Determination of the Electron Impact Fragmentation Pattern of Methamidophos via Stable Isotope Enrichment

Charles M. Thompson, Takaaki Nishioka, and T. Roy Fukuto*

The mass spectra of isotopically enriched [*O*-*methyl*-^{13}C]- and [*S*-*methyl*-^{13}C]methamidophos (*O*,*S*-dimethyl phosphoramidothioate) have been recorded and fragmentation patterns have been interpreted. The use of the ^{13}C isotope showed that the *O*-methyl and *S*-methyl groups give different ion decomposition products. The neutral methyl radical was lost only from the *S*-methyl moiety and the methyl cation was predominantly formed from the *O*-methyl moiety. An ion decomposition pathway for methamidophos is proposed.

The use of stable isotope enrichment has been a common practice to aid in the elucidation of mass spectral fragmentation patterns. In particular, carbon-13 incorporation achieves the labeled product without the undesirable scrambling effects encountered when deuterium is employed (Budziekiewitz et al., 1967). Carbon-13 labeling of organophosphates has seen limited, if any, application in mass spectrometry (MS) for the analysis of fragmentation patterns and metabolite identification. Usually metastable ion and high-resolution MS are of value in these determinations. However, these methods are not useful when attempting to differentiate between two exact groups. In our study, differentiation between the *O*-methyl and *S*-methyl could only be achieved by selective isotope enhancement.

Previous reports from this laboratory have dealt with the mechanism of action of methamidophos. Quistad et al. (1970) was first to suggest that the P–S linkage is cleaved during the inhibition of acetylcholinesterase (AChE). Fahmy et al. (1974) demonstrated through hydrolysis studies that either the P–O or P–S linkage is cleaved, depending on solvent conditions, and consequently left this question open. In a recent paper, Thompson and Fukuto (1982) have shown that the in vitro inhibition of electric eel acetylcholinesterase proceeds with cleavage of the P–S bond. Toxicokinetic studies (Gray et al., 1982) have also supported this claim. It then became our intention to acquire spectral data to (a) provide pure physicochemical data free from such solvent effects (Fahmy et al., 1974) or ambiguous side reactions and (b) assist metabolite identification. Information obtained in objective a may provide relative bond strengths and patterns of degradation which could support results of previous investigations. This report is concerned with the mass spectral analysis of isotopically enriched [*O*-*methyl*-^{13}C]- and [*S*-*methyl*-^{13}C]methamidophos.

MATERIALS AND METHODS

General. Silica gel 60F-254 sheets (E. M. Reagents) of 0.2-mm thickness were used for analytical thin-layer chromatography (TLC). Solvent systems used for TLC were ethyl acetate–methanol (9:1 v/v) and dichloromethane–acetone (1:1 v/v). Spots were located by use of 0.5% 2,6-dibromoquinone-4-chloroimide (DBQ) in ether as a spray reagent (Menn et al., 1957). Silicic acid (Mallinckrodt, Inc., CC-7 Special) was used for column chromatography, eluting with a diethyl ether–acetone gradient.

Electron impact mass spectrometry (EIMS) was conducted by using a Finnigan Model 3300 mass spectrometer. The ion source temperature was 100 °C and the electron energy of the ion source was 70 eV. The samples were introduced by the direct insertion technique. EIMS data acquisition and reduction were performed by a System 150 computer (System Industries) at an integration time of 6 s/scan, scan range m/z 10–200, with a 1-s delay between scans.

Purity of the isotopically enriched samples (>99.6%) was obtained on the mass spectrometer itself as well as on a Varian 1440 gas chromatograph utilizing a 2 ft × 2 mm i.d. glass column packed with 1% OV-101 on Gas-Chrom Q. The gas chromatograph was equipped with an alkali ($RbSO_4$) flame ionization detector (AFID). Gas flows for hydrogen, helium, and air were 40, 28, and 300 mL/min, respectively.

Proton magnetic resonance (^{1}H NMR) spectra were recorded on a Varian EM-390 spectrometer by using tetramethylsilane (Me_4Si) as the internal standard and chloroform-d_1 as the solvent. Carbon-13 nuclear magnetic resonance (^{13}C NMR) spectra were recorded on a Bruker 90-MHz multinuclear facility by using dioxane vs. Me_4Si as the internal standard in D_2O.

