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ROOT UPTAKE AND LEAF DISTRIBUTION OF ^{14}C-PHORATE IN BEAN PLANTS [1]

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(With 3 text-figures)

Phorate, 0,0 – diethyl S (ethylthio) methyl phosphorodithioate, is a plant systemic insecticide used in a wide range of agricultural crops against the attack of soil insects and leaf-feeding pests, particularly aphids and mites. Its metabolism in plants has been extensively investigated (Metcalf *et al.*, 1957; Bowman and Casida, 1957, 1958; Bowman *et al.*, 1969). Five toxic metabolites are produced by oxidation of the sulphide in the side chain to the sulphoxide and sulphone, and by the formation of a P=O analogue, phoratoxon, from the P=S group of the parent compound with its respective sulphoxide and sulphone.

In the present paper, the distribution of phorate, two of its toxic metabolites and the non-toxic water soluble fraction are investigated in bean plants after a 24 hour translocation period.

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MATERIAL AND METHODS

Bean plants, *Vicia faba* L. (var. Sutton), grown in sand in a glasshouse, were transferred to 100 mm x 36 mm specimen tubes containing nutrient solution (Hewitt, 1966). ^{14}C–methylene labelled phorate was added to 30 ml of nutrient and the plants were kept in a constant temperature room at 20°C with a light regime of eight hours dark and 16 hours light provided by four 85 Watt fluorescent tube lights mounted 60 cm above the bench. The relative humidity fluctuated from 60% during the daylight period to 80% at night.

Isotope extraction procedure. Individual leaves were crushed in a hand homogenizer with 10 ml of acetone-water 1:1 v/v and transferred to a 50 ml conical flask containing 1.5 g of finely divided activated charcoal (Norit GSX). The slurry was shaken for five minutes and filtered through a 4.5 cm Buchner funnel. Both homogenizer and conical flask were washed, first with 10 ml of the same acetone-water mixture and then with 20 ml chloroform. The washings were added to the Buchner funnel to facilitate the removal of any insecticide residue adsorbed on the charcoal. The filtrate was transferred to a 50 ml separating funnel and shaken. The two phases were allowed to separate; the solvent fraction was collected in a 50 ml round-bottomed flask and the aqueous residue re-extracted with a further 20 ml chloroform. The solvent extracts were pooled and concentrated to near dryness in a rotary film evaporator under reduced pressure in a water bath at 50 – 55°C. The water fraction was similarly concentrated at a temperature of 80°C.

Analyses of phorate, its sulphoxide and sulphone followed the procedure described by Galley and Foerster (1973).

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^{14}C was counted in a Beckman LS 250 scintillation counter. The activity in each sample was counted for 50 minutes; counting efficiency was assessed by the external standard method and the results were corrected for background and efficiency.

Autoradiography. The leaf sample was placed in contact with a Kodak "Kodirex" X-ray film in a dark room, using a Kodak safe light with a 6B filter. The film was removed after 90 days and developed following conventional methods in Ilford ID-19. The film was fixed in a solution of Amfix for 10 minutes before the final washing and drying.

RESULTS AND DISCUSSION

The distribution of radio-activity in the first three leaves after adding 20 μg of ^{14}C-phorate to each of four plants followed an apically dominant pattern, with more label accumulating in the young upper leaves (fig. 1).

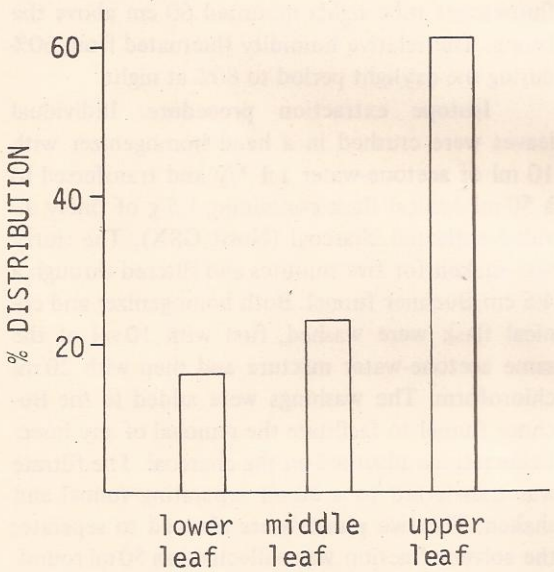

Fig. 1 – Per cent distribution of radio-label in the first three leaves of bean plants, after 24 hours uptake from the nutrient solution.

