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# Short communication Effect of 2,4-D residues on soil microflora

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### Abstract

A field experiment was conducted to assess the persistence of 2,4-dichlorophenoxy acetic acid (2,4-D) and the consequent changes in microbial populations in the wetland paddy soils of Thrissur, Kerala (India). Results indicate that 2,4-D does not persist in the paddy field beyond 30 days after spraying. Application of 2,4-D benefited soil fungi while the bacterial populations were depressed initially. By 30 days after spraying, however, the populations in the treated and untreated plots were similar and this period coincided with the disappearance of the 2,4-D residues in the soil.

Keywords: Herbicide persistence, Wetland paddy, Thrissur.

Persistence of 2,4-dichloro phenoxy acetic acid (2,4-D) in the soil is generally determined by microbial activity (Audus, 1960) and biochemical reactions (Smith, 1989). Although accumulation of 2,4-D is generally not a problem at recommended rates of application  $(1.0 \text{ kg a.i. ha}^{-1})$ , its indiscriminate use may lead to residue accumulation in the soil (Tejada et al., 1995). Long persistence herbicides also adversely affect the soil microflora as 2,4-D is mainly adsorbed on the soil organic fraction (Adams, 1973). Eventhough many studies on the persistence of 2,4-D and its effects on soil microflora are available, it is difficult to predict the persistence time without a detailed knowledge on the type and history of soil and its microbiota (QueHee and Sutherland, 1981). In particular, such studies are lacking from the Kole lands, a unique lowland ecosystems and major rice production area of Kerala (India). The present investigation was carried out to determine the effects of graded doses of 2,4-D application on the persistence of this chemical in the soil and its impact on soil microflora in the Kole lands of Thrissur district, Kerala.

(Pidikkaparambu) at Vallachira, a representative Kole location of Thrissur. The soils are derived from riverine alluvial deposits and belong to the USDA subgroup Oxyaquic Eutropepts (Ambili, 1995). Rice var. 'Jyothi' was direct-sown in the puddled land during the second crop season of 2001. Treatments consisted of four levels of 2,4-D (0.5, 1.0, 2.0 and 4.0 kg ha<sup>-1</sup>) and a control, replicated four times in a randomized block design. The herbicide was sprayed as per treatment protocol in plots of  $20 \text{ m}^2$  (5 x 4 m) at three weeks after sowing. Required quantities of 2,4-D sodium monohydrate equivalent to 83.90% of 2,4-D acid (obtained from M/s Atul Limited, Agro & Pharma Division, Gujarat) for each plot as per treatments were weighed accurately, dissolved in measured quantities of water (at the recommended spray volume of 500 L ha<sup>-1</sup>), and sprayed using an ASPEE knapsack sprayer fitted with a flood jet nozzle.

Duplicate soil samples were collected from each plot (0 to15 cm depth) using an aluminium scoop at 0, 1, 3, 6, 9, 15, 30, and 60 days after spraying for monitoring the herbicide residues. The soil sample collected in the scoop was spread on a filter paper and kept for 1 h to drain the excess water. The duplicate samples from

The experiment was laid out in a paddy field

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the same plot were composited and 20 g was taken for residue analysis. Another 20 g soil sample was used for gravimetric moisture determination. The soil samples were also analysed for organic carbon using Walkley and Black's rapid titration method after drying and sieving. Soil residues of 2,4-D up to 15 days after spraying were estimated by colorimetric procedure (Devi et al., 2001). The samples taken thereafter (i.e., 30 and 60 days) were analyzed by the more sensitive gas chromatographic methods (Sankaran et al., 1993).

To evaluate the effects of 2,4-D application on the population of soil bacteria and fungi, soil samples were drawn at 3, 6, 15 and 30 days after herbicide application. Quantitative estimation of soil microflora was carried out by serial dilution technique (Rangaswami, 1988) with dilutions of  $10^{-4}$  and  $10^{-6}$  for fungi and bacteria respectively. The required dilutions were prepared and plated on the respective enrichment media (Martin's rose Bengal streptomycin agar for fungi and soil extract agar for bacteria). Changes in microfloral counts with 2,4-D application compared to untreated control were worked out by taking the population in the control plot as 100%. The data on soil residues and microfloral population were statistically analyzed by applying the analysis of variance for randomized block design.

A comparison of the data in Table 1 indicates that residue

levels at the lower doses (0.5 and 1.0 kg ha<sup>-1</sup>) of 2,4-D application were similar and at higher rates of application (2.0 and 4.0 kg ha<sup>-1</sup>) higher concentration of residues were noted with the variation between treatments remaining significant only up to six days after spraying. At 15 days after spraying, however, only the highest rate of application viz., 4.0 kg ha-1 registered detectable residue levels in the colorimetric estimations. Bevond this, even the GC method failed to detect residues. The soils of the study area being homogenous in respect of organic carbon (0.96 to 1.19%) and moisture (35.93 to 42.64%), these parameters can probably be overruled as factors causing variations in the persistence of 2,4-D. It could thus be concluded that the rate of application of the chemical and time after spraying are key factors determining the dissipation pattern of 2,4-D in the soils.

Soil bacteria and fungi showed divergent trends in their population growth patterns following 2,4-D application (Fig.1). While the herbicide exerted a negative influence on soil bacteria up to 15 days after spraying, total number of fungal colonies increased. Up to 89.5% reduction in bacterial population was observed at 6 DAS in the treatment receiving 4.0 kg ha<sup>-1</sup> 2,4-D. Other treatments registered a decline in population (61 to 84%) at this sampling interval. This is presumably because of the inhibitory effect of 2,4-D on denitrifying bacteria (Audus, 1960), which predominates the submerged

Quantity of 2, 4-D applied (kg ha <sup>-1</sup> )	2, 4-D recovered (µg g <sup>-1</sup> ) from soil at days after application							
	0.0	ND	ND	ND	ND	ND	ND	ND
0.5	0.062	0.046	0.039	0.041	0.028	ND	ND	ND
	(0.749)	(0.739)	(0.734)	(0.735)	(0.727			
1.0	0.178	0.125	0.115	0.102	0.086	ND	ND	ND
	(0.822)	(0.790)	(0.784)	(0.775)	(0.765)			
2.0	0.439	0.264	0.240	0.157	0.153	ND	0.00014	ND
	(0.966)	(0.873)	(0.860)	(0.810)	(0.806)			
4.0	0.957	0.710	0.446	0.326	0.202	0.021	0.0011	ND
	(1.192)	(1.086)	(0.969)	(0.908)	(0.836)			
CD(0.05)	0.175	0.160	0.101	0.124	0.072	%	%	%

Table 1. Persistence of 2, 4-D in the paddy soil (sandy loam Oxyaquic Eutropept) at Vallachira, Kerala (India) during the second crop season.

Values in parentheses indicate sqrt(x+0.5) transformed values; % Not statistically analyzed; ND Not detected.



*Figure 1.* Change in the population of bacteria and fungi consequent to 2,4-D application at different levels (equating the population of control (0.00 kg ha<sup>-1</sup>) as 100 %) in the paddy soil (sandy loam Oxyaquic Eutropept).

soils. However, at 30 DAS, there was an increase in the bacterial population in all treatments suggestive of dissipation of the herbicide. Higher doses of 2,4-D led

to a corresponding increase in the fungal colonies and the maximum population was noticed at 6 DAS (344 to 615%; Fig. 1). This period could be considered as the lag phase during which the organisms develop adaptive capacity to degrade 2,4-D and proliferate.

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