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MICROBIOLOGICAL APPROACH AND PHYSIOCHEMICAL ANALYSIS OF PESTICIDES POLLUTED SOIL FROM CUDDALORE DISTRICT

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ABSTRACT

Microbes are an important group of organisms and they thrive in wide variety of soil habitats. In modern agricultural practices variety of synthetic chemical compounds including pesticides, e.g., zoocides, fungicides, herbicides and others are used for crop protection. Microorganisms are first to respond to those synthetic compounds let in to the environment, the natural aquifers through the runoff water. These changes will affect the process of primary production, nutrient circulation and decomposition of matter, as in which bacteria serve as an important function. The present investigation suggests that the \ influence of physiochemical analysis from ten different areas of Cuddalore district for one year of 2014-2015. The soil samples from Cuddalore distict of Thanjavur were collected one time and analyzed for parameters such as pH, electrical conductivity, organic carbon, available nitrogen, phosphorus, potassium, zinc, copper, iron, manganese, calcium, magnesium, sodium and potassium. Maximum pH was 8.26 in the Panruti village (site 7) and minimum 7.26 in T. mannalur (site 3) respectively. The higher concentration of carbon was also from Panruti village (site 7) i.e., 0.58 percentage when compared to other site areas. The bacterial count showed that Streptococcus sp. Proteus in constants (80%) had the highest count followed by *Streptococcus* sp, *Nitrosomonas, B.subtilis* (77%). Due to this organic carbon is considered as a recognizing factor for

microbial population especially for bacteria including the nitrifying bacteria which were isolated and identified in each samples.

KEYWORDS: Pesticide, physiochemical analysis, soil bacteria.

INTRODUCTION

Nitrogen is a major nutrient for the growth of crop plants, which is being applied as chemical fertilizer in the agricultural practices. Besides this considerable quantity of nitrogen is generated within the ecosystem by the involvement of microorganism present there. But the effects of applications of pesticides on these beneficial micro-organisms are unknown. Physiochemical parameters of the environment in the nature and distribution of flora and fauna of a particular ecosystem were determined. Some of the parameters in the total ecosystem affect the entire ecosystem that leads to toxicity of the agricultural area. Pesticides are broadly defined as substance or mixtures intended to prevent, destroy, repel or mitigate any pest, including insects, rodents and weeds. They include pesticide but also herbicide, fungicides, disinfectants and growth regulators. These agrochemicals have become inevitable in intensive agriculture to improve production and protect stored crops. Although pesticides use benefits the human population, health risks have been suggested in humans that are occupationally and environmentally exposed to these agrochemicals. The era of modern synthetic pesticides largely dates from 1939 when the insecticidal properties of 1, 1, 1-trichloro-2, 2-bis (p-chlorophenyl) ethane (DDT) were discovered. Unlike naturally occurring organic pesticides properties such as DDT are extremely resistant to biodegradable by native micro flora. In most cases, the persistence can be explained by the chemical structure and by the degree of water solubility. In addition, some of these pesticides tend to accumulate in organisms at different trophic levels of the food chain. Chlorinated organic pesticides are one of the major groups of toxic chemicals responsible for environmental contamination and an important potential risk to human health.

Must attention has been directed towards the use of microorganisms for bioremediation of all industries, including pesticides. Microbial mediated decomposition, often through eco metabolism is the major, and sometimes the only mechanism for the permanent removal are modification of pesticides in soils, in contrast fungi, bacteria have been extensively studied and exploited for use in the degradation of pesticides. This is primarily because of their ease of culture, more rapid growth rates and amenity to microorganisms, evaluate the enriched microbial isolates to in situ degrade pesticides in the complex environment of soils.

MATERIAL AND METHOD

Sample collection

Soil sample were collected with clean sample materials from the adjoining soils of rhizosphere. Ten area soil samples were collected from leguminous plants. The experimental site was at cuddalore district of tamilnadu that is located at 23°c-15'.00"N to 23°c-45'-00"N and 88°-45'-00" E to 89°-45'-00"E. Soil samples were taken from 0-15 cm depth in rhizosphere along the diagonals. The monsoon data's were collected from meteorology departments from cuddalore district. Physiochemical characterization from the soil samples were carried out using standard method (APHA 1995).The soil was characterized such as pH, electrical conductivity (EC), organic carbon, available nitrogen, phosphorus, potassium, zinc, copper, iron, manganese, sodium, potassium and total number of colonies were determined from ten site area of cuddalore district.

