Available online at http://www.journalijdr.com

IJDR

International Journal of
DEVELOPMENT RESEARCH

ISSN: 2230-9926

International Journal of Development Research
Vol. 06, Issue, 07, pp.8770-8774, July, 2016

Full Length Research Article

MICROBIAL DIVERSITY OF ALLUVIAL BROWN SOIL FROM NAGAPATTINAM TALUK, NAGAPATTINAM DISTRICT

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ARTICLE INFO

Article History:

Received 28th April, 2016
Received in revised form
19th May, 2016
Accepted 14th June, 2016
Published online 31st July, 2016

Key Words:

Diversity,
Aspergillus niger,
Trichoderma viride.

ABSTRACT

The present study deals with the diversity, of soil fungi from different Agricultural fields and un cultivated soils of Nagapattinam Taluk Nagapatinam District, Tamil Nadu. Soil samples were collected at various seasons (Monsoon, Premonsoon, Summer, Postmonsoon) from three villages. The physico chemical parameters of the soils were identified. The physical parameter includes the analysis of pH, moisture content and temperature of the soils. The chemical parameter includes the macronutrients such as Carbon, Nitrogen, Pottasium, Phosphorus, calcium, magnesium and micronutrients such as Zinc, Iron, manganese and copper present were analysed crop land soils of three different villages. Totally 17 different species of soil bacteria were identified. Among the bacterial species were identified, *Staphylococcus spp, E.coli, Bacillus subtilis* and *Streptococcus spp* were dominant bacteria in the collected soil samples. The chemical parameters and microbial population of rhizosphere soils of crop plant have suggested as future course work. Totally 16 different species of soil fungi were observed from the soil samples collected from Nagapattinam, Sikkal and Vadagudi villages. Among the fungal species identified, *Aspergillus niger, Aspergillus terrus*, and *Trichoderma viride* were predominant in all the soil samples.

INTRODUCTION

Soil is a most precious natural resource and contains the most diverse assemblages of living organisms. Indigenous microbial populations in soil are of fundamental importance for ecosystem functioning in both natural and managed agricultural soils (O'Donnell *et al*. 1994; Doran and Zeiss 2000) because of their involvement in such key processes as soil structure formation, organic matter decomposition, nutrient cycling and toxic removal (Van Elsas, 1997; Doran and Zeiss, 2000). The community of soil flora and fauna is influenced directly or indirectly by management practices, e.g. cultivation and the use and application of organic and inorganic fertilisers (Bloem *et al*. 1994; Matson *et al*., 1997). A growing number of studies show that organic farming leads to higher soil quality and more biological activity (microbial populations and microbial respiration rate) in soil than conventional farming (Droogers and Bouma, 1996; Mader *et al*. 2002; Girvan *et al.,* 2004).

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Microbial population size and community structure are sensitive to changes in chemical properties of the surrounding soil (Pansombat *et al.,* 1997; Tokuda and Hayatsu, 2002). Further, considerable evidence indicates that changes in the composition of a microbial community can be used to predict and dictate alteration in soil quality (Van Brugen and Semenov, 2000; Breure, 2005). Microbial communities, particularly bacteria and fungi constitute an essential component of biological characteristics in soil ecosystems. It has been estimated that 1.5 million fungal species are present in natural ecosystems, but only 5 –10% have been described formally (Hawksworth, 2001). Schmit and Mueller (2007) estimated that there is a minimum of 7, 12,000 fungal species worldwide. The actual number of fungi is still unknown; however, only 5-13 % of the total estimated global fungal species have been described (Wang *et al.* 2008). Research on fungal diversity provides a basis for estimating the functional role of fungi in ecosystems. Microbial diversity has received particular attention in environmental studies since 1960's, but its functional significance in ecological processes is still a subject of debate and analysis. The present study was undertaken to throw a light on the diversity and abundance of

fungal and bacterial species to reveal the characteristics distribution and diversity with special reference to fungi and bacteria. The physic- chemical parameters of such soil fertility status were determined by analysed the physical parameters includes the analysis of pH and moisture content of soil. The chemical such as macronutrients (Nitrogen, Phosporus, Magnesium, Calcium) and micronutrients (Iron, Copper, Zinc, Manganese) were analysed in Nagapattinam taluk of Nagapattinam District.

