphere to improving our life style (Hunter-Cevera, 1998).

Biodiversity is the variety of nature. It is manifested at several levels of biological organization and should be studied at the levels of individuals, populations and biocoenoses (plant, animals and microbial communities). In evolutionary terms, the ultimate cause of diversity exists at higher levels of genetic variation. Any change at one level can affect other levels, leading either to increased or decreased biodiversity (Romke *et al.*, 1996). Soil biodiversity should not only mean, however, to maximize the number of species in a certain ecosystem, more likely it is the ability to maintain current macro- and micro-flora and emphasizing the various species and processes so as to accomplish specific objectives. The term species diversity consists of two components; the first component is the total number of species richness. In other words, it refers to the quantitative variation among species. The second component is the distribution of individuals among these species, which is referred to as evenness or equitability. One

problem is that evenness is often unknown in bacterial assemblages because individuals very seldom are identifiable to the species level. The term "bacterial diversity" has been used to describe complexity and variability at different levels of biological organization, including genetic diversity within bacterial taxons (species), diversity of bacterial taxons in assemblages or habitats and ecological diversity including variability in community structure, complexity of interactions, number of trophic level and number of guilds (functional diversity). As the soil microbial community is a complex picture of interwoven relationships between organisms of different trophic levels, this will lead to many indirect effects. There are two important implications of microbial diversity in this regard. Firstly, a decrease in diversity will generally result in the risk that there is a decrease in ability of the biological system to respond to perturbations (Ekschmitt and Griffiths, 1998). Secondly, bacterial diversity reflects the state and history of influences on the microenvironment, the diversity itself gives an indication as to how stressed the ecosystem has been. Biodiversity is affected by both qualified and quantified factors: in less diverse habitats a relatively slight damage is able to cause a more completed destruction, where less type of species will be found. So, the general assessment should be based on the status of the species: conservation value of a given species is higher if it occurs exclusively in endangered habitats.

In an ecosystem, spatiotemporal complexity is of fundamental importance in maintaining species diversity (Tilman, 1994; White *et al.*, 1998). Although it is not possible to be sure that different species are using the same resource, the following well-documented patterns are suggestive of this: (1) seasonal variations in fungal populations following variations in temperature and moisture (Christensen 1969; Bisset and

Parkinson, 1979a and b; Widden and Arbitol, 1980; Widden, 1981, 1986b; Eastburn and Butler, 1988; Cooke and Whipps, 1993); and (2) short-term rhythmic behavior, e.g. diurnal cycles of growth and reproduction which are often controlled by light, temperature or water (Cooke and Whipps, 1993). Microorganisms play an integral and often unique role in ecosystem functions, yet, little is known about dominant populations that presumably play vital roles in these functions, nor do we know much about how these populations differ with habitat. The greatest microbial diversity at small scales appears to reside in the soil. Soil microbial communities are among the most complex, diverse and important assemblages in the biosphere diversity (Zhou *et al.*, 2003).

Diversity can vary with a number of factors such as disturbances and stress, in addition to ecological interactions (predator-prey interactions). The major underlying principle of diversity studies is probably the assumption that interactions between populations in a habitat lead to an organized and stable community (Atlas, 1984).

Microbial diversity indices can function as bio-indicator to show community stability and describing the ecological dynamic of community (Atlas, 1984) and analysis of soil microbial diversity is important to evaluate the importance of perturbations in soil systems (Turco *et al.*, 1994). Microbial populations can also provide an early indication of changes in soil long before it can be measured by changes in organic matter (Powlson *et al.*, 1987). It has been generally hypothesized that reduction in soil microbial diversity will result in reduction in the functional capability of soil (Giller *et al.*, 1997). However, decline in soil microbial diversity does not consistently result in reductions in the functional diversity of microbial communities (Klein *et al.*, 1986; Atlas *et al.*, 1991).