Isolation of nitrifying bacteria from soil

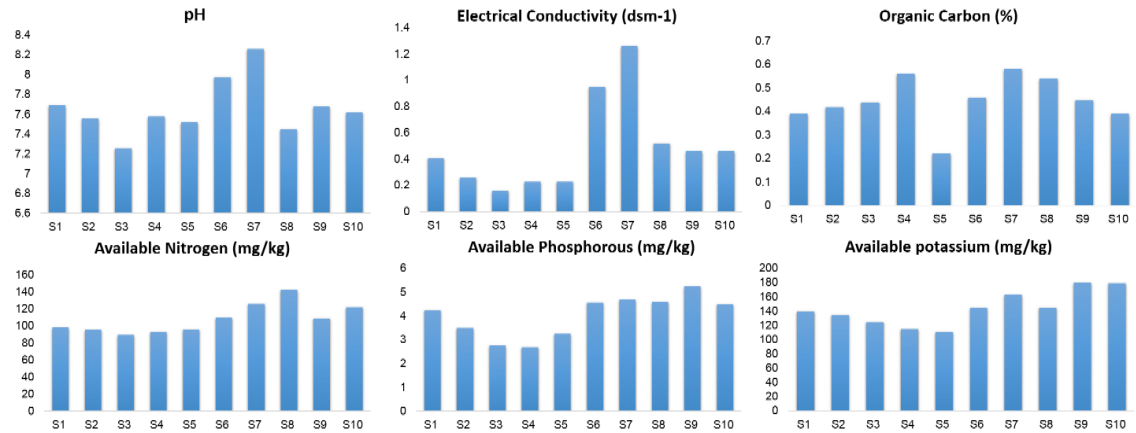
The nitrogen fixing bacteria was isolated directly from the soil sample using yeast extract Mannitol selective culture media (YEM). Isolation of nitrogen fixing bacteria from ten different soil samples and bacteria were confirmed by bromothymol blue (BTB) that was used as an indicator in order to detect the multiplication of the nitrogen fixing bacteria. The microbial population from the soil and larger in number of colonies were found in the soil.

