Wiley, New York, N.Y., 1970, p. 507.

(4) J. P. Scovill, D. L. Klayman, T. S. Woods, and T. R. Sweeney, J. Med. Chem., 22, 1164 (1979).

(5) L. H. Schmidt, R. Fradkin, D. Vaughan, and J. Rasco, Am. J. Trop. Med. Hyg., 26, 1116 (1977).

(6) M. P. LaMontagne, A. Markovac, and J. R. Menke, J. Med. Chem., 20, 1122 (1977).

(7) L. H. Schmidt, A. H. Rossan, R. Fradkin, and J. Woods, Bull. WHO, 34, 783 (1966).

(8) "World Health Organization (1972b)," WHO/MAL/72763 (cyclostyled report), World Health Organization, Geneva, Switzerland.

(9) T. S. Osdene, P. B. Russell, and L. Rane, J. Med. Chem., 10, 431 (1967).

(10) G. R. Coatney, W. C. Cooper, N. B. Eddy, and J. Greenberg, "Survey of Antimalarial Agents," Public Health Monograph No. 9, U.S. Government Printing Office, Washington, D.C., 1953.

(11) R. C. Elderfield, W. J. Gensler, T. A. Williamson, J. M. Griffing, S. M. Kupchan, J. T. Maynard, F. J. Kreysa, and J. B. Wright, J. Am. Chem. Soc., 68, 1584 (1946).

(12) R. H. F. Manske and M. Kulka, in "Organic Reactions," vol. 7, R. Adams, Ed., Wiley, New York, N.Y., 1953, p. 59.

(13) A. Maercker, in "Organic Reactions," vol. 14, A. C. Cope, Ed., Wiley, New York, N.Y., 1965, p. 270.

(14) L. Friedman and H. Sheckter, J. Org. Chem., 26, 2522 (1961).

(15) K. Kindler and K. Lührs, Chem. Ber., 99, 227 (1966).

(16) K. Kindler and K. Lührs, Ann. Chem., 685, 36 (1965).

(17) F. L. Cook, C. W. Bower, and C. L. Liotta, J. Org. Chem., 39, 3416 (1974).

(18) A. McKillop and D. P. Rao, Synthesis, 1977, 759.

(19) R. C. Elderfield, H. E. Mertel, R. T. Mitch, I. M. Wempen, and E. Werble, J. Am. Chem. Soc., 77, 4816 (1955).

(20) E. H. Chen, A. J. Saggiomo, K. Tanabe, B. L. Verma, and E. A. Nodiff, J. Med. Chem., 20, 1107 (1977).

(21) E. A. Chen, A. J. Saggiomo, and E. A. Nodiff, U.S. pat. 4,167,638 (1979).

(22) M. Carmack, L. W. Kissinger, and I. Von, J. Am. Chem. Soc., 68, 1551 (1946).

(23) W. C. Hanson, W. L. Chapman, Jr., and K. E. Kinnamon, Int. J. Parasitol., 7, 443 (1977).

(24) K. E. Kinnamon, E. A. Steck, P. S. Loizeaux, W. L. Hanson, W. L. Chapman, Jr., and V. B. Waits, Am. J. Trop. Med. Hyg., 27, 751 (1978).

(25) H. Mort and W. Montouri, ibid., 24, 179 (1975).

(26) K. E. Kinnamon and D. S. Rane, ibid., 28, 937 (1979).

(27) C. C. Price and R. M. Roberts, J. Am. Chem. Soc., 68, 1204 (1946).

Aporphines XXVI: GLC and Mass Spectrometric Properties of Trifluoroacetyl Derivatives of N-Methyl-, N-Propyl-, and Noraporphines

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Abstract □ The O-trifluoroacetyl and N,O-trifluoroacetyl derivatives of a series of aporphine and noraporphine alkaloids were prepared, and their GLC and mass spectrometric characteristics were determined. The positional isomers apocodeine and isoapocodeine were resolved chromatographically as their trifluoroacetyl derivatives, and their mass spectra were distinctly different. Mass spectrometric fragmentation processes apparently unique to N-trifluoroacetyl derivatives of noraporphines were observed. Plausible mechanisms for the formation of the major ions in the mass spectra of the various compounds are given.

Keyphrases □ Aporphines—trifluoroacetyl derivatives, GLC and mass spectrometry □ Noraporphines—trifluoroacetyl derivatives, GLC and mass spectrometry □ GLC—trifluoroacetyl derivatives of aporphines and noraporphines

The need for sensitive and quantitative methods for elaborating the metabolism of apomorphine (Ia) (Scheme I) has increased due to the biological activity of apomorphine and related alkaloids (1–3). Although conjugation was reported as a primary route for the metabolism of Ia in the rat (4), apocodeine (Id) and isoapocodeine (Ie) formed from O-methylation have been reported as possible metabolites in rat liver preparations (5). N-Dealkylation also was reported to give an in vitro metabolite of several naturally occurring aporphine alkaloids (6). More recent studies in liver microsomal preparations indicated that several aporphine analogs could be converted metabolically to their O-demethylated derivatives (i.e., Id → Ia and Ie → Ia) (7).

BACKGROUND

N-n-Propylnorapomorphine (Ib) was reported to be several times more potent than Ia in several biological systems (3). Monohydroxyaporphines substituted in either the 10- or 11-position were found to be active dopamine agonists, although they were less potent than Ib (8, 9). It is generally accepted that catechol analogs of apomorphine are more potent dopamine agonists than are the monohydroxy analogs (10). However, the possibility that monohydroxy analogs such as Ij and Ik are converted in vivo into a catechol-like compound (e.g., Ib) cannot be excluded (11). Preliminary studies showed no evidence for the microsomal hydroxylation of the monohydroxy aporphines Ij and Ik to the corresponding catechol, Ib, using TLC to distinguish monophenols from catecholic metabolites (7).

To develop more definitive techniques, specifically GLC–mass spectrometric methods for the detection, identification, and quantitation of aporphines, the O-trifluoroacetyl derivatives of a series of N-methyl- and N-propylaporphines were prepared. Since N-dealkylation is a common metabolic process, the mass spectra of a series of trifluoroacetyl noraporphine derivatives also were considered. It was hoped that this examination might reveal unique fragmentation processes which could aid in the recognition of noraporphine metabolites or natural products in general.