The diversity of soil fauna and microorganisms is influenced by vegetation, soil factors, climate and management practices (Gupta and Malik, 1996). Soil microorganisms play a very important role in fertility of soil not only because of their ability to carry out biochemical transformation but also due to their importance as a source and sink of mineral nutrients (Jenkinson and Ladd, 1981). There have been studies on the distribution of microbial diversity in various environmental niches (Stolp, 1988). Limited work has been carried out on microbial diversity of various management practices (Wardle, 1995).

The knowledge of complex relationship between plant and microorganisms and also among different microorganisms is an important aspect to understand plant behavior. There is a growing interest in the relationships among ecosystem diversity, structure, function and a number of theories have been formulated concerning how species diversity is related to ecosystem function (Muller *et al.*, 2001). Enhanced species diversity is beneficial to ecosystem function (Naeem *et al.*, 1995). In contrast, other authors suggest that the properties of an ecosystem depend more upon the functional abilities of a particular species than on the total number of species (Tilman *et al.*, 1997 and Wardle *et al.*, 1997). The diversity and functional importance of soil organisms present an excellent opportunity to test currently tropical aspects of ecological theory, the hypotheses relating ecosystem diversity and function have mainly been developed by plant ecologists with only a few studies having concerned the soil community (Mikola and Setälä, 1998; Griffiths *et al.*, 2000). Hypotheses from the aboveground communities may not easily be applied to belowground, since there are differences. The microbial diversity in soil is enormous (Torsvik *et al.*, 1990) and there may be substantial overlap in function between

microbial species (Chapin *et al.*, 1997). Furthermore, it is likely that microorganisms within a functional group differ in their response to the environment. As microorganisms are fast growing, they can quickly fill out empty niches occurring when the environment is changing (Giller *et al.*, 1998). These circumstances could create a high degree of stability, but it is unknown what level of diversity is necessary to maintain stability (Wardle and Giller, 1996).

Soil life is highly diverse and consists of interacting population of microorganisms and soil fauna whose activities influence physical, chemical and biological properties of the soil. Soil, the most important natural resource harbors a variety of microorganisms and is considered to be the most dynamic site of interactions in nature. Determination of the optimum diversities of soil microbial populations of both natural and agricultural systems for their sustainable management is very important.

Soil microorganisms are of great importance for soil ecosystems because they affect plant available nutrients and soil structural stability (Paul and Clark, 1989). The abundance, size and activity of the microbial populations depend on quantity and quality of organic matter, texture, and other environmental factors (e.g. soil type, nutrient status, pH, moisture) as well as plant factors e.g. species, age (Insam *et al.*, 1989; Kaiser *et al.*, 1992). Microbial growth in soil is carbon limited and therefore, the presence of organic matter has the greatest influence on microbial populations (Lynch and Whipps, 1990; Wardle, 1992).

The impact of burgeoning human populations has destroyed the soil physico-chemical environment and the soil's species through activities such as: inputs of chemicals from the atmosphere, disposal of waste products in soils, ground water

contamination and physical modification or removal of soil by cultivation and erosion. Soil among the vast unknown life on our planet, is a dark frontier, despite their critical importance to understanding ecosystem function. For example, thousands of species of microbes and invertebrates inhabit just a square meter of temperate grassland soil, organisms whose identities and contributions to sustaining our biosphere are largely undiscovered. The elucidation of species diversity of soils in conjunction with sustainability assessments of soil-mediated ecosystem processes must be a high priority in global biodiversity efforts.

It is also suggested that the total functional diversity of microorganisms in different soils may be similar and that a combination of environment and plant factors influence organisms which are active, become culturable (Colwell *et al.*, 1985) and proliferate under different conditions. Many fungi and more than half of the species of bacteria at present classified can be found in soil. Soil microorganisms are largely hidden underground population; frequently have greater mass than the plants and animals aboveground (Jenkinson and Ladd, 1981 and Sparling, 1985). Despite their comparable mass, we know much less about microorganisms than we do about higher plants and animals (Wardle and Giller, 1996). The analysis of the functional diversity of soil populations is a useful way to characterize and compare the microbial community. The high degree of functional redundancy in soil microbial communities may result in there being no effect of changes in microbial diversity on the function of the communities (Andren *et al.*, 1995; Giller *et al.*, 1997). This is likely because, in the case where a large number of species conduct similar functions, a reduction in any group of species has little effect on overall soil processes since other organisms fill the functional role (Lawton and