Chemicals. Methamidophos, 99.6% pure, was supplied as an analytical standard by the Chevron Chemical Co., Richmond, CA. Purified methamidophos was obtained by repeated recrystallization from ethyl acetate–diethyl ether samples partially purified by column chromatography.

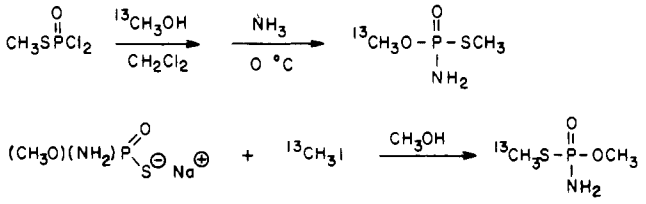
[*O*-*methyl*-^{13}C]Methamidophos was prepared according to Lubkowitz et al. (1974). [^{13}C]Methanol (0.5 g, 90% enriched, Merck and Co., Teterboro, NJ) was added to 2.64 g (0.016 mol) of *S*-methyl phosphorothioic dichloride in 30 mL of dichloromethane at 0 °C. After being stirred for 0.5–1.0 h, the solution was quenched with anhydrous ammonia gas and filtered through Celite to remove ammonium chloride. The reaction mixture was concentrated by rotary evaporation and the yellowish oil was purified by column chromatography. The resulting [*O*-*methyl*-^{13}C]-methamidophos was lyophilized to a fine powder and subjected to repeated recrystallization to yield clear needles (1.161 g, 51%): mp 42–43 °C; ^{1}H NMR ($CDCl_3$) δ 4.58 and 2.94 ($^{13}CH_3O$, dd, J_{H-P} = 12.5 Hz, J_{C-H} = 147 Hz), 2.30 (SCH_3, d, J_{H-P} = 14 Hz); ^{13}C NMR (D_2O) δ 54.1 (OCH_3, d, J_{C-P} = 6.2 Hz).

[*S*-*methyl*-^{13}C]Methamidophos was prepared according to Mel'nikov and Zen'kovich (1955) by reacting 1.0356 g (7.05 mmol) of sodio-*O*-methyl phosphoramidothioate with 7.05 mmol of [^{13}C]methyl iodide (1.0 g, 90% enriched;

Department of Chemistry, University of California, Riverside, California 92521 (C.M.T. and T.R.F.), and Department of Agricultural Chemistry, Kyoto University, Kyoto, Japan 606 (T.N.).

0021-8561/83/1431-0696$01.50/0

Scheme I. Selective Labeling of [^{13}C]Methamidophos



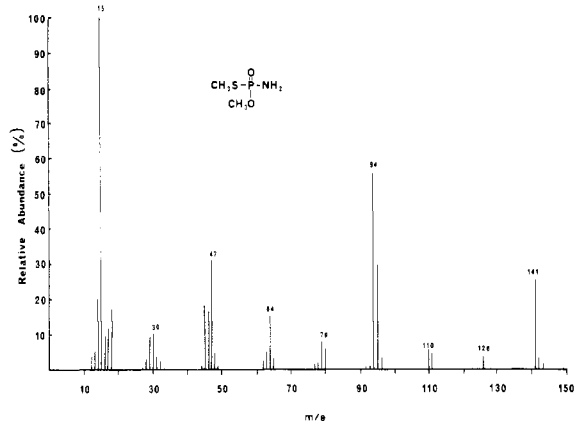


Figure 1. Mass spectrum of methamidophos $(CH_3O)(CH_3S)P(O)NH_2$.

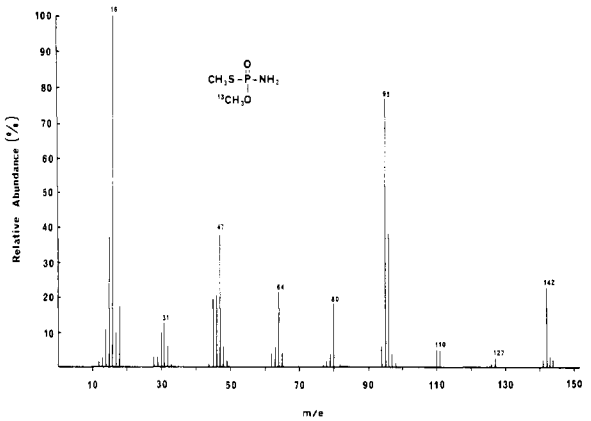


Figure 2. Mass spectrum of [^{13}C]methamidophos $(^{13}CH_3O)(CH_3S)P(O)NH_2$.

