obvious difference in fine structure in the two curves. Such fine structure differences are usually noted with changing solvent polarity when the transition involved is $n \rightarrow \pi^*$.[15]

The locations of the Cotton effects of compound IV at 237 mμ ($\Theta = 5.7 \times 10^4$) and 267 mμ ($\Theta = -3.3 \times 10^4$) in cyclohexane are shifted to higher wavelengths from those observed in the more polar trifluoroethanol. Such a shift is normally strong evidence for the assignment of an $n \rightarrow \pi^*$ transition. However, Sidman[16] points out that $\pi \rightarrow \pi^*$ transitions which shift to shorter wavelengths in polar solvents are known in heteropolar systems such as pyridine N-oxide. In view of this and because of the other pieces of evidence already cited, we feel that we are dealing with a $\pi \rightarrow \pi^*$ transition.

In addition to the Cotton effects already discussed, compound IV in cyclohexane exhibits an additional Cotton effect at 207 mμ ($\Theta = -1.2 \times 10^4$). We have no ready explanation of this effect other than that the transition involved could be olefinic in nature as suggested by Sandman and Mislow,[17] who observed a similar Cotton effect at 207.5 mμ in their CD studies on (+)-2-methylenebenznorbornene.

Experimental Section

The CD studies were carried out at room temperature using a Cary 60 spectropolarimeter with CD attachment. The compound LL-BH872α acetate Ib was examined in trifluoroethanol at a concentration of 1.18 mg/ml using a 0.2-mm cell. Compound III, reduced LL-BH872α acetate, was studied at a concentration of 2.10 mg/ml in trifluoroethanol in a 0.2-mm cell while the CD curve of elaimocyin was made on a 2.60 mg/ml solution in trifluoroethanol in a 0.2-mm cell.

Oxidation of Elaiomycin. Approximately 54 mg (just over 0.2 mmole) of elaiomycin in 4–5 ml of ether was stirred with a solution consisting of 50 mg of potassium dichromate and 0.1 ml of concentrated sulfuric acid in 1.0 ml of water. After 1.5 hr stirring, thin-layer chromatography (tlc) on silica gel using the developing system hexane–ethyl acetate (60:40) showed that about 20% reaction had occurred. The reaction mixture was stored overnight in the refrigerator. Stirring at room temperature was continued the following day for 3.5 hr during which time the ether volume was replenished several times and two lots of 20 mg of potassium dichromate were added. Tlc then showed about 80% reaction had occurred so the ether phase was recovered and concentrated to 60 mg of faintly yellow oil which was chromatographed over 20 g of acid-washed silica gel (Davison grade 62) using the solvent system hexane–ethyl acetate (95:5). The desired product was eluted in the fourth and fifth holdback volumes. Evaporation of the solvent yielded 30 mg of colorless oil. The molecular ion by mass spectrum is *m/e* 256. The ir spectrum has a sharp carbonyl absorption band at 1715 cm^{-1} while the 3100–4000-cm^{-1} area is free of absorption bands. The nmr spectrum in carbon tetrachloride is definitive for the structure 4-methoxy-3-(1′-*cis*-octenylazoxy)-2-butanone. The 8′ terminal methyl signal is at δ 0.87. The methylene protons of the 4′, 5′, 6′, and 7′ carbon atoms appear as a broad singlet at 1.30. The sharp singlet at 2.12 integrating for three protons belongs to the ketonic methyl group of C_1. The multiplet at 2.60 which accounts for two protons is due to the allylic methylene group of C_3′. The methoxy substituent of carbon 4 appears at 3.32. The doublet (J = 6 Hz) at 4.45 is attributable to the single proton of carbon 3. The vinyl proton of C_1′ appears as a doublet of triplets at 6.77 (J = 9–10 Hz indicative of *cis* coupling) while the remaining vinyl proton of C_2′ is a multiplet at 5.70. The uv of oxidized elaiomycin has λ_{max} at 238 mμ (ϵ 9000) in methanol; $[\alpha]^{25}D$ −87.8° ± 2.0 (c 0.148, methanol). The CD curves of oxidized elaiomycin IV were made on a 1.04 mg/ml solution in trifluoroethanol and on a 0.97 mg/ml solution in cyclohexane. The cell width in each case was 0.2 mm.

Acknowledgments. We wish to thank Professor Murray Goodman of the Polytechnic Institute of Brooklyn for the CD curves and our colleagues, Mr. W. Fulmor and his associates, for the spectral and rotational data.

(15) W. D. Closson and P. Haug, *J. Amer. Chem. Soc.*, **86**, 2384 (1963).
(16) J. W. Sidman, *Chem. Rev.*, 689 (1958).
(17) D. J. Sandman and K. Mislow, *J. Amer. Chem. Soc.*, **91**, 645 (1969).

Chemical Ionization Mass Spectrometry of Complex Molecules. II. Alkaloids

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Abstract: The chemical ionization mass spectra of 29 alkaloids, representing nine of the eighteen major alkaloid families, have been measured using methane as reactant gas. These spectra are discussed in terms of the structure of the compounds, with particular reference to their conventional electron impact mass spectra. The quasi-molecular ion $(M + 1)^+$ is invariably more relatively abundant in the chemical ionization mode than is the molecular ion in the electron impact mode. Both spectra supply structural data that is often of a complementary nature, *via* fragmentation.

No area of organic chemistry has remained unaffected by mass spectrometry, which during the last decade has been developed into a routine analytical technique of uniquely high sensitivity, having particular value in the general field of structure determination. The high sensitivity of the method has led to its rapid acceptance and exploitation by those working on the characterization of natural products. In this area alkaloids have been the subject of a very large number of reports.[1]

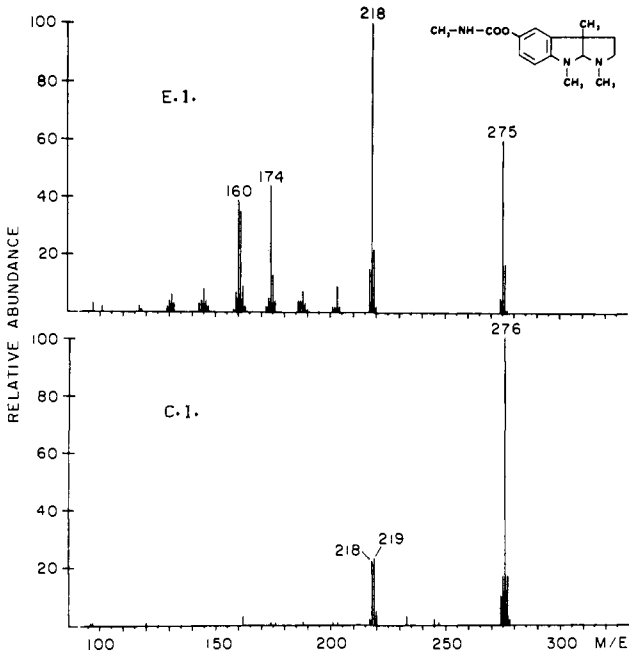


Figure 1. Mass spectra of physostygmine (I).