Brown, 1993; Andren *et al.*, 1995). However, broad functional diversity may be additionally important in influencing the resilience of soils (Elliot and Lynch, 1994; Pankhurst *et al.*, 1996; Giller *et al.*, 1997). Soil resilience principally concerns the capacity for soil to function after different disturbance treatments. However, an important element of soil resilience should also include the capacity for soils to continue functioning under a range of environmental conditions. It is, therefore, relevant that an assessment of the functional capability of microbial communities with reduced diversity be conducted under a range of environmental conditions.

Soil microbial communities remain some of the most difficult communities to characterize, because of their immense phenotypic and genotypic diversity. Activity and growth of microorganisms is restricted by soil environmental factors, e.g., temperature, moisture, pore size, distribution and nutrient availability and therefore, indirectly by cultivation practices.

The structure of soil microbial communities is determined by a hierarchical series of interacting variables. Climate and microclimate set the physiological limits for microbial activities. The quality and quantity of plant resource inputs influence the abundance of different fungal and bacterial species. At the micro-scale, physico-chemical environment and biotic interactions refine the composition and dynamics of specific species associations or unit communities. Vegetation cover directly and indirectly influences all these variables and often has a dominant effect on the spatial patterning of soil microbial communities. This is particularly evident for trees that modify the environment and underlying soil properties at scales that are amenable to systematic analysis. The application of geostatistical technique has shown that the spatial

distribution of microbial communities in forest is related to the location of different tree species and the associated ground flora (Pennanen *et al.*, 1999; Saetre, 1999).

Fungi and bacteria control many of the vital processes on which the very maintenance and survival of tropical forests depend (Hawksworth and Colwell, 1992). An overview of the role of microorganisms in ecosystem functioning as a whole has been presented by Allsopp *et al.* (1995). Their goal is to identify functional attributes of microorganisms in tropical forests and to identify those processes that are most likely to be sensitive to losses of diversity especially in the face of disturbance or broad environmental changes. In some cases, microbes may influence ecosystem processes indirectly by altering the diversity of other organisms.

Humans are altering global nutrient cycles as well as reducing biological diversity. It is of paramount importance to understand the effect of these global changes on ecosystem functions, especially those on which human life directly depends. Microbes are key players in ecosystem functions such as decomposition of organic matter and nitrogen cycling. It has been demonstrated that microbial community composition can influence certain ecosystem process rates. However, little is known about the factors that determine microbial community composition and/or functioning, including anthropogenic disturbance.

The structure and functional diversity of the microbial communities in the soil is tightly related to plant species composition above - ground, thus providing an important link between above and below ground processes in terrestrial ecosystems (Grayston and Campbell, 1996; Westover *et al.*, 1997; Priha *et al.*, 1999; Grayston *et al.*, 2001).

Soil management practices affect soil microbial communities, which mediate many processes essential to the productivity and sustainability of soil. Microbes also play a major role in the formation of good soil structure. Bacterial mucigel and hyphal threads produced by fungi and actinomycetes bind the soil particles together. Microbial activity helps to aggregate the soil, which reduces soil erosion, allows for good water infiltration, and maintains adequate aeration of soil. Soil microbes also affect the persistence of organic compounds applied to soil (Biro *et al.*, 1983).

The effect of disturbance on microbial community function depends on its duration and specificity. After a transient disturbance, system function may eventually return to its former state, whereas a permanent disturbance will result in a new altered state (Rykiel, 1985). Disturbance with a specific mode of action only alter a few groups of organisms, whereas, those that act non-specifically affect a wide range of organisms. Investigation of how different kinds of disturbances affect the system function will enhance our knowledge of the relationships among diversity, structure and function.

