

## Effect of chlorpyrifos on soil microbial populations and enzyme activities

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Chlorpyrifos is an organophosphate pesticide that acts as an insecticide-acaricide by ingestion, contact, and inhalation, and is now widely used in greenhouse for the control of pests. The dissipation of chlorpyrifos in crops such as orange fruit (Martínez-Vidal *et al.*, 1998), tomatoes (Aysal *et al.*, 1999), soil and its effects on soil microbial characteristics (Singh *et al.*, 2002) has been reported frequently. However, little information is available to describe impacts of chlorpyrifos on soil microbial population and enzyme activity in greenhouse. In this study, the effect of chlorpyrifos on soil bacteria, fungi, actinomycetes, and soil enzyme activity was investigated.

The effect of chlorpyrifos on soil microorganism is given in Table 1. Total population numbers of soil bacteria and actinomycetes were decreased by chlorpyrifos at a concentration of 2.0 mg/kg one day after treatments. Subsequently, the corresponding populations were recovered to the levels that are similar to those of the controls. However, the population of soil fungi was consistently increased by

the addition of chlorpyrifos at the same concentration. It seems to be likely that fungi is more resistant to chlorpyrifos than bacteria and actinomycetes. It is interesting to note that a fungal strain capable of utilizing chlorpyrifos as sole carbon and energy sources was isolated from the tested soil (data not shown). The effects of chlorpyrifos at a concentration of 4.0 mg/kg on the total numbers of soil microorganisms were similar to those at the low concentration. Nevertheless, chlorpyrifos at a high concentration of 10.0 mg/kg obviously has inhibitory impacts on soil bacteria, fungi and actinomycetes. The populations were significantly different from the control at 1, 7 and 14 d after treatment. However, the population numbers of soil microbes were recovered at the end of the experiments. It could therefore be concluded that the inhibitory effect of chlorpyrifos is a temporal phenomenon even chlorpyrifos presents in soil at higher concentration up to 10.0 mg/kg. The results reported here are similar to the observations of Pandey and Singh (2004), who indicated that

**Table 1** Effect of chlorpyrifos on total counts of soil bacteria( $10^5 \cdot g^{-1}$ ), fungi( $10^4 \cdot g^{-1}$ ) and actinomycetes( $10^5 \cdot g^{-1}$ )

| Soil microorganisms | Chlorpyrifos, mg/kg | Days after treatments       |                           |                            |                            |                            |
|---------------------|---------------------|-----------------------------|---------------------------|----------------------------|----------------------------|----------------------------|
|                     |                     | 1                           | 7                         | 14                         | 21                         | 30                         |
| Bacteria            | 0                   | 41.93 ± 5.99 <sup>a</sup>   | 41.92 ± 5.99 <sup>a</sup> | 38.75 ± 3.63 <sup>a</sup>  | 36.30 ± 0.30 <sup>a</sup>  | 39.70 ± 2.93 <sup>a</sup>  |
|                     | 2                   | 24.74 ± 5.49 <sup>b</sup>   | 39.12 ± 3.01 <sup>a</sup> | 44.73 ± 3.16 <sup>a</sup>  | 40.31 ± 7.01 <sup>a</sup>  | 40.22 ± 3.07 <sup>a</sup>  |
|                     | 4                   | 18.84 ± 0.61 <sup>bc</sup>  | 28.22 ± 6.98 <sup>b</sup> | 41.29 ± 6.81 <sup>a</sup>  | 51.10 ± 3.81 <sup>b</sup>  | 46.23 ± 2.57 <sup>b</sup>  |
|                     | 10                  | 12.71 ± 1.21 <sup>c</sup>   | 18.64 ± 0.60 <sup>c</sup> | 27.55 ± 3.02 <sup>b</sup>  | 53.40 ± 3.03 <sup>b</sup>  | 46.24 ± 3.35 <sup>b</sup>  |
| Fungi               | 0                   | 38.93 ± 3.00 <sup>a</sup>   | 35.93 ± 5.99 <sup>a</sup> | 33.61 ± 3.14 <sup>ab</sup> | 27.51 ± 3.12 <sup>a</sup>  | 12.40 ± 1.47 <sup>a</sup>  |
|                     | 2                   | 42.59 ± 6.09 <sup>b</sup>   | 44.14 ± 9.20 <sup>a</sup> | 42.64 ± 5.74 <sup>a</sup>  | 27.22 ± 3.22 <sup>a</sup>  | 12.58 ± 2.85 <sup>a</sup>  |
|                     | 4                   | 21.27 ± 3.04 <sup>b</sup>   | 33.26 ± 3.03 <sup>a</sup> | 33.03 ± 6.96 <sup>ab</sup> | 33.61 ± 2.73 <sup>b</sup>  | 15.67 ± 2.27 <sup>a</sup>  |
|                     | 10                  | 11.50 ± 2.42 <sup>c</sup>   | 18.64 ± 5.14 <sup>b</sup> | 22.83 ± 6.81 <sup>b</sup>  | 15.61 ± 2.44 <sup>c</sup>  | 16.93 ± 2.87 <sup>a</sup>  |
| Actinomycetes       | 0                   | 29.95 ± 5.99 <sup>a</sup>   | 37.93 ± 3.46 <sup>a</sup> | 33.72 ± 3.09 <sup>a</sup>  | 15.40 ± 2.58 <sup>a</sup>  | 15.45 ± 2.54 <sup>bc</sup> |
|                     | 2                   | 24.74 ± 10.20 <sup>ab</sup> | 52.16 ± 3.48 <sup>b</sup> | 52.90 ± 3.92 <sup>b</sup>  | 19.12 ± 5.00 <sup>ab</sup> | 16.40 ± 2.18 <sup>ac</sup> |
|                     | 4                   | 17.42 ± 5.96 <sup>ab</sup>  | 50.40 ± 6.99 <sup>b</sup> | 35.15 ± 3.83 <sup>a</sup>  | 22.47 ± 3.41 <sup>b</sup>  | 22.18 ± 2.22 <sup>b</sup>  |
|                     | 10                  | 14.93 ± 8.12 <sup>bc</sup>  | 27.06 ± 3.01 <sup>c</sup> | 26.49 ± 3.61 <sup>c</sup>  | 16.23 ± 2.10 <sup>ab</sup> | 16.31 ± 2.25 <sup>c</sup>  |