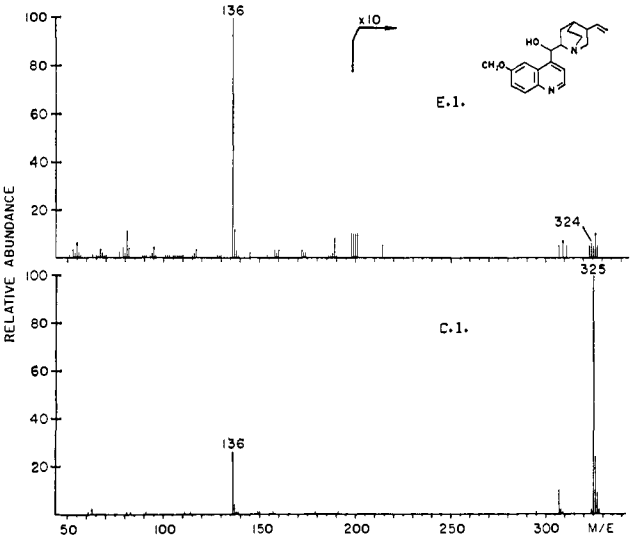


Figure 2. Mass spectra of quinine (VII).

The effectiveness of electron impact (EI) mass spectrometry in the alkaloid field is probably related to the fact that these compounds are usually cyclic amines, often containing aromatic rings. The first of these factors ensures that the extensive β cleavage expected from the molecular ions of the amines will not automatically result in complete loss of the molecular ion. In an ideal case, when the nitrogen atom is located at a bridgehead position, as in the pyrrolizidine alkaloids, an abundant molecular ion may be expected. Compounds containing aromatic rings also tend to give more abundant molecular ions in most cases and also serve as intact entities whose presence can be traced throughout the mass spectral pattern. In spite of these points however, the high energy content and radical nature of molecular ions derived by electron impact is often a serious drawback, in that cleavage of carbon–carbon bonds is common, and leads to very complex fragmentation patterns further complicated by skeletal rearrangement.[2] In view of this, it seemed reasonable to consider other methods of forming ions, and of the various possibilities such as field ionization, photoionization, etc. we chose to investigate chemical ionization with methane,[3] a technique in which ions are formed in ion–molecule collisions, involving protonation or hydride abstraction as the primary process. The basicity of most alkaloids and consequent stability of their protonated forms suggest that quasimolecular $(QM)^+$ ions formed in chemical ionization (CI) mass spectrometry should be generally stable.

We have previously reported[4] preliminary results of this work which suggested promise for the method in analysis of complex molecules and this present report deals with the methane chemical ionization mass spectra of some 29 alkaloids in which (i) a QM^+ ion is never absent, (ii) identification of aliphatic hydroxyl and methoxyl groups is always possible, and (iii) skeletal information from rearrangement reactions is generally absent. The results show that CI and EI mass spectra often complement one another well.

Experimental Section

All CI and EI mass spectra were measured at low resolving power (~2000) on an AEI MS-9 mass spectrometer equipped with a dual EI/CI ion source.[4] In both modes, the samples were admitted *via* a direct insertion probe at ion source temperatures of 100–200°. In the CI mode, quantities similar to those used in the EI mode were employed. Methane was the reactant gas in all cases. Pressures between 0.8 and 1.1 mm were obtained as noted on a source housing ion gauge indicating $\sim 2 \times 10^{-4}$ mm which had been previously calibrated both by a McLeod gauge in the ion chamber and by the methane ion–pressure dependence curves as given by Field.[3] The ratio of CH_5^+ (*m/e* 17) to H_3O^+ (*m/e* 19) was always maintained greater than 1 to ensure that the former and not the latter was the main reactant species.

Although the small number of peaks in the CI mass spectra shown in Figures 1–7 would suggest difficulty in counting the spectra, this is not the case. At high gain, ions from either the sample or background are seen at about every mass unit, particularly below *m/e* 300. At higher masses, internal standards such as hydrocarbons, which give peaks at least every 14 mμ[5] may be useful, but were not in fact employed in this work.

The following alkaloids were studied: physostygmine,[6] calycanthine,[7] dicentrine,[8] strychnine,[9] glaucine,[8] caffeine,[10] quinine,[6] protopine,[8] colchicine,[11] L-α-isosparteine,[12] lupanine,[12] 13-hydroxylupanine,[13] 13-epihydroxylupanine,[12] cytisine,[12] N-methylcytisine,[14] anagyrine,[12] angustifoline,[13] ephedrine,[11] cassine,[15] homatropine,[16] hyoscyamine,[16] tecomanine,[17] galanthine,[18] lycorine,[18] haemanthidine,[18] buphanisine,[18] tazettine,[18] and yohimbine.[9]

(1) K. Biemann, *Fortschr. Chem. Org. Naturstoffe*, **24**, 1 (1966); H. Budzikiewicz, C. Djerassi, and D. H. Williams, "Structural Elucidation of Natural Products by Mass Spectrometry," Vol. 1, Holden-Day, San Francisco, Calif., 1964.
(2) R. G. Cooks, *Org. Mass Spec.*, **2**, 481 (1969).
(3) F. H. Field, *Accounts Chem. Res.*, **1**, 42 (1968).
(4) H. M. Fales, G. W. A. Milne, and M. Vestal, *J. Amer. Chem. Soc.*, **91**, 3682 (1969).
(5) M. S. B. Munson and F. H. Field, *ibid.*, **88**, 2621 (1966).
(6) Merck & Co., Rahway, N. J.
(7) Pierce Chemical Co., Rockford, Ill.
(8) Eastman Kodak Co., Rochester, N. Y.
(9) Mallinckrodt Chemical Works, St. Louis, Mo.
(10) National Biochemical Corp., Cleveland, Ohio.
(11) Fisher Scientific Company, Silver Spring, Md.
(12) K & K Laboratories, Plainview, N. Y.
(13) H. A. Lloyd, *J. Org. Chem.*, **26**, 2143 (1961).
(14) H. A. Lloyd and E. C. Horning, *ibid.*, **23**, 1074 (1958).
(15) R. J. Highet and P. F. Highet, *ibid.*, **31**, 1275 (1966).
(16) Gifts from the Meer Corp., N. Bergen, N. J.
(17) G. Jones, H. M. Fales, and W. C. Wildman, *Tetrahedron Lett.*, 397 (1963).
(18) From the alkaloid collection of this laboratory. See W. C. Wildman, *Alkaloids*, **6**, 289 (1960).