Recognition of the importance of soil microorganisms in the functioning of ecosystems has led to an increased interest in measuring the nutrients held in soil biomass (Brookes *et al.*, 1982; Martikainen and Palojarvi, 1990). The microbial biomass is a potential source of nutrients for plants especially of their ability to bind temporarily the nutrition to microbial cells. The microbial biomass and its activity are important indicators of changes in content and vertical distribution of the organic matter in a soil profile. Numerous studies on the measurement of microbial C, N and P in different natural and disturbed ecosystems have been shown that the soil microbial biomass is among the most labile pools of C and mineral nutrients which are liberated after the death

of microorganisms (Anderson and Domsch, 1980; Smith and Paul, 1990; Diaz-Ravina *et al.*, 1993). It serves as an important reservoir of plant nutrients such as N and P (Jenkinson and Ladd, 1981; Marumoto *et al.*, 1982; Van Veen *et al.*, 1987; Duxbury *et al.*, 1989; Jenkinson and Parry, 1989; Smith and Paul, 1990; Lodge, 1993).

Large annual fluctuations in the microbial biomass has been reported by Lynch and Panting (1980a and b) while others observed only small annual changes (Schnurer *et al.*, 1986; Patra *et al.*, 1990). Soil microbial biomass plays a key role in maintaining soil fertility because its activity is the primary driving force for cycling of elements such as carbon and nitrogen.

With increased study, more precise information has been gained on the influence of the microbial biomass on carbon and nutrient cycling, controlling turnover and mineralization of organic substrates (Sparling, 1985) and influencing forest productivity (McGill *et al.*, 1986). Estimation of soil microbial biomass is now frequently made because of the importance of soil organisms in nutrient cycling and their role as a source and sink of plant nutrients (Smith and Paul, 1990). Study of microbial communities and biomass in forest soil may give insight into the role of microbes in restoring soil fertility. Microbial biomass is the characteristic of microorganisms which participate in the biochemical cycles and are living part of the soil organic matter (Cengel, 1990; Srivastava, 1992).

The microbial biomass content of the soil depends on the quality and quantity of resource and its distribution of the C-input varies with time and depth (Kaiser and Heinemeyer, 1993). Generally, microbial biomass C comprises about 2-4 % of total organic C (Anderson and Domsch, 1989) but variation within this range is influenced by

the quantity and quality of organic inputs to the soil (Wardle, 1992). A recent review by Nannipieri *et al.* (2002) has focused on the inter-relationship between the biomass, its diversity and function in soil. The microbial biomass is a sensitive indicator of changes resulting from agronomic practices and other perturbations of the soil ecosystem (Doran, 1987; Smith and Paul, 1990).

Though, microbial biomass is relatively small fraction of the total biomass in terrestrial ecosystem, the microbial activity is of paramount importance in the nutrient cycling and energy flow (Diaz- Ravina *et al.*, 1993b). Usually, the microbial biomass of the tropical environments is sensitive to land use changes and even appears to be a sensitive indicator of both soil carbon and background nitrification. The changes in microbial biomass can provide an early indication for a slower and less detectable soil organic matter and soil fertility as well (Srivastava, 1992; Henrot and Robertson, 1994; Maithani *et al.*, 1996; Taylor *et al.*, 1999).

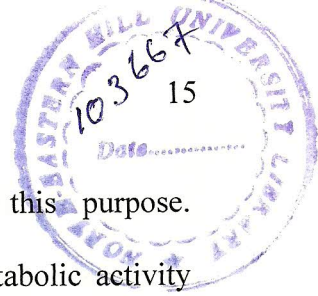
The physico-chemical characteristics of soil influence the level of biomass and activity of microorganisms. Values of microbial biomass can provide one of the most satisfactory estimates of the restoration of soil microbial populations (Maithani *et al.*, 1996). The close relationship between pH and the microbial biomass is indicative of complex feedback mechanisms which are activated by the fauna. Less acidic environments stimulate the microbial biomass as determined by substrate-induced respiration (Anderson and Domsch, 1993). The stimulation leads to higher enzymes production, as shown by the close correlation between microbial biomass and N turnover. Higher enzyme production, in turn, may raise the soil pH by releasing NH_4^+ .

