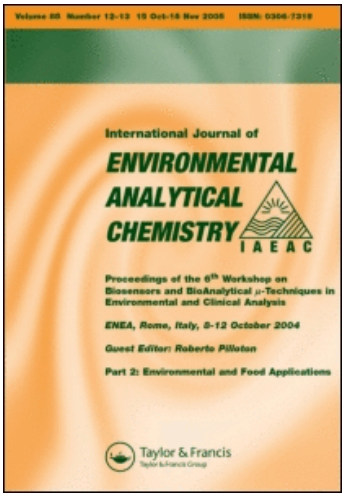


International Journal of Environmental Analytical Chemistry

Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713640455

Electron Donor-Acceptor Complexing Reagents for the Detection of Pesticides on TLC

James D. Macneil[a]; R. W. Frei[a]; O. Hutzinger[b]
[a] Dalhousie University Halifax, Nova Scotia, Canada [b] Atlantic Regional Laboratory National Research Council of Canada, hali, Nova Scotia, Canada

To cite this Article Macneil, James D. , Frei, R. W. and Hutzinger, O.(1972) 'Electron Donor-Acceptor Complexing Reagents for the Detection of Pesticides on TLC', International Journal of Environmental Analytical Chemistry, 1: 3, 205 — 220
To link to this Article: DOI: 10.1080/03067317208076372
URL: http://dx.doi.org/10.1080/03067317208076372

Intern. J. Environ. Anal. Chem., 1972, Vol. 1. pp. 205–220

Printed in Great Britain

Electron Donor-Acceptor Complexing Reagents for the Detection of Pesticides on TLC†

II. Carbamates, Anilides and Ureas. Spray Reagents, Chromatography and Instrumental Techniques

JAMES D. MACNEIL and R. W. FREI
Dalhousie University
Halifax, Nova Scotia, Canada

O. HUTZINGER
Atlantic Regional Laboratory
National Research Council of Canada
Halifax, Nova Scotia, Canada

(*Received March* 12, 1971)

The formation of π-complexes to detect and identify a representative group of carbamate, anilide and urea pesticides was investigated. Results obtained with several electron-acceptor reagents used by previous workers are compared with those obtained with a group of nitro-substituted fluorene derivatives. The pesticides studied were chosen for their potential ability to act as electron donors. Detection limits after thin-layer chromatography ranged from 0.5 to 5.0 micrograms, depending upon the strength of the complex formed. Various chromatographic systems were studied and applications of *in situ* reflectance spectroscopy and mass spectrometry were investigated. The method combines a reasonable sensitivity with good selectivity and the pesticide may be recovered from the complex for further study. This non-destructive method of analysis should be of particular use in the separation and identification of these pesticides and their metabolites.

†First presented at the Symposium on Recent Advances in the Analytical Chemistry of Organic Pollutants; 54th Canadian Chemical Conference, Halifax, May 31—June 2, 1971.

INTRODUCTION

Carbamates and structurally related compounds, such as ureas, comprise a major group of pesticides. Thin-layer chromatography is particularly suitable for the analysis of these compounds, especially the *N*-methyl carbamates which are known to decompose when examined by GLC using the recommended conditions.[1,2] Thus the development of TLC methods for rapid screening with a sensitivity and selectivity of GLC would seem attractive for these compounds.

The thin-layer chromatographic separation of a number of pesticides, including some carbamates, on silica gel G has been reported by Walker and Beroza.[3] Fishbein and Fawkes investigated the separation of 3,4-methylenedioxyphenyl and 3,4-methylenedioxybenzyl carbamates on silicic acid layers and on prepared Eastman Chromagram silica-gel sheets.[4] Henkel studied the separation of insecticidal carbamates on silica gel G and on polyamide layers.[5] Thin-layer chromatographic methods for a number of carbamate and urea pesticides have been reported by Abbott *et al.*[6] and by Finocchiaro and Benson.[7] Suitable solvent systems for the separation of urea herbicides on silica-gel plates were investigated by Hance.[8] More recently, the thin-layer chromatographic separation of carbamate pesticides has been reported by Ramasamy,[9] El-Dib,[10] and Mendoza and Shields.[11]

While some of these methods are highly sensitive,[11] and generally most give detection limits in the microgram range or lower after chromatography, there are certain disadvantages associated with their use. These problems lie primarily in the detecting reagents, many of which serve only to visualize the pesticide, so that the chromatographic R_f value must be used as the parameter by which identification is made. Thus, the development of reagents having higher specificity to provide a further means of identifying any pesticide in these groups should be of interest.