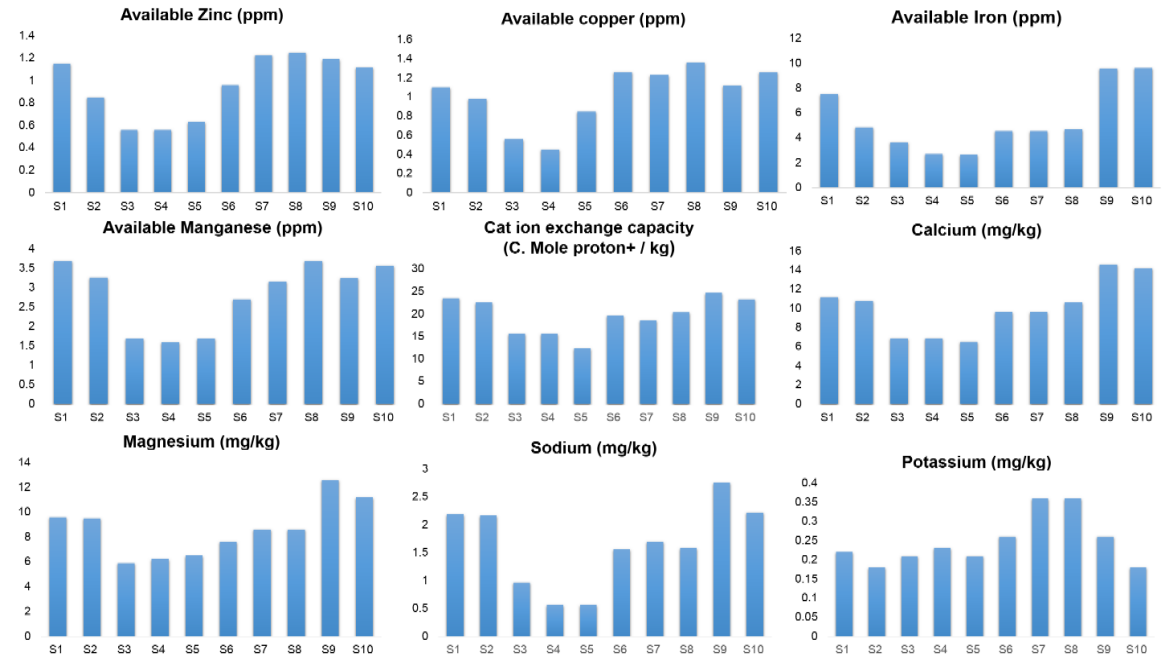
RESULT AND DISCUSSION

Dubey, *et al*., (1982) reported that the seeds inoculated with rhizobium strains have increased the nitrogen content of root, shoot, grain and straw over uninoculated seeds *Vigna radiata*. Jarak, *et al*. (1989) the minimum amount of nitrate content in the month of august 2013 as (4.3 µg/L) and maximum nitrate content in the month of October 2013 as (9.3µg/L) were observed. These findings agreed with the observation made by Qasim (1980), Murugan and Ayyakannu (1993) and Jagadeesan (1986) in cochin backwater and uppanar back waters of coleroon estuary and total phosphorous concentration varied between 1.8 and 4.3µg/g. minimum concentration (1.8µg/g) in march, 2013, maximum concentration (4.3 µg/g) in October ,2013 were recorded in phosphate concentration increased from summer to post-monsoon period and this increase to may be due to liberation of inorganic phosphate (under high oxygen concentration) from freshwater in flow in the river (Chandran and Ramamoorthy 1984, Das *et al*., 1997).

Generally the growth of physiochemical and biological parameters as similar reports have been published from Maharashtra coast by Dhargalkar *et al*.,(2001) nutrient studied that the nitrate and inorganic phosphate were abundant by during monsoon due to moonsonal flow of fresh water and land runoff and decrease nutrient level during summer and post-monsoon was observed. In the soil where herbicide was applied, soil and increase of bacterial number were observed in most experimental combination. The only exception was yellow lupine, under which a fall in the mean number of bacteria was found in the presence of herbicide as it compared to the control soil, it was reflected in the mean number of microorganisms from almost all analyzed soils where herbicides were applied.

In the present investigation suggests that the physiochemical parameters were analysed from the soil sample of various places of Cuddalore district. The parameters such as pH, electrical conductivity , organic carbon, available nitrogen, phosphorus, potassium, zinc, copper, iron, manganese, cation exchange capacity ,calcium, magnesium, sodium, potassium and total number of colonies with 7.69, 0.41 dsm-1, 0.39%, 9.35 mg/kg 1.15ppm, 1.10ppm, 7.52ppm, 3.68ppm,23.5 c.mole proton/ kg, 11.2 mg/kg, 9.6 mg/kg, 2.18mg/kg, 0.22mg/kg, 0.22mg/kg and 70 colonies recorded in koolapadi site respectively. The Veiyalur samples was 7.56, 0.26, dsm-1 0.42%, 95.6 mg/kg, 3.50 mg/kg, 1.35 mg/kg, 0.85mg/ppm, 0.98ppm, 4.85ppm, 3.26ppm, 22.5c.mole proton+/kg, 10.8mg/kg, 9.5mg/kg, 2.16 mg/kg, and total number of 72 colonies was recorded from the second site. The maximum physiochemical parameters of Panrutti were analyzed such as pH 8.26, EC-1.26, organic carbon 0.58%, available nitrogen 126.3 mg/kg, phosphorus 4.69 mg/kg, potassium 163 mg/kg, zinc 1.23 ppm, copper 1.23 ppm, iron 4.56 ppm, manganese 3.15 ppm, cation exchange capacity 18.6 C.mole proton/kg, calcium 9.6 mg/kg, magnesium 8.6 mg/kg, sodium 1.69 mg/kg, potassium 0.36 mg/kg and total number of colonies (63) also highly represented in the Panruti sample site (Fig 1).