All the biochemical reactions in soil are carried out in the presence of enzymes. Degree of enzymes activity is usually the important indicator of soil bio-activity and fertility. Major biological processes such as mineralization, immobilization, nitrification, nitrate reduction etc. are the result of microbial activities and are catalyzed by enzymes. Chemical reaction in soil is highly influenced by enzymatic activity. The biological activity in soil provides better insight in the understanding of transformation of organic matter (Pietikainen and Fritze, 1995). Therefore, knowledge about enzymes activities and their temporal and seasonal variations in soil has considerable biological significance.

Enzymes in soils can persist for an extended period of time, because the organic fractions have a protective influence on soil enzyme activity (Skujins, 1976; Coyne, 1999). The decomposition of organic matter is a chain reaction in which enzymatic reactions control the supply of substrate and energy for microbial growth, while the latter controls the microbial processes. Soil enzymes have an essential role in integrating the effects of climate, cultivation, soil amendments and edaphic properties; therefore, they may be considered as important indicators of the total biological activity and also the fertility of soil. The activity of a particular enzyme in the soil is a composite of activities associated with various biotic and abiotic components (Burns, 1982). These soil enzymes have been considered as potential component of group of indices to assess soil quality as the level of soil enzymes can serve as an estimate of the ecosystem disruption (Tate, 1995). Measurements of enzymatic activities have been used as a measure of total microbiological activity and soil fertility levels (Stevenson, 1959; Tiwari *et al.*, 1988a and b; Chander and Brookes, 1991). There appears to be a direct relationship between number of microorganisms and the enzymatic activity.

Enzymes in soils originate not only from microbial sources but also from animals, plants and the resulting soil biological activity includes the metabolic processes of all these organisms (Ladd, 1978; McKay, 1991; Tabatabai, 1994; Schinner *et al.*, 1995; Sarapatka, 2003). During recent times, much emphasis is laid on the activity of microorganisms in the soil. Generally, the rates of microbial mediated biochemical reactions are used for this purpose as they provide an index of microbial activity (Skujins, 1978). Soil enzymes play a key role in releasing nutrients. It is useful to consider some of the soil properties that may influence phosphomonoesterase (PME) and phosphodiesterase (PDE) activity. Evaluation of soil organisms is closely related to soil enzyme activity. Soil enzymes have been suggested as potential indicators of soil quality because of their relationship to soil biology, ease of measurement and rapid response to changes in soil management (Dick, 1994; Dick *et al.*, 1996). Such an index would integrate chemical, physical and biological characteristics and be used to monitor the effects of soil management on long-term productivity (Doran and Parkin, 1994). The activity of soil microorganisms is strongly linked to the activity of enzymes and soil management (including crop rotations, fertilization, tillage and crop residue placement) strongly influences the activity of soil enzymes (Miller and Dick, 1995; Deng and Tabatabai 1996a and b, 1997; Klose *et al.*, 1999). Most of the enzymes are added to soils by decaying microbial tissues, plant residues and animal remains.

The physiological and metabolism of soil microorganisms are driven by enzymes and the microbial habitat in soil is affected by these soil enzymes. The enzymes whose activities are most widely studied are dehydrogenase (oxido-reductase), urease (hydrolases) and phosphatase due their importance in various management practices.



Dehydrogenase, urease and phosphatase are commonly used for this purpose. Dehydrogenase, being a respiratory enzyme, provides a measure of catabolic activity (Lenhard, 1956; Stevenson, 1959; Casida *et al.*, 1964). The dehydrogenase activity in the soil may be used as a measurement of overall microbial activity. Dehydrogenase enzymes are considered to play an essential role in the initial stage of the oxidation of soil organic matter by transferring hydrogen or electron from substrates to acceptors. This aspect has been studied by several workers since the introduction by Lenhard (1956). Being a respiratory enzyme, it provides a measure of catabolic activity of soil (Skujins, 1976) and is considered to be caused by broad group of endocellular enzymes (Skujins, 1978). It acts as an indicator of the microbiological system in soils and can be considered a good measure of microbial oxidative activity. It is a sufficiently reliable method of measuring metabolic activity of the soil microorganisms.