A more serious problem with most reagents previously used for the detection of these compounds is that the process of detection destroys the pesticide. A reagent which would permit the recovery of the pesticide for confirmation by mass spectrometry after preliminary identification from the R_f of the pesticide and the colour it produced with the reagent would have obvious advantages.

The use of reagents which form π-complexes with pesticides has previously been reported and these meet the requirements outlined above.[12] The colour of the complex formed aids in the identification of the pesticide, which may then be regenerated from the complex for further analysis. As most phenylcarbamate and phenyl-urea pesticides are, from structural considerations, potentially electron donors, they seemed highly suitable for the study of this method of detection. The work was carried out on Eastman Chromagram

silica-gel and cellulose sheets, for which suitable solvent systems had to be found. The pesticides were also spotted and sprayed on alumina sheets, but no chromatographic separations were attempted on alumina.

EXPERIMENTAL

Pesticide standards

Standard solutions of the pesticides described in Table I were prepared (1000 ppm in ethanol). (Acetone was the solvent originally chosen, but at least some of the compounds, notably Zectran and Matacil, degrade in acetone solution within a few days.) The chromatographic sheets were spotted using a 10-mcl Hamilton microsyringe or Drummond microliter pipets. Results were obtained on Eastman Chromagram silica-gel sheets 6061 and Eastman Chromagram cellulose sheets 6064.

Reagents

The spray reagents 2,4,5,7-tetranitro-9-fluorenone (TetNF), tetracyanoethylene (TCNE) and 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) were obtained from Aldrich Chemical Co. 2,4,7-Trinitro-9-fluorenone (TNF) and 9-dicyanomethylene-2,4,7-trinitrofluorene (CNTNF) were obtained from Eastman Organic Chemicals.

9-Dicyanomethylene-2,4,5,7-tetranitrofluorene (CNTetNF) and 7,7,8,8-tetracyanoquinodimethane (TCNQ) were synthesized in our laboratory.[12] The latter is also available from Eastman Organic Chemicals as 2,5-cyclohexadiene-Δ1,α:4,α'-dimalononitrile.

Freshly prepared 1% solutions of the reagents in acetone were made before use. DDQ was prepared in methylene chloride.

Chromatographic solvent systems

The following solvent systems were studied for the separation of various representative carbamates and ureas on Eastman Chromagram silica-gel sheets 6061:

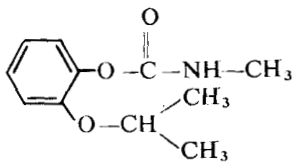
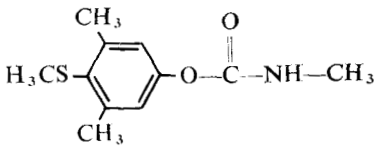
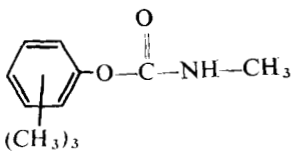
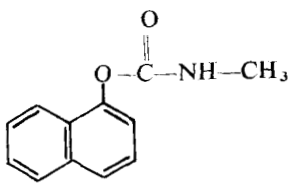
1) isopropyl ether–toluene (1:3)
2) ethyl ether–toluene (1:3)
3) benzene–acetone (14:1).

The same group of compounds was also studied on Eastman Chromagram cellulose sheets 6064 using the following solvent systems:

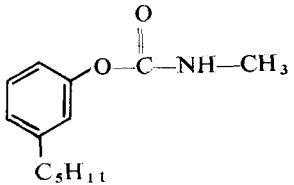
4) heptane–isopropyl ether (5:1)
5) isooctane–acetic acid (100:1).