MATERIALS AND METHODS

Site description

Nagapattinam soil series comes under Agro climatic zone IV – Cauvery Delta zone. Coastal eco – Eastern coastal plain hot sub humid to semi – arid eco system with a growing period 90 – 210+ days. Nagapattinam taluk is one of the 8 taluks of Nagapattinam District of Tamilnadu that lies between Lat. 110 3'N; Long 790 50' E, surrounded by karaikal, Nannilum, Thiruvarur , kivelur and Bay of Bengal. It covers an extant of 31181 ha land area.

Sample Collection

Soil samples were collected from the three villages viz, Nagapattinam, Sikkal, Vadagudi at Nagapattinam Taluk, Nagapattiinam District – Tamil Nadu. The soil samples were taken during the four seasons in agricultural field and uncultivated soils. Samples were collected from 10 – 15 cm deep pits dug in the area to be sampled. The samples were collected in polythene bags. Soil from 8 – 10 pits was pooled together and mixed in the same polythene bag.

Soil physico-chemical properties

Collected samples were brought to the laboratory sieved through 2mm sieve at field moist conditions and determination of soil moisture content and pH was done. Air dried ground and sieved (0.25 mm) samples were used for the estimation of organic C, total N, available P and K content. Three replicate samples were used for each analysis. Moisture content (MC) was determined by weight loss after drying 10 g of soil at 105°C for 24 hours and expressed as percentage dry weight. Soil pH was measured in a 1:5 water suspension using a portable digital pH meter. Colorimetric method (Anderson and Ingram, 1993), micro Kjeldahl distillation and titration method (Jackson, 1973), Molybdenum blue method (Allen *et al.*, 1974) and the ammonium acetate flame photometry method (Jackson, 1973) were applied to estimate organic carbon (C_{org}), total nitrogen (N), available phosphorus (P) and exchangeable potassium (K) respectively. The soil samples were ground, passed through 2 mm sieve and analyzed for DTPA (Diethylene Triamine Penta Acetic acid) extractable micronutrients (Fe, Mn, Zn and Cu) as per method proposed by Lindsay and Norvell (1978) and the consentrations of Fe, Mn, Zn and Cu were determined using a Atomic Absorption Spectrophotometer.

Isolation Of Bacteria From Soil: Soil samples were taken from each container and subjected to serial dilution followed by pour plate method.

Pour Plate Method: Nutrient agar medium was used for pour plate method. Nutrient Agar Medium was sterilized at 121°c for 15 minutes. Petriplates were sterilized and labelled as control A, B, C and 1ml of sample from 10^{-3}, 10^{-5} and 10^{-7} dilution was transferred into the respective plates. Finally, the cooled medium was poured into the sample containing plates and incubated at 37°C for 24 hours and the colonies were counted.

Identification of bacterial isolates

The isolated species were identified using with some modifications also done by Nopparat *et al.* (2007) based on characters such as morphology, staining reactions, nutritional, cultural characteristics , physiology and biochemical test results for specific metabolic end products. Also following criteria based identification conformed viz., Gram staining, Motility Test, Starch hydrolysis, Gelatin hydrolysis, Lipid hydrolysis, Carbohydrate fermentation test, Urea hydrolysis test, Hydrogen Sulphide Production test, Indole production test, Methyl Red test, Voges-Proskaeur test, Citrate utilization test, Oxidase test and Catalase test (Dubey and Maheshwari, 2000).

