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Research Article

FUNGICIDES EFFECT ON SOIL MICRO FLORA IN TEKKALI MANDAL, SRIKAKULAM (DIST.)

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ABSTRACT

In this study was examined the response of microbial populations in soil after the application of several fungicides. The soil samples were collected from experimental plots field at village tekkali in and around the circumstance, localized at 20 kilometres, on January 2015-November 2015. The soil was collected from upper 10 cm of the soil profile. In the laboratory plant material and soil macro fauna were removed and the soil samples were sieved (<2mm) and mixed. Each soil sample was treated with one of the following fungicides at recommended dosages: Copper Oxy Chloride (COC) at the rate of 0,0025g/, Carbendzim at the rate of 0,01g/ and Cmmodity Mancozeb at the rate of 0,01 ml. Controls, consisting of soil only, were included within all tests. The numbers of total microorganisms were determined using the dilution method. By these fungicides effect on total viable fungi were investigated in laboratory experiments, these results compared with my previous studies, fungal colonies were dropped, graphically and statistically analyzed.
Keywords: soil fungi, effects of fungicides, common fungicides.

INTRODUCTION

Fungicides are used extensively in modern agriculture for the control of fungal pathogens. The chemical alters the number and activity of micro organisms and these effects biochemical process and fertility of soil[1] .Fungicides are often applied several times during one crop season and apart always reaches the soil. Fungicides treated soil harboured less population of fungi in comparison to control. Micro-organisms are of assistance in increasing the soil fertility and plant growth as they are concerned in several biochemical conversions and mineralization actions in soils. The soil micro-organisms ensure the permanence of element cycles in nature due to the array of their metabolic behaviour. The consequence of their activities is not only mineralization of organic compounds but also the changes of mineral compounds, which have an immense impact on the development of the plants. Micro fungi play an imperative role in nutrient cycling by regulating soil biological activity, these fungi grow in different pH, moisture, temperature and aeration and nutrient availability. Hence fungi are major components of the soil micro-biota characteristically constituting more of the soil biomass than bacteria, depending on soil depth and nutrient conditions[2]. Fungi benefit most plants by suppressing plant root diseases and promoting healthier plant by attacking plant pathogens with fungal enzymes[3]. The pace at which organic matter is decomposed by the microbes is interrelated to the chemical composition of the substrate as well as environmental conditions. Fungi can efficiently grope around for N and P better than the plant root hairs and wholly increase the plant root nutrient extraction efficiency. Fungi get dominated over other micro-organisms by secreting enzymes. They also have the ability to survive and propagate in various extreme conditions of the environment. They also use antagonisms to

reduce competition by producing antibodies, which suppress other micro-organisms from growing[3]. By the use of large quantity of fungicide can also damage the beneficial micro organisms.

STUDY SITE AND LOCATION

Tekkali is a town and a mandal in Srikakulam District in the state of Andhra Pradesh in India. It lies on the coast of Bay of Bengal and is located at 18.6167^{0}N 83.2333^{0}E. The temperature ranges from 18-42^{0}C. Deltaic Alluvial soils, Red Sandy soils and Latirate soils are the major soil types existing. It receives total rainfall of 1162mm with 60% of annual rainfall (705mm normal) during South-West Monsoon season from June to September, and North-East Monsoon provides 277 mm (23.8%) between October and December months. Farmers take up double cropping of paddy with monsoon rainfall and a second crop of sunflower or ground-nut with North-East monsoon rainfall and supplemental irrigation in rabi season.

Effect of Fungicides on growth of fungus:

In this study three fungicides were used list given Table 1. The effect of Fungicides on growth of fungi (A. niger) studied on agar plates methods

MATERIALS AND METHODS

Soil Sample Collection:

Soil samples used in this investigation were collected randomly in demonstration farm with auger from 0 – 20 depth[4]. Three sample points were selected designated in and around the Tekkali mandal srikakulam district (a, b and c) and soil samples collected were used as control without Fungicide application after which Fungicides are applied recommended dosages in interval. After two weeks of Fungicide application, four other soil samples were collected making a total of five (S1- S5) samples where fungi were isolated. The samples were taken to the laboratory and stored at temperature of 4^{0}C until the analyses were conducted and completed.

In this study used fungicides are mentioned in table no.1. Erlenmeyer flasks (250 ml) containing 100ml of Potato Dextrose (PDA) Agra and autoclaved at 121°C for 20min. In one set of experiment, actively grown four days A. niger (10^6 cell per ml) was aseptically inoculated with PDA plates were allowed to solidify. The PDA were taken by using a sterile cork borer and Fungicide solution were placed on the agar diffusion method (8.0 mm diameter) and the inhibitory zones of restricted growth of A. niger were evaluated after 3 to 5 days. Without Fungicide amended PDA plates were taken as control.