TABLE I

Classification and structures of pesticides investigated

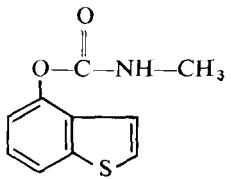
Pesticide	Formula	Chemical name
Group I, aryl N-methyl carbamates:		
Baygon		2-Isopropoxyphenyl-*N*-methylcarbamate
Bay 37344 (esurol)		4-(Methylthio)-3,5-xylyl-*N*-methylcarbamate
Landrin		Isomer mixture: 2,3,5-Trimethyl- and 3,4,5-trimethylphenyl *N*-methylcarbamate
Carbaryl (Sevin)		1-Naphthyl *N*-methylcarbamate

BUX



Isomer mixture:
m-(1-Methylbutyl)phenyl *N*-methylcarbamate (75%) and *m*-(1-ethylpropyl)phenyl *N*-methylcarbamate (25%)

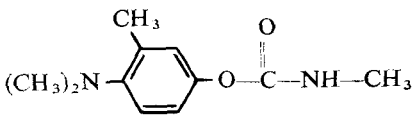
Mobam



4-Benzothienyl *N*-methylcarbamate

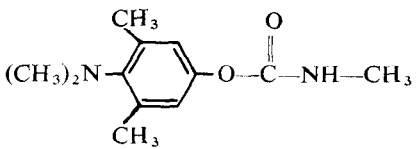
Group II, N-methyl carbamates containing tertiary amines:

Matacil (Bay 44646)



4-(*N*,*N*-Dimethylamino)-*m*-tolyl *N*-methylcarbamate

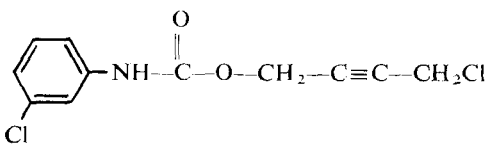
Zectran



4-(*N*,*N*-Dimethylamino)-3,5-xylyl *N*-methylcarbamate

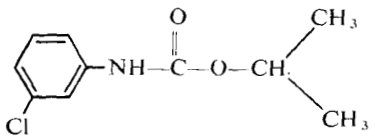
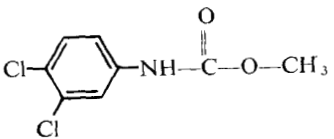
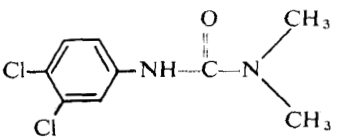
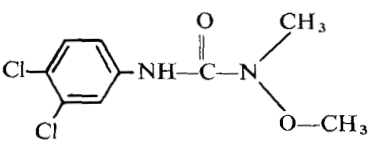
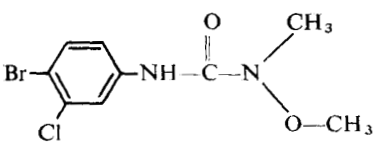
Group III, chlorine containing carbamates and ureas:

Barban



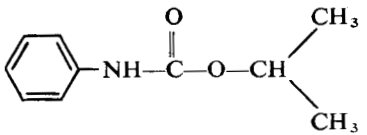
4-Chloro-2-butynyl *N*-(3-chlorophenyl)carbamate

TABLE I—(*contd.*)

Pesticide	Formula	Chemical name
CIPC		Isopropyl *N*-(3-chlorophenyl)carbamate
Swep (NIA 2995)		Methyl *N*-(3,4-dichlorophenyl)-carbamate
Diuron		3-(3,4-Dichlorophenyl)-1,1-dimethylurea
Linuron		3-(3,4-Dichlorophenyl)-1-methoxy-1-methylurea
Maloran (C-6313)		3-(4-Bromo-3-chlorophenyl)-1-methoxy-1-methylurea

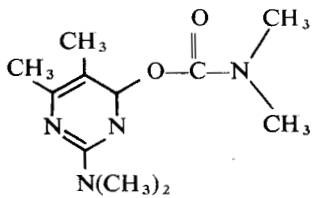
Group IV, Miscellaneous carbamates, ureas and thioureas:

IPC



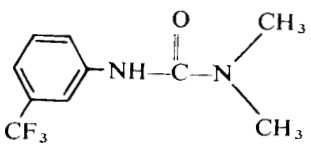
Isopropyl *N*-phenylcarbamate

Pirimicarb



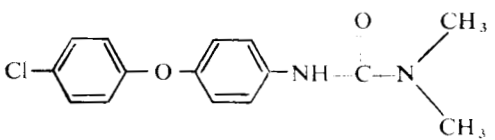
5,6-Dimethyl-2-dimethylamino-4-pyrimidinyl *N*,*N*-dimethylcarbamate