Merck and Co., Teterboro, NJ) in 10 mL of anhydrous methanol. The reaction mixture was stirred at room temperature for 1.0 h. The resulting [*S-methyl*-^{13}C]methamidophos was purified in the same manner as the *O*-methyl analogue. Recrystallization resulted in 0.980 g of clear needles (98% yield): ^{1}H NMR ($CDCl_3$) δ 3.75 (OCH_3, d, J_{H-P} = 15 Hz), 3.06 and 1.50 ($^{13}CH_3S$, dd, J_{H-P} = 14 Hz, J_{C-H} = 141 Hz); ^{13}C NMR (D_2O) δ 12.4 ($^{13}CH_3S$, d, J_{C-P} = 4.4 Hz). The synthetic route to the labeled materials is shown in Scheme I.

RESULTS

Figures 1, 2, and 3 contrast the EIMS of (a) $(CH_3O)(CH_3S)P(O)NH_2$, (b) $(^{13}CH_3O)(CH_3S)P(O)NH_2$, and (c) $(CH_3O)(^{13}CH_3S)P(O)NH_2$, respectively. The mass/charge (m/z) ratios are listed in Table I for the compounds investigated. Since this is a study of the direct nature of the fragmentation mechanisms, each fragment ion will be discussed individually and not each separate compound. The general characteristics of the spectrum of methamidophos agreed with that previously reported (Desmarchelier et al., 1976).

Percent Abundance of [^{13}C]Methamidophos. The quantity of nonlabeled methamidophos which contaminated the ^{13}C-labeled samples was calculated from the ratio of the intensity of the molecular ion peak of [^{13}C]methamidophos, m/z 142, to that of nonlabeled, m/z 141. The ion m/z 142 contains the isotopic contribution from the natural abundance ^{13}C, ^{33}S, and ^{15}N isotopes present in nonlabeled methamidophos. This contribution was estimated to be 11.9% from the m/z 142/m/z 141 ratio of the spectrum of standard methamidophos. After correction from this contribution, the relative abundance of ^{13}C-labeled compounds was determined to be 91.1% for [$^{13}CH_3O$]methamidophos and 90.3% for [$^{13}CH_3S$]meth-

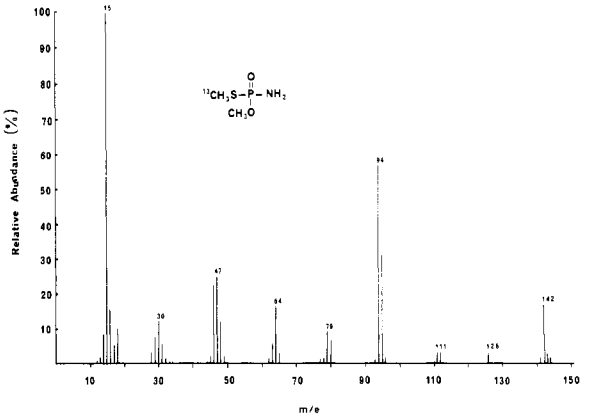


Figure 3. Mass spectrum of [^{13}C]methamidophos $(CH_3O)(^{13}CH_3S)P(O)NH_2$.

Table I. Relative Intensity[a]

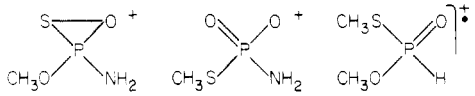
m/z	$(CH_3O)(CH_3S)P(O)NH_2$	$(^{13}CH_3O)(CH_3S)P(O)NH_2$	$(CH_3O)(^{13}CH_3S)P(O)NH_2$
12	3.90	1.82	1.11
13	5.11	3.20	2.14
14	20.60	10.97	8.50
15	100.00	37.05	100.00
16	9.72	100.00	16.26
17	11.45	10.33	5.61
18	16.83	17.47	9.89
28	2.87	2.74	3.22
29	9.21	2.65	7.53
30	9.91	10.06	12.14
31	3.51	12.53	5.58
32	1.79	5.76	1.41
44	1.08	1.18	0.58
45	17.97	19.67	2.25
46	16.31	20.12	22.68
47	31.41	37.96	25.10
48	4.41	6.03	12.31
49	1.34	2.01	2.16
62	2.62	3.93	1.47
63	5.37	6.03	5.58
64	14.77	21.13	16.37
65	2.81	3.75	2.97
77	1.59	0.73	1.11
78	1.72	1.55	1.50
79	7.62	3.38	8.39
80	5.63	17.93	6.56
94	55.72	6.67	56.24
95	29.23	77.21	31.27
96	3.13	37.96	1.66
97	0.38	3.93	0.19
98		1.37	
110	5.31	4.84	0.47
111	4.47	4.84	3.41
112	0.70	0.45	3.30
126	3.77	0.82	2.97
127	0.31	3.31	0.08
141	24.66	1.98	1.60
142	2.95	22.82	16.78
143	1.81	2.79	2.84
144	0.17	2.03	0.91

