RESEARCH ARTICLE

Influence of Selective Herbicide 2, 4-D on Rhizosphere Mycoflora of *Psoraleacorylifolia*

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ABSTRACT

Population of microorganisms colonizing root surface of the plant forms and population pertaining to an ecological niche can be studied in rhizosphere. The present investigation deals with the mycoflora associated with the root surfaces of *Psoraleacorylifolia.* The fungal population was assessed after spraying the different concentrations of herbicide 2, 4-D from rhizosphere regions of root of *Psoraleacorylifolia*. The highest 52 colonies/gm were recorded in control. The flora belonging to Deutoromycetes was dominated as compared to Oomycets, Zygomycetes and Ascomycetes. In 100, 200, 400, 600 and 800 ppm 23, 21, 19, 15 & 12 colonies/gram were isolated respectively.

Keywords: 2, 4-D, herbicide,mycoflora, rhizosphere, *Psoraleacorylifolia*.

INTRODUCTION

The weeds very much interfere not only with the growth and development of agricultural crop but also decrease the productivity by utilizing water, light, macro and micro-elements from the soil. Beside this, they influence the soil media and the microbes responsible for maintenance and improvement of fertility of the soil. Herbicides are applied either to foliage or to the soil. The effects of herbicides on soil microorganisms have been extensively investigated from the point of view of herbicides degradation, persistence and effect on soil fertility (Cullimore, 1971). Many of the investigated microorganisms are important components of the saprophytic microflora and play a major role in maintaining the biological equilibrium in soil. It has been suggested by Audus (1964); Bollen (1961); Eno (1962); Newman and Downing (1958) that at normal rates most of the herbicides have no pronounced adverse effect on the soil microflora, at least as far as their total population is concerned. 2, 4-D is the common name approved by ISO for 2,4-dichlorophenoxy acetic acid. It belongs to aromatic compound, where phenyl ring is attached to an oxygen which in turn is attached to a carboxylic group. The first reference on 2,4-D had appeared in an article by Pokorny (1941). In USA, 2,4-D was discovered independently by Zimmerman and Hitchcock (1942) at Boyce Thompson Institute. They were the first to demonstrate 2,4-D including physiological activities (elongation, morphogenesis, root development and parthenocarpy) on different plants.

MATERIAL AND METHOD

The seeds of *Psoraleacorylifolia* were sown in 4×4 m^2 plots arranged in randomised block design using three replicates.The different concentrations of 2, 4-D *i.e.* 100, 200, 400, 600, 800 and 1000 ppm were prepared. To ascertain the lethal dose, the plants were sprayed with Aspy-hand sprayer at the age of 45 days. The roots with soil samples were collected with the help of sterile scissors.

The rhizosphere mycoflora was studied by soil dilution plat method (Timonin, 1940). Plants were carefully removed from the soil, shaken to remove excess soil and cut at the crown to separate the roots which were then transported to the laboratory in sterile petridishes. The roots were put into 500ml of sterile distilled water in 1000ml conical flask. The soil still clinging to the roots was removed by shaking the flask. 20ml of Martin's rose Bengal agar was plated into triplicate with one ml of this dilution.Further observations for rhizosphere mycoflora were made by the serial root washing technique (Harley and Waid, 1955). The original root system was removed from the dilution flask. Root pieces (1cm) randomly selected from different root regions, were placed in a sterile test tube. They were washed 20 times with sterile distilled water and were plated (5 pieces in each petridish) on martin's rose Bengal medium to allow fungi to grow on the root surface.

The plate kept for incubation to study the rhizosphere for 7 days at room temp. (26± 3^0c). Observations were made for numbers and kinds of fungi growing on the plates over a period of 7 days. The identified and unidentified species were isolated on PDA (Potato dextrose agar) slants. The number of colonies per gram of soil suspension were recorded.

RESULT&DISCUSSION

The data in table 1 revels that the colonies/gram recorded were 23,21,19,15 and 12 in 100, 200, 400, 600 and 800 ppm, respectively. The highest 52 colonies/gram were recorded in control. This indicated that herbicide had fungicidal effect. The flora belonging to Deuteromycetes was dominant as compared to Oomycetes, Zygomycetes and Ascomycetes. The total number of taxa isolated were 8 fromDeuteromycetes, 1 from Oomycetes, from Zygomycetes and only 1 from Ascomycetes.