Fluometuron



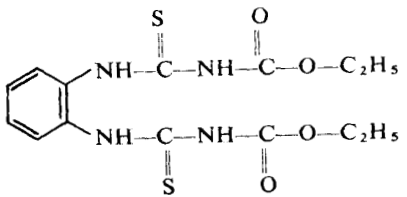
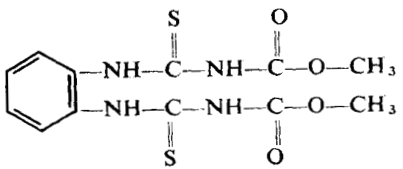
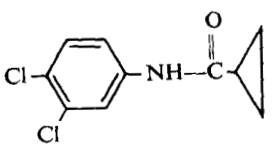
1,1-Dimethyl-3-(α,α,α-trifluoro-*m*-tolyl)urea

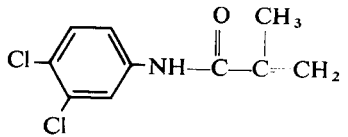
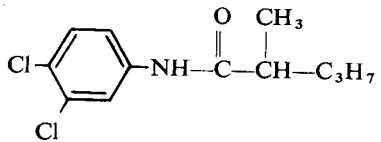
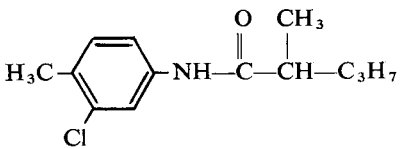
Chloroxuron



3-[*p*-(*p*-Chlorophenoxy)phenyl]-1,1-dimethylurea

TABLE I—(*contd.*)

Pesticide	Formula	Chemical name
Thiophanate		1,2-Bis-(3-ethoxycarbonyl-2-thioureido)benzene
Thiophanate-methyl		1,2-Bis-(3-methoxycarbonyl-2-thioureido)benzene
Group V, Anilides:		
Cypromid		*N*-(3,4-Dichlorophenyl)cyclopropancarboxamide

Dicryl		*N*-(3,4-Dichlorophenyl)-2-methylpropenamide
Karsil		*N*-(3,4-Dichlorophenyl)-2-methylpentanamide
Solan		*N*-(3-Chloro-4-methylphenyl)-2-methylpentanamide

Chromatograms were developed at room temperature using an Eastman Chromagram Developing Apparatus 6071.

Instrumentation

Reflectance spectra were obtained on the Farrand UV-VIS chromatogram analyzer. The monochromator on the analyzer leg was removed and instead a suitable auxiliary filter (7-54, 3-73, 3-74, or 3-75) was used. The 325-800 u.v.-vis. lens was placed in the exciter drawer with a 0.625 screen mesh aperture reducer as recommended by the company for operation in this mode. A IP28 phototube was used as a detector, with a xenon lamp being the source of radiation. Spectra were obtained by plotting the reflectance characteristics of the sprayed plate background and of the complex at 10-nm intervals. Spectra were recorded on the instrument's 45% scale.

Mass spectra were obtained with a DuPont/CEC 21-110B instrument using standard probes for direct introduction of the sample into the ion source. Complexes were transferred directly from the cellulose chromatograms or after elution with acetone from silica-gel chromatograms to a mass spectrometer sample tube. From 2–4 mcg of pesticide were used for the experiment.

RESULTS AND DISCUSSION

Complex formation

All colours observed after spraying with the various complexing agents are recorded in Table II. No great variations in colour were observed between silica-gel and cellulose thin layers. The complexes were viewed under u.v. lamps (Ultra-Violet Products Inc. UVS-11 and UVS-22). Fluorescence quenching of the background fluorescence of the spray was observed for CNTNF and CNTetNF. While tests with other pesticides have shown that this is not specific to carbamates and ureas, all the compounds tested for this paper produced an absorption band (fluorescence quenching) at 370 nm with CNTNF. As the fluorescence quenching is more sensitive than the visible colours, this could be used to lower detection limits for the carbamates tested here to the submicrogram level.

Visual detection limits of the coloured complexes on the plate were between 0.5 mcg and 5 mcg, depending upon the reagent used, the structure of the pesticide and the nature of the layer, with best results being obtained on cellulose.

The general order of efficiency of the acceptors tested, considering both acceptor strength and stability of the complex formed was: CNTNF > CNTetNF > TNF > TetNF > DDQ > TCNQ > TCNE.

