

# THE CHROMATOGRAPHIC DETERMINATION OF ORGANOPHOSPHORUS PESTICIDES

## PART III. THE EFFECT OF IRRADIATION ON THE PARENT COMPOUNDS\*,\*\*

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The effect of light on pesticides is of considerable interest to the residue analyst as it is one of the mechanisms by which break-down products are produced under natural conditions<sup>1-3</sup>. The analyst confronted with the problem of analyzing for pesticide residues in foodstuffs must obviously be prepared to look for such break-down products in addition to the parent compounds. Fortunately this light effect can be conveniently simulated and studied under laboratory conditions.

The ultra-violet irradiation of organophosphorus pesticides has been studied by several workers<sup>2</sup>. COOK<sup>4,5</sup> showed that irradiation converted several pesticides to more polar compounds but was unable to characterize these. Subsequently COOK AND OTTES<sup>6</sup> reported the formation of less polar compounds when the pesticides carbo-phenothion, ethion and phorate were irradiated. A comprehensive study by MITCHELL<sup>7</sup> indicated that most organophosphorus pesticides break down to produce a variety of new compounds on irradiation but no attempt at characterization was made.

The majority of commercially important organophosphorus pesticides contain sulphur in the molecule, either bonded directly to the phosphorus atom as a thiono or thiolo group or occurring as a thio-ether group in the side chain. Compounds containing both groupings can theoretically form a wide range of stable oxidation products and several compounds of this type were studied.

Whereas paper chromatography was used in all the above studies to separate irradiation products from the parent compounds, in this study the speed and versatility of thin layer chromatography has been applied to the problem. In addition gas chromatography using a "specific" phosphorus detector has enabled characterization of the irradiation products to be made.

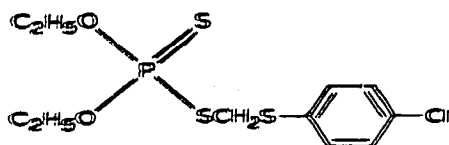
### EXPERIMENTAL

#### *Irradiation*

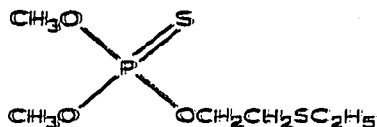
If irradiation is to cause appreciable break-down, thin films of pesticide must be exposed. This can be done either by coating a glass plate or paper with the pesticide or alternatively can be simulated by exposure of the pesticide at the surface of a

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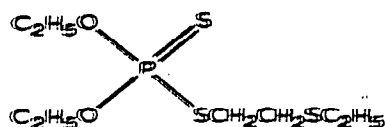
\*\* For Parts I and II of this series, see refs. 8 and 9.



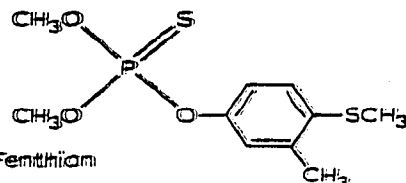
Carbophenothion



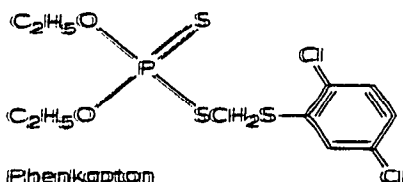
Demeton-O-methyl



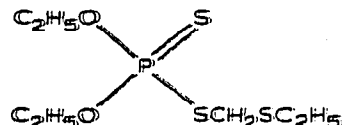
Disulfoton



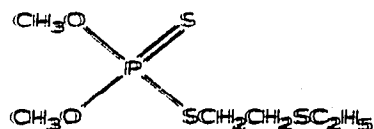
Fenitrothion



Phenkapton



Phorate



Thiometon

solution. Preliminary experiments indicated that the latter technique was technically much easier to carry out and gave results of greater reproducibility. Acetone was found to be preferable to hexane as a solvent, the latter being a poor solvent for the more polar compounds produced during irradiation. The following irradiation procedure was used:

Stock solutions containing 100  $\mu\text{g}$  of pesticide per ml in acetone were prepared and 10 ml aliquots placed in glass beakers of 3 cm diameter. A series of beakers was placed under a Camag Universal U.V. lamp (Type T.L. 900) and irradiated with light of wavelength maximum 2540 Å. The surface of the pesticide solutions was 2 cm from that of the lamp. Evaporation of acetone occurred during the course of the irradiation as the beakers were not covered in any way. The volume was readjusted at the end of each day's irradiation by addition of more solvent. Samples were removed at timed intervals, transferred to test tubes and stored in a refrigerator. The samples obtained were subsequently examined by gas and thin layer chromatography.

### Gas chromatography

The chromatographic equipment and columns used have been previously described<sup>8,9</sup>. As compounds of longer retention time than the parent pesticides were frequently produced by irradiation, column temperatures higher than those previously recommended<sup>8</sup> were used in some cases. Oxidation products of the pesticides were prepared using potassium permanganate in acetone. Preparation of this oxidant and the procedure for its use were described previously<sup>8</sup>. Chromatographic comparison of the oxidized and irradiated pesticides was then made by injecting 5  $\mu\text{l}$  aliquots of suitably diluted solutions on to the two columns. The decay pattern of each pesti-

cide was also determined by injecting aliquots of each of the irradiated samples in turn and comparing the peak heights of the various component peaks on the chromatogram.

#### *Thin layer chromatography*

Several visualization reagents are available for the detection of organophosphorus pesticides on thin layer chromatograms; ABBOTT AND EGAN<sup>10</sup> have recently reviewed this topic. Prior to the work reported here three visualization reagents were examined, brilliant green/bromine<sup>11</sup>, palladous chloride<sup>12</sup> and 4-(*p*-nitrobenzyl)-pyridine<sup>13</sup>. The first of these reagents was found to provide a sensitive and rapid method for detecting the compounds encountered in this study and was used subsequently in the examination of the U.V. irradiated pesticides. The palladous chloride reagent required a long period of development and the 4-(*p*-nitrobenzyl)-pyridine reagent proved insensitive to some of the irradiation products.

BLINN<sup>14</sup> has recommended the use of pH 6.0 buffer in preparing thin layer plates for this type of study, to prevent the hydrolytic decomposition of organophosphorus esters. In this work the plates were prepared by shaking 30 g Silica Gel G with 60 ml of a pH 6.0 buffer (10.2 g potassium hydrogen phthalate and 1.82 g sodium hydroxide per litre). The plates were coated with a 250  $\mu$  thick layer and were oven dried at 120° for at least 2 h before use.

Aliquots of the irradiated and oxidized samples, equivalent to 100  $\mu$ g of the original pesticide, were evaporated to low volume and spotted onto the plates in duplicate, together with the standards available. The plates were developed with one of the following solvent systems:

(I) Acetone-chloroform (1:9, v/v),

(II) Methyl ethyl ketone-light petroleum (boiling range 40–60°) (1:1, v/v).

One portion of the developed plates was sprayed with the brilliant green reagent and visualized by treatment with bromine vapour. The second portion of the plate was covered during the visualization process and those areas corresponding to the irradiation products were removed, eluted with chloroform and examined by gas chromatography.

#### RESULTS

Typical chromatograms of the parent pesticides, the irradiated pesticide and the oxidized compounds, all at the same dilution and sensitivity setting, are shown in Fig. 1. Table I lists the thin layer characteristics of the compounds. The decay curves for each pesticide are shown in Fig. 2.

#### DISCUSSION

From the gas chromatographic and thin layer data (Fig. 1, Table I), it is apparent that the organophosphorus pesticides decompose on irradiation to produce compounds identical in their characteristics with the  $\text{KMnO}_4$  oxidation products, however additional irradiation products are produced which do not correspond to known oxidation products. The decay curves (Fig. 2) show that the parent compounds are fairly rapidly decomposed but the irradiation products are more persistent.