[a] All the ions listed are of intensity greater than 1.0%.

amidophos. These values correspond to those of ^{13}C-labeled methanol and methyl iodide used as synthetic reagents.

Assignment of Fragment Ions. Assignment of elemental composition for the major fragment peaks was made via the differences in the isotopic shift which appeared in the three spectra. Plausible structures are given in terms of tetravalent (Haake, 1968) and pentavalent (McLafferty, 1956, 1959; Occolowitz and Swan, 1966) phosphorus, a degree of resonance being understood. Consecutive processes are shown for convenience and are not necessarily factual.

Fragment Ion *m/z* 126. There are three possible structures, 1–3, which may contribute to the mass of ion



1 2 3

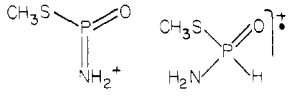
[*S*-methyl-^{13}C]

126. The [*S-methyl*-^{13}C]methamidophos did not show any shift of ion 126 to 127. Loss of 16 mass units relative to the 15 mass units for the [*O-methyl*-^{13}C]methamidophos or the standard then implies that this ion is unambiguously formed by the loss of a methyl radical from the *S*-methyl and not the *O*-methyl group. This fragment ion is assigned to 1. Contribution from the ion 3 which corresponds to the loss of the –NH group was negligible.

In the paper by Desmarchelier et al. (1976), the small peak observed at *m/z* 126 is attributed to cleavage of the P–N bond. We now have ascertained that methamidophos follows traditional ion fragmentation among the unsubstituted phosphor- and phosphonamidothioates where few, if any, demonstrate fission of the P–N bond. The stability of the P–N bond in unsubstituted amidates to fission is described by Roesky and Kloker (1970) and Jakobsen et al. (1972). The P=S isomer of methamidophos also exhibits a small peak at *m/z* 126 (Desmarchelier et al., 1976); however, correct assignment cannot be attributed to O–C cleavage by analogy at this point and, therefore, P–N cleavage may be evident for this compound.

Fragment Ions *m/z* 110 and 111. These ions are typical fragments of the type $(M - 31)^+$ for organophosphorus esters containing an *O*-methyl group (Jakobsen et al., 1972; Pritchard, 1970; Bafus et al., 1966).

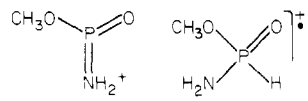
In this study, ^{13}C labeling at the *S*-methyl group induced a concomitant shift of ions *m/z* 110 and 111 by one mass unit (Table I). Conversely, ^{13}C labeling at the *O*-methyl did not. They were assigned to ions formed by the loss of *O*-methyl radical (**4**) and formaldehyde (**5**), respectively.



m/z 110 *m/z* 111

4 5

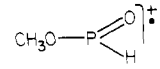
Fragment Ions *m/z* 94 and 95. These ions, $(M - 47)^+$ and $(M - 46)^+$, are typical fragments of organophosphorus esters containing an *S*-methyl group; that is, they correspond to fragment ions formed by the loss of an *S*-methyl radical and $CH_2{=}S$, respectively. The spectrum of [*S-methyl*-^{13}C]methamidophos was identical with that of standard methamidophos in the intensity of peaks *m/z* 94–96. However, the spectrum of [*O-methyl*-^{13}C]methamidophos showed a shift of 1 mass unit. Ions *m/z* 94 and 95 were assigned to **6** and **7**, respectively.