Paper chromatography, GLC, and TLC were used previously to define
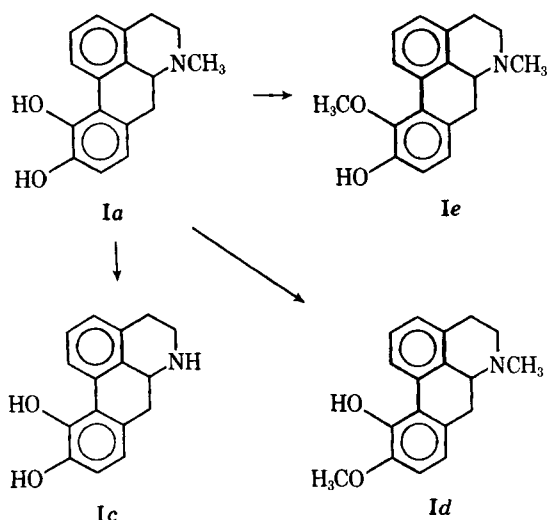
Scheme I—Possible metabolic pathways of apomorphine

the metabolism of such compounds *in vitro* (5, 7) and *in vivo* (12). These techniques *per se* do not necessarily define specificity for such studies. Findings obtained in these laboratories indicated that GLC–mass spectrometry permitted unambiguous analysis of a series of hydroxynoraporphines as their N-heptafluorobutyryl-O-trimethylsilyl derivatives (13), as well as the trimethylsilyl derivatives for N-methyl- and N-propylaporphines (14). GLC–mass spectrometry of the trifluoroacetyl derivatives described in this report offers specific advantages over the use of both their trimethylsilyl and heptafluorobutyryl-O-trimethylsilyl counterparts.

EXPERIMENTAL

The compounds were available in these laboratories as their hydrochloride or hydroiodide salts. A solution of 1 mg/ml or less of the aporphine and noraporphine in trifluoroacetic anhydride was prepared and heated for 10 min at 60°. The excess reagent was evaporated under a nitrogen stream, and the derivative was dissolved in methylene chloride to yield a 1-mg/ml solution. An aliquot (1–2 μl) was injected into the gas–liquid chromatograph–mass spectrometer system[1] for analysis.

A 183-cm × 2-mm i.d. glass column packed with 1% OV-17 on 100–120-mesh Supelcoport and temperature programmed for 6°/min from 200° was used. The gas chromatograph injection port, detector, and
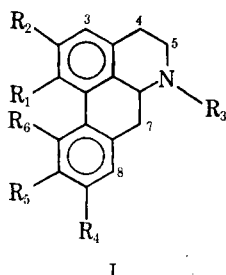
I

Ia: $R_1 = R_2 = R_4 = H, R_3 = CH_3, R_5 = R_6 = OH$
Ib: $R_1 = R_2 = R_4 = H, R_3 = n\text{-}C_3H_7, R_5 = R_6 = OH$
Ic: $R_1 = R_2 = R_3 = R_4 = H, R_5 = R_6 = OH$
Id: $R_1 = R_2 = R_4 = H, R_5 = OCH_3, R_6 = OH, R_3 = CH_3$
Ie: $R_1 = R_2 = R_4 = H, R_5 = OH, R_6 = OCH_3, R_3 = CH_3$
If: $R_1 = R_2 = R_4 = H, R_5 = OCH_3, R_6 = OH, R_3 = n\text{-}C_3H_7$
Ig: $R_1 = R_4 = H, R_2 = R_5 = R_6 = OH, R_3 = CH_3$
Ih: $R_1 = R_4 = H, R_2 = R_6 = OH, R_5 = OCH_3, R_3 = CH_3$
Ii: $R_1 = R_4 = H, R_2 = R_5 = OCH_3, R_6 = OH, R_3 = CH_3$
Ij: $R_1 = R_2 = R_4 = R_6 = H, R_5 = OH, R_3 = C_3H_7$
Ik: $R_1 = R_2 = R_4 = R_5 = H, R_6 = OH, R_3 = C_3H_7$
Il: $R_1 = R_3 = R_4 = H, R_2 = R_5 = OCH_3, R_6 = OH$
Im: $R_1 = R_3 = R_4 = H, R_2 = R_6 = OH, R_5 = OCH_3$
In: $R_1 = R_3 = R_4 = H, R_2 = R_5 = R_6 = OH$
Io: $R_1 = R_5 = OH, R_2 = OCH_3, R_3 = R_4 = R_6 = H$
Ip: $R_1 = OCH_3, R_2 = OH, R_3 = R_4 = R_5 = R_6 = H$
Iq: $R_1 = R_2 = OCH_3, R_3 = R_4 = R_6 = H, R_5 = OH$

[1] Varian Aerograph model 2700 interfaced *via* a 0.15-mm, 25-cm capillary tube to a Nuclide model 12-90-G mass spectrometer.

Table I—GLC Retention Data of Aporphine Derivatives [a]

Compound	Trimethylsilyl Derivatives	Trifluoroacetyl Derivatives
Ia	27.56	23.78
Id	28.50	26.62
Ie	27.79	25.56
IIa	31.60	31.00
2,10,11-Trihydroxy-N-methylaporphine	29.12	24.33
2,11-Dihydroxy-10-methoxy-N-methylaporphine	30.24	26.83
2,10-Dimethoxy-11-hydroxy-N-methylaporphine	30.88	29.33
Ib	28.67	25.04
If	29.52	27.67
Ij	29.08	25.80
Ik	27.07	24.33

[a] Values are in methylene units.

transfer line were maintained at 250, 250, and 200°, respectively. The mass spectrometer ion source was heated to 225°. Mass spectra were recorded in the electron-impact mode (70 ev) at an accelerating voltage of 4.5 kv and a trap current of 50 μamp.

The following compounds were prepared as their O-trifluoroacetyl derivatives and studied by GLC–mass spectrometry: apomorphine (Ia), N-n-propylnorapomorphine (Ib), apocodeine (Id), isoapocodeine (Ie), N-n-propylnorapocodeine (If), 2,10,11-trihydroxyaporphine (Ig), morphothebaine (Ih), 11-hydroxy-2,10-dimethoxyaporphine (Ii), 10-hydroxy-N-n-propylnoraporphine (Ij), and 11-hydroxy-N-n-propylnoraporphine (Ik).

Mass spectrometric data also were acquired on the following noraporphines as their N-trifluoroacetyl or N,O-bis(trifluoroacetyl) derivatives: 11-hydroxy-2,10-dimethoxynoraporphine (Il), 10-methoxy-2,11-dimethoxynoraporphine (Im), and 2,10,11-trihydroxynoraporphine (In). The naturally occurring aporphine alkaloids sparsiflorine (Io), asimilobine (Ip), tuduranine (Iq), bulbocapnine (IIa), nandigerine (IIb), cassyfiline (IIc), xylopine (IId), anolobine (IIe), and actinodaphnine (IIf) also were examined[2].