The effect of 2,4-D on rhizosphere of *Psoraleacorylifolia* at different concentrations showed the lethal dose at 1000ppm. The total number of colonies/gram in different concentrations of 2,4-D continuously decrease with increase in concentrations. The maximum number had been noted in 600 ppm and 800 ppm, respectively. The qualitative and quantitative variations observed in the rhizosphere mycoflora of *Psoraleacorylifolia*. These results about variation in mycoflora without considering the weedicides are in agreement with the observations of Sreeramula*et al*. (1995) who studied the rhizosphere mycoflora of ten different dryland weeds namely *Amaranthus viridis, Acanthospermumhispidum, Ageratum conyzoides, Commelina benghalensis, Cyperusrotundus, Eupatorium gladulosum, Mimosa pudica, Parthenium hysterophorus, Phyllanthusniruri* and *Tridaxprocumbens* wherein he recorded maximum 20 fungal population in *Mimosa pudica* and minimum in *Cyperusrotundus*. In 100 ppm, in all 23 colonies/gram were isolated. Out of these, 21 from Deuteromycetes and 2 from Oomycetes were found but Ascomycetes were absent. This also reveals that the Deuteromycetes were dominant as compared to other two groups.

The important taxa isolated from this concentration were *Phytophthoraphaseoli, Aspergillus terreus, Fusarium incarnatum, Phomaherbarum* and unidentified species. The 200 ppm concentration of 2,4-D also showed the fungicidal effect. Only 21 colonies/gram were isolated. The important taxa isolated were only three. These were *Aspergillus terreus, Fusarium incarnatum*and *Phomaherbarum*.

Mall (1978) also reported variation in rhizosphere and rhizoplane microflora of the three potato varieties and noted the differences in flora of these varieties. Gangawane and Deshpande (1977) also reported qualitative and quantitative differences in the fungal flora of *Arachis hypogea* in rhizosphere and rhizoplane where they recorded 54 species. Srivastava and Dayal (1980) also observed varietal differences in rhizosphere mycoflora of eight varieties of *Abelmoschusesculentus*. Mishra and Kanaujia (1972) studied rhizosphere microbial population of 56 varieties of wheat and observed maximum number (11) of colonies/gram of dry root on safedlarma (5307) and minimum (5) of resistant and susceptible varieties. The study revealed quantitative variations in the mycoflora of different weeds under study. Zain *et*

Table 1: Effect of 2,4-D at different concentrations on the rhizosphere mycoflora of *Psoraleacorylifolia*.

S.N	Name of the Organisms	Number of colonies/gram at different concentrations in ppm					
		C	100	200	400	600	800
	Oomycetes:						
1	*Phytophthoraphaseoli*	4(7.69)	2(8.69)	-	-	-	-
	Zygomycetes:						
2	*Mortierella sp.*	5(9.61)	-	-	-	-	-
	Deuteromycetes:						
3	*Aspergillus terreus*	15(28.84)	5(21.73)	14(66.66)	-	3(20.00)	6(50.00)
4	*Fusarium incarnatum*	18(34.61)	7(30.43)	2(9.52)	11(57.89)	7(46.66)	4(33.33)
5	*Phomaherbarum*	10(19.23)	7(30.43)	5(23.80)	3(15.78)	5(33.33)	2(16.66)
6	Unidentified	-	2(8.69)	-	5(26.31)	-	-
	Total no. of colonies	**52**	**23**	**21**	**19**	**15**	**12**
	Total no. of taxa	**5**	**5**	**3**	**3**	**3**	**3**

al. (2013) suggested that herbicide applications in oil palm plantations induced transient effects on the growth and development of fungal community in soil.Adhikary*et al*. (2014) assessed the impact of three commonly used herbicides on soil microbial populations in chilli. It showed that the herbicide treatments significantly inhibited the development of microbial populations in the soil and the degree of inhibition varied with the type of herbicide.