m/z 94 *m/z* 95

6 7

Fragment Ions *m/z* 79 and 80. Examination of Table I implies there are two different ions which have the same mass of 79. Approximately 75% of the ions that appeared at *m/z* 79 in the spectrum of methamidophos shifted to *m/z* 80 in the spectrum of [*O-methyl*-^{13}C]methamidophos. This predominating ion contains an *O*-methyl group and is assigned structure 8. The other ion at *m/z* 79 is assigned the structure $NH_2PO_2^{+\cdot}$ since it did not shift with ^{13}C substitution. The ion at *m/z* 80 did not shift with either ^{13}C substitutions. Therefore, this ion does not contain any carbon atom and is assigned to $HO_2PNH_2^+$.



m/z 79

8

Fragment Ions *m/z* 62–65. Table I reveals that substitution of ^{13}C in either of the two methyl groups did not alter the spectra throughout this mass range. This suggests that these fragments do not contain any carbon atoms. Phosphor- and phosphonamidothioates also give this series of ions at 62–65 (Desmarchelier et al., 1976; Jakobsen et al., 1972). The P–N bond in unsubstituted amidates is stable under mass spectral conditions. Therefore, the fragment ions at *m/z* 62–65 are most likely assigned to $HNPO^+$ (*m/z* 62), $PS^+/NH_2PO^{+\cdot}$ (*m/z* 63), $HPO_2^{+\cdot}/NH_2POH_2^{+\cdot}$ (*m/z* 65).

Fragment Ions *m/z* 45–48. Although *m/z* 47 remains predominant regardless of ^{13}C incorporation, there is a definite change in the relative abundance of the fragments. For example, isotopic substitution of the *S*-methyl carbon atom decreased the relative abundance of the ion *m/z* 45 (Table I). In addition, one-third of the relative abundances of both ions *m/z* 46 and 47 shifted by 1 mass unit. It is deduced that most of the ions that appeared at *m/z* 45 and one-third of the ions at *m/z* 46 and 47 contain the carbon atom from the *S*-methyl group. Ions at *m/z* 45 and 46 are assigned to HCS^+ (*m/z* 45), $CH_2{=}S^{+\cdot}$ (1/3, *m/z* 46), and PNH^+ (2/3, *m/z* 46).

From high-resolution studies, fragment ion *m/z* 47 in the spectra of alkylthiophosphate esters was assigned to PO^+ and CH_3S^+ (Desmarchelier et al., 1976; Santoro, 1973). The spectra shown in Figures 1–3 are consistent with this assignment. ^{13}C-substitution at the *S*-methyl carbon induced a shift of the peak at *m/z* 47 by 1 mass unit but ^{13}C-substitution at the *O*-methyl did not. These ions were assigned to CH_3S^+ (1/3, *m/z* 47) and PO^+ (2/3, *m/z* 47). The spectrum of [*S-methyl*-^{13}C]methamidophos did not show any clear shift of the peak *m/z* 48 to *m/z* 49 which was previously assigned to $CH_4S^{+\cdot}$ (Santoro, 1973). The ion *m/z* 48, therefore, does not contain a carbon atom and could be either NH_2PH^+, $POH^{+\cdot}$, or $SO^{+\cdot}$.

Fragment Ions *m/z* 28–32. Table I reveals the similarity of methamidophos with [*S-methyl*-^{13}C]methamidophos for these fragment ions. For these same ions the [*O-methyl*-^{13}C]methamidophos showed a shift by 1 mass unit. They are readily assigned to CHO^+, $CH_2{=}O$, and CH_3O^+, respectively.

Fragment Ion *m/z* 15. The most obvious change in the spectrum (Figure 2) illustrates the shift of the methyl cation to *m/z* 16 for the [*O-methyl*-^{13}C]methamidophos. It was first assigned by Bafus et al. (1966) as the base peak

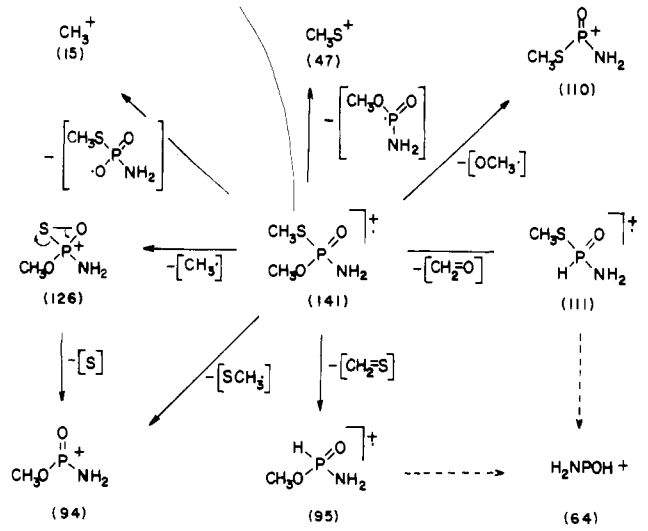


Figure 4. Proposed ion decomposition pathway of methamidophos.

in his study. It suggests that more than 80% of the methyl cation is derived from the $-OCH_3$ group.