RESULTS AND DISCUSSION

GLC—N-*Propyl-* and N-*Methylaporphines*—The GLC retention data of the O-trifluoroacetyl derivatives of N-propyl- and N-methylaporphines are given in Table I. For comparison, the methylene unit values for the corresponding trimethylsilyl derivatives reported previously (13) have been included.

In general, the O-trifluoroacetyl derivatives of these compounds exhibited good GLC characteristics. The retention data indicate a marked increase in volatility for the O-trifluoroacetyl derivatives compared to the corresponding O-trimethylsilyl derivatives. For example, this increase in volatility for apomorphine represents a 40° difference in the elution temperature (280 *versus* 240°). This difference not only decreases analysis time but also eliminates the problems of thermal sensitivity associated with these compounds (14).
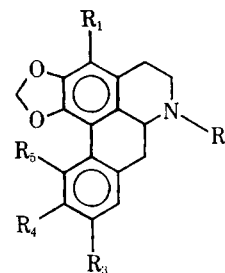
II

IIa: $R_1 = R_3 = H, R_2 = CH_3, R_4 = OCH_3, R_5 = OH$
IIb: $R_1 = R_2 = R_3 = H, R_4 = OH, R_5 = OCH_3$
IIc: $R_1 = R_4 = OCH_3, R_2 = R_5 = H, R_3 = OH$
IId: $R_1 = R_2 = R_4 = R_5 = H, R_3 = OCH_3$
IIe: $R_1 = R_2 = R_4 = R_5 = H, R_3 = OH$
IIf: $R_1 = R_2 = R_4 = H, R_3 = OH, R_5 = OCH_3$

[2] The aporphine alkaloids Io–Iq and IIb–IIf were kindly supplied by Dr. M. Shamma, Pennsylvania State University.

Table II—GLC Retention Data of Noraporphine Derivatives [a]

| Compound	Mixed Derivative [b]	Trifluoroacetyl Derivative
I*m*	30.48	30.60
I*n*	30.00	28.90
I*o*	28.96	26.30

[a] Values are in methylene units. [b] N-Heptafluorobutyryl-O-trimethylsilyl derivative.

Of even greater importance is the ability to separate chromatographically, with baseline resolution, the O-trifluoroacetyl derivatives of apocodeine (I*d*) and isoapocodeine (I*e*), as substantiated by the retention data in Table I. By contrast, the O-trimethylsilyl derivatives of these compounds coelute, a phenomenon that was observed previously (7). Because these two compounds were suggested as possible metabolites of apomorphine, it is essential to identify and to distinguish these products in a biological medium.

Noraporphines—The retention data for the trifluoroacetyl derivatives of noraporphines (I*l*–I*n*) are given in Table II. These data can be compared to the methylene units of the corresponding mixed derivatives (O-trimethylsilyl-N-heptafluorobutyryl) reported previously (13). The general improvement in volatility when trifluoroacetyl derivatives are used is apparent. Moreover, temperature variation of the injection port, column, detector, and gas chromatograph–mass spectrometer transfer line over a wide range gave no indication of thermal sensitivity for the trifluoroacetyl derivatives as opposed to their trimethylsilyl counterparts.

Mass Spectrometry—N-*Methyl-* and N-*Propylaporphines*—The complete mass spectra of the trifluoroacetyl derivatives of apomorphine (I*a*), N-*n*-propylnorapomorphine (I*b*), apocodeine (I*d*), and isoapocodeine (I*e*) are shown in Figs. 1–4, respectively. Partial mass spectra of the compounds in the series are summarized in Tables III and IV.

M[+·] *and* [*M* − *1*][+] *Ions*—The mass spectra of the O-trifluoroacetyl derivatives of N-methyl- and N-propylaporphines were noted for the high intensity of the molecular ion peaks or those corresponding to the loss of a hydrogen at [M − 1][+]. The [M − 1][+] ion presumably is formed by the loss of the 6α-hydrogen to yield a quaternary nitrogen (Scheme II). In general, there were few other ions of any significance in the spectra of the

I*d*

[M − 17][+]

I*e*

[M − 99][+]
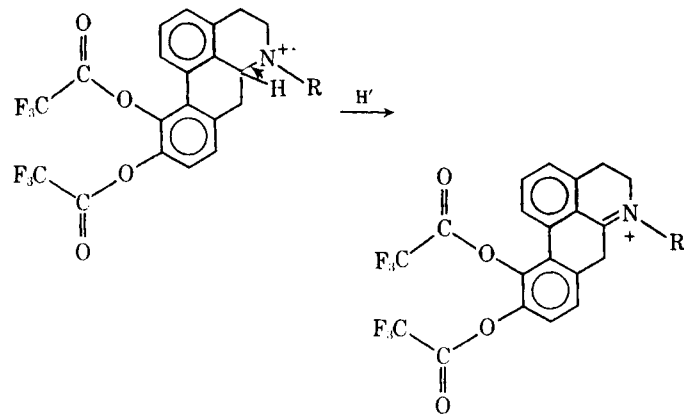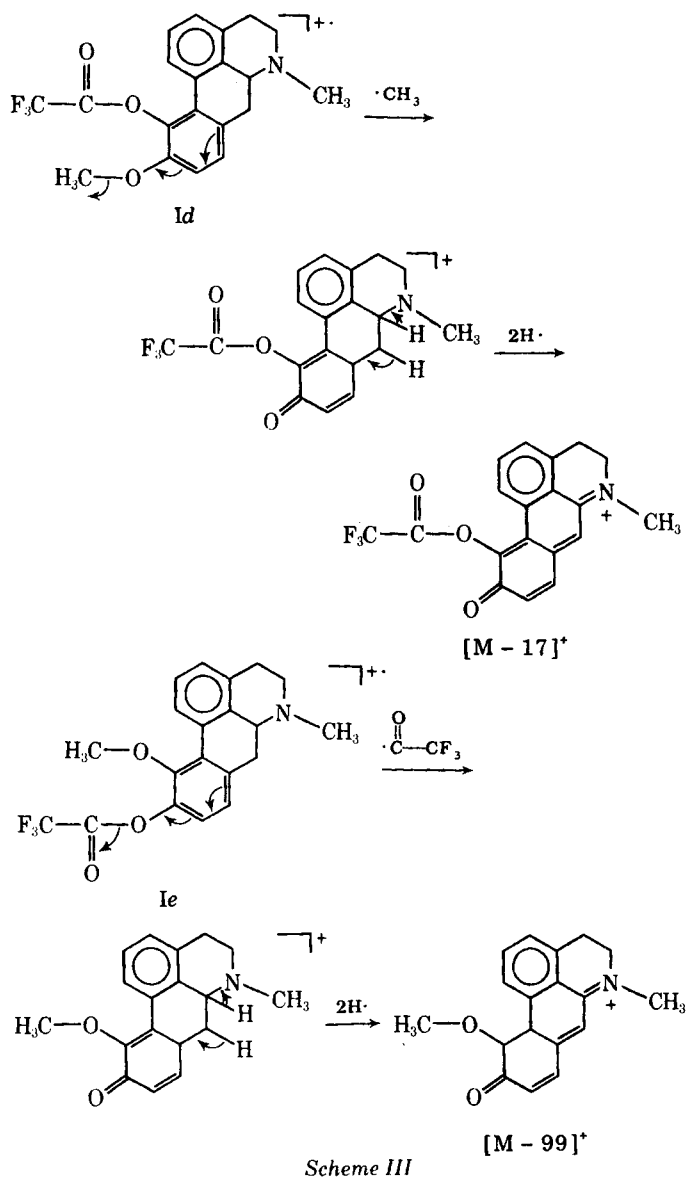
Scheme III