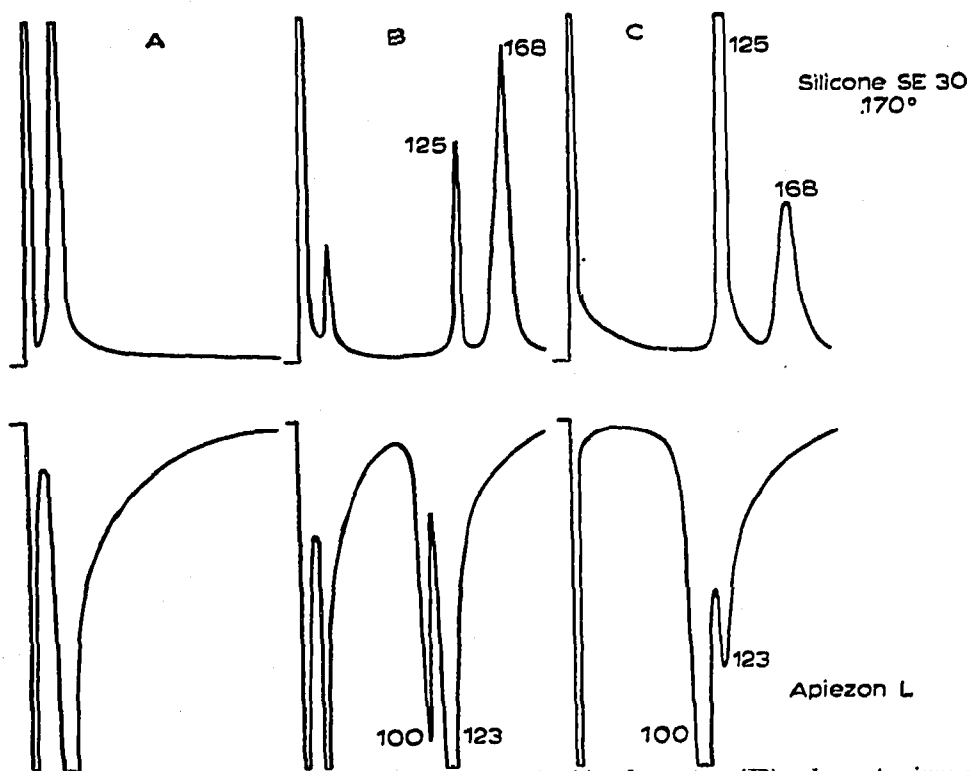


Fig. 1. Gas-liquid chromatography of (A) phorate, (B) phorate irradiated 24 h, and (C) phorate oxidized with  $KMnO_4$ .