DISCUSSION

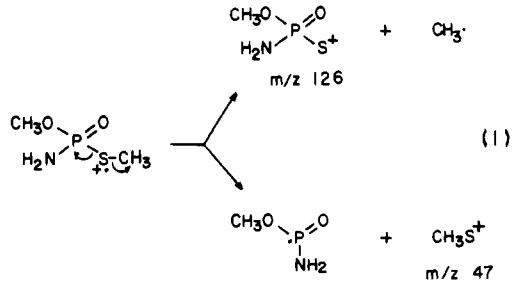
Selective isotope enrichment at either one of the two methyl groups of methamidophos has clearly shown that the *O*-methyl and *S*-methyl groups gave different ion decomposition products. Two decomposition pathways in particular were observed in this study: (1) the neutral methyl radical is lost only from the *S*-methyl group, (2) the methyl cation is predominantly formed from the *O*-methyl group. These results suggest that the fission of the O–C and S–C bonds proceeds exclusively under mass spectral conditions. Fragment ions other than m/z 126 $(M - CH_3\cdot)^+$, and m/z 15, CH_3^+, are the products of decomposition pathways which do not effect the O–C or S–C bond.

The proposed ion decomposition pathway of methamidophos is summarized in Figure 4. Although there are some limitations to our knowledge about thermochemical parameters of phosphoramidothioates, the ion decomposition products of methamidophos can be reasonably explained by empirical rules accumulated from the spectra of amides, ethers, and thioethers. We present here a qualitative, yet somewhat empirical, discussion of the factors that control these selective fragmentations of methylthio and methoxy groups present in the methamidophos molecule.

Loss of Neutral Methyl Radical. In the mass spectra of *O,O,S*-trimethyl phosphorothioate and *O,O,S*-trimethyl and *O,S,S*-trimethyl phosphorodithioates, the $(M - CH_3\cdot)^+$ ion was observed in significant abundances, but this same peak was absent in the spectrum of *O,O,O*-trimethyl phosphate (Santoro, 1973). From these observations, Santoro suggested the loss of the methyl radical from the *S*-methyl group of organophosphorus esters.

The sulfur atom is initially ionized by the loss of a nonbonding d-shell electron; then the C–S bond is cleaved via substantial σ-ionization which resembles an alkane σ-ionization reaction (McLafferty, 1980). Decomposition due to charge retention on sulfur is favored because stabilization of the resulting positive charge is aided by participation of the inner shell electrons.

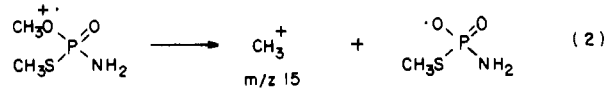
With charge retention on the sulfur atom, two competitive cleavages are possible in methamidophos (eq 1). The relative intensity of the ion SCH_3^+ [(31.4% × 1/3)/10%] is more abundant than the ion $(M - CH_3\cdot)^+$ (3.8%). This suggests that the decomposition to SCH_3^+ is preferred to



(1)

the loss of a methyl radical. The spectra of *S*-methyl thiophosphorus esters previously reported (Desmarchelier et al., 1976; Santoro, 1973) shows a peak at m/z 47 with significant intensities, but only 11 among 18 compounds showed a small peak associated with the loss of a methyl radical. The presence of an $(M - CH_3\cdot)^+$ peak is limited to *S*-methyl thiophosphorus esters though the relative intensity is small. Exceptions are *N*-methyl phosphorus amides and aryl phosphorus esters, some of which gave an $(M - CH_3\cdot)^+$ ion in spite of the absence of an *S*-methyl group in the molecule.