N-methylaporphines. The only peak of any significant intensity in most cases was [M − 97][+], arising from the loss of CF₃C(=O)· from M[+·].

Apocodeine and Isoapocodeine—Two significant differences may be noted (Figs. 3 and 4 and Table III) when the mass spectra of these positional isomers are compared. The spectrum of apocodeine shows a peak at [M − 17][+], which was not detected in the spectrum of the trifluoroacetyl derivative of isoapocodeine. In addition, the relative intensities of the peaks at [M − 99][+] in the spectra of these two compounds are significantly different, the [M − 99][+] ion being much more (10 times)

Scheme II

Table III—Partial Mass Spectra of N-Methyl-O-trifluoroacetyl Derivatives of Aporphines

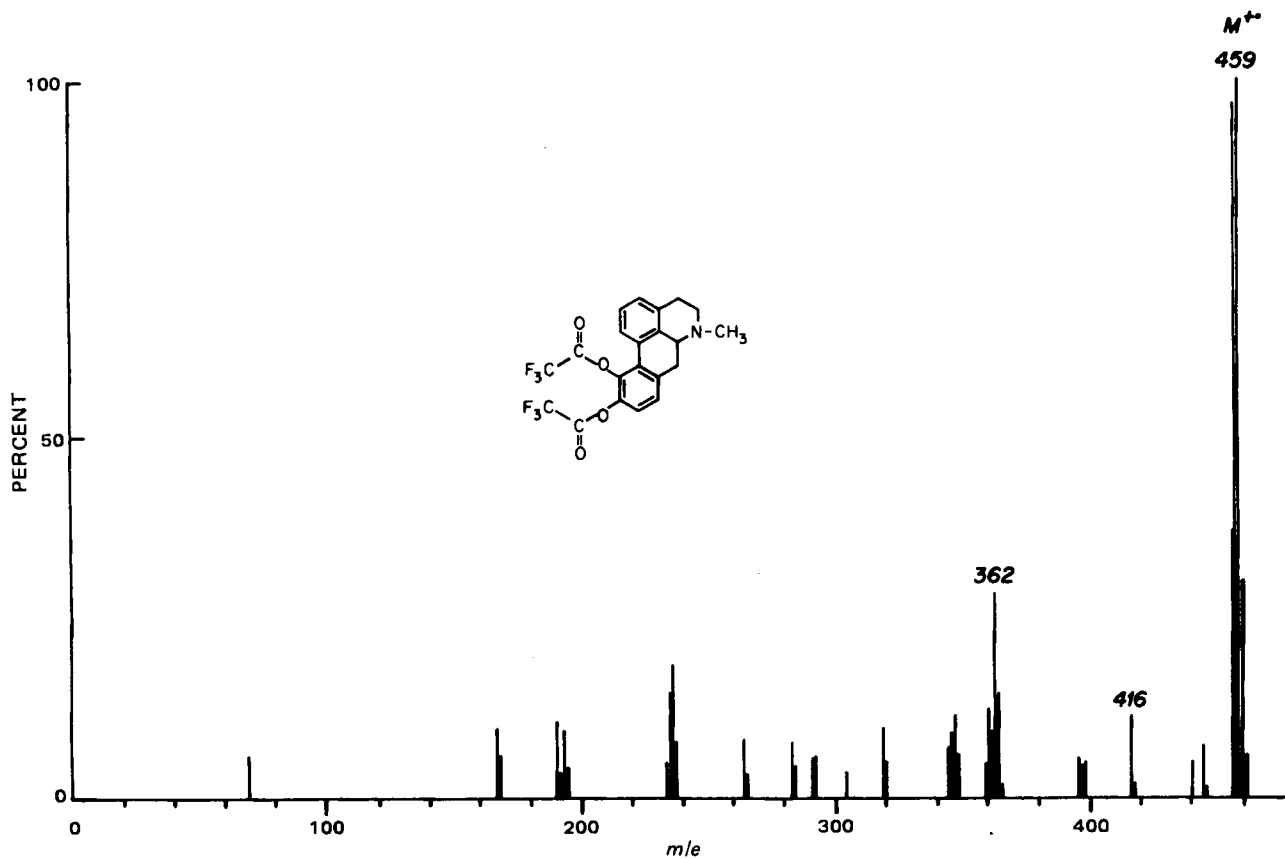| Compound	M[+·]	[M − 1][+]	[M − 15][+]	[M − 17][+]	[M − 97][+]	[M − 99][+]
I*a*	459[a]	458	444	442	362	360
	(100)[b]	(97)	(5)	(—)	(26)	(10)
I*d*	377	376	362	360	280	278
	(91)	(100)	(26)	(21)	(—)	(5)
I*e*	377	376	362	360	280	278
	(76)	(100)	(3)	(—)	(10)	(34)
II*a*	421	420	406	404	324	322
	(100)	(56)	(65)	(12)	(—)	(—)
I*g*	571	570	556	554	474	472
	(100)	(85)	(6)	(—)	(40)	(6)
2,11-Dihydroxy-10-methyoxyaporphine	489	488	474	472	392	390
	(100)	(86)	(34)	(8)	(9)	(4)
2,10-Dimethoxy-11-hydroxyaporphine	407	406	392	390	310	308
	(87)	(100)	(42)	(30)	(—)	(—)

[a] The m/e values. [b] Relative intensity.