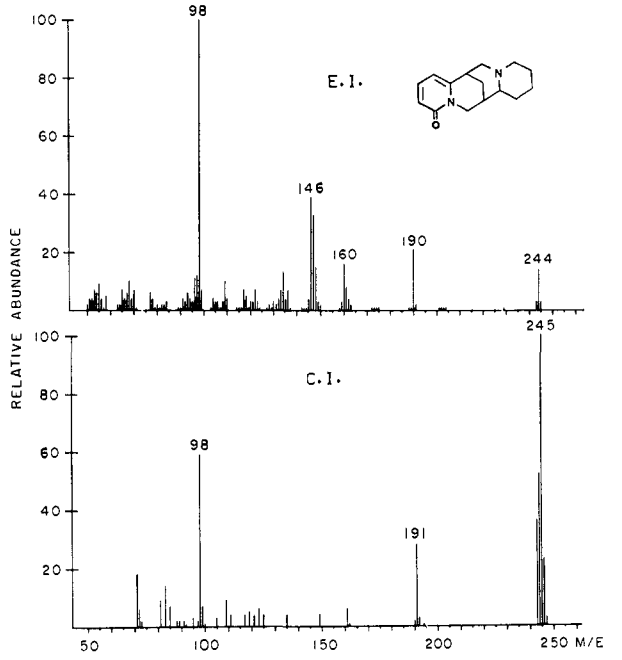


Figure 3. Mass spectra of anagyrine (XIV).

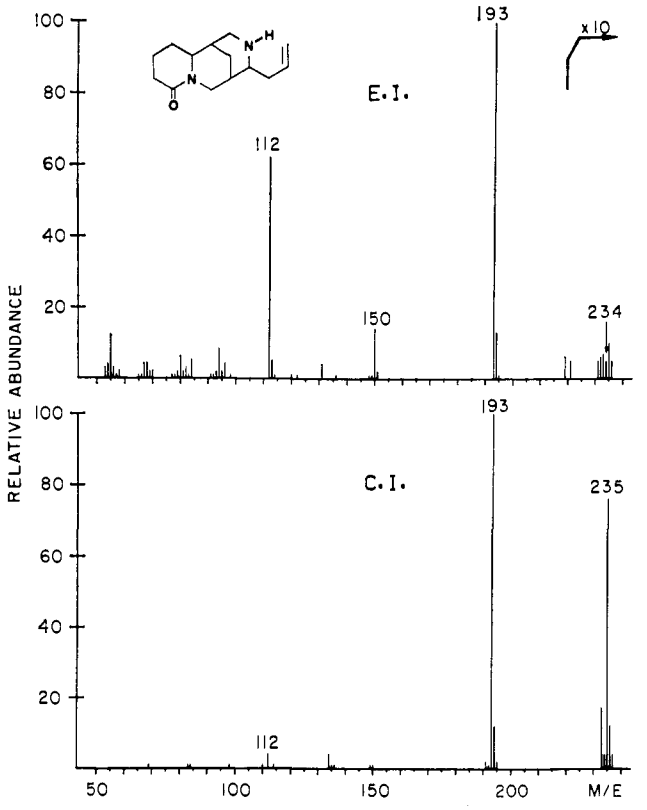


Figure 4. Mass spectra of angustifoline (XV).

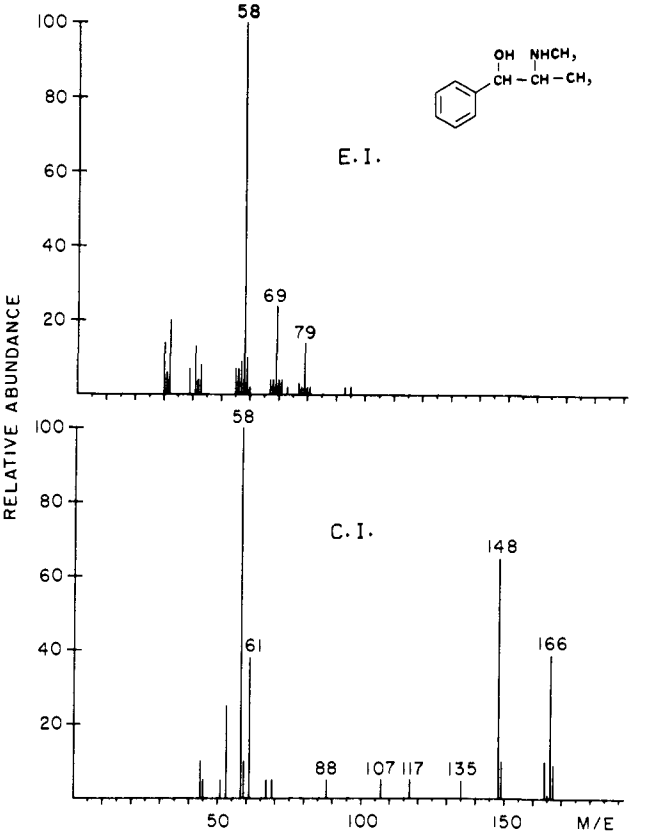


Figure 5. Mass spectra of ephedrine (XVI).