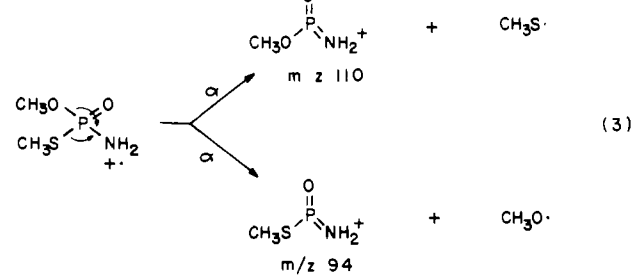
Formation of Methyl Cation. Methyl cation (m/z 15) was the base peak for methamidophos. Formation of methyl cation from the *O*-methyl group of methamidophos is due to the ionization of the oxygen atom, followed by the fission of the O–C bond by charge migration to the methyl carbon atom (eq 2). This tendency for charge-site



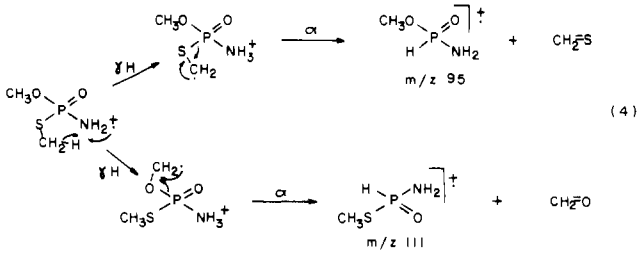
(2)

cleavage is expected from the higher inductive withdrawal of electrons by the oxygen atom (McLafferty, 1980). The cleavage of the O–P bond partly competes with that of the O–C bond.

Loss of Methoxy and Methylthio Groups. The loss of methoxy and methylthio groups as a neutral radical is presumably initiated by the ionization of nitrogen which has the lowest ionization potential among the four charge sites of the methamidophos molecule; nitrogen, sulfur, and the two oxygen atoms. Ionization of a nitrogen atom provides a powerful α-cleavage which leads to ions $(M - OCH_3\cdot)^+$ and $(M - SCH_3\cdot)^+$ (eq 3) or provides a rearrangement of hydrogen followed by α-cleavage which leads to ions $(M - CH_2{=}O)^{+\cdot}$ and $(M - CH_2{=}S)^{+\cdot}$ (eq 4).



(3)

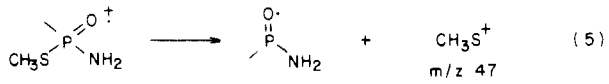


(4)

Intensities of peaks m/z 94 and 95 were much more abundant than those of peaks m/z 110 and 111. This ease

of loss of SCH_3· and CH_2=S rather than loss of OCH_3· and CH_2=O is in general agreement with phosphoramidothioate and phosphorothioate esters (Desmarchelier et al., 1976). This was attributed to the difference in the bond energies between the P–S and P–O bond rather than the difference in the product stabilities (Santoro, 1973).

This difference in the bond energies is also reflected in the relative abundances of SCH_3^+, m/z 47 = 31.4%, and OCH_3^+, m/z 31 = 3.5%. The SCH_3^+ ion is derived from a molecular ion or $(M - CH_2{=}O)^+$· ion by inductive cleavage with charge migration to the electronegative oxygen atom (eq 5). In general, the CH_3O^+ ion shows a very



(5)

small peak except in the case of low molecular weight esters (McLafferty, 1980).

In summary, the cleavage of the P–S (or P–O) bond gave products corresponding to inductive cleavage in which charge retention on the sulfur (oxygen) competes with charge migration from the sulfur (oxygen) to the carbon at some degree. On the other hand, the cleavage of the C–S (C–O) bond gave the methyl radical and the $S(CH_3O)P(O)NH_2$ ion [the CH_3^+ ion and the $O(CH_3S)P(O)NH_2$· radical] corresponding to charge retention on the sulfur (charge migration from the oxygen to the carbon). This reaction is *not* competing with charge migration (charge retention). We cannot directly apply these results to the in vivo or in vitro enzymatic reactions; however, they demonstrate the possibility that such specific reactivities of two methyl groups could play an important role in the intoxication mechanism.

Registry No. Methamidophos, 10265-92-6.