Figure 1—*Mass spectrum of the* O-*bis*(*trifluoroacetyl*) *derivative of* Ia.

abundant in the spectrum of the isoapocodeine derivative. These differences can be explained by the formation of a quinone-type ion structure (Scheme III). The O-trimethylsilyl derivatives of these compounds have virtually identical spectra.

N-n-*Propyl Compounds*—In addition to the [M − 1]+ and [M − 99]+ ions, there are fragmentation pathways specific for compounds containing an N-n-propyl substituent. The base peak in the spectra of the N-n-propyl compounds studied occurred at [M − 29]+, which results from
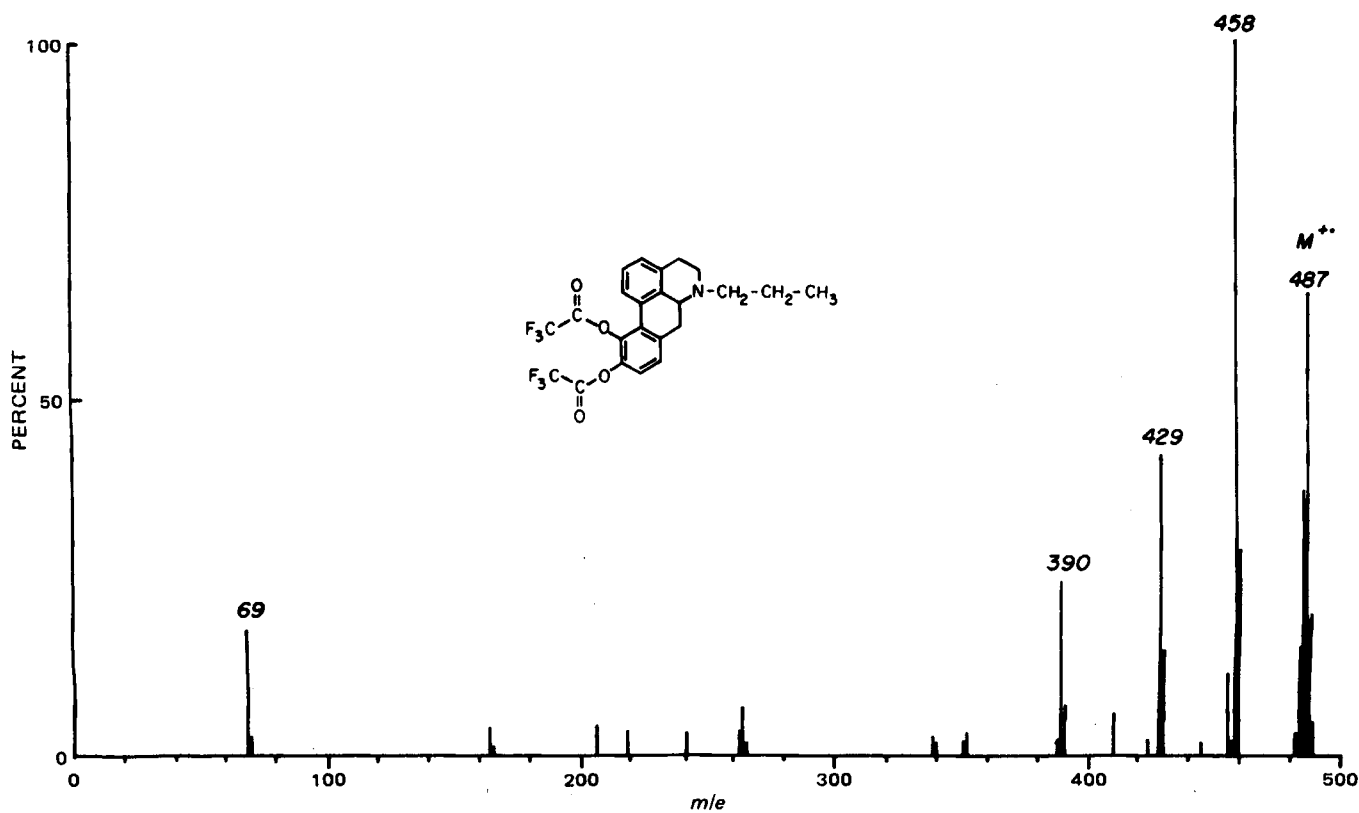

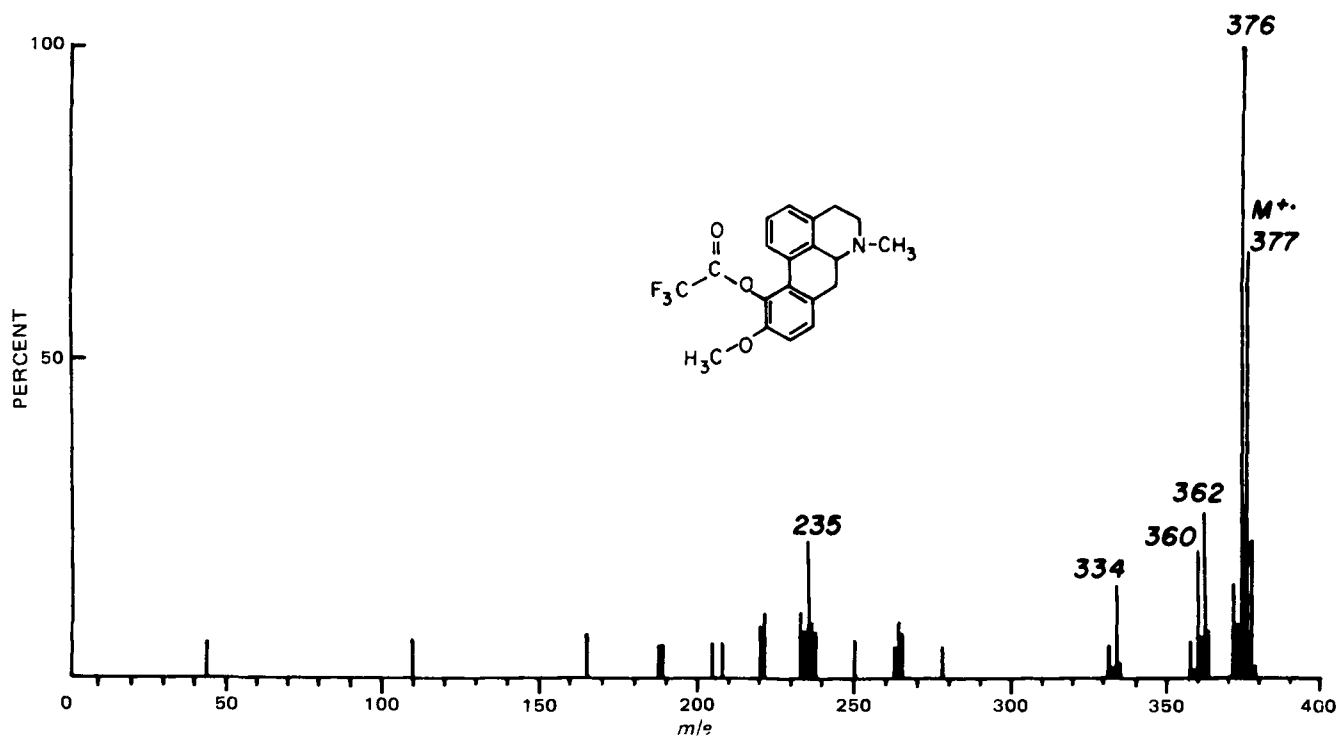Figure 2—*Mass spectrum of the* O-*trifluoroacetyl derivative of* Ib.

