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## A METHOD FOR SCREENING THE VAPOUR-PHASE OF VOLATILE COMPOUNDS AS NEMATICIDES

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Di-butyl phthalate (DBP) is widely used as a plasticiser and concern has been expressed about the effect of phthalate pollution on plants and animals (Hardwick *et al.*, 1986). During a study examining the potential toxicity of DBP to plant parasitic nematodes (Alphey and Brown, 1987) we devised a method to test DBP vapour for possible nematotoxic effects. DBP has a very low vapour pressure (Table I) and the method, which permits a mixture of air and DBP vapour to be actively drawn through a segmented column of soil containing nematodes, is more efficient than the passive diffusion method used by McKenry and Thomason (1974).

The apparatus (Figure 1) was developed from that described by Evans (1969) and was constructed from two 120 mm lengths of perspex tubing, one tube forming a friction-fit inside the other. The tube diameters were 32 mm and 26 mm externally and 26 mm and 17 mm internally. The smaller diameter tube was cut into 10 mm lengths forming rings on which 15  $\mu$ m aperture nylon bolting cloth (Henry Simon, Stockport, England) was glued at one end to retain soil and nematodes, thus creating a series of sample cells each with an internal capacity of 2.3 cm<sup>3</sup>. The terminal 40 cm at one end of the larger diameter tube was machined to form a friction-fit inside a 60 cm<sup>3</sup> Sintaglass funnel (5  $\mu$ m to 10  $\mu$ m sinter pore; Gallenkamp, Loughborough, England).

To load the sample cells they are first one-third filled with a 2:1 mixture of air-dried sand and steam-sterilized soil (*ssm*) with a particle and aggregate size  $< 1500 \ \mu m$  and  $> 250 \ \mu m$ . These cells were placed in a petri dish containing water to a depth of *c*. 7 mm and the nematodes hand-picked

	Nemati		Di-butyl-phthalate	
	fumigant		non-fumigant	
	dichloropropene	carbamate aldicarb	organophosphate phenamiphos	
Vapour pressure (mm Hg)	55	0.0001	0.000001	0.00001
Water solubility (g dm <sup>-3</sup> )	1.0	6.0	0.7	0.4
Boiling/Melting point (°C)	104-112 (BP)	100 (MP)	49 (MP)	340 (BP)

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Table I - Some physico-chemical characteristics of fumigant and non-fumigant nematicides and di-butyl phthalate



Fig. 1 - A, Apparatus for screening the vapour-phase of volatile compounds as nematicides. B, Hypodermic syringe for adding water to the sand and soil mixture. C, perspex tube. D, Sintaglass funnel with s, 5-10  $\mu$ m pore sinter. E, Posterior rings. F, Sample cells with m, 15  $\mu$ m bolting-nylon bases. G, Anterior-rings. Inset sample cell in petri dish prepared to receive nematodes.

directly into the water overlying the *ssm* in the cells. The cells were then removed from the petri dish and carefully filled with *ssm*.

To assemble the apparatus the outer tube is inserted into the funnel which is clamped vertically in a retort stand. Four rings are placed into the bottom of the tube and filled with *ssm*, next three sample cells containing nematodes are inserted and additional sample cells filled with dry *ssm*, but without nematodes, are added. Water is added by pipette until all the *ssm* in the apparatus is saturated. The outer tube is closed with a rubber bung containing a central glass tube. A length of medical catheter tubing is inserted inside the glass tube so that one end lies in contact with the *ssm* in the uppermost ring. This catheter is attached to a syringe needle inserted through a rubber-tube which connects the apparatus to a flask containing the chemical sample (see Fig. 1). The syringe can be used, if necessary, to add water to the *ssm* in the apparatus during vapour perfusion.

During operation the apparatus is clamped horizontally in a retort stand. A vacuum/pressure station (CP. Instruments, Bishop's Stotford, England), connected to the Sintaglass funnel by rubber tubing, is used to draw air or vapour through the apparatus. The vapour being tested is drawn from a flask to which air is admitted by a tube extending below the surface of the test chemical and open to the atmosphere at its other end. The air-flow through the apparatus may be adjusted by altering the vacuum pressure and in practice a flow rate of 0.7 cm<sup>3</sup> s<sup>-1</sup> was used. A simple gas pipette can be used to extract vapour samples from the rubber tubing connecting the apparatus with the vacuum/pressure station if required. Samples can be analyzed by gas chromatography and thus a concentration time product (C.T.P.; Page and Lubatti, 1963) can be calculated for the chemical being tested. Furthermore, such a pipette can be used to donate a specific concentration of volatile, as a single dose, in the air passing through the *ssm* column.

The method described here is particularly useful when chemicals with low vapour pressure characteristics are being tested for nematoxic effects. Also, it can be easily modified to facilitate the exposure of soil insecta to the vapour-phase of chemicals. Several sets of apparatus can be operated simultaneously from a single vacuum/pressure station. Care must be taken to exhaust volatiles emanating from the system safely out of the laboratory. Some vapours may react with the perspex and/or rubber components in the apparatus e.g. petroleum vapours will necessitate the replacement of the rubber by neoprene and the perspex by glass components. It should also be noted that under certain conditions some rubber and other tubings can liberate volatiles.

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