In another set different concentration (viz., 1ppm and 0.5ml) of these three fungicides were amended aseptically in fungicide well as in PDA medium and vigorously shaken. The PDA plates were allowed to solidify. 4 –days old agar blocks with mycelia were inoculated into funicides amended PDA and also in control plate (without fungicides amended) respectively (Table-3). All the Petri plates were incubated at room temperature at 28 ± 2°C for 7 days. Each experiment were carried out in triplicates and replicated twice. The growth patterns of the respective fungus were observed in plates.

ANALYTICAL PROCEDURE

Microbiological analyses of the soil samples were made as per the following procedures:

For the enumeration of the micro-fungal population dilution plate count technique (DPCT) (Waksman, 1922) was followed. The primary suspension of the soil was prepared from 1 gram of soil which was diluted up to 10^{-9} times using sterile water as diluting fluid. For microfungal population, 1ml of aliquots from 10^{-4} diluted suspension was transferred to Petri dishes and to it 25 ml of molten PDA medium was poured. Petri dishes were incubated at room temperature till the development of fungal colonies. Colonies developed on the Petri dishes were identified following Gilman (1957), Barnett and Hunter (1973).

Statistical Analysis:

Mean:

Mean = Sum of X values / N (Number of values)

Standard Deviation:

$$S=\sqrt{\frac{\sum(X-M)^2}{n-1}}$$

Population Standard Deviation:

$$S=\sqrt{\frac{\sum(X-M)^2}{n}}$$

Variance = S^2

RESULTS

The changes in activity of soil enzymes characterized the effect of fungicides on microbes in the soil. Therefore fungicides treated soils have considerably lower enzymatic

Table 1: List of the Fungicides

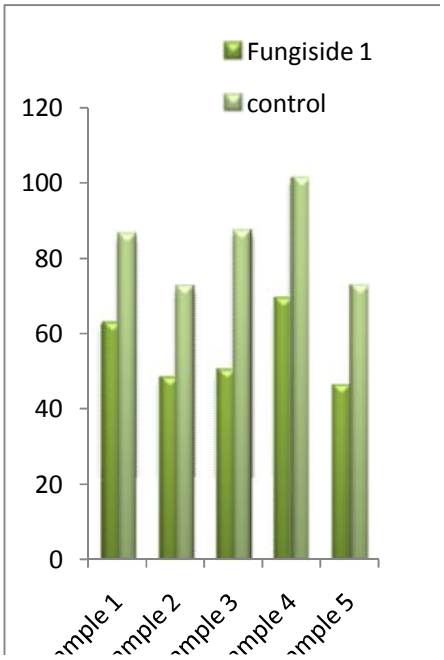
Sample Code	Fungicides Name	Concentration
F1	*Carbendzim	1ppm
F2	*Copper Oxy Chloride (COC)	1ppm
F3	*Cmmodity Mancozeb	1ppm

Table 2: Fungi from soil treated with Fungicides

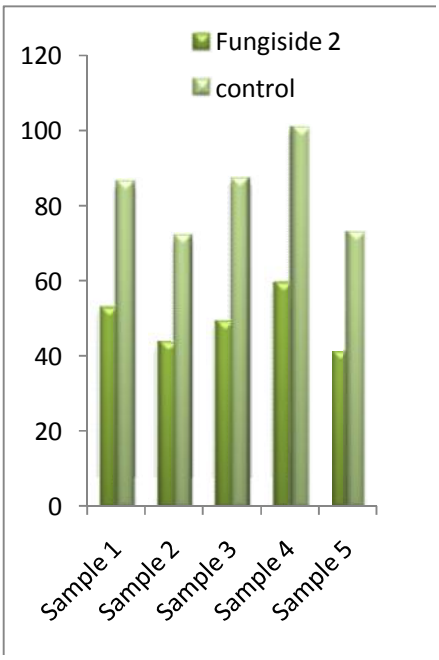
Soil Sample Code	F_1 CFU/ml				F_2 CFU/ml				F_3 CFU/ml			
	A	b	c	Mean	A	B	c	Mean	a	b	c	Mean
S1	65	60	63	62.6 ± 2.51	58	55	46	53.0 ± 6.2	52	52	40	48.0 ± 6.9
S2	52	45	48	48.3 ± 3.5	50	41	40	43.6 ± 5.5	46	38	38	40.6 ± 4.6
S3	59	46	45	50.0 ± 7.8	52	52	44	49.3 ± 4.6	46	42	48	45.3 ± 3.0
S4	75	64	69	69.3 ± 5.5	52	64	62	59.3 ± 6.4	52	62	58	57.3 ± 5.0
S5	46	44	50	46.6 ± 3.0	38	41	45	41.3 ± 3.5	52	40	40	44.0 ± 6.9