Figure 3—*Mass spectrum of the O-trifluoroacetyl derivative of I*d.

cleavage of the carbon–carbon bond beta to the nitrogen. Subsequent fragmentation of this ion gave rise to a peak at [M − 58]+ (Scheme IV).

Noraporphines—The partial mass spectra of the trifluoroacetyl derivatives of the noraporphines (I*l*–I*n*) are listed in Table V. A complete mass spectrum of the N,O-bis(trifluoroacetyl) derivative of anolobine (II*e*) is shown in Fig. 5. The mass spectrum consists of only a few major ions associated with the fragmentation of the trifluoroacetyl group. This pattern typifies the general spectral characteristics of the trifluoroacetyl derivatives of the noraporphines studied. The ion peaks at [M − 69]+ and [M − 97]+ correspond to the loss of CF₃· and CF₃C(=O)· from M+·.

The [M − 126]+ ion is unique to the noraporphine derivatives, and a plausible mechanism for its formation is given in Scheme V. It is suggested that its formation is initiated by a six-membered ring hydrogen transfer (McLafferty rearrangement) from C-7 to the carbonyl oxygen,

followed by cleavage of a carbon–carbon bond beta to the heteroatom. The proposed γ-hydrogen transfer is consistent with the general behavior of the structurally related N-acetates of biogenic amines (14). Alternatively, the possibility of formation of the [M − 126]+ ion by the loss of a hydrogen from the ion formed by a retro-Diels–Alder fragmentation of M+· cannot *a priori* be ruled out.

In a previous publication, Green *et al.* (15) reported on the GLC–mass spectrometry of mixed N-heptafluorobutyryl-O-trimethylsilyl derivatives of noraporphines. By comparison, the mass spectra of the trifluoroacetyl derivatives reported here are noted for their overall simplicity. The intensity of the peak at [M − 126]+ is rather striking, and this peak usually dominates the spectra of these derivatives. The analogous peak is of significantly lower relative intensity in the spectra of the N-heptafluorobutyryl-O-trimethylsilyl derivatives.

In view of the apparent uniqueness of the [M − 126]+ ion, determina-

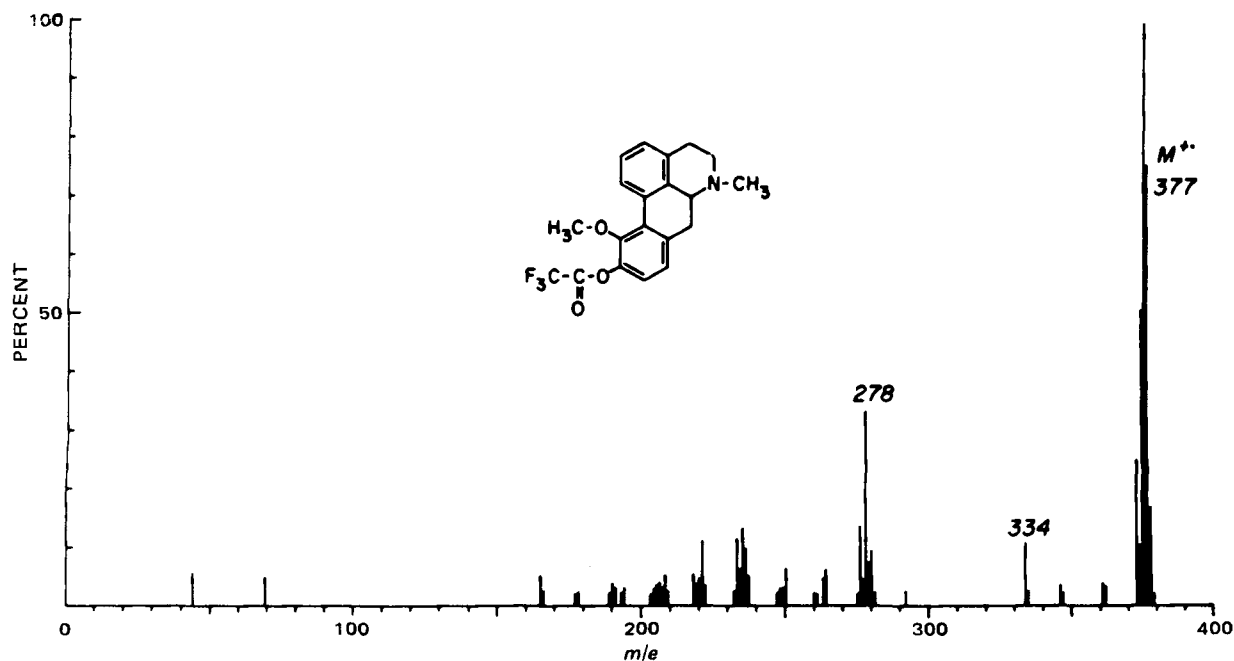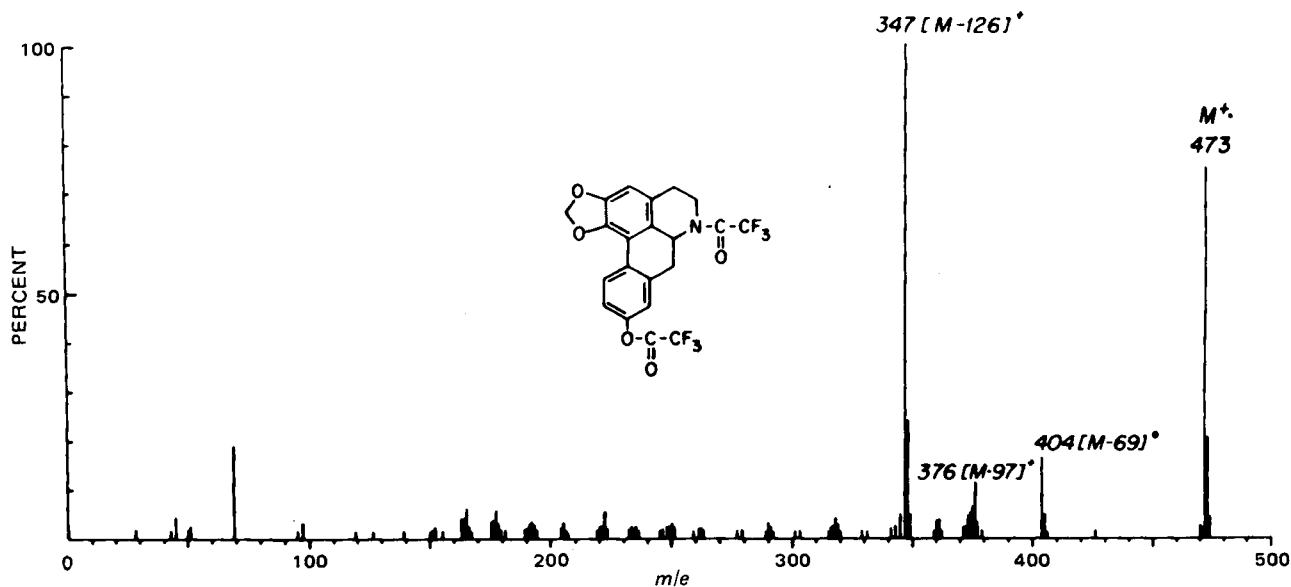**Figure 4**—*Mass spectrum of the O-trifluoroacetyl derivative of I*e.