Fungal population count

For fungal population analysis, serial dilution plate method (Johnson and Curl, 1972) was followed using Rose Bengal Agar medium (Martin, 1950) supplemented with streptomycin sulphate. The inoculated Petri plates were incubated in a sterile culture room at 25˚ ± 1˚C. Colony forming units (CFU) were estimated by counting the number of colonies after five days. Fungal colonies formed were calculated on per gram dry soil basis. Fungi were identified according to their macroscopic and microscopic features. Identification at the species level was carried out according to the morphological characters found principally in publications by Gillman (1957), Barnett and Hunter (1972), Domsch *et al.* (1980), Subramanian (1983), Ellis (1993) and Watanabe (1994). Pure cultures of fungi were maintained in test tubes slants containing Czapex Dox agar medium (Raper and Thom 1949) and preserved in deep freezer at 20˚C.

RESULTS AND DISCUSSION

Soil Physico- chemical properties

Nagapattinam taluk of Tamilnadu has deep and fertile soils. It can be grouped into two categories namely 1) alluvial soil and 2) lateritic soil. Alluvial soil is found to occur in the old delta region comprising the major portion of the zone lying in the northern part whereas, lateritic soil covers new delta region lying in the southern part of the district The alluvial soils are clayey in texture with 40 – 45 percent clay fraction (Cauvery Delta zone - Status paper: Proper Literature not found). In the present study, showed that the soils of Nagapattinam District were alkaline in nature. The maximum pH (7.85) was recorded at Nagapattinam, whereas minimum pH (7.06) was recorded at Nagapattinam soils. The present study also recorded average pH of the soil as 7.54 from three locations of Nagapattinam District. The maximum organic carbon (1, 84) was recorded at Nagapattinam, whereas minimum organic carbon (0.12) was recorded at Vadagudi soils.

Table 1. Physico- Chemical parameters of the soil

Name of the parameters	Monsoon			Post monsoon			Summer			Pre monsoon		
	S_1	S_2	S_3	S_1	S_2	S_3	S_1	S_2	S_3	S_1	S_2	S_3
pH	7.35	7.85	7.12	7.54	7.14	7.31	7.13	7.06	7.35	7.74	7.44	7.18
Moisture	40.2	42.1	40.1	40.2	41.1	40.7	52.70	52.2	40.6	35.7	38.5	40.0
Temperature	25	27	28	37	38	39	45	48	45	38	31	38
carbon(%)	0.96	1.6	1.84	0.32	0.48	0.9	0.36	0.50	0.73	0.12	0.29	0.90
Nitrogen(Kg/ac)	85.5	87.5	84.7	82.3	86.8	84.6	87.6	81.6	90.1	87.8	83.4	87.8
Potassium(kg/ac)	76.5	74.5	73.5	70.3	69.1	68.5	65.5	71.2	71.6	72.1	69.8	70.1
Phosphorus(kg/ac)	4.5	1.25	2.78	4.35	4.38	4.37	4.78	2.56	2.65	3.24	2.85	1.9
Magnesium(ppm)	10.5	10	9.7	8.6	8.2	8.5	9.5	9	8.8	9.8	9.2	9
Calcium(ppm)	3.7	4.2	4.5	3.5	3.7	4.0	3.6	3.8	3.9	4.1	3.6	3.8
Copper(ppm)	0.88	0.79	0.94	1.5	1.4	1.6	0.98	0.95	0.92	1.8	1.6	1.2
Iron(ppm)	2.44	2.52	2.56	3.3	3.1	3.4	4.7	4.6	4.5	2.5	2.37	2.46
Zinc(ppm)	0.78	0.75	0.67	0.87	0.81	1.4	1.8	0.92	2.5	0.7	1.8	1.3
Manganese(ppm)	2.5	2.2	2.7	2.1	2.5	2.4	3.1	3.2	1.1	1.3	1.5	1.8