Table 3: Total Fungal Count from soil not treated Fungicide

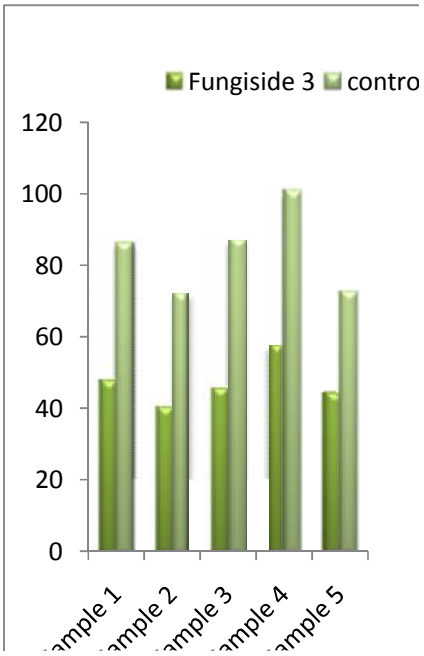
Soil Sample Code	TFC a CFU/ml	TFC b CFU/ml	TFC c CFU/ml	Mean	Standard deviation(SD)	Variance (SD)	Population (SD)	Population SD Variance
S1	96	81	83	86.6	8.14	66.33	6.65	44.22
S2	77	64	75	72	7	49	5.71	32.66
S3	91	84	86	87	3.6	13	2.94	8.66
S4	111	92	100	101	9.53	91	7.78	60.66
S5	76	69	73	72.6	3.51	12.33	2.86	8.22



Graph-1: Mancozeb graphical variation



Graph-2: Copper oxy chloride graphical variation



Graph-3: Carbendzim graphical variation

Fig. 1

Fig. 2

Fig. 3

Figure: Fungicides effect on Aspergillus nigur.

activities. Biochemical reaction in soil, which include the cycling of plant nutrients, depend on the activity of different enzymes[5,1]

Effect of fungicides	Zone of Inhibition in mm
F1	35
F2	23
F3	12

Sample 1

In sample1 the total plate count was 86.6CFU's/ml. The sample 1 showed maximum inhibition with the Commodity Mancozeb (48CFU's/ml) followed by Copper Oxy Chloride (53 CFU's/ml) and Carbendzim (62.6CFU's/ml).

Sample 2

In Sample 2 the total plate count was 72 CFU's/ml. The sample 2 showed maximum inhibition with the commodity Mancozeb (40.6 CFU's/ml) followed by Copper oxy chloride (43.6CFU's/ml) and Carbendzim (48.3CFU's/ml).

Sample 3

In Sample 3 the total plate count was 87 CFU's/ml.The sample 3 showed maximum inhibition with the commodity Mancozeb (45.3 CFU's/ml) followed by Copper oxy chloride (49.3CFU's/ml) and Carbendzim (50 CFU's/ml).

Sample 4

In Sample 4 the total plate count was 101CFU's/ml.The sample 4 showed maximum inhibition with the commodity Mancozeb (57.3 CFU's/ml) followed by Copper oxy chloride (59.3CFU's/ml) and Carbendzim (69.3 CFU's/ml).

Sample 5

In Sample 5 the total plate count was 72.6 CFU's/ml.The sample 5 showed maximum inhibition with the commodity Mancozeb (44 CFU's/ml) followed by Copper oxy chloride (41.3CFU's/ml) and Carbendzim (46.6 CFU's/ml).

Above sample variations showed graphically as Graph-1, 2 &3.

DISCUSSION

Agriculture is the backbone of Indian economy. In India around 70% of the population earns its livelihood from agriculture. It still provides livelihood to the people in our country. It fulfils the basic need of human beings and animals. It is an important source of raw material for many agro based industries. India's geographical condition is unique for agriculture because it provides many favourable conditions. There are plain areas, fertile soil, long growing season and wide variation in climatic condition etc. According to the Primary Census Abstract 2011, India's population over the decade has increased from 102.8 crore in 2001 to 121.1crore in 2011. The increasing global population and higher demand of food leads to increasing and sustainability of food production through intensive agriculture, attention of public health and proper utilization natural resources. The improvement of agriculture with advanced agricultural technology to meet this demand, keeping soil in its productive quality plays a dominant role for much of today's productivity. To solve the food crisis in the country the "Green revolution" began in 1960s, but it's confirmed that it began