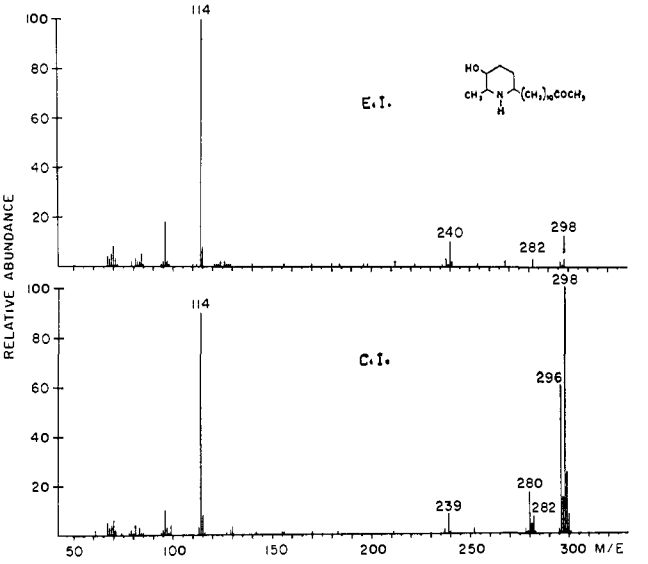
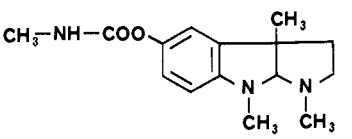


Figure 6. Mass spectra of cassine (XVII).

Results and Discussion

The EI mass spectrum[19] of physostygmine (I), shown in Figure 1, has an abundant molecular ion (63%) and a small ion at m/e $(M - 1)^+$. The base peak of the spectrum, at m/e 218, is formed by loss of the elements of methyl isocyanate from the urethane system. In the CI mass spectrum of I, the base peak is the QM^+ ion (m/e 276) accompanied by ions at m/e 275 (14%) and 274 (10%) as well as a ^{13}C satellite at m/e 277 (14%). The only two significant fragment ions

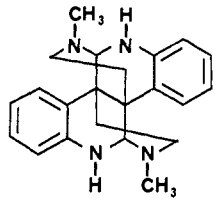


I

are at m/e 218 (21%) and 219 (22%). These six ions together account for 87% of the total ion current.

(19) E. I. Clayton and R. I. Reed, *Tetrahedron*, **19**, 1345 (1963); G. Spiteller and M. Spiteller-Friedmann, *Tetrahedron Lett.*, 147 (1963).

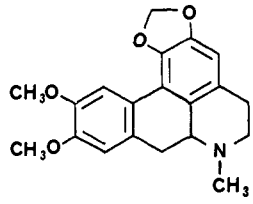
No skeletal fragmentation has occurred and the only fragment ions are those formed by loss of either protonated or unprotonated methyl isocyanate, from the QM^+ ion. The stability of this type of skeleton in CI mass spectrometry is seen again in the CI mass spectrum of the related alkaloid calycanthine (II), where the QM^+ ion (*m/e* 347, 100%) is virtually the only ion in the spectrum. Together, the ions at *m/e* 346, 347, and



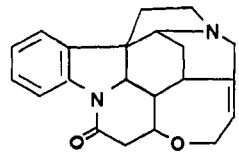
II

348 account for 76% of the total ion current above *m/e* 60. This stands in contrast to the EI mass spectrum of II[20] in which extensive fragmentation of the skeleton is observed.

Such great stability of the QM^+ ion appears to be general in alkaloids which do not possess aliphatic functional groups. Thus in the CI mass spectrum of dicentrine (III) and strychnine (IV), the QM^+ ions (*m/e* 340 and 335, respectively) and their satellites at *m/e* ($QM^+ - 2$), ($QM^+ - 1$), and ($QM^+ + 1$) account for the majority of the total ionization. Strychnine



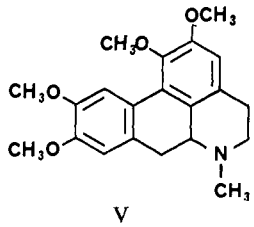
III



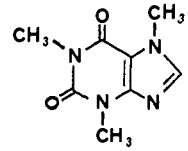
IV

(IV) is known[21] for its reluctance to fragment even in EI mass spectrometry, but the EI mass spectrum of III[22] shows a considerable amount of informative fragmentation, major ions being observed from straightforward processes at *m/e* 339 (M^+, 61%), 338 ($M^+ - 1$, 100%), 324 ($M^+ - 15$, 10%), 307 ($M^+ - 32$, 13%), 296 ($M^+ - 43$, 28%), and 265 ($M^+ - 74$, 20%).

The CI mass spectra of glaucine (V) and caffeine (VI) similarly consist only of the QM^+ ions (*m/e* 356 and 195, respectively), both of which are base peaks, and the usual fragment ions at *m/e* ($QM^+ - 2$), ($QM^+ - 1$).



V



VI

The EI mass spectra of both V[23] and VI[24] are considerably more complicated. It is clear that in this

(20) E. I. Clayton, R. I. Reed, and J. M. Wilson, *Tetrahedron*, **18**, 1495 (1962).

(21) K. Biemann, "Mass Spectrometry," McGraw-Hill Book Co., Inc., New York, N. Y., 1962, p 332.

(22) A. H. Jackson and J. A. Martin, *J. Chem. Soc., C*, 2222 (1966).

(23) A. H. Jackson and J. A. Martin, *ibid.*, 2061 (1966).

(24) G. Spiteller and M. Spiteller-Friedmann, *Monatsh. Chem.*, **93**, 634 (1962).

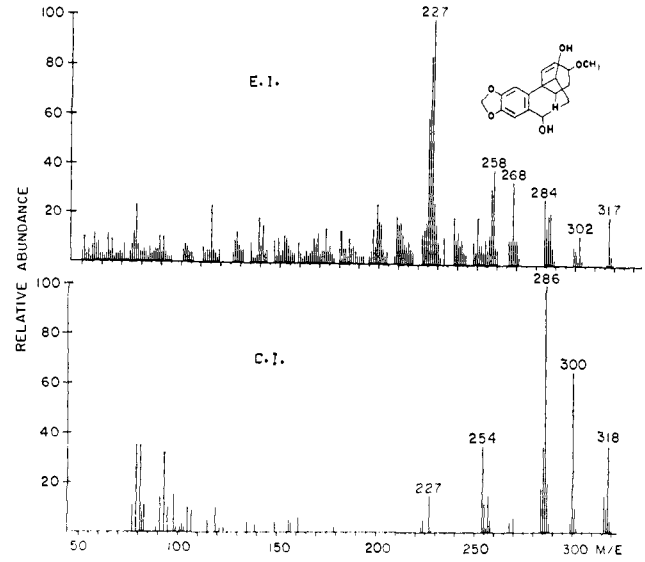
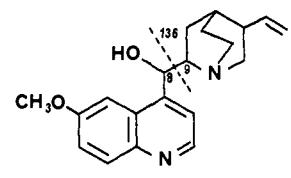


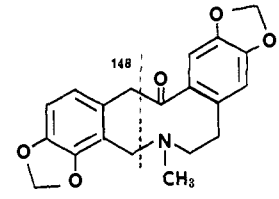
Figure 7. Mass spectra of haemanthidine (XXIV).

type of alkaloid, the main use of CI mass spectrometry will be to confirm the identity of a molecular ion.