In 400ppm, three taxa were isolated. These were *Fusarium incarnatum, Phomaherbarum* & unidentified species. The total number of colonies/gram was 19. The *Fusarium incarnatum* showed increased number of colonies/gram but there was decrease in *Phoma herbarum.* The unidentified species was absent in control but appeared in 100 and 400 ppm. In this concentration also the flora had decreased both qualitatively and quantitatively. In 600 ppm, total 15 colonies/gram representing only Deuteromycetes were present. There were only three taxa. They were *Aspergillus terreus, Fusarium incarnatum and Phoma herbarum. Aspergillus terreus*was isolated in treatments of 400 ppm and above but Fusarium *incarnatum* had decreased while *Phoma herbarum* had increased. This shows that this concentration had stimulatory effect on *Aspergillus terreus* and *Phoma herbarum* while inhibitory effect on *Fusarium incarnatum.*

Bhowmick and Choudhary (1982) observed 45 fungal species belonging to 30 genera in the rhizosphere of *Triticum vulgare* in relation to physico-chemical nature of the soil. A good correlation was noticed between the qualitative and quantitative proportion of the mycoflora and the variation in the physico-chemical factors of the soil of the different localities. With an increase in moisture, organic carbon, nitrogen and phosphorus contents, the micropopulation increased considerably whereas it decreased with the rise in pH of the soil. In the present study investigation the physico-chemical factors added to the increase or decrease in the rhizosphere mycoflora. Singh (1982) studied the effect of indole acetic acid on the colonization of microflora under the rhizosphere of *Phaseolus aureus* and reported that the rhizosphere micropopulation increased with increase in IAA concentration upto 100 ppm beyond which the micropopulation decreased.

In the present investigation different concentrations of herbicide affected on the micropopulation. Mohiuddin and Mohammed (2013) studied that the herbicides (2,4-D, Metribuzin and Atrizin) and fungicide (Carbendazim) in agricultural soil particularly tomato crop cultivating soil leads to decrease the total numbers of soil microorganisms at initial period. After gradually diluted, the pesticides may not much affect on soil microbial populations and lead to selection pressure in existing microorganisms to promote the functional groups of microorganisms adapted to the new conditions. In 800 ppm, 12 colonies/gram were isolated. The taxa recorded were *Aspergillus terreus, Fusarium incarnatum andPhomaherbarum.* This concentration also shows fungicidal effect on the flora. The flora also decreased both qualitatively and quantitatively over the control. Except the three species, rest of species had shown sensitivity to this herbicide. Randhawa *et al.* (1979) studied the rhizosphere of *Allium cepa* in relation to the effect of herbicide singly or in combination. The application of pre-plant, pre-emergence and post-emergence herbicides were done four days sowing, one day after sowing and fifteen days after sowing. The pre-plant

herbicides were incorporated into soil through harrowing. The soil samples were collected after two months of sowing the crop to study the population of bacteria, actinomycetes and fungi under the influence of different treatments. The herbicides used and their doses Kg/ha were Nitrofen (pre-em, 2.00), Alachlor (pre-em, 250), Fluchloralin (pre-em, 0.09), Fluchloralin (pre-plant, 1.20), Propanil (post-em, 1.36), Chloroxaron (post-plant, 1.50), Nitrofen (pre-em, post-em, 2.00+2.00) etc. They observed that there was a large spectrum of variation in fungal population under the unweeded and weeded control while in some there were significant reductions from the controls, unweeded and weeded. Hsia and Christensen (1951) observed effect of 2,4-D on seedling blight of wheat caused by *Helminthosporiumsativum*. The concentrations of ammonium and sodium salts of 2,4-D were 1000, 5000, 10,000 ppm and those of the amines and ethyl ester were 100, 1000, 5000, 10,000 ppm. Treated plants when inoculated with *Helminthosporiumsativum were* more heavily infested than those of untreated. Further, growth of *H. sativum* on culture medium was in most cases inhibited by 2,4-D and in a few cases it was stimulated. Joshi and Gupta (2008) observed that the 2,4-D has toxic effect on soil mycoflora. In contrast to the all fungal suppression, only *Aspergillus*sp showed enhancement at higher concentration of herbicide, whereas, the *Mucor*sp was suppressed by higher concentration of 2,4-D. Since *Aspergillus*sp utilized this herbicide as a carbon source, this fungus can be used as a bio-agent for the remediation of pesticide, concentrated soil.

CONCLUSION

The inhibitory effect of 2,4-D increased with the increase in the concentration. In general in the present observations also with exceptions the mycoflora was found to be decreased at higher concentrations. The response of fungal species to the herbicide was variable as per the habitats. Interestingly the importance of the investigation lies in the fact that some potential pathogens were stimulated in their occurrence due to these herbicidal compounds indicating their role in the control of these weeds.

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