Figure 5—*Mass spectrum of the* N,O-*bis(trifluoroacetyl) derivative of* IIe.

tion of whether this fragmentation could be used diagnostically to recognize the presence of a noraporphine was desired. The type of substitution on the secondary nitrogen of the underivatized aporphine generally is determined on the basis of the retro-Diels-Alder fragmentation. However, in noraporphines, this fragmentation is of reduced intensity (10% relative intensity) compared to that of the N-alkyl-substituted alkaloids. Accordingly, the mass spectra of the trifluoroacetyl derivatives of the following naturally occurring noraporphines were studied: sparsiflorine (Io), asimilobine (Ip), tuduranine (Iq), nandigerine (IIb), cassyfiline (IIc), xylopine (IId), anolobine (IIe), and actinodaphnine (IIf).

The partial mass spectra of these derivatives are listed in Table V, and the complete mass spectrum of anolobine is shown in Fig. 5. As with the other noraporphines, the mass spectra again were characterized by few major ions, of which the [M − 126]$^+$ ion was the most prominent. These

[M − 126]$^+$
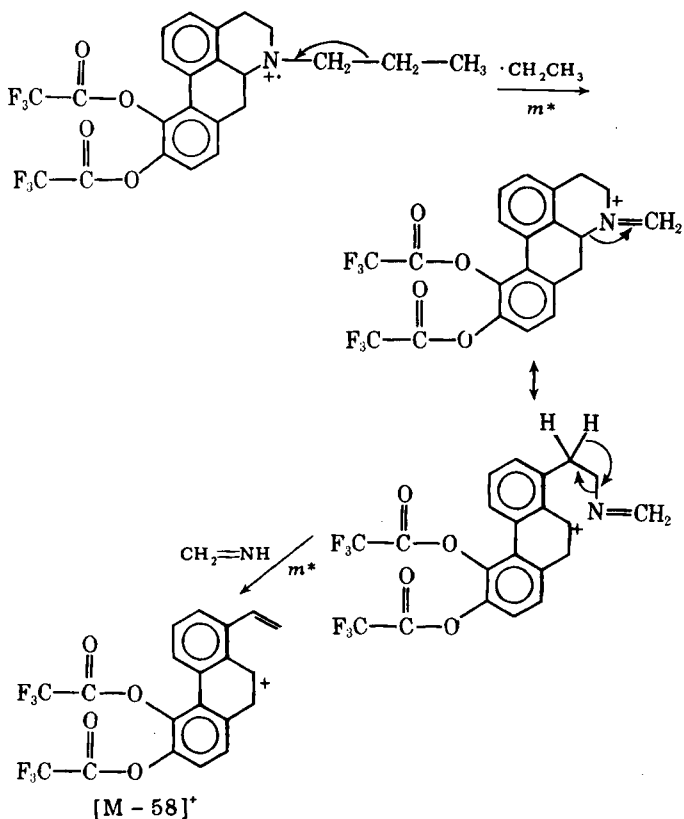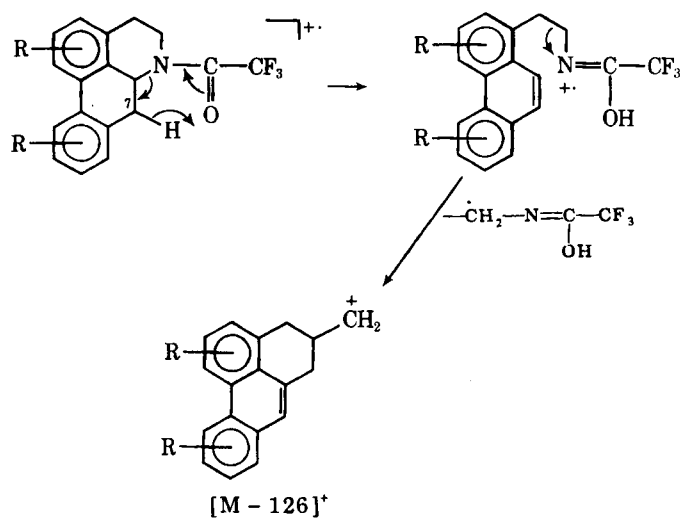
Scheme V

findings support the earlier supposition that this fragmentation can be used as a diagnostic tool to determine the degree of substitution on the nitrogen of aporphine alkaloids isolated from natural products or as possible metabolites in biological fluids.

CONCLUSIONS

The trifluoroacetyl derivative formation for aporphine and noraporphine alkaloids represents a reliable technique which has application in the analysis of biological fluids in drug metabolism studies, as well as in the characterization of naturally occurring aporphine alkaloids.