On the other hand, the complementary nature of CI and EI mass spectra is very clear in the case of quinine (VII), whose spectra are shown in Figure 2. In this case, the predictable lability toward electron bombardment of the C_8–C_9 bond leads to the formation of the base peak at *m/e* 136,[25] relative to which the abundance of the molecular ion at *m/e* 324 is only 0.5%. In the CI mass spectrum of VII however, the base peak is the QM^+ ion while the peak at *m/e* 136 has a relative abun-



VII



VIII

dance of only 30%. The CI mass spectra of quinine and quinidine differ quantitatively from each other as do those of cinchonine and cinchonidine. These differences, which are temperature dependent, are the subject of further study in this laboratory.

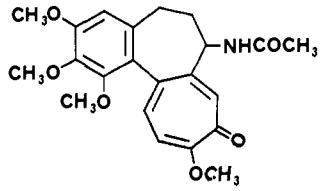
The EI mass spectrum of protopine (VIII) similarly has a major fragment ion at *m/e* 148 as the base peak[26a] and the molecular ion at *m/e* 353 has a relative abundance of 3%. In addition to these two ions, there are fourteen other ions of significant abundance at *m/e* 163 (21%), 177 (3%), 190 (9%), 204 (2%), 209 (3%), 224 (2%), 237 (2%), 252 (4%), 267 (6%), 281 (4%), 295 (2%), 309 (2%), 322 (1%), and 338 (1%). In the CI mass spectrum of VIII, the QM^+ (*m/e* 352, 84%) is relatively stable, perhaps as a result of protonation on the carbonyl oxygen with transannular interaction by the nitrogen.[26b] The nature of the ion at *m/e* 148 (100%) is unclear because by analogy with the EI spec-

(25) G. Spiteller and M. Spiteller-Friedmann, *Tetrahedron Lett.*, 153 (1963).

(26) (a) L. Dolejs, V. Hanus, and J. Slavik, *Collect. Czech. Chem. Commun.*, **29**, 2479 (1964); (b) F. von Bruchhausen, *Arch. Pharm.*, **260**, 97 (1922). See also, N. J. Leonard and R. R. Sauers, *J. Org. Chem.*, **22**, 63 (1957), and references cited therein.

trum, it is unlikely to contain nitrogen and must therefore be an odd electron ion.

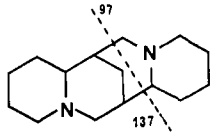
The very informative EI mass spectrum of colchicine (IX) has an ion of *m/e* 312 which is considered to be formed by successive loss of CO and C_2H_5NO (acetamide) from the molecular ion at *m/e* 399.[27] Such loss of CO has never been observed[28] in CI mass spectra of ketones and does not occur here. It is noted that although the ester linkage in physostygmine cleaves in both CI and EI mode, the amide linkage in IX is stable



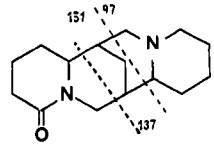
IX

to methane CI. The CI mass spectrum of colchicine shows only a QM^+ ion at *m/e* 400.

The CI mass spectra of a series of quinolizidine alkaloids were measured. Spectra were obtained from 1-α-isosparteine (X), lupanine (XI), 13-hydroxylupanine (XII, R_1 = H; R_2 = OH), 13-epihydroxylupanine (XII, R_1 = OH; R_2 = H). In addition, the spectra of the related alkaloids cytisine (XIII, R = H), N-methylcytisine (XIII, R = Me), and anagyrine (XIV) were

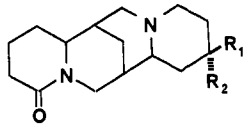


X

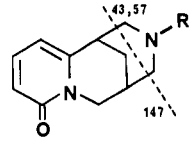


XI

measured. The CI mass spectrum of 1-α-isosparteine

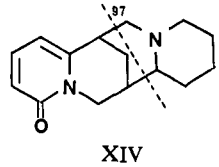


XII



XIII

(X) shows, as expected, relatively little fragmentation, with the result that the QM^+ ion (*m/e* 235, 77%) and the ($QM^+ - H_2$) ion (*m/e* 233, 100%) together account for over 60% of the total ionization. The high abundance of the $QM^+ - H_2$ ion is noteworthy as is the relatively high abundance of the $(M - H)^+$ ion (38% of the molecular ion) in the EI spectrum.[29] The two most abundant ions in the EI spectrum are at *m/e* 98 (100%) and 137 (90%). These ions, which presumably result from cleavage as shown in X with hydrogen transfer are also

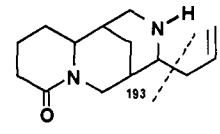


XIV

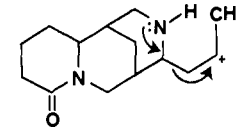
present in the CI mass spectrum but with somewhat reduced abundances, 52% and 48%, respectively. Attachment of a carbonyl group as in lupanine (XI) does not sponsor any new fragmentation route in either the CI or the EI spectrum. In the former, the ions at *m/e* 247, 248, 249, and 250 account for 82% of the total ionization. In the latter, major fragment ions appear at *m/e* 136 and 149, resulting from cleavage analogous to that in 1-α-isosparteine, again with hydrogen transfer.

The spectra of the epimeric 13-hydroxylupanines are dominated by the hydroxyl group. In both cases, the QM^+ ion (*m/e* 265) is the base peak of the CI mass spectrum and the only major fragment ion is that at *m/e* 247 (33% in 13-hydroxylupanine and 59% in 13-epihydroxylupanine).