in 1953 through the introduction of high-yield crop varieties and application of modern agricultural techniques, and led to an increase in food production. The introduction of high-yielding varieties of seeds and the increased use of chemical fertilizers and irrigation led to the increase in production needed to make India self-sufficient in food grains, thus improving agriculture in India. Thus the use of fertilisers for agriculture in India has risen astronomically in the last 60 years, resulting in deterioration of soil health in many parts of the country. In 1951-52, fertilizer usage in the country averaged less than one kg per hectare, which has now risen to 133 kg per hectare, according to information given on the Department of Fertilisers website. However, despite this increase, the consumption of fertilisers is still less in India than many developed countries. According to World Bank data, per hectare fertiliser consumption (kilogram per hectare of arable land) in India, China, Japan, Bangladesh, USA, Pakistan, and Israel in 2007 stood at 142.3 kg/ha, 331.1 kg/ha, 171.2 kg/ha, 166.2 kg/ha and Israel 524 kg/ha, respectively.

Soil microorganisms can have both positive and negative effects on plant growth. They can facilitate nutrient absorption by plants[6] ; promote plant growth or stimulate seedling development by producing hormone-like substances[7,8,9] suppress and control plant pathogens and diseases through various antagonistic activities[10] or adversely affect plant growth through their pathogenic behaviour[11]. The increasing use of chemical fertilizers is deteriorating the soil microbial community. This soil microbial community plays a crucial role in numerous ecosystems. Thus the change in the soil microbial community in turn affects the ecosystem.

The fertilizers used in this study had a significant effect on soil microbial characteristics. The fungal populations in each of the five agricultural soils were significantly decreased in the presence of three different fungicides such as Carbendzim, Copper Oxy Chloride and Cmmodity Mancozeb (fig 1, 2 & 3). Several studies[12,13]. On soil micro flora showed that soil characteristics may modify the effect of pesticides on microbial numbers and their biological activity.

The effects due to fungicide on soil microflora and especially on microorganisms involved in Nitrogen cycling of agricultural soils deserve a great deal of attention. Application of this fungicide to an agricultural soil may affect the composition of the microbial communities and thus disturb the fertility of the soil, especially the Nitrogen budget. In view of the deterioration in soil health, the government had in 2008-09 launched a new scheme, namely the National Project on Management of Soil Health and Fertility, to promote soil test-based balanced and judicious use of chemical fertilisers in conjunction with organic manure. In addition, the National Project on Organic Farming was started in 2004-05 to promote the use of organic fertilisers.

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REFERENCES

1. Wainwright M, 1978. A review of the effects of pesticides on microbial activity in soil. J Soil Sci, 29(3): 287–298.
2. Ainsworth, G.C. and Bisby, G.R. 1995. Dictionary of the Fungi. 8th edn. Common Wealth Mycological Institute. Kew, Surrey. p.445
3. James J. Hoorman. 2011. The role of soil fungus. Agriculture and Natural Resources pp 1-6.
4. Behera, N and Mishra, P. C. (2001): Insecticide Pollution and Soil Fertility. Soil Pollution and Soil Organisms (ed.) P. C. Mishra, Ashish Publishing House, Punjabi Bagh, New Delhi, pp. 63 – 83.
5. P K Shetty and S.P.Magu (1998) Invitro effect of pesticides on carbon dioxide evolution and dehydrogenase activities in soil. Journal of Environmental Biology 19(2): 141-144.
6. Bowen G. D and Rovira, A. D.1966. Microbial factors in short term phosphate uptake studies with plant roots. Nature (London) 211: pp 665-666.
7. Rubenchik L.I. (1963). Azotobacter and its use in agriculture. Israel Program for Scientific Translations, Jerusalem, p. 278.
8. Mishustin E. N. 1970. The importance of non-symbiotic microorganisms in agricultural plants. Plant and Soil. 32: pp 545-554.
9. Brown, M. E. 1974. Seed and root bacterisation. Ann. Rev. Phytopathol. 12: pp 181-197.
10. Marios J. J., D. J. Mitchell and R. M. Sonoda. 1981. Biological control of Fusarium crown root of tomato under field conditions. Phytopathol. 71: 1257-1260.
11. Elad Y. 1985. Mechanism of interactions between rhizosphere microorganisms and sol borne plant pathogens p 49-72. In V. Jansen, A . Kjoller and L.H Sorensen (ed.) Microbial communities in soil, Elsevier Applied Science, New York.

12. Wingfield G I, Davies H A, Greaves M P. the effect of soil treatment on the response of the Herbicides dalapon. J Applied Bacteriol, 1977, 43: pp 39-46.
13. Lal, R. & Saxena, D.M. (1982). Accumulation, Metabolism, and Effects of Organochlorine Insecticides on Microorganisms, Microbiological reviews, Vol. 46, No. 1, (March 1982), pp.95-127, ISSN 0146-0749.