Of the total label detected, 61% was concentrated in the upper leaf, in comparison with 23% and 16% present in the middle and lower leaves respectively.

Phorate was rapidly oxidised to its sulphoxide and sulphone (fig. 2). Phorate sulphoxide was the major metabolite found in all the leaves, accounting

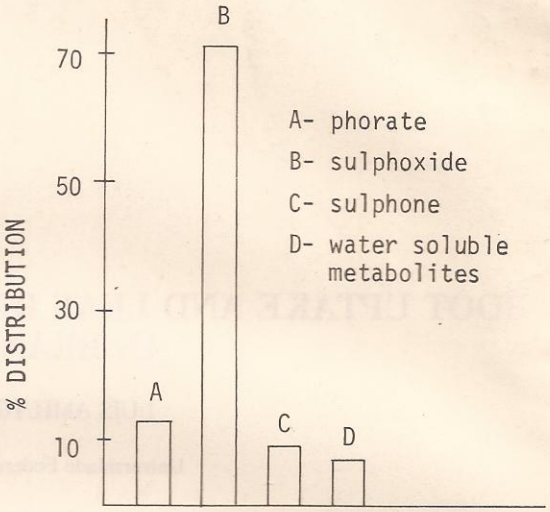

Fig. 2 – Per cent distribution of phorate and its metabolites in bean plants.

for 71% of the total label translocated. Unchanged phorate represented 13% and phorate sulphone nine per cent of total radio-activity transported to the leaves.

Due to its lipophilic properties, phorate is readily absorbed by the roots and held in the lipoprotein layers, where it is rapidly oxidised to its sulphoxide and to a lesser extent to its sulphone (Metcalf, 1967). The sulphoxide, being approximately twice as hydrophilic as the parent compound (Bowman *et al.*, 1969), is easily translocated to the foliage with the ascending transpiration stream; however phorate and its sulphone, which have similar partition properties, are less readily translocated.

Hydrolysis was also rapid; after 24 hours, seven per cent of the radio-activity was present in the water soluble fraction of the extract (fig 2). It is likely that some hydrolysis products were formed in the nutrient solution and were subsequently absorbed by the plants. Hacskaylo *et al.* (1961) found that after two days in nutrient solution, 32% of the phorate present had been hydrolysed.

The oxygen analogue, phoratoxon, its sulphoxide and sulphone, according to Bowman *et al.* (1969) are readily hydrolysed and therefore do not persist long enough to be of significant toxicological importance.

The autoradiograph (fig. 3) shows that the isotope was evenly distributed within the leaf, after 24 hours translocation.

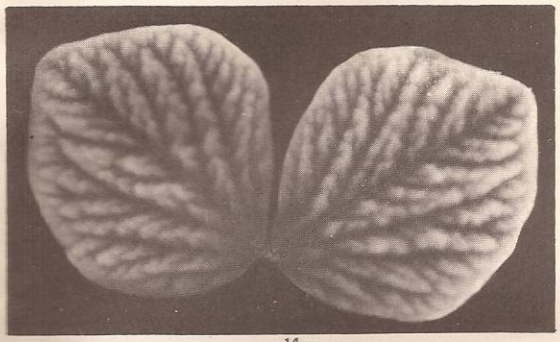

Fig. 3 – Distribution of ^{14}C-phorate and its metabolites within the leaf.

SUMMARY

The amounts of ^{14}C-phorate and its metabolites present in bean leaves were quantitatively estimated after a 24-hour translocation period. It is concluded that the labelled insecticide is rapidly oxidised to its sulphoxide and sulphone, the former accumulating in greater proportions due to its hydrophilic properties. Some detoxification occurred after 24 hours uptake. More label was found in the growing upper leaf than in the lower leaves.

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