TABLE II

Colors of pesticides obtained with complexing reagents

Pesticide	TNF	TetNF	CNTNF	CNTetNF	TCNE	TCNQ	DDQ
Barban	Y	Y	O	LO	P	Y-Br	P
Baygon	Y	LBr	LR	LBr	LBr	Y	P
BUX	Y	LBr	—	—	T	—	Br
Carbaryl	Y-Br	O	R	Gy-Br	P	LGr	Bu
Chloroxuron	Br	Br	Gy	Gy	Gy	LGr	Bu
CIPC	Y	Y	O	R-Br	Br	LGr	P
Cypromid	Y	Y-Br	LR	Br	R-Br	Br	P
Dicryl	Y-Gr	Y	Go	Br	Br	LGr	LMv
Diuron	Y-Br	Br	R	Gy	Gr	LGr	Gy
Fluometuron	Y	Y-Br	LBr	T	LBr	T	P
IPC	Y	Y	O	R-Br	Br	T	P
Karsil	Y-Gr	Y	Go	Br	Br	LGr	Mv
Landrin	Y	Y	O	O	Br	Br	R-P
Linuron	Y	O	R	R-Br	Y-Gr	LGr	Gy
Maloran	Y	Y-Br	O	Gy	R-Br	LGr	Mv
Matacil	Br	Gy	Gy	Gy	Gr	LGr	DBr
Mesurol	Y	Y	LO	LO	LBr	Y-Br	P
Mobam	Y	O	R	LBr	Br	LGr	P
Pirimicarb	R	P	Mv	LGy	Gr	LGr	Bu
Solan	Y-Gr	Y-Br	LR	Br	DR	Ol	DBu-Gy
Swep	Br	Gy	O	Gy	LGr	LGr	DBr
Thiophanate	Y	Y-Br	Y	R-Br	O	Br	P
Thiophanate-Methyl	Y	Y	Y	LBr	O	Br	P
Zectran	Gy	Gy	Gy	Gy	Gr	Br	Gy

Colour abbreviations: Y=yellow; O=orange; P=purple; Br=brown; R=red; Gy=grey; Gr=green; Bu=blue; Mu=mauve; Go=gold; Ol=olive; L=light; D=dark; T=trace; – =no colour development.

DDQ, TCNQ and TCNE complexes lose colour rapidly as they dry, indicating that the complex formation seen with these compounds may be a preliminary step to a reaction. The complexes formed with the nitro-fluorene derivatives were found to be very stable and retained their colour with only a small loss in intensity for days.

Chromatographic separations

Generally, better separations were obtained on silica-gel sheets than on cellulose sheets, but for most applications the solvent systems used on cellulose should be acceptable. R_f values for a selection of pesticides using the five chromatographic systems selected for study are given in Table III.

TABLE III

R_f values of some selected pesticides

Pesticide	$R_f \times 100$[a] Solvent system (1)[b]	(2)[b,c]	(3)[b]	(4)[d,e]	(5)[d,f]
Barban	34	49	78	79	55
Baygon	79	82	90	75	71
Carbaryl	35	46	73	67	63
Chloroxuron	5	9	38	17	16
Diuron	6	12	41	23	19
Matacil	26	40	70	77	67
Mesurol	42	54	79	76	70
Mobam	35	45	68	62	56
Pirimicarb	17	33	70	87	75
Swep	58	65	79	89	72
Thiophanate-methyl	7	18	44	3	3

[a] Average of four determinations.
[b] On silica-gel thin-layer sheets.
[c] Used previously on alumina layers.[7]
[d] On cellulose thin-layer sheets.
[e] Ureas streak in this system, while carbamates give defined spots with some slight tailing.
[f] Ureas streak in this system, while carbamates show a tendency to tail.

Reflectance spectra

Reflectance spectra were obtained for a number of complexes and some typical maxima are presented in Table IV. Complex bands measured *in situ* with the nitro-fluorene derivatives as acceptors tended to exhibit very broad maxima. Complexes formed with DDQ, TCNQ and TCNE as acceptors tended to have narrower maxima, but were more difficult to measure due to their rapid fading. Representative spectra obtained for several complexes are given in Figure 1.

TABLE IV

In situ reflectance data for selected CNTNF pesticide complexes

Pesticide	λ_{max}[a] (cellulose) (nm)	λ_{max}[a] (silica) (nm)
Carbaryl	500	490
Mobam	500	490
Diuron	500	490
Linuron	490	480
Maloran	490	470
Barban	490	470
Thiophanate	470	450

[a] λ_{max} is reproducible to within ± 5 nm.