Urease and Phosphatase have been the most commonly studied soil enzymes along with dehydrogenase activity, which provide an index of total biological activity. Urease enzyme is responsible for the break-down of urea into carbon dioxide and ammonia. Due to this property it has got an applied importance in the N-economy of soil. Soil ureases are microbial products that can accumulate in cell free form in the soil because they are highly resistant to environmental degradation (McNaughton *et al.*, 1997). Urease acts as an intermediary enzyme in the transformation of organic nitrogen, while phosphatase provides an estimate of breakdown of organic phosphate compound and release of phosphate in the soil (Cosgrove, 1967). The term 'phosphatase' has been widely used to describe a group of enzymes that catalyze and hydrolyse both esters and anhydrides of phosphoric acid (Schmidt and Laksowski, 1961). The phosphatases are

involved in transformation of organic phosphorus compounds in soil. Their activity may play a significant role in release of phosphorus compounds (Rastin *et al.*, 1988). The phosphatase activity of soil has been shown to vary with the standing vegetation (Neal, 1973). Phosphatase has been found to be a good indicator of the organic matter in soil (Hattori, 1988). The estimation of the activity of these enzymes provides an assessment of three different microbe-mediated processes in soil and gives the most reliable measure of microbial activity.

Enzymes are markedly dependant on pH, ionic strength, inhibitors (or pollutants), moisture regimes, temperature and other environmental factors (Tabatabai and Dick, 1979; Frankenberger and Johanson, 1982; Dick and Tabatabai, 1983), but they may become stabilized in the soil by forming humus-enzyme or clay-enzyme complexes (Makboul and Ottow, 1979).

Soil microbes and plants roots are sources of extracellular enzymes, mainly through either secretion from living cells or from lysed cells (Burns, 1982). Once in the soil, enzyme may be protected from denaturation by being absorbed into organic or inorganic surfaces (Pang and Kolenko, 1986). In this absorbed state, extracellular enzymes developed stability to desiccation and heat and can remain active for several years (Miller and Dick, 1995). Under favourable conditions, microorganisms increase most of the enzyme activity. The effect of plants on soil enzymatic activity is due to changes in organic matter content and microbial populations, but is also formed by accumulated enzymes and by continuous release of extracellular and endocellular enzymes; all of which originate in the plant root. The overall enzyme activity of the soil

is derived from the activity of accumulated enzymes and from that of proliferating microorganisms (Burns, 1982; Tabatabai and Fu, 1992).

Changes in soil chemical and physical conditions influence microbial activity and population structure. On a primary basis, the natural environment provides long-term and seasonal fluctuations in temperature, moisture and plant growth to which microorganisms must respond (Schimel, 1995). Microorganisms are intimately associated with their physical and chemical environment and it is, therefore, conceivable that the temporal dynamics observed will partly reflect adaptation to environmental variables rather than competition between different components of the microbial biomass. Many aspects of soil microbial communities are affected by prevailing conditions with respect to substrate quality and stresses (Soren *et al.*, 2002).

Although changes in land growth and allocation in response to changing supplies of light, water, or nutrients are widely recognized (Mooney, 1972; Chapin III, 1980; Tilman, 1985; Ingestad and Agren 1988; Rastetter and Shaver, 1992), the nature of environmental limitation of soil microbial activity is less clearly understood. Energy supply is often viewed as the major factor limiting soil microbial activity (Richards, 1987; Tate III, 1995) because of the heterogeneity nature of decomposition. However, most soils contain 30-100 times more dead organic carbon than live microbial C (Jenkinson and Ladd, 1981). Consequently other factors such as substrate quality, nitrogen (N) availability, physical environment (e.g. temperature and moisture) and physical protection by clays have been invoked as additional controls over microbial processing of soil organic matter (Swift *et al.*, 1979; Richards, 1987; Tate III, 1995; Mary *et al.*, 1996).

