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Effects of Paraquat and Alachlor on Soil Microorganisms in Peat Soil

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ABSTRAK

Satu kajian telah dijalankan untuk melihat kesan alachlor dan paraquat ke atas aktiviti mikrob dalam tanah gambut. Kesan racun rumpai ke atas pembebasan CO_2 dan aktiviti phosphatase dimonitor selama 12 minggu. Hasil yang diperolehi menunjukkan paraquat dan alachlor yang disembur kepada tanah menyebabkan peningkatan pembebasan CO_2 di peringkat awal pengeraman tetapi berkurangan selepas 53 hari. Lebih banyak CO_2 dibebaskan dari tanah yang dirawat dengan alachlor berbanding dengan tanah yang dirawat dengan paraquat. Aktiviti phosphatase meningkat di peringkat awal pengeraman bagi tanah yang diperlakukan dengan sama ada alachlor atau paraquat. Aktiviti phosphatase meningkat di peringkat awal pengeraman bagi tanah yang diperlakukan dengan sama ada alachlor atau paraquat tetapi aktiviti phosphatase berkurangan selepas 12 hari eraman. Populasi kulat dan bakteria dipengaruhi oleh kedua-dua racun rumpai yang diuji. Pada kepekatan 250 ppm, alachlor dan paraquat, masing-masing menyebabkan pengurangan populasi bakteria kira-kira 78 dan 95%. Alachlor didapati lebih toksik terhadap kulat berbanding paraquat.

ABSTRACT

A study was carried out to investigate the effects of alachlor and paraquat on microbial activities in peat soil. Effects of the herbicides on CO_2 evolution and phosphatase activity were monitored for 12 weeks in ambient conditions. The results showed that paraquat and alachlor caused an initial increase in CO_2 released and subsequently decreased after 53 days of incubation. Comparatively, more CO_2 was released from the soil treated with alachlor than that treated with paraquat. An initial increase in phosphatase activity was observed for both herbicides but the level of activity was substantially reduced after 12 days of incubation. Fungal and bacterial populations in the soil were also affected by both herbicides. At 250 ppm, alachlor and paraquat caused a reduction in bacterial population of 78% and 95% respectively. Alachlor was shown to be more toxic to fungal populations in the soil than paraquat.

INTRODUCTION

Microorganisms play important roles in soil processes, among which are the recycling of essential plant nutrients, humus formation, and pesticide detoxification. In agriculture, a major concern over the usage of herbicides is the possible harmful effects exerted on the soil microflora, which contribute to soil fertility. Herbicides are equally toxic to plants as well as to many soil microorganisms. Problems in assessing the impact of herbicides on the soil microflora are numerous and complicated. This is largely due to the highly complex nature of multiple interactions occurring simultaneously between herbicide, soil, and micro-organisms.

There are several ways of assessing the effects of herbicides on microbial activities and these include detecting CO_2 evolution, measurement of O_2 uptake, estimation of microbial population levels and assaying soil enzyme activities. Each parameter has its advantages and disadvantages. In recent years, the measurement of soil biological activity has increasingly relied on assays for soil-borne microbial enzymes such as phosphatase (Marsh 1980; Davies and Greaves 1981). The present study describes the effects of alachlor and paraquat on soil microflora.

MATERIALS AND METHODS

Soil

Peat soil was obtained from the top 0-5 cm of an uncultivated plot at MARDI Research Station, Jalan Kebun, Kelang, Selangor. The physico-chemical properties of the soil are shown in Table 1. Before use, the soil was passed through a 5 mm sieve, placed in black polythene bags and stored at 4°C.

TABLE 1
Physico-chemical characteristics of soil used in the study.

pH	4.8
% water content	80
% carbon	34.2
% organic matter	59.3
% N	3.1
% clay	44.6
% silt	11.7
% sand	43.7
CEC (M equiv/100 g soil)	145.0

Herbicides

The two herbicides tested were paraquat (Gramoxone®, ICI) containing 20% w/v of 1,1'-dimethly-4,4'-bipyridylium chloride, and alachlor (Lasso®, Monsanto) containing 48% w/v of chloro 2', 6'-diethyl-N- methoxymethyl.

Soil treatment

Soil samples were treated with both alachlor and paraquat according to the commercial formulation of either Lasso® or Gramoxone®. Moist soil equivalent to 4 kg oven-dry soil was placed in a cylindrical metal drum (30 x 27.5 cm) containing a polythene liner. Each herbicide was applied separately by spraying onto the soil to give a mean final concentration of 0, 100 or 250 ppm of the active ingredient (calculated on an oven-dry basis).