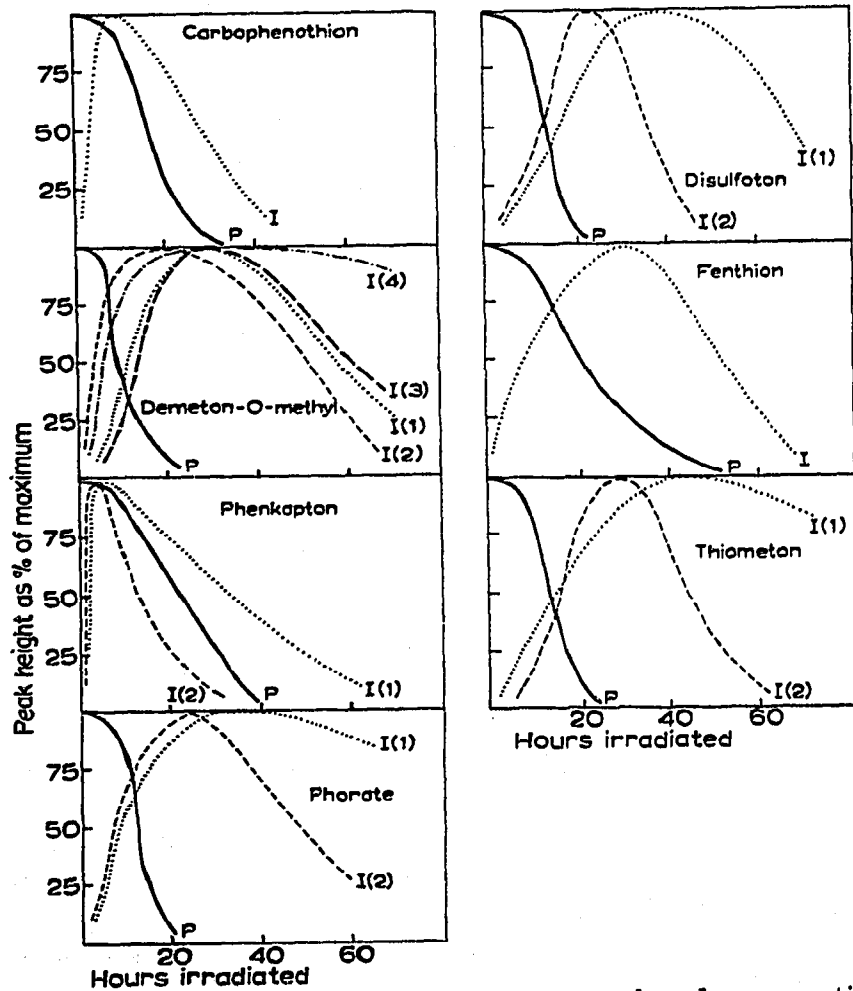


Fig. 2. Effect of irradiation on the organophosphorus pesticides. P refers to parent compound.

TABLE I

 $R_F$  VALUES OF PESTICIDES AND THEIR IRRADIATION AND OXIDATION PRODUCTS

I indicates an irradiation product; O indicates an oxidation product; \* indicates the major component in terms of peak height.

Pesticide	$R_F \times 100$		Retention time	
	Solvent I	Solvent II	SE 30 column	Apiezon L column
Carbophenothion	94	98	290	450
I	86	94	1020	1120
O(1)	89	95	785	875
O(2)	86	94	1020	1120
Demeton-O-methyl	91	97	15	8
Demeton-S-methyl	79	71	32	20
Demeton-S-methyl sulphoxide	10	4	Not detected	Not detected
Demeton-S-methyl sulphone	40	25	Not detected	Not detected
* I(1)	81	86	Not detected	58
* I(2)	41	13	Not detected	91
I(3)	41	22	Not detected	190
I(4)	Not detected	Not detected	Not detected	101
O(1)	81	86	Not detected	58
O(2)	41	22	Not detected	190
O(3)	Not detected	Not detected	Not detected	101
Disulfoton	93	95	41	42
* I(1)	87	87	280	210
I(2)	55	19	Not detected	290
I(3)	13	35	Not detected	Not detected
O(1)	87	87	280	210
O(4)	55	19	Not detected	290
Fenthion	94	96	92	108
I	51	28	680	485
O(1)	87	92	470	375
O(2)	51	28	Not detected	485
Phenkapton	93	98	510	820
* I(1)	89	97	1200	1450
I(2)	Not detected	Not detected	Not detected	720
O	89	97	1200	1450
Phorate	93	99	25	25
Phorate-O-analogue	87	82	24	19
Phorate-O-analogue sulphone	54	49	Not detected	93
I(1)	86	94	125	100
* I(2)	62	45	168	123
O(1)	86	94	125	100
O(2)	62	45	168	123
Thiometon	98	96	29	28
* I(1)	95	87	Not detected	180
I(2)	45	17	Not detected	255
I(3)	10	4	Not detected	Not detected
O(1)	95	87	Not detected	180
O(2)	45	17	Not detected	255

### Phorate

The oxidation of phorate has been discussed previously<sup>8</sup> and there is strong evidence to suggest that the oxidation products are the sulphoxide and sulphone of the parent compound. The sulphoxide, being more polar, would be expected to show thin layer characteristics corresponding to O(2) (Table I) and the sulphone to O(1). The retention time data of BACHE AND LISK<sup>15</sup>, who examined pure standards by gas chromatography, support this characterization.