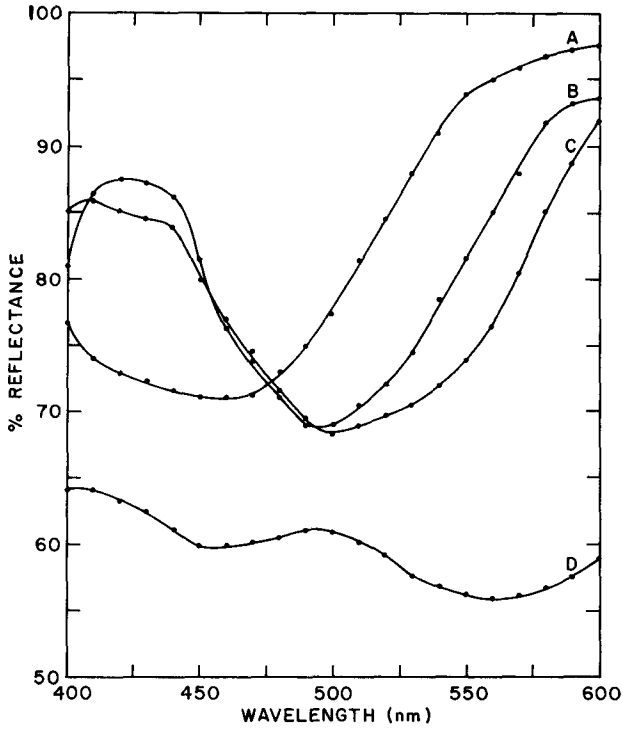


FIGURE 1 Reflectance spectra of complexes measured *in situ* A, Thiophanate-methyl–CNTNF; B, Carbaryl–CNTNF; C, Mobam–CNTNF; D, α-naphthol–CNTNF.

Mass spectrometry

An added advantage gained with the use of the nitrofluorene derivatives is that these complexes may be decomposed thermally in the probe of a mass spectrometer so that two separate spectra, one at a lower temperature for the pesticide and one at a higher temperature for the complexing agent, are obtained.[13] Spectra of carbamates obtained from these complexes were compared with spectra previously tabulated for these compounds.[14] A mass spectrum of carbaryl obtained in this fashion is presented in Figure 2. Detection at the microgram level and even lower is possible, with the visual detection limit of the complex on the thin layer being the limiting factor.

CONCLUSIONS

The techniques described in this paper are suitable for the analysis of carbamate and urea residues where a sample of 0.5 mcg or more (depending