Table 2. Isolated bacteria in Nagapattinam taluk

Organisms	Monsoon			Postmonsoon			Summer			Premonsoon		
	S_1	S_2	S_3	S_1	S_2	S_3	S_1	S_2	S_3	S_1	S_2	S_3
Staphyloccus spp	+		–	–	–	–	–	–	–	+	–	+
Streptococcus	+	–	–	–	–	–	–	–	–	–	–	+
Staphylococcus auicularis	+	–	–	–	–	–	–	–	–	–	–	–
Azotobacter spp	–	+	–	–	–	+	–	–	–	–	–	–
Azospirillum spp	–	+	–	–	–	–	+	–	–	–	–	–
E.coli	–	–	+	–	–	–	–	+	–	–	–	–
Enterobacter spp	–	–	+	–	+	–	–	–	–	–	–	–
Vibrio spp	–	–	+	–	–	–	–	–	–	–	–	+
Bacillus licheniformis	–	–	+	–	–	–	–	–	–	–	–	–
Bacillus subtilis	–	–	–	+	–	–	–	–	+	+	–	–
Micrococcus	–	–	–	+	–	–	–	–	–	–	–	–
P.putida	–	–	–	+	–	–	–	–	–	–	–	–
Rhizobium spp	–	–	–	+	–	–	–	–	–	–	–	–
Rhizobium meliloti	–	–	+	–	+	–	–	–	–	–	–	–
Proteus spp	–	+	+	–	+	–	+	–	–	+	–	+
S.epidermis	–	–	–	–	–	+	–	–	–	–	–	–
Pseudomonas aeroginosa	–	–	–	–	–	–	+	–	–	–	–	–
B. cereus	–	–	–	–	–	–	–	+	–	–	–	–
E. aerogens	–	–	–	–	–	–	–	–	+	–	–	–
Rhizobium meliloti	–	–	–	–	–	–	–	–	–	+	–	–

Table 3. Isolated fungai in Nagapattinam taluk

Organisms	Monsoon			Postmonsoon			Summer			Premonsoon		
	S_1	S_2	S_3	S_1	S_2	S_3	S_1	S_2	S_3	S_1	S_2	S_3
Aspergillus granulates	+	–	–	–	–	–	–	–	–	+	–	+
Aspergillus flavus	–	–	–	–	–	–	–	–	–	–	–	+
Aspergillus niger	–	+	–	+	–	–	–	–	–	–	–	–
Saccharomyces spp	–	+	–	–	–	+	–	–	–	–	–	–
Rhizopus oryzae	–	+	–	–	+	–	+	–	+	–	–	–
Verticillium spp	–	–	+	–	+	–	–	–	–	–	+	–
Penicillium lividum	–	–	+	–	–	–	–	–	–	–	–	+
Alternaria spp	–	–	–	–	+	–	+	–	+	–	+	–
Rhizopus stolonifer	–	–	–	+	–	–	–	–	–	–	–	–
Penicillium janthinellum	–	–	–	–	–	+	–	–	–	–	–	–
Penicilliumturbatum	–	–	–	+	–	–	–	–	–	+	–	–
Torulaallii	–	–	–	–	–	–	–	–	+	–	–	–
Trichoderma lignorum	–	–	–	–	+	–	–	–	–	+	–	–
Gliocladium virens	–	–	–	–	–	–	–	–	–	–	–	+

The present study also recorded average organic carbon of the soil as 0.50 from three locations of Nagapattinam District. The maximum Nitrogen (90.1 kg/ac) was recorded at Nagapattinam, whereas minimum Nitrogen (81.6kg/ac) was recorded at soils. The present study also recorded average Nitrogen of the soil as 85.5 kg/ac from three locations of Nagapattinam District. The maximum Potassium (76.5 kg/ac) was recorded at Nagappattinam, whereas minimum Pottasium (69.1 kg/ac) was recorded at Vadagudi soils.