The mixture was mixed thoroughly in a rotating drum. The moisture content was then adjusted to 80% of field capacity as described by Grossbard and Wingfield (1975). Field capacity was determined at a suction pressure of 75 cm of water on tension tables (Clements 1966). Three 4 kg replicates per treatment were prepared and incubated in air-filled double polyethylene bags at 27°C. The bags were opened once a week to prevent the soil becoming oxygen-deficient. The soil moisture levels were checked regularly by weighing and adjusted to 80% of field capacity by adding deionized water as necessary.

Carbon dioxide evolution

Carbon dioxide evolution was measured using a continuous gas flow system as described by Grossbard and Marsh (1974). Two samples of 100 g of soil were taken from each replicate one day after spraying and incubated in 500 ml respiration flasks attached to a manifold distributing a slow flow of moist CO_2-free air. This was passed through the layer of soil in the flask from an inlet close to the bottom of the vessel and the CO_2 was absorbed in 40 ml M-NaOH in Drechsel bottles and measured periodically by titration with 0.05 M H_2SO_4. The soil samples were incubated at 27°C. By replacing fresh NaOH in Dreschel bottles, CO_2 evolving from the soil was recorded during 3 months' incubation. The soil moisture in the flasks was adjusted to, and maintained at, 80% field capacity.

Microbial count and phosphatase activity

Soil samples were removed from the polythene bags at day 2, 12, 32, 60 and 90 for microbial population counts and phosphatase assay.

Total counts of fungal and bacterial populations were determined by the plate count method using Potato Dextrose Agar for fungal, and nutrient agar (NA) for bacterial growth. Ringer's solution was used as diluent at all times. Plates were incubated at 30±3°C. Dilutions of soil samples were made in triplicate.

Phosphatase activity was measured on 1 g samples of soil by the method of Tabatabai and Bremner (1969). The production of p-nitrophenol was determined spectrophotometrically at 420 nm.

RESULTS AND DISCUSSION

Addition of alachlor or paraquat to soil initially increased the rate of CO_2 evolution during the first 7 days' incubation. The initial stimulation of CO_2 evolution in herbicide-treated soil may be due to direct stimulation of respiratory activity of the soil microbes. A similar observation has been reported by Quilt *et al.* (1979) in barban-treated soil. The rate of CO_2 released was affected in soil treated with 250 ppm alachlor during the first 50 days' incubation *(Fig. 1)*. The CO_2 from soil treated with alachlor was higher than from control soil after 70 days of incubation. CO_2 evolution from paraquat-treated soil at 100 ppm was not significantly different from control between day 7

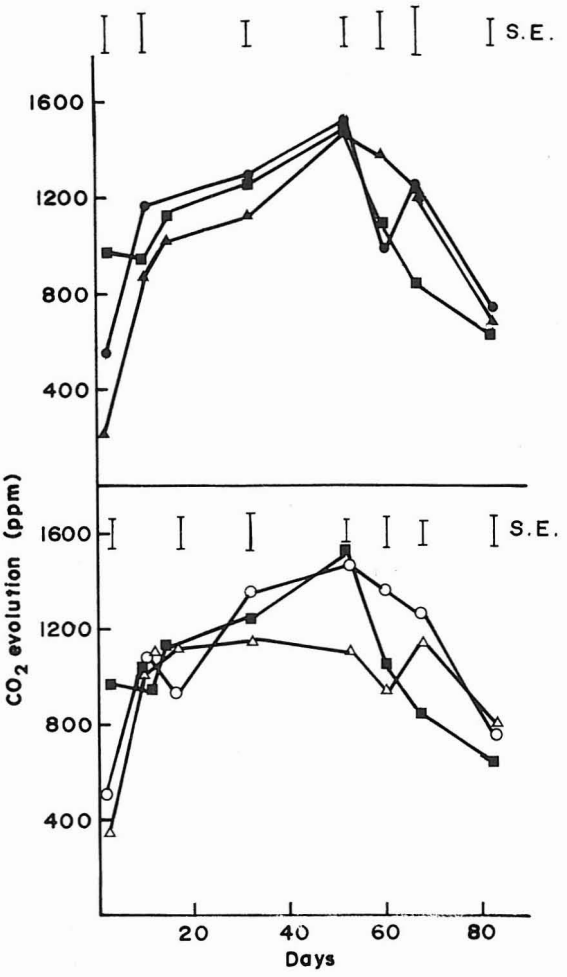

Fig. 1: Effect of herbicides on CO_2 evolution from the soil. ■ control, • 100 ppm alachlor; ▲ 250 ppm alachlor, o 100 ppm paraquat, Δ 250 ppm paraquat.

until day 50 but thereafter it showed significantly higher readings than control. The higher evolution of CO_2 at day 60 may be due to an increase of microbial activity. At certain concentrations, paraquat may serve as a possible carbon and/or nitrogen source for certain bacteria and fungi (Camper *et al.* 1973). But at higher doses, paraquat inhibited growth and activity of certain fungi (Tu and Bollen 1968). Therefore, CO_2 evolution from soil treated with 250 ppm paraquat did not show an increment after 7 days of incubation.