The QM^+ ion (*m/e* 191, 100%) is the only important ion in the CI mass spectrum of cytisine (XIII, R = H), but in the EI mass spectrum of the same compound, the base peak is actually a fragment ion (*m/e* 146) and the molecular ion (*m/e* 190) has an abundance of 69%. In the EI mass spectra of N-methylcytisine (XIII, R = Me) however, the other fragment from the same cleavage (*m/e* 58) is the base peak, the molecular ion having an abundance of 25%. The QM^+ ion at *m/e* 205 is the base peak of the CI mass spectrum of N-methylcytisine. In the case of anagyrine (XIV), the CI and EI mass spectra complement each other as can be seen in Figure 3. The CI mass spectrum has the QM^+ ion as the base peak and an ion of *m/e* 98 (57%) as the other major peak. In the latter, the molecular ion (*m/e* 244, 14%) is subordinate to the base peak at *m/e* 98. In both cases, the loss of the elements of cyclopropenone from the dienoid ring takes place but this process, predictably, is more important in the EI mass spectrum. A final alkaloid from the quinolizidine family is angustifoline (XV), whose allyl side chain is such a labile point in the molecule that upon electron bombardment, its loss is virtually complete. The molecular ion at *m/e* 234 is of very low abundance (1%), loss of C_3H_5 giving the base peak at *m/e* 193, presumably by a simple cleavage. In the CI mass spectrum shown in Figure 4, the QM^+ ion (*m/e* 235, 76%) is relatively stable perhaps because only in species where the double bond is protonated (XVa) can loss of C_3H_6 ensue giving the base peak at *m/e* 193, with a very intense metastable ion at 158.5.



XV



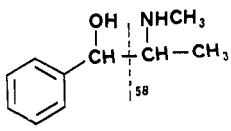
XVa

The biologically important ethanolamine, ephedrine (XVI), is a substituted benzyl alcohol and thus the absence of a molecular ion in its EI mass spectrum (Figure 5) is not surprising. The CI mass spectrum of XVI on the other hand could stand alone as a complete structure proof. The QM^+ ion at *m/e* 166 permits identification of a molecular formula; loss of water to give the ion at *m/e* 148 or of methylamine to give that at *m/e* 135 reveals the presence of the two functional groups, –OH and $-NHCH_3$, and the ions at *m/e* 58 ($CH_3CH{=}NH^+CH_3$) and 107 ($C_6H_5CH{=}OH^+$) can be

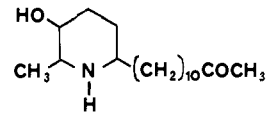
(27) J. M. Wilson, M. Ohashi, H. Budzikiewicz, F. Santavy, and C. Djerassi, *Tetrahedron*, **19**, 2225 (1963).
(28) See accompanying paper: H. Ziffer, G. W. A. Milne, H. M. Fales, and F. H. Field, *J. Amer. Chem. Soc.*, **92**, 1597 (1970).
(29) N. Neumer-Jehle, H. Nesvadba, and G. Spiteller, *Monatsh. Chem.*, **95**, 687 (1964).

simply and unambiguously reassembled to give the correct structure.

The piperidine alkaloid cassine[15] (XVII) gives an EI mass spectrum shown in Figure 6, which is rather unhelpful from the point of view of structure determination. The most abundant ion in the molecular ion



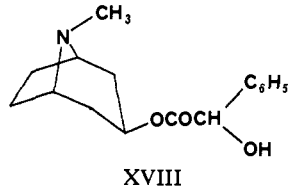
XVI



XVII

region, at *m/e* 298 (3%), is actually an (M + 1) ion. Such ions at *m/e* $(M + 1)^+$ have been noted repeatedly[30] in EI mass spectrometry and are often helpful. Their intensity usually varies as the square of the pressure in the source and they are therefore considered to be the result of ion–molecule reactions. Probably, in cases such as that of cassine (XVII), the alkaloid is its own source of protons and this is thus a special case of chemical ionization. There is no ion in the EI mass spectrum of XVII corresponding to the loss of the hydroxyl group and the origin of the ion at *m/e* 282 is unclear in the absence of certainty about the molecular weight. The base peak at *m/e* 114 is due to the ion formed by loss of the entire C_{12} side chain. Study of the CI spectrum of cassine (Figure 6) serves to clarify the situation considerably. The appearance of a QM^+ ion at *m/e* 298 (100%) and a $QM^+ - H_2$ ion at *m/e* 296 (60%) establishes the molecular weight as 297. Loss of water from the QM^+ ion gives the ion at *m/e* 280 (16%) and loss of methane from the same ion gives an ion at *m/e* 282 (7%). As in the EI mass spectrum, an ion at *m/e* 114 (89%) is formed by the loss of the entire C_{12} side chain. As is discussed later, such cleavage is probably a second step, following formation of the QM^+ ion by protonation at some point on the side chain, which is then released as a neutral molecule. The CI mass spectrum of cassine permits the identification of the three substituents on the nucleus and is therefore of considerably greater value from the point of view of structure determination than is the EI mass spectrum.

In the tropane alkaloids the EI and CI mass spectra tend to be complementary as in the quinine case. Thus, although the EI mass spectrum of a homatropine (XVIII)[31] shows a very small molecular ion (*m/e* 275,

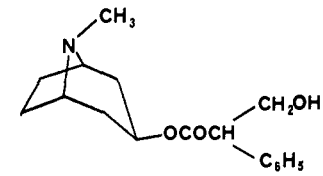


XVIII

2%), there is no ion at *m/e* 258 and thus the presence of a hydroxyl group passes unnoticed. The CI mass spectrum, however, has a QM^+ ion (*m/e* 276, 12%) and an ion corresponding to $(QM^+ - H_2O)$ (*m/e* 258, 2%). In both spectra the easy cleavage of esters is again seen and accounts for the major fragmentation. Similar behavior is exhibited by hyoscyamine (XIX), although

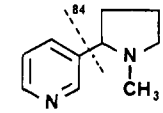
(30) Reference 21, p 220.

(31) E. C. Blossey, H. Budzikiewicz, M. Ohashi, G. Fodor, and C. Djerassi, *Tetrahedron*, 20, 585 (1964).



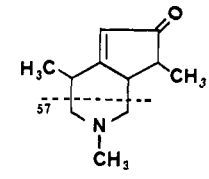
XIX

in this case, loss of OH is seen in the EI spectrum. The EI mass spectrum of nicotine (XX)[32] has only three major ions, one of which is derived by a rearrangement



XX

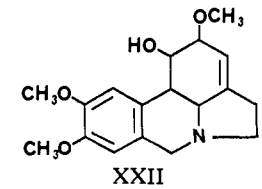
process. As befits a molecule so devoid of functional groups however, its CI mass spectrum is extremely simple. The QM^+ ion (*m/e* 163, 100%) and its satellites account for 84% of the total ionization. A further 9% is due to the single fragment ion (*m/e* 84) which is formed by cleavage of the bond joining the two rings and provides the base peak in the EI mass spectrum. A somewhat similar picture emerges from the spectra of the related alkaloid tecomanine (XXI). The extensive fragmentation in the EI mass spectrum of this



XXI

compound is the result of considerable rearrangement, undoubtedly due, in part, to the presence in the molecule of the unsaturated ketone system. Ketones do not appear to have the same labilizing influence in methane–CI mass spectrometry[28] and the CI spectrum of XXI is relatively simple, having a QM^+ ion at *m/e* 180 (100%) and a $QM^+ - H_2$ ion of much lesser abundance. The base peak in the EI spectrum is at *m/e* 57, but is at *m/e* 180 in the CI spectrum.