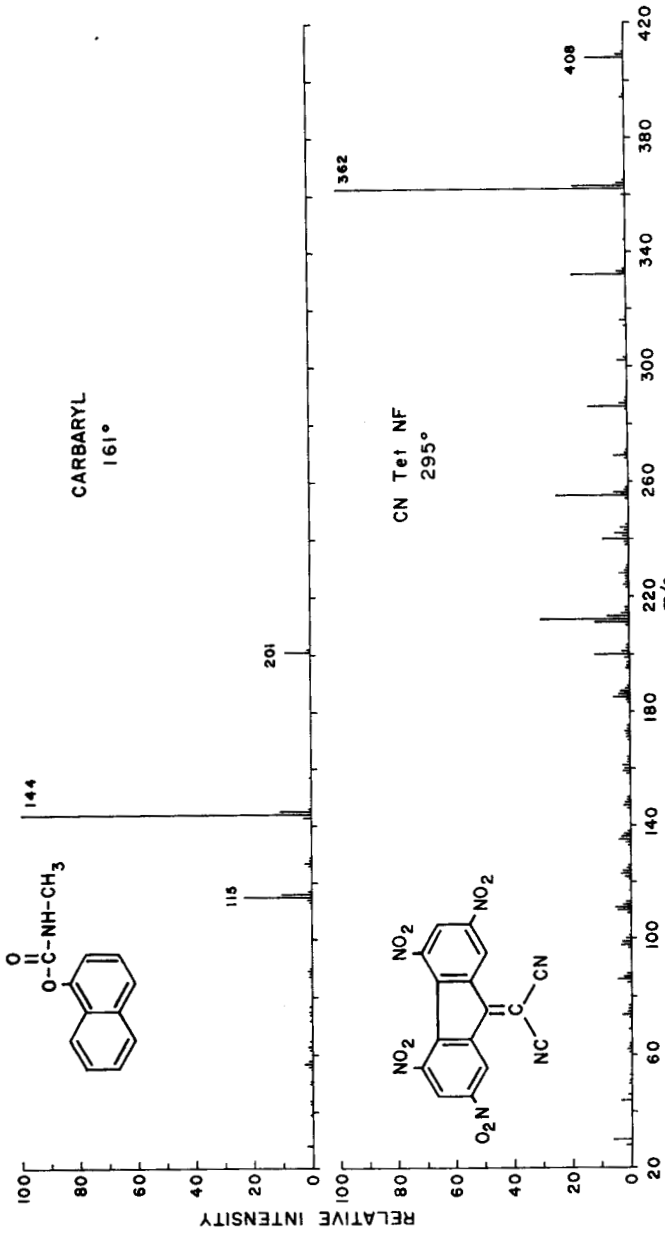


FIGURE 2 Mass spectra of carbaryl CNTetNF complex at different sample temperatures.

on the pesticide) is to be determined. With most of the pesticides studied (carbaryl and mobam excepted), best results would be obtained where at least 5 mcg of the pesticide may be spotted if the separation is on silica gel, whereas 1.0–2.0 mcg of most of these pesticides may be readily detected on cellulose thin layers. However, CNTNF could be used to detect these pesticides under a u.v. lamp in submicrogram amounts.

The methods described have excellent possibilities for metabolic studies, as many of the products of the decomposition of carbamate and urea pesticides are anilines and phenols, and therefore are actually better electron donors than the parent pesticides. The detection of anilines and phenols with the reagents used in this study has been reported previously[13] and it is our intention to test the application of the techniques described in this paper to a metabolic problem.

The technique has the advantages that it is sensitive to microgram quantities of the pesticides investigated; it is selective to a greater degree than some other spray methods; it is nondestructive; and instrumental techniques such as *in situ* reflectance spectroscopy and mass spectrometry may be used to identify and analyze the pesticide after complexation. The development of better chromatographic solvent systems for the separation of carbamates and ureas on cellulose sheets would be of advantage, as these reagents have a considerably better sensitivity on cellulose.

The nitro-fluorene derivatives studied are generally better acceptors than such compounds as DDQ, TCNQ and TCNE, yielding complexes with much greater stability.

Acknowledgements

The authors are grateful to the companies which supplied the analytical-grade pesticide samples used in this study. The loan of a Farrand UV-VIS chromatogram scanner by the Farrand Company is much appreciated. J. D. M. and R. W. F. are grateful for support through grants from the Canada Department of Agriculture and the National Research Council of Canada. Thanks are also due to Dr. W. D. Jamieson and D. J. Embree for the mass spectra.

References

1. W. R. Benson, *J. Ass. Offic. Anal. Chem.* **53**, 351 (1970).
2. *Pesticide Analytical Manual* (Food and Drug Administration, Washington, D.C., 1968), Vol. I, Table 311.5A.
3. K. C. WALKER and M. BEROZA, *J. Ass. Offic. Anal. Chem.* **46**, 250 (1963).
4. L. Fishbein and J. Fawkes, *J. Chromatog.* **20**, 521 (1965).
5. H. G. Henkel, *J. Chromatog.* **21**, 346 (1966).
6. D. C. Abbott, K. W. Blake, K. R. Tarrant, and J. Thompson, J. Chromatog. **30**, 136 (1967).

7. J. M. Finocchiaro and W. R. Benson, *J. Ass. Offic. Anal. Chem.* **50**, 888 (1967).
8. R. J. HANCE, *J. Chromatog.* **44**, 419 (1969).
9. M. Ramasamy, *Analyst* **94**, 1075 (1969).
10. M. A. El-Dib, *J. Ass. Offic. Anal. Chem.* **53**, 756 (1970).
11. C. E. Mendoza and J. B. Shields, *J. Chromatog.* **50**, 92 (1970).
12. J. D. MacNeil, R. W. Frei, O. Hutzinger, and W. D. Jamieson, Submitted to *J. Ass Offic. Anal. Chem.*
13. O. Hutzinger, *Anal. Chem*, **41**, 1662 (1969).
14. J. N. Damico and W. R. Benson, *J. Ass. Offic. Anal. Chem.* **48**, 344 (1965).