Notes: All data are means ± SD of triplicate samples; values in horizontal rows followed by the same letter (a, b, c) are not statistically different ( $P < 0.05$ ) from the control

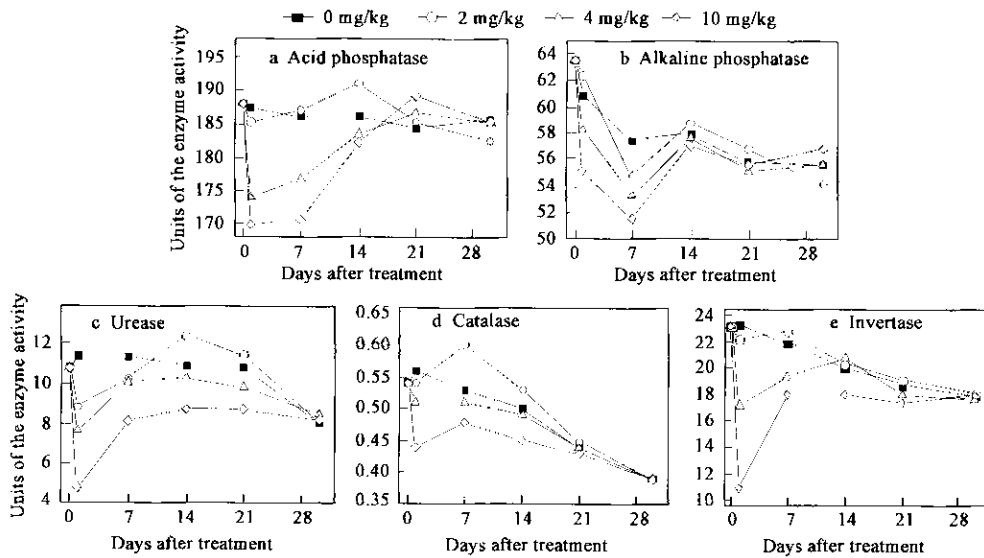


Fig.1 Effect of chlorpyrifos on soil enzyme activity

chlorpyrifos (2.2 mg/kg) had a short-term inhibitory effect on the soil bacterial population, and the fungal population was significantly enhanced during the initial days(0—15 d) in the groundnut fields.

Acid phosphatase activity in the soil treated with chlorpyrifos is presented in Fig.1a. The enzyme activity was significantly reduced by chlorpyrifos at concentrations of 4.0 and 10.0 mg/kg till 14 d, and then recovered to and exceeded to the levels of the controls at 21 d after treatment. Chlorpyrifos at 2.0 mg/kg showed a slightly effect on acid phosphatase, it enhanced the activity at 7 and 14 d. Alkaline phosphatase was significantly inhibited at 7 d as a consequence of the addition of chlorpyrifos (Fig.1b). After incubation for 14 d, the activity was recovered to the level of the control. An enhancement in enzyme activity was observed at the end of the experiment. These data are in agreement with the results reported by Pozo *et al.* (1995), who indicated that chlorpyrifos decreased initially the activities of phosphatase, but recovered after 14 d. Urease activity in the soil treated with chlorpyrifos at a concentration of 2.0 mg/kg was decreased at the day 1 and 7, whereas it recovered to the level of control after incubation for 14 d (Fig.1c). However, a significant reduction in urease activity was observed in the soil treated with chlorpyrifos at two

higher concentrations. Catalase activity was negatively affected by the addition of chlorpyrifos at high concentrations of 4.0 and 10.0 mg/kg (Fig.1d). This negative effect on catalase activity disappeared at 21 d. The insecticide had no adverse effect on soil catalase at concentration of 2.0 mg/kg. The change of invertase activity was similar to that of catalase (Fig. 1e).

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