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MOSS RHIZOSPHERE AND ITS MICROFLORA

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Abstract

Microorganisms associated with the rhizosphere of mosses were isolated. From the initial isolation of 84 bacterial and 24 actinomycetes strains; 15 bacterial and 8 actinomycetes strains were selected and studied. The organisms included *Bacillus, Sporolactobacillus, Micrococcus, Paracoccus, Frankia, Intrasporangium, Micromonospora, Promicromonospora* and *Streptomyces*. *In vitro* experiments showed positive stimulation of some of the bacterial isolates by the extract of rhizoids. Major components of the tested rhizosphere soil *viz.* organic matter, total nitrogen, available phos-phorus and potassium were also measured and reported.

Introduction

Moss, in general grows in shady moist places (Parihar 1973) on a thin layer of rather immature and loose soil having coarse granules of minerals, decomposing materials, variable amount of moisture and a consortium of microorganisms. The moss rhizoids spread out through the heterogenous system of the soil not only to absorb nutrients but also to interact with the non living components as well as the living microflora. While rhizosphere microflora of higher plants have been studied and reported by many workers (Alexander 1977, Rovira 1965, Atlas and Bartha 1981) the moss rhizosphere remained rather unexplored.

Just as plant roots have a direct effect on the surrounding microbial popula-tions, microorganisms in the rhizosphere have also a marked influence on the growth of plants. In the absence of appropriate microbial populations in the rhizosphere, plant growth may be impared (Lochhed and Chase 1943). Microbial populations in the rhizosphere may benefit the plant in a variety of ways, including increased recycling and solubilization of mineral nutrients; synthesis of vitamins, amino acids, auxins and gibberellins which stimulate plant growth and antagonism with potential plant pathogens through competition and development of amensal relationships based on production of antibiotics. Rhizosphere microorganisms increased the availa-

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bility of phosphate through solubilization of minerals that would be otherwise unavailable to plants. Plants have been shown to exhibit higher phosphate uptake when associated with rhizosphere microorganisms than in sterile soils (Campbell *et al.* 1987). Bangladesh as a humid tropical country with plenty of organic materials available for decomposition, offer a paradise for different types of microorganisms. The population of moss rhizosphere plays an important role in humus formation by decomposing organic materials and thus prepare the stage for the next scenario i.e. the "fern stage" of on going plant succession.

Materials and Methods

Soil samples were collected from rhizosphere of mosses at different times from different places of Dhaka Metropolitan City and Netrokona town. The temperature of the rhizosphere soil sample as well as ambient temperature was recorded. The pH and moisture content of each soil sample were determined in the laboratory. After drying the soil samples were passed through a 0.5 mm sieve and organic matter content, total nitrogen, available phosphorus and available potassium were determined (Didar *et al.* 1991). Serial dilution technique was followed for the isolation of microorganisms from the rhizosphere soil samples and direct inoculation of rhizoids were also done using different culture media *viz.* nutrient agar medium for bacteria, Oat meal agar, starch casein agar, yeast and malt extract agar media were used for actinomycetes. The R.S. ratio for bacteria were also determined.

The selected isolates were screened by physiological and biochemical tests including IMViC, catalase test, casein hydrolysis, tyrosine decomposition, nitrate reduction. Growth responses were recorded at different environmental conditions viz. temperature, pH and NaCl concentration. Bergeys Manual of Systematic Bacteriology (Krieg and Holt 1984) (9th ed.) was followed for the identification of the selected strains.

Results and Discussion

Out of 15 selected bacterial strains 12 were found to be rods of various sizes and 3 strains were spherical. Among them 14 strains were gram positive and I was gram negative. The rod shaped bacteria were spore formers. All the tested stains closely resembled standard strains cited in the Bergeys Manual (9th ed.) except a few minor differences (Table 1).

The organisms of *Micrococcus varience* and *M. variance* were found to be related. The former strain differed from the standard strain in its inability to hydrolise starch and gelatin while the later strains differed from its inability to hydrolise casein and degrade tyrosine. It was observed that most of the strains grew well between 30 and 40°C. However, *Bacillus globisporus* and *B. sterothermofilus* survived even at 60°C. The collected samples had a pH range of 7.7 to 8.3 (Table 2). All the strains showed moderate growth within this range of pH.

Table 1. Biochemical characters of the selected organisms.

Organisms	Catalase activity	V.P. test	Anaerobic growth	Strach hydro-lysis	Casein hydro-lysis	Citrate utiliza-tion	Tyrosine decompo-sition	Nitrate reduc-tion
Bacillus alcalofillus	+	–	–	+	+	+	–	+
B. alvei	+	+	+	+	+	+	–	–
B. cereus	+	+	–	+	+	+	+	+
B. circulans	+	–	–	+	+	+	–	+
B. licheniformis	+	+	–	+	+	+	–	+
B. polymyxa	+	+	+	–	+	+	+	+
B. pantothenticus	+	–	+	+	+	–	–	+
B. globisporus	+	+	–	+	–	+	–	+
B. sphaericus	+	–	–	+	–	+	–	–
B. subtilis	+	+	–	+	–	+	–	–
B. stearothermophillus	+	–	–	+	+	+	+	–
Micrococcus variance	+	+	+	–	+	+	+	+
M. variance	+	+	+	+	–	+	–	+
Paracoccus sp.	+	–	–	–	–	+	–	–
Sporolactobacillus inolinus	–	+	–	+	+	–	–	–

Due to the availability of carbonaceous and nitrogenous materials from the sloughed off root hairs or exudates from healthy roots the rhizosphere is a highly favourable and very interesting niche for the growth of various microorganisms. It is well known that microorganisms, break down organic matter and release inorganic nutrients which may then be used for plant growth. Without the intervention of microorganisms, organic matter would accumulate and nutrients would gradually become unavailable to plants.