The EI mass spectra of the alkaloids of the amaryllidaceae, particularly those of the 5,10b-ethanophenanthridine system, are rather complicated and have proved to be difficult to interpret.[33] In this series, a great deal of information regarding functional groups can be easily derived from the appropriate CI mass spectrum. Thus the CI mass spectrum of galanthine (XXII) has a QM^+ ion at *m/e* 318 (65%), which loses methanol or water to give ions at *m/e* 286 (71%) and 300 (54%), respectively. In this case, the base peak is at *m/e* 316 $(QM^+ - H_2)$.

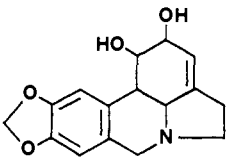


XXII

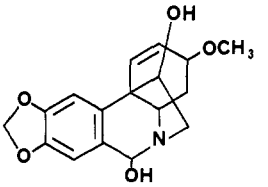
(32) W. F. Kuhn, C. J. Varsel, and W. A. Powell, Proceedings ASTM Committee E-14, New Orleans, La., 1962; F. W. McLafferty, *Anal. Chem.*, 28, 306 (1956); A. M. Duffield, H. Budzikiewicz, and C. Djerassi, *J. Amer. Chem. Soc.*, 87, 2926 (1965).

(33) A. M. Duffield, R. J. Aplin, H. Budzikiewicz, C. Djerassi, C. F. Murphy, and W. C. Wildman, *ibid.*, 87, 4902 (1965).

Lycorine (XXIII) gives a QM^+ ion at *m/e* 288 (85%), which loses a molecule of water to give the ion at *m/e* 270 as the base peak of the CI mass spectrum. As in the case of galanthine (XXII) the consecutive loss of both these oxygen functions does not occur, a point of some mechanistic significance, as is discussed later.

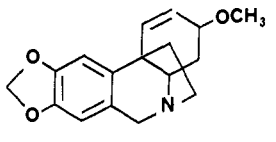


XXIII

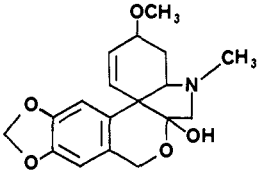


XXIV

In Figure 7 are shown the EI and CI mass spectra of haemanthidine (XXIV). In the latter, the QM^+ ion (*m/e* 318, 25%) loses either methanol, giving an ion at *m/e* 286 (100%), or water, giving the other major fragment ion, at *m/e* 300 (65%). As can be seen from Figure 7, such information is far less reliably derived from the EI mass spectrum. An important process in the CI mass spectrum of buphanisine (XXV) is the loss of methanol from the QM^+ ion (*m/e* 286, 100%) giving the ion at *m/e* 254 (77%). The same general behavior is exhibited by tazettine (XXVI) whose QM^+ ion (*m/e* 332, 40%) loses water to give an ion at *m/e* 314 (80%) or methanol to give one at *m/e* 300 (31%). Other alkaloids in this series give similar CI mass spectra, as



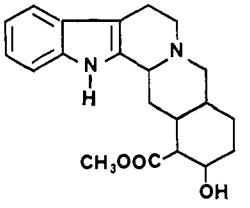
XXV



XXVI

has been previously reported.[4]

The indole alkaloid yohimbine (XXVII) yields little information beyond its molecular weight upon electron bombardment.[34] The molecular ion at *m/e* 354 is the base peak of the EI mass spectrum and the very low abundance of the fragment ions makes their significance tenuous. The CI mass spectrum of yohimbine however has a QM^+ ion (*m/e* 355) as the base peak and shows quite clearly the loss from this ion of water, giving the ion at *m/e* 337 (25%), methanol, giving that at *m/e* 323 (15%), and acetic acid, giving that at *m/e* 295 (3%). This spectrum permits therefore the definite identification of the two functional groups in the alkaloid.

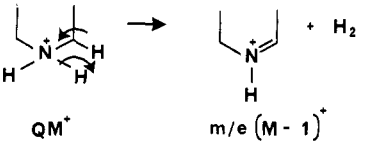


XXVII

All the foregoing CI mass spectra were measured using a plasma derived from methane at pressures between 0.8 and 1.1 mm. Under these conditions, the major active species are CH_5^+ and $C_2H_5^+$.[35] Upon collision of either of these ions with a neutral alkaloid molecule, a proton, CH_3^+ or $C_2H_5^+$, is transferred to the latter giving, respectively, a QM^+ ion $(M + 1)^+$, an ion at *m/e* $(M + 15)^+$, or one at *m/e* $(M + 29)^+$. The latter two ions are present in most alkaloid CI mass spectra with low abundance and for the sake of brevity they will not be considered here.[36]

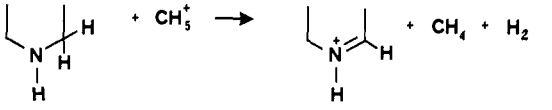
Transfer of a proton to the molecule under study however gives the QM^+ ion and in view of the presence in most alkaloids of a basic nitrogen atom, the high abundance (*i.e.*, stability) of the QM^+ ion is not unexpected. Of the compounds discussed above, none give, upon electron impact, a molecular ion that is more abundant than its corresponding QM^+ ion.

The common formation of an ion at *m/e* $(M - 1)^+$ in addition to the QM^+ ion may be interpreted either as loss of H_2 from the QM^+ ion (*e.g.*, XXVIII)—a process for which a metastable is sometimes, but not always observed—or as the direct abstraction of a hydride ion



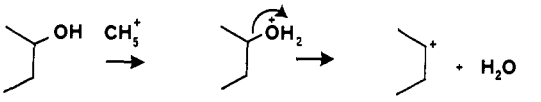
XXVIII

from the neutral molecule by CH_5^+ acting as a Lewis acid (*e.g.*, XXIX).[5] These two processes might be distinguishable by experiments with CD_4; possibilities of this sort are under investigation in this laboratory.