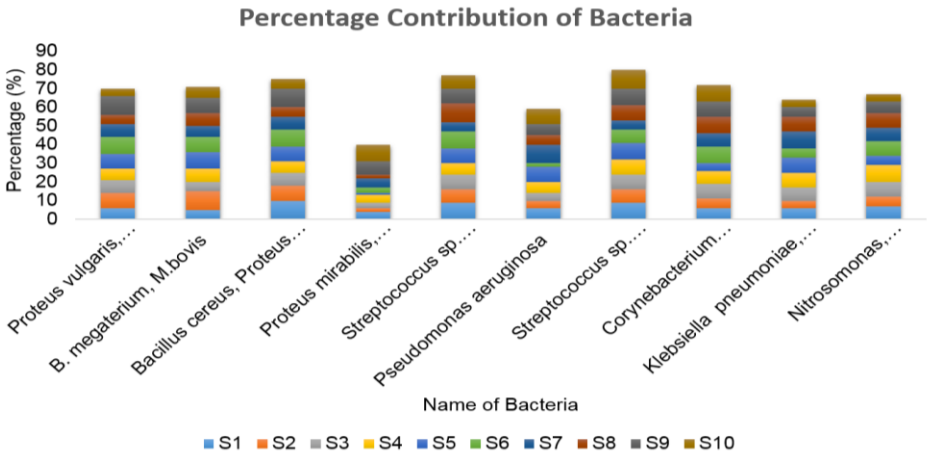




S1-Koolapadi, S3-T.mannalur, S5-Old town, S7-Panruti, S9-Kattumannarkoil
S2-Veiyalur, S4-Tittakudi, S6-Semmandalam, S8-Kurinjipadi, S10-Virudhachalam

Fig. 1: Analysis of physico chemical parameters of pesticide polluted soil sample.

Sudbakar *et al*., (2000) found that there was a variable effects of pesticides on the growth of nitrogen fixing bacteria. The results showed that fungicides, carbendazim reduced the bacterial population at all concentrations, but dimethoate and wettable sulphur stimulated and these concentrations are supported by Gallori *et al*. (1991), Revellin *et al*.,(1993), Taiwo and Oso (1997) and Dunfield *et al*.,(2000) who reported that bacterial growth inhibition due to agrochemical, and they contain similar active ingredient that reduce the number of nitrogen fixing bacteria.



S1-Koolapadi, S3-T.Mannalur, S5-Old town, S7-Panruti, S9-Kattumannarkoil
S2-Veiyalur, S4-Tittakudi, S6-Semmandalam, S8-Kurinjipadi, S10-Virudhachalam

Fig. 2: Isolation of bacteria from different regions of Cuddalore soil samples.

Similar trend was reported by Martensson(1992) who observed that the fungicide treatment decreased the number of viable nitrogen fixing bacteria. dimethoate decreased the growth of rhizobium population (Castro *et al*., 1997). The fixing of nitrogen was parallel to the population of both nitrogen fixing bacteria in soil treated with pesticides, Martinez *et al.* (1992) and Pozo *et al.* (1995) found that organo phosphorous insecticides profenofos and chloropyrifos reduced the number of aerobic nitrogen fixers and significantly decreased nitrogen fixation. Mubeen (2004) found negative effect of fungicides on nitrogen fixation. Primerpair nifH1, nifH2, nifH3, nifV, nifU, FV genes of *A.chroococcum*, and nifH, nifK, nifD, nifM, and FV genes in A. vinelandii were used to amplified nif genes of *A.chrococcum* and *A.vinelandii* from three pot soil samples. In my present study that the Population dynamics of soil nitrifying bacteria were isolated and identified was done by using standard manual of Bergy's manual of determination bacteriology 12th edition was followed. The bacterial isolates such as *Proteus vulgaris, Pseudomonas flourescens* was isolated from koolapadi area soil sample, Veiyalur village soil was represented in *Bacillus megaterium, Micrococcus bovis* was observed, the *Bacillus cereus*, *proteus vulgaris*, was isolated from T.mannalur village. In the village of Tittakudi soil was *Proteus mirabilis*, *Nitrobacter* sp, *B. subtilis* were recorded. The Kurijipadi soil was *Corynebacterium xerosis*, and *Klebsiella pneumoniae*, isolated. The Kattumannarkoil soil was *Klebsiella pneumoniae, Azotobacter* sp, resulted. The *Nitrosomonas* sp and Streptococcus sp, was isolated from Virudachalam soil polluted with pesticide applied by former from the cultivated leguminous plants (Table-2). It is concluded from this study that the pesticides have differential effect on the growth of nitrogen fixing bacteria with reference to physiochemical character were determined.