LITERATURE CITED

Bafus, D. A.; Gallegos, E. J.; Kiser, R. W. *J. Phys. Chem.* **1966**, *70*, 2614.
Budziekiewitz, H.; Djerassi, C.; Williams, D. H. "Mass Spectrometry of Organic Compounds"; Holden-Day, Inc.: San Francisco, 1967; pp 55–59.
Desmarchelier, J. M.; Wustner, D. A.; Fukuto, T. R. *Residue Rev.* **1976**, 76–185.
Fahmy, M. A.; Khasawinah, A.; Fukuto, T. R. *J. Org. Chem.* **1974**, *36*, 617.
Gray, A. J.; Thompson, C. M.; Fukuto, T. R. *Pestic Biochem. Physiol.* **1982**, *18*, 28.
Haake, P.; Ossip, P. S. *Tetrahedron* **1968**, *24*, 565.
Jakobsen, P.; Treppendahl, S.; Wieczorkowski, *J. Org. Mass Spectrom.* **1972**, *6*, 1303.
Lubkowitz, J. A.; Revilla, A. P.; Baruel, J. *J. Agric. Food Chem.* **1974**, *22*, 151.
McLafferty, F. W. *Anal. Chem.* **1956**, *28*, 306.
McLafferty, F. W. *Anal. Chem.* **1959**, *31*, 82.
McLafferty, F. W. "Interpretation of Mass Spectra"; University Science Books: Mill Valley, CA, 1980; p 50.
Mel'nikov, N. N.; Zen'kovich, A. G. *J. Gen. Chem. USSR (Engl. Transl.)* **1955**, *25*, 793.
Menn, J. J.; Erwin, W. R.; Gordon, H. T. *J. Agric. Food Chem.* **1957**, *5*, 601.
Occolowitz, J. L.; Swan, J. M. *Aust. J. Chem.* **1966**, *19*, 1187.
Pritchard, J. G. *Org. Mass Spectrom.* **1970**, *3*, 163.
Quistad, G. B.; Fukuto, T. R.; Metcalf, R. L. *J. Agric. Food Chem.* **1970**, *18*, 189.
Roesky, H. W.; Kloker, W. *Z. Anorg. Allg. Chem.* **1970**, *375*, 140.
Santoro, E. *Org. Mass Spectrom.* **1973**, *7*, 589.
Thompson, C. M.; Fukuto, T. R. *J. Agric. Food Chem.* **1982**, *30*, 282.

Received for review July 21, 1982. Accepted March 21, 1983.

Effect of Dockage on the Degradation of [^{14}C]Malathion in Stored Wheat

Barbara N. Anderegg* and Linda J. Madisen

The degradation of [^{14}C]malathion (1,2-ethyl-^{14}C label) in whole wheat containing 0, 2.5, 5.0, and 10.0% dockage (ground wheat) was investigated. The effect of storage time on the degradation of [^{14}C]malathion in wheat containing 2.5% dockage was also studied. The total quantity of [^{14}C]malathion residues recovered in the dockage fraction increased significantly both as the ratio of dockage to whole grain increased and as the storage time increased. A large proportion of the radiocarbon recovered from the dockage fraction was in the form of unextractable (bound) residues. As the proportion of dockage in the grain increased, the recovery of volatile ^{14}C compounds decreased.

Much of the grain stored in the United States contains varying quantities of dockage, or small particles of broken grain, weed seeds, dust, and other foreign material. Not only does dockage increase the chances of grain heating and deteriorating, but several reports have indicated that the presence of dockage reduces the effectiveness of stored grain protectants. Strong and Sbur (1960) reported that malathion applied to clean wheat remained toxic to *Sitophilus granarius*, *Sitophilus oryzae*, and *Tribolium confusum* longer than malathion applied to wheat containing a high percentage of damaged kernels, dust, and insect fragments. Minett and Williams (1971) found that malathion was less effective against *T. confusum* when applied to a 50% mixture of whole and kibbled wheat than when applied to 100% whole grain. Conversely, Kadoum and LaHue (1969) found that the presence of dockage had a negligible effect on the degradation of malathion in grain sorghum. Quinlan (1982) pipetted malathion onto wheat containing 2.0% cracked wheat at a rate of 10 ppm and found that the concentration of malathion per unit weight of wheat particle increased as particle size decreased. Godavari Bai et al. (1964) found that the toxicity of malathion decreased as the particle size of the insecticide-

U.S. Grain Marketing Research Laboratory, Agricultural Research Service, U.S. Department of Agriculture, Manhattan, Kansas 66502 (B.N.A.), and Department of Entomology, Kansas State University, Manhattan, Kansas 66506 (L.J.M.).