The amount of CO_2 released is correlated with the presence of different species of microbes in the soil. The populations of soil fungi and bacteria were affected by treatment with either alachlor or paraquat (*Fig. 2*). An increase of paraquat concentration was less affective on fungal count in soil. Paraquat treatment appeared to cause a greater reduction of bacterial than fungal counts. In previous studies, two fungal species, *Aspergillus* sp. and *Penicillium* sp. were found to be dominant in soil treated with paraquat and they have been reported to be paraquat-resistant (Smith *et al.* 1976). The behaviour of paraquat in soil may explain the differences between the two herbicides. Paraquat was also found to be readily absorbed by soil particles through its ionic exchange capacity (Weber and Coble 1968). The adsorption of paraquat molecules onto soil particles may reduce their deleterious effects on soil microbes (Weber and Coble 1968). In contrast, alachlor is soil-active and this may be the factor contributing to the reduction of both fungi and bacterial populations. At a higher concentration (250 ppm), alachlor caused 81% reduction of the fungal population, whereas paraquat resulted in only 29% reduction.

From the data reported here, it is evident that paraquat and alachlor are equally toxic to bacteria and to fungi. The bacterial population in untreated soil was approximately 7.5×10^4/g dry soil. At 250 ppm, alachlor and paraquat caused 78% and 75% reduction, respectively in the bacterial and fungal populations *(Fig. 2)*. Since microbial flora contribute significantly toward the improvement of the soil, these effects on bacteria and fungi could give a negative response to the usage of herbicides.

Phosphatase activity was generally very low in control and treated soil during 7 days of incubation but reached a maximum at day 10. The level of phosphatase in soil treated with 100 ppm paraquat was higher than the activity in alachlor-treated soil at the same concentration, 32 days after spraying. Consequently, phosphatase activity was still higher in soil treated with paraquat as compared to alachlor-treated soil when incubation period was prolonged to 60 days. The difference in behaviour between the two herbicides in soil explained the differences in phosphatase activity. Phosphatase activity in soil treated with either herbicide at 32 days showed correlation with microbial population in which lower microbial population was observed at the higher herbicide concentrations. Earlier reports have shown that herbicides may enhance or inhibit soil enzyme activities (Quilt *et al.* 1979; Davies and Greaves 1981). For instance, Quilt *et al.* (1979) showed that after an initial inhibition, there was a consistent increase in phosphatase activity in soil treated with barban at 200 ppm. In contrast, atrazine significantly reduces soil enzymatic activity (Voets