Hence I(1), which is probably phorate sulphone, and I(2), phorate sulphoxide, are the two irradiation products, with the sulphone being the more persistent, but the sulphoxide being in greater quantity during the early stages of the irradiation.

### Disulfoton

By analogy with phorate it seems probable that

O(1) = disulfoton sulphone = I(1),

O(2) = disulfoton sulphoxide = I(2).

The sulphone again is the persistent irradiation product. Essentially similar results have been found in field trials in which phorate and disulfoton were sprayed on kale<sup>16</sup>.

Irradiation product I(3) could not be characterized, but may well be a further oxidation product (disulfoton-O-analogue sulphone or sulphoxide) which does not give a gas chromatographic peak under the conditions used:

### Thiometon

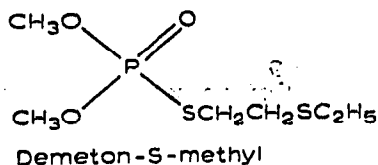
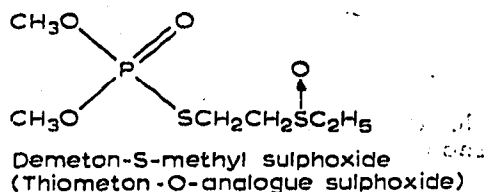
Again by analogy with phorate it seems likely that

O(1) = thiometon sulphone = I(1);

O(2) = thiometon sulphoxide = I(2).

The decay pattern is similar to the two compounds above with the sulphone persisting for longer periods than the parent compound or sulphoxide.

Irradiation product I(3) has the same characteristics as demeton-S-methyl sulphoxide.



### Demeton-O-methyl

Demeton-O-methyl as such is not listed as a pesticide<sup>8</sup>. It can, however, occur as an impurity in demeton-S-methyl, to which it can isomerize.

It is apparent from Table I, however, that irradiation could produce new compounds three at least of which (I(1), I(3), I(4)), are apparently oxidation products of greater stability than the parent compound. These compounds might well be detected on crops sprayed with the demeton-O/demeton-S-methyl mixture.

### Fenthion

The irradiation product of fenthion, which corresponds to O(2), may well be the sulphoxide, but the presence of a substituted phenyl group in the side chain does

not allow a direct comparison to be made with other organophosphorus pesticides, e.g. phorate.

#### *Carbophenothion and phenkapton*

The thin layer separation of these compounds from their oxidation products was very poor with the two solvents used in this study. In the case of carbophenothion the two oxidation products are only just separable but the irradiation product does apparently correspond to the more polar of the two (sulphoxide = I(1) = O(2)). In the case of phenkapton no correlation can be made.

The chromatograms of several of the irradiated pesticides show peaks of shorter retention time than the parent compound, indicating the formation of more volatile and possibly less polar break-down products. Neither the gas chromatographic or thin layer conditions used were suitable for the assessment of these materials and they have not been investigated. In addition, the possibility that materials formed were undetectable both by the thin layer visualization reagent and gas chromatography cannot be ruled out.

#### CONCLUSION

The irradiation of seven pesticides resulted in their decomposition, with new compounds being formed which corresponded to their oxidation products in gas and thin layer chromatographic characteristics. The irradiation products were more stable than the parent pesticides in most cases and if similar behaviour occurs under field conditions the residue analyst must be prepared to determine these products. Gas chromatography with the "specific" phosphorus detector provides a convenient method for the detection of many of these products.

#### SUMMARY

One of the mechanisms by which metabolites of the organophosphorus pesticides are produced is by the effect of light. In this part of the series the authors have studied the products produced by the action of light on the pesticides and correlated these products with the products of oxidation.

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