The physico-chemical properties of soils can directly influence the structure, spatial distribution and activity of microbial populations and enzymes in soils, which are potential early indicators of soil health and quality (Schnurer *et al.*, 1985; Dick, 1994). Each of the organic and microbial fractions in soil has special influence on enzyme activity (McLaren, 1975; Skujins, 1976). Soil microbial activity contributes to the regulation of soil carbon storage and ecosystem productivity (Bauhus *et al.*, 1998).

Most of the forest lands on the earth are under tremendous pressure of developmental activities such as urbanization and industrialization which cause undesirable changes in various physico-chemical, biological and biochemical characteristics leading to the problem of soil degradation globally. It has been reported that many of the world's ecosystems are in various state of decline as evident by erosion, low productivity and poor water quality caused by forest clearing, intensive agricultural production and continuous use of soil resources that are not sustainable (Kennedy and Smith, 1995). This includes loss of structure and an increase in soil compaction (Vazquez *et al.*, 1993) commonly associated with soil organic matter reduction and ash formation (Prieto-Fernandez *et al.*, 1993). In the past, the disturbance of natural forest was less and was confined to areas of some country only. But at present, the condition has been changing due to increase in human population, environmental pollution, decline in soil quality, lower fertility level and encroachment of public to protected areas, environmental effects such as global warming. The problem of soil degradation and disturbance becomes the subject of environmental debate which attracts attention from the scientific communities of the world (Sehgal and Abrol, 1994) for successful restoration and rehabilitation of the disturbed areas.

It is widely accepted that each type of vegetation community harbors characteristic soil mycoflora population, particularly the assemblage of the more predominant species. However, there is no evidence that the microfungi form discrete communities; rather they constitute a continuum, the species composition gradually changing with differences in the vegetational cover and soil characteristics. Several studies have shown that there is a shift in the species composition of the soil mycoflora which parallels pioneer vegetational succession (Brown, 1958; Cooke and Lawrence, 1959; Pugh, 1963; Wohlrab *et al.*, 1963).

Soil physical conditions play an important role in determining the environment in which biological processes take place (DeVos *et al.*, 1994) while chemical characteristic determines maximum quality of a particular soil (Hassink, 1997). Nowadays, much attention is paid to the study of biological processes in soil because of the reason that nutrient transformation processes make soil a dynamic part of the biosphere with the vital role of soil microorganisms and invertebrates (DeVos *et al.*, 1994). The biological and biochemicals are important indicators which can sensitively respond to anthropogenic and environmental stresses on soil as dynamic system (Filip, 1998). Therefore, research studies in the last two decades have revealed that soil quality may be assessed using selected indicators related to soil microorganisms (Staddon *et al.*, 1998). The important microbiological parameters consist of population dynamics, diversity, soil respiration, microbial biomass carbon and nitrogen and enzyme activities. These parameters are considered as bio-indicators of soil quality and use as group of indices as they are quickly responsive and sensitive to changes occurring in the soil environment and could illustrate the effects of anthropogenic activities and other disturbances in soil (Dick, 1994; Turco *et*

al., 1994; Filip, 1998; Trasar-Cepeda *et al.*, 1998; Bending *et al.*, 2000; Palma *et al.*, 2000). Of the environmental factors which define the fundamental niche of terrestrial fungi, water is probably the most important (Cooke and Whipps, 1993). Water potential affects fungal "individual" through both impact on germination, radial growth, sporulation (e.g. Griffin, 1994; Dix and Webster, 1995; Bruchl and Kaiser, 1996) and mycelial cord development (Donnelly and Boddy, 1997). Water potential also affects the distribution and abundance of fungal population (Shameemullah *et al.*, 1971; Dowding and Widden, 1974; Bissett and Parkinson, 1979a and b; Widden, 1986a; Eastburn and Butler, 1988; Cooke and Whipps, 1993; Domsch *et al.*, 1993; Dix and Webster, 1995) and the interactions between fungal species (Eastburn and Butler, 1991; Marin *et al.*, 1998a and b).

The microbial communities are influenced by soil moisture and temperature (Campbell and Biederbeck, 1976), physical disturbance of soil (Doran, 1987) and interaction with soil fauna (Beare *et al.*, 1992).