Table 1 Analysis of physico chemical parameters of pesticide polluted soil sample

Name of the Parameters	**S1**	**S2**	**S3**	**S4**	**S5**	**S6**	**S7**	**S8**	**S9**	**S10**
pH	7.69	7.56	7.26	7.58	7.52	7.98	8.26	7.45	7.68	7.62
Electrical conductivity (dsm^{-1})	0.41	0.26	0.16	0.23	0.23	0.95	1.26	0.52	0.46	0.46
Organic carbon (%)	0.39	0.42	0.44	0.56	0.22	0.46	0.58	0.54	0.45	0.39
Available nitrogen (mg/kg)	98.5	95.6	89.6	93.5	96.3	110.2	126.3	142.6	109.2	122.3
Available Phosphorous (mg/kg)	4.25	3.50	2.75	2.69	3.25	4.56	4.69	4.58	5.25	4.50
Available potassium (mg/kg)	140	135	125	115	111	145	163	145	180	179
Available zinc (ppm)	1.15	0.85	0.56	0.56	0.63	0.96	1.23	1.25	1.19	1.12
Available copper (ppm)	1.10	0.98	0.56	0.45	0.85	1.26	1.23	1.36	1.12	1.26
Available iron (ppm)	7.52	4.85	3.65	2.69	2.65	4.56	4.56	4.69	9.58	9.63
Available manganese (ppm)	3.68	3.26	1.69	1.59	1.69	2.69	3.15	3.68	3.25	3.56
Cat ion exchange capacity (C. Mole $proton^{+}$ / kg)	23.5	22.5	15.6	15.6	12.3	19.6	18.6	20.3	24.6	23.2
Calcium (mg/kg)	11.2	10.8	6.9	6.9	6.5	9.6	9.6	10.6	14.6	14.2
Magnesium (mg/kg)	9.6	9.5	5.9	6.2	6.5	7.6	8.6	8.6	12.6	11.2

Sodium (mg/kg)	2.18	2.16	0.96	0.56	0.56	1.56	1.69	1.58	2.75	2.22
Potassium (mg/kg)	0.22	0.18	0.21	0.23	0.21	0.26	0.36	0.36	0.26	0.18

S1-Koolapadi, S2-Veiyalur, S3-T.mannalur, S4-Tittakudi, S5-Old town, S6-Semmandalam, S7-Panruti, S8-Kurinjipadi, S9-Kattumannarkoil, S10-Virudhachalam.

Table 2 Isolation of bacteria from different regions of cuddalore soil samples

S.No	Name of the bacteria	S1	S2	S3	S4	S5	S6	S7	S8	S9	S10	Total number of colonies
1	*Proteus vulgaris, P.fluorescens*	6	8	7	6	8	9	7	5	10	4	70
2	*B. megaterium, M.bovis*	5	10	5	7	9	8	6	7	8	6	71
3	*Bacillus cereus, Proteus vulgaris*	10	8	7	6	8	9	7	5	10	5	75
4	*Proteus mirabilis, Nitrobacter sp , Nitrococcus sp*	4	2	3	4	1	3	5	2	7	9	40
5	*Streptococcus sp. Nitrosomonas, B. subtilis*	9	7	8	6	8	9	5	10	8	7	77
6	*Pseudomonas aeruginosa*	6	4	4	6	8	2	10	5	6	8	59
7	*Streptococcus sp. proteus inconstans*	9	7	8	8	9	7	5	8	9	10	80
8	*Corynebacterium xerosis, Neisseria mucosa, E. aerogens*	6	5	8	7	4	9	7	9	8	9	72
9	*Klebsiella pneumoniae, Azotobacter sp.*	6	4	7	8	8	5	9	8	5	4	64
10	*Nitrosomonas, Streptococcus sp.*	7	5	8	9	5	8	7	8	6	4	67
	Total number of colonies	68	60	65	67	68	69	68	67	76	66	674

S1-Koolapadi, S2-Veiyalur, S3-T.mannalur, S4-Tittakudi, S5-Old town, S6-Semmandalam, S7-Panruti, S8-Kurinjipadi, S9-Kattumannarkoil, S10-Virudhachalam.

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