The trifluoroacetyl derivative offers the following advantages over the corresponding trimethylsilyl derivative:

[M − 58]$^+$

Scheme IV

Table IV—Partial Mass Spectra of N-n-**Propyl-**O-**trifluoroacetyl Derivatives of Aporphines**

Compound	M$^+$	[M − 1]$^+$	[M − 29]$^+$	[M − 58]$^+$	[M − 97]$^+$
Ib	487[a]	486	458	429	390
	(65)[b]	(38)	(100)	(42)	(24)
If	405	404	376	317	308
	(77)	(45)	(100)	(37)	(4)
Ij	375	374	346	317	278
	(49)	(41)	(100)	(46)	(7)
Ik	375	374	346	317	278
	(62)	(55)	(100)	(48)	(4)

[a] The m/e values. [b] Relative intensity.

Table V—Partial Mass Spectra of Trifluoroacetyl Derivatives of Noraporphines

Compound	M+·	[M − 69]+	[M − 97]+	[M − 126]+
I*l*	489[a]	420	392	363
	(100)[b]	(21)	(5)	(46)
I*m*	571	502	474	445
	(100)	(8)	(36)	(85)
I*n*	653	584	556	527
	(0)	(27)	(17)	(100)
I*o*	571	502	474	445
	(100)	(11)	(12)	(57)
I*p*	459	390	362	333
	(53)	(6)	(4)	(100)
I*q*	489	420	392	363
	(71)	(10)	(12)	(100)
II*a*	503	436	406	377
	(100)	(6)	(30)	(99)
II*c*	533	464	436	407
	(43)	(2)	(8)	(100)
II*d*	391	322	294	265
	(56)	(7)	(8)	(100)
II*e*	473	404	376	347
	(76)	(17)	(11)	(100)
II*f*	503	434	406	377
	(49)	(2)	(3)	(100)

[a] The m/e values. [b] Relative intensity.

1. The O-trifluoroacetyl derivatives have increased volatility.
2. The O-trifluoroacetyl derivatives of apocodeine and isoapocodeine can be separated by GLC.
3. The mass spectra of the O-trifluoroacetyl derivatives of apocodeine and isoapocodeine show characteristically different fragmentation patterns that can be applied to the characterization of 10- or 11-hydroxylated aporphines.
4. A fragmentation leading to the formation of an ion at a mass value corresponding to [M − 126]+ can be used diagnostically to determine the degree of substitution on the nitrogen of aporphine alkaloids.

REFERENCES

(1) G. C. Cotzias, P. S. Papavasiliou, E. S. Papavasiliou, E. S. Tolosa, J. S. Mendez, M. Bell-Midura, and B. Kaufman, *N. Engl. J. Med.*, **294**, 597 (1976).
(2) P. S. Papavasiliou, G. C. Cotzias, V. L. Rosal, and S. T. Miller, *Arch. Neurol.*, **35**, 787 (1978).
(3) M. K. Menon, W. G. Clark, and J. L. Neumeyer, *Eur. J. Pharmacol.*, **52**, 1 (1978).
(4) P. N. Kaul, E. Brochmann-Hanssen, and E. L. Way, *J. Pharm. Sci.*, **50**, 244 (1961). *Ibid.*, **50**, 248 (1961).
(5) K. Missala, S. Lal, and T. L. Sourkes, *Eur. J. Pharmacol.*, **22**, 54 (1973).
(6) R. V. Smith and S. P. Sood, *J. Pharm. Sci.*, **60**, 1654 (1971).
(7) R. V. Smith, P. W. Erhardt, J. L. Neumeyer, and R. J. Borgman, *Biochem. Pharmacol.*, **25**, 2106 (1976).
(8) J. L. Neumeyer, F. E. Granchelli, K. Fuxe, V. Ungerstedt, and H. Corrodi, *J. Med. Chem.*, **17**, 1090 (1974).
(9) J. L. Neumeyer, J. F. Reinhard, W. P. Dafeldecker, J. Guarino, D. S. Kosersky, K. Fuxe, and L. Agnati, *ibid.*, **19**, 25 (1976).
(10) R. I. Schoenfeld, J. L. Neumeyer, W. Dafeldecker, and S. Roffler-Tarlov, *Eur. J. Pharmacol.*, **30**, 63 (1975).
(11) J. L. Neumeyer, N. J. Uretsky, and R. R. Charest, *J. Pharm. Pharmacol.*, **29**, 179 (1977).
(12) R. V. Smith and R. M. Cook, *J. Pharm. Sci.*, **63**, 161 (1974).
(13) J. V. Evans and P. Vouros, *Anal. Biochem.*, **90**, 389 (1978).
(14) R. T. Coutts, G. R. Jones, A. Benderly, and A. L. C. Mak, *J. Chrom. Sci.*, **17**, 350 (1979).
(15) J. F. Green, J. V. Evans, J. L. Neumeyer, and P. Vouros, *Biomed. Mass Spectrom.*, **6**, 282 (1979).

Decomposition of Aminophylline in Suppository Formulations

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Abstract □ An aminophylline suppository product, when stored at room temperature, was found to be deficient in ethylenediamine content by the USP XIX assay and by a specific method for primary amines. The product also had a melting point that was considerably higher than body temperature. An accelerated decomposition experiment, conducted on normal suppositories of identical original composition, yielded a product refractory at steam bath temperatures and containing no ethylenediamine measurable by the USP assay. The suppositories from both the original sample and the decomposition experiment contained considerable amounts of a white material, which melted at ~150° and which consisted of the diamide products formed by the reaction of ethylenediamine and the fatty acids present in coconut and palm kernel oils. The results, which confirmed the work of Cieszynski, showed that the ethylenediamine constituent of aminophylline can react with suppository base materials to produce insoluble amide decomposition products.

Keyphrases □ Aminophylline—decomposition in suppository formulations □ Suppositories—aminophylline, decomposition in suppository formulations □ Decomposition—aminophylline in suppository formulations

During a study by the Food and Drug Administration which required the analysis of various dosage forms of xanthine derivative drugs, an aminophylline suppository formulation was encountered that unexpectedly failed to melt when heated in a beaker on a steam bath. In fact, the suppositories barely softened, and a composite was prepared for assay only with great difficulty. Analysis by the USP XIX method (1) showed that the suppository contained approximately the labeled amount of theophylline but considerably less than the compendial requirement for ethylenediamine. The suppository sample had been stored at room temperature for a month before analysis, so the original condition of the product was unknown.

An accelerated decomposition experiment was devised to study the phenomenon of hardening and to elucidate the decomposition that apparently had occurred. Ci-