Forest harvesting alters the amount of soil organic matter (Mattson and Smith, 1993), soil temperature, soil moisture and pH (Bormann and Likens, 1979) all of which affect microbial activity (Harvey *et al.*, 1980; Hendrickson *et al.*, 1985; Entry *et al.*, 1986).

The accumulation of organic nitrogenous compounds is more evident in forest soils (Pritchett and Fisher, 1987) and nitrogen mineralization is markedly retarded in soils like those developed over granite due to the abundance of organo-aluminium compounds, which present a high resistance to microbial mineralization (Gonzalez-Prieto *et al.*, 1991,

1996). It is, therefore, important to know the composition, variability and bioavailability of the nitrogenated substrate of soils.

Inputs of acidifying substances to forest ecosystems have increased in recent decades (Fowler *et al.*, 1999) and have led to increased acidification processes in forest soils on sensitive sites (Blaser *et al.*, 1999). In particular, inputs of N can alter forest soil chemistry significantly and in addition to acidification, nitrate can leach into the ground water resulting in relative shortages of nutrient elements (Magill *et al.*, 1997; Emmett, 1999; Rennenberg and Gessler, 1999).

Organic C and N are associated in soil organic matter and their pattern of distribution down the soil profiles studies are similar and closely followed the texture and bulk density data. The depth wise distributions of C and N underline the importance of sampling at depth to characterize soil organic storage and also indicate that C and N accumulation down the profile can be subject to mathematical analysis and modelling once the pattern of vertical distribution is established (Arrousays and Pelissier, 1994).

Understanding of the variation in fungal population in time and space has paramount importance due to its relevance in biodiversity and role of fungi in regulating population of other organisms and ecosystem processes. Therefore, research on microbial diversity and activity may provide some advance evidence of ecosystem degradation. Little is known of the importance of microbial diversity in the functioning of soils (Klein *et al.*, 1986; Pankhurst *et al.*, 1996; Giller *et al.*, 1997). The amount and diversity of microorganisms are extremely important for soil metabolic processes, because they act on the organic matter decomposition contributing for the soil fertility (Andrea *et al.*, 2000). It is important to determine optimum diversities of soil microbial populations of both

natural and agricultural systems for their sustainable management. In order to maximize the beneficial effects of microbial activity, there is a need for greater understanding of factors influencing microbial diversity and activity. The relationship between microbial diversity, microbial activity, plant quality, and ecosystem sustainability of disturbed and rangelands are still poorly understood.

4. *Phys* North eastern region of India is unique in many respects such as, rich floristic composition, high annual precipitation, undulated topography and varying types of forest ecosystems. This region experiences high rainfall of a monsoonic type which is followed by a dry winter and a brief summer. It is known to possess diverse forest types from Tropical Evergreen forests to Moist Alpine Shrub Forest and occupy 59.9 % of its geographical area, which is much higher than all India average of 28 %. The state of Meghalaya, in particular, is endowed with rich natural vegetation, which ranges from tropical type to sub-tropical type of vegetation or evergreen to mixed deciduous types of forests. It lies between 25° 00' and 26°15' N latitude and 89° 45' and 90°47' E longitude.

Most of the original vegetation cover of Meghalaya have been interfered with and modified by man to a great extent and if the process continues unabated, very soon the state will be devoid of true natural vegetation. The result is that the entire ecosystem of the region would undergo changes and create a chaotic condition.

The information on microbial diversity from northeast India is sporadic, as compared to other parts of India and no comprehensive study has so far been made to explore and conserve the microbial diversity of northeast India as a whole particularly in disturbed and undisturbed forests of Meghalaya. It was, therefore, proposed to investigate

the microbial diversity and their activities of disturbed and undisturbed forest soil of Meghalaya. The present studies were carried out under the following heads:

1. Diversity of soil microorganisms (fungi and bacteria)
2. Soil microbial biomass carbon (C_{mic})
3. Soil enzymes activities (dehydrogenase, urease and phosphatase)
4. Physico-chemical characteristics of the soil viz., temperature, moisture content, pH, organic carbon, total nitrogen, available phosphorus and exchangeable potassium