The present study also recorded average Potassium of the soil as 68.5 kg/ac from three locations of Nagappattinam District. The maximum Phosphorus (4.78 kg/ac) was recorded at Nagapattinam, whereas minimum Phosphorus (1.9kg/ac) was recorded at Vadagudi soils. The present study also recorded average Phosphorus of the soil as 2.56 kg/ac from three locations of Nagapattinam District. The maximum Magnesium (10.5ppm) was recorded at Nagapattinam, whereas minimum Magnesium (8.2 ppm) was recorded at Vadagudi soils. The

present study also recorded average Magnesium of the soil as 8.8 ppm from three locations of Nagapattinam District. The maximum Calcium (4.5ppm) was recorded at Nagapattinam, whereas minimum Calcium (3.5 ppm) was recorded at Vadagudi soils. The present study also recorded average Calcium of the soil as 3.8 ppm from three locations of Nagapattinam District. Similar type of work has been reported by many workers physic- chemical properties of the rhizosphere soil of the *Curcuma longa* L. was analysed by Sumathi *et al*., (2008); rehabited secondary forests soil physic-chemical properties by Akbar *et al*., (2010). In addition, most of the rhizosphere soils of the present study appeared blackish brown in colour and texture was alluvial brown soil in nature. The lime content of the soil was present only in Nadapattinamtaluk. The heavy metals (ppm) also present in all the three sampling stations (Table 1). The crop soils were also analysed for its micronutrients such as available Zn, Cu, I and Mg. The maximum content available in Zn (2.5ppm) , Cu(1.8 ppm), I (4.7ppm) and Mg (3.2 ppm) was recorded in soils of the Nagapattinam, Sikkal, and Vadagudi respectively (Table 1). Similarly Praveen *et al*., (1993) studied micronutrient status of some agriculturally important soil series of the Northwest Frontier Province, Pakistan and their relationship with various physic chemical properties for 30 soil series. Most sandy soils (coarse texture) are deficient micronutrients. Clay soils (fine texture) are not comparatively to low plant available micronutrients. Chabra *et al*., (1996) studied that available Mg and I decreased with soil pH and available Cu increased with clay and organic carbon content. Hence, the correlation co – efficient analysis between the soil physico chemical parameters and microbial population of rhizosphere soils of cop plant have suggested as future course work.

Bacterial isolates

Totally 17 different species of soil bacteria were observed from soil samples. The bacterial species are identified their morphological character and Bergey's manual of determinative bacteriology. The predominant bacterial species are *Staphylococcus spp, E.coli, Bacillus subtilis, Streptococcus spp.* Generally fungi and bacteria found in deep layer or slow growing due to unavailability of mineral nutrients and compaction of soil along depth (Dkhar and Mishra, 1992).One gram of the soil sample thus prepared was added to 10 ml sterilized distilled water and serial dilutions were made (Johnson, 1957).

Fungal isolates

Totally 16 different species of soil fungi where observed from the soil samples collected from three different villages. The colonies showed a characteristic colour of black, green, white and brown and they were confirmed by identifying their morphological characters and by Ellis Manual. The predominant microbes are *Aspergillus niger, Aspergillus terrus, Trichoderma viride.* The fungal species of *Aspergillus* have been reported that they were the most tolerant one to the adverse conditions in the laboratory (Venkataraman and Rajyalakshmi, 1971) and species of *Aspergillus* and *Penicillium* were tolerant to wide range of environmental conditions.

Conclusion

Soil fertility is important factor, which determines the growth of plant. Soil fertility is determined by the presence or absence of nutrients i.e., macro and micro nutrients. A basic soil test will provide information on soil texture, organic matter, pH, buffer index, phosphorus, potassium and nitrate. Most of the soil tests will give a range for the nutrients, such as low range, medium and high, to give an indication of relative amounts of nutrients in the soil. When a nutrient is in the low range, it means that added inputs of that nutrient will likely show a strong growth response in the next crop planted. A conventional soil laboratory will provide fertilizer recommendations based on the next crop. On the whole , the soil will influenced by the annual crop rotation practice, quality of water used for irrigation and application of chemical fertilizer and so on.

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