Nitrogen is particularly important to plants.for the formation of proteins. The immobilization of phosphate by microorganisms is well illustrated by Lochhead and Chase (1943). They showed that when the concentration of phosphate external to the root was low (below 0.5 ppm), microorganisms accumulated phosphate at the plants expense.

Table 2. Soil properties of rhizosphere region.

Sample No.	pH	Temperature (°F)		Moisture content	% of organic matter	% of total nitrogen	Available phosphos-rus	Available potassium
		Soil	Air					
1.	7.7	28	38	62	5.912	0.157	0.95	0.93
2.	7.8	32	35	63	4.902	0.109	1.90	0.87
3.	7.7	30	36	67	1.424	0.129	1.19	0.95
4.	7.9	28	36	61	6.204	0.081	1.90	1.02
5.	7.9	29	34	52	1.383	0.166	1.70	0.02
6.	8.3	31	36	54	1.424	0.089	1.07	0.97
7.	7.8	30	34	62	1.424	0.095	1.30	0.88
8.	8.1	30	34	60	6.790	0.157	1.30	0.87
9.	8.3	29	35	55	1.428	0.109	1.30	0.89
10.	8.3	30	36	58	6.790	0.159	1.90	0.95

Table 3. Total counts of bacteria in collected rhizosphere and non-rhizosphere soil and the R.S. ratio.

No. of samples	No. of bacterial colony per gram of soil		Approximate R. S. ratio
	Rhizosphere soil	Non-rhizosphere soil	
1.	106×10^6	53×10^6	2 : 1
2.	24×10^5	2×10^5	12 : 1
3.	12×10^6	6×10^6	2 : 1
4.	49×10^6	7×10^6	7 : 1
5.	39×10^4	3×10^4	13 : 1
6.	7×10^5	1×10^5	7 : 1
7.	9×10^3	3×10^3	3 : 1
8.	10×10^4	5×10^4	2 : 1
9.	12×10^5	3×10^5	4 : 1
10.	14×10^3	2×10^3	7 : 1

The effect of soil moisture has been irregularly studied. Campbell and Peterya (1987) showed that over a moisture range of 12 to 20% the R/S values did not vary (R/S = 9) but at 24.5% moisture the R/S value fell to 4.5. In this study it was found that the range of moisture content of the collected samples were 52 to 62% and the R. S. ratio were 2:1 to 13:1 (Table 3).

Rovira (1965) noted that there were increased rhizosphere populations in soils when the moisture content was 30% of the total moisture holding capacity than where the moisture content was 60%. Selected actinomycetes strains were identified up to their generic level on their morphological characters (Table 4) as described in the Bergey's Manual (Krieg and Halt 1984). (9th ed.).

Table 4. Morphological characters of the selected actinomycetes.

Organisms	Vegetative mycelium	Aerial mycelium and spore
Frankia	Vegetative mycelium with limited branching	Oval and lemon shaped vesicles are formed terminaly
Intrasporangium	Vegetative mycelium with limited branching	The mycelium bears intercalary vesicles that do not contain spores
Micromonospora	Hair-like thin vegetative mycelium	Spore borne on aerial mycelium singly
Promicromonospora	Hyaline hair like vegetative mycelium	Arrangement of spores in aerial mycelium in short chain
Streptomyces	Extensively branching thin vegetative mycelium	Arrangement of pores in aerial mycelium in long chain

It is well established that there is a prominent effect of microbes on soil structure. A good soil structure is essential to plant growth, as it promotes gas exchange, penetration of water and resistance to erosion. Evidently the best structure is achieved by the aggregation of soil particles into crumbs. The optimum size of the aggregates is between 1.0 and 5.0 mm in diameter (Gerretson 1943).

It was interesting to note that the microflora associated with the moss rhizosphere was somewhat different from the microflora reported from other rhizospheres. In case of moss rhizosphere, the percentage incidence of gram positive rods dominated among the bacterial population while according to Alexander (1977) the short gram nagative rods predominate the rhizosphere soil.

Evolution, the constant and dynamic phenomenon, is not only effected by selection pressures like nutrient supply, temperature and moisture but also by the important role of microbes. Lately when environmental scientists are showing increasing concern about pollution and scientists are working on the ability of mosses to clear our environment (Molla *et al.* 1983, Thomas 1986, Hertz *et al.* 1984), the moss rhizosphere should be examined more closely and carefully to have an insight of the pollutant-microbe interaction which could possibly help us in a better understanding of the problem of pollution management, beside biogeo-chemical transformation so closely related with plant succession. The purpose of the work described here was to study the composition and nature of the moss rhizosphere along with its microflora.

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