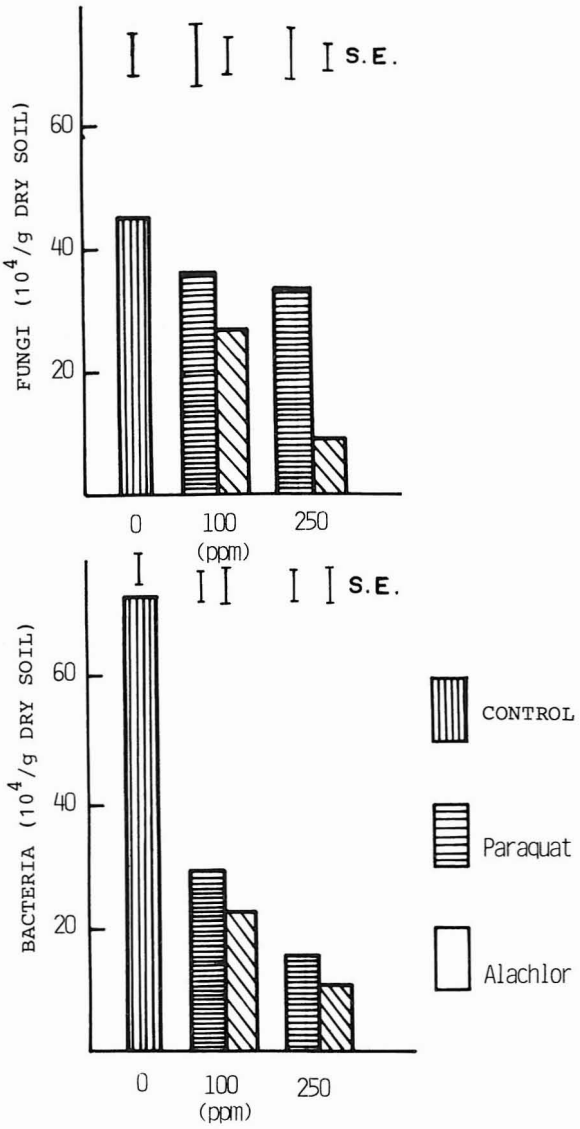

Fig. 2: Effect of herbicides on microbial population in peat soil

et al. 1974). Under field conditions, it was suggested that the reduction of soil enzyme activity results partially from the indirect effect of the herbicide treatment, namely the elimination of the direct vegetative cover and the concomitant decrease in the soil organic matter (Voets *et al.* 1974). Organic content and other factors have an effect on soil microbe populations (Marsh *et al.* 1978). Therefore, reduction in phosphatase activity was also observed in control soil when incubation periods were prolonged.

The concentrations of herbicides used in this study were higher than normal application rates applied in the field, which are less than 10 ppm for the top 5 cm of soil. Therefore, the low concentration of paraquat or alachlor, which is characteristic of normal field applications, is unlikely to have any effect of agronomic importance. However, it is possible that normal application in the field may lead to uneven distribution and thus localized high concentrations of herbicide. How far these may influence plant growth and crop production will depend on how the specific microbial activity affected is related to soil fertility.

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REFERENCES

CAMPER, N. D., E.A. MOHEREK and J. HUFFMAN. 1973. Changes in Microbial Populations in Paraquat-treated Soil. *Weed Res.* **13:** 231-233.

CLEMENT, C. R. 1966. A Simple and Reliable Tension Table. *J. Soil Science* **17:** 133-135.

DAVIES, H. A. and M.P. GREAVES. 1981. Effect of some Herbicide on Soil Enzyme Activities. *Weed Res.* **21:** 205-209.

GROSSBARD, E. and J.A.P. MARSH. 1974. The Effect of Seven Substituted Urea Herbicides on the Soil Microflora. *Pestic. Sci.* **5:** 609-623.

GROSSBARD, E. and G.I. WINGFIELD. 1975. Techniques for the Assay of Effects of Herbicides on the Soil Microflora II. The Effect of Herbicides on Cellulose Decomposition. In: *Some Methods for Microbiological Assay* ed. R.G. Board and D.W. Lovelock. Soc. Appl. Bact. Ser. No. 8. London: Academic Press. 236-256.

MARSH, J.A.P. 1980. Effect of Asulam on some Microbial Activities of Three Soils. *Bull. Environm. Contam. Toxicol.* **25:** 15-22.

MARSH, J. A. B., G. I. WINGFIELD, H. A. DAVIES and E. GROSSBARD. 1978. Simultaneous Assessment of Various Responses of the Soil Microflora to Bentazone. *Weed Res.* **18:** 293-300.

QUILT, P., E. GROSSBARD and S. J. L. WRIGHT. 1979. Effects of the Herbicide Barban and its Commercial Formulation Carbyne on Soil Microorganisms. *J. Appl. Bacteriol.* **46:** 431-444.

SMITH, S.N., A.J.E. LYON and ISMAIL SAHID. 1976. The Breakdown of Paraquat and Diquat by Soil Fungi. *New Phytol.* **77**: 735-740.

TABATABAI, M.A. and J.M. BREMNER. 1969. Use of P-nitrophenyl Phosphate for Assay of Soil

Phosphatase Activitiy. *Soil Biol. Biochem.* **1:** 301-307.

TU, C.M. and W.B. BOLLEN. 1968. Effect of Paraquat on Microbial Activities in Soils. *Weed Res.* **8:** 28-37.

VOETS, J.P., P. MEERSCHMAN and W. VERSTRAETE. 1974. Soil Microbiological and Biochemical Effects of Long-term Atrazine Applications. *Soil Biol. Biochem.* **6:** 149-152.

WEBER, J.B. and H. C. COBLE. 1968. Microbial Decomposition of Diquat Adsorbed on Montmorillonite and Kaolinite Clays. *J. Agric. Food Chem.* **16:** 475-478.

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