XXIX

It seems clear however, from the CI spectra reported here, that this loss of H_2 is the only important fragmentation following protonation of the basic nitrogen of an alkaloid. Protonation elsewhere in the molecule is really of more interest from the point of view of structure determination because it is often followed by fragmentation which affords information as to the nature of the functional groups present in the alkaloid molecule. The ubiquitous loss of water from the QM^+ ions of aliphatic alcohols is best explained in terms of protonation at the alcohol oxygen, followed by spontaneous loss of water as in XXX. A metastable ion for the second of these two steps is often observed indicating



XXX

(34) L. D. Antonaccio, N. A. Pereira, B. Gilbert, H. Vorbrueggen, H. Budzikiewicz, J. M. Wilson, L. J. Durham, and C. Djerassi, *J. Amer. Chem. Soc.*, **84**, 2161 (1962); G. Spiteller and M. Spiteller-Friedmann, *Monatsh. Chem.*, **93**, 795 (1962).

(35) F. H. Field and M. S. B. Munson, *J. Amer. Chem. Soc.*, **87**, 3289 (1965).

(36) Although they might be considered to constitute an annoyance, obscuring the parent ion region, this is not the case. In fact these ions always bear a fixed intensity relationship to each other (at a given reactant gas pressure) and to the QM^+ ion and so can be used to check that the QM^+ ion is indeed correctly assigned. If a question does arise, the pressure or nature of the reactant gas could be varied causing a predictable variation in the intensity of these peaks.

that it is a relatively slow process. Obviously, any structural feature, such as the double bond in lycorine (XXIII), which serves to stabilize the final carbonium ion, will greatly facilitate a sequence such as XXX and increase the relative abundance of the resulting fragment ion.

Loss of methanol from aliphatic methyl ethers is presumably a completely analogous process but in molecules such as haemanthidine (XXIV), which contains a methoxyl group and a hydroxyl group, the activating proton is presumably carried away with whichever function is lost and the charge left behind is stabilized by some other feature of the molecule so there is less tendency for a second fragment to be lost. Thus is observed the loss of 32 or 18 mass units from the QM^+ ion of XXIV, but there is no ion at m/e ($QM^+ - 50$).

In the light of the data reported here, methane CI mass spectrometry would appear to have great promise as a method for the mass spectrometric investigation of functional groups in organic molecules. Information regarding those groups that are more labile under conditions of electron bombardment may well be available by this technique and the complementary nature of the two types of spectra increased the value of both techniques.

Chemical Ionization Mass Spectrometry of Complex Molecules. III.[1] The Structure of the Photodimers of Cyclic α,β-Unsaturated Ketones

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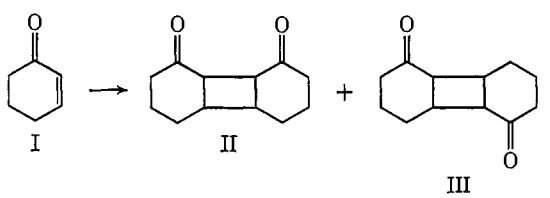
Contribution from the Laboratory of Physical Biology, National Institute of Arthritis and Metabolic Diseases, and Molecular Disease Branch, National Heart Institute, Bethesda, Maryland 20014, and the Corporate Research Laboratory, Esso Research and Engineering Company.
Received August 8, 1969

Abstract: The chemical ionization mass spectra of several pairs of head-to-head and head-to-tail photodimers of α,β-unsaturated cycloalkenones have been examined. The most abundant ion in the spectra of the head-to-head dimers is the quasimolecular ion while in the spectra of the head-to-tail dimers the ion at m/e $((M/2) + 1)^+$ is the most abundant ion. These results have been rationalized in terms of possible fragmentation paths of the protonated photodimers.

It has long been known[2] that irradiation of cyclic α,β-unsaturated ketones leads to dimerization with the formation of the so-called "photodimers."

A number of isomeric dimers is possible; however, these can be divided into two groups, head-to-head (h–h) and head-to-tail (h–t), which differ in that the former is a 1,4-diketone while the latter is a 1,5-diketone. Several isomers are frequently isolated from the irradiation of cyclohexenones,[3] cyclopentenones,[4] and cycloheptenones.[5] Cyclohexenone (I) for example, gives the h–h dimer (II) and the h–t dimer (III). Determination of the structure and stereochemistry about the cyclobutane ring is difficult. An unequivocal assignment of the structure greatly facilitates the remaining stereochemical assignments. Only in special cases do physical measurements such as ir or nmr spectroscopy aid in distinguishing between the two groups of dimers.[6,7] Since chemical ionization (CI) mass spectrometry has been shown[8] to be of value in differentiating between isomeric structures, the CI spectra of a series of h–h and h–t dimers were examined.



I II III

Experimental Section

In all the chemical ionization mass spectra reported here, the reactant gas was methane, at an ion chamber pressure of approximately 0.8 mm.[1] Thus the major protonating species are[9] CH_5^+ and $C_2H_5^+$. Samples of the photodimers were admitted to the source *via* a direct insertion probe with a source temperature of 50–100° except where otherwise noted.

(1) Part II: H. M. Fales, H. A. Lloyd, and G. W. A. Milne, *J. Amer. Chem. Soc.*, **92**, 1590 (1970).
(2) N. J. Turro, "Molecular Photochemistry," W. A. Benjamin, New York, N. Y., 1965, p 194.
(3) (a) D. Valentine, N. J. Turro, and G. S. Hammond, *J. Amer. Chem. Soc.*, **86**, 5202 (1964); (b) H. Ziffer, submitted for publication; (c) O. L. Chapman, *Rec. Chem. Progr.*, **28**, 167 (1967).
(4) P. E. Eaton, *J. Amer. Chem. Soc.*, **84**, 2344 (1962).
(5) P. E. Eaton, private communication.
(6) H. Ziffer, N. E. Sharpless, and R. D. Kan, *Tetrahedron*, **22**, 3011 (1966).
(7) We have also found that gas chromatography may be used to differentiate the isomers. The h–h isomer is always retained longer than its h–t counterpart on a nonpolar OV-1 phase. Thus Rt (h–h)/(h–t) for II/III = 1.14; VI/VII = 1.70; V/IV = 1.15; VIII/IX = 1.54; VIII/X = 2.12; XVI/XV = 1.43. The results are reasonable in view of the increased dipole moment of the h–h isomer and as expected, the effect is magnified on the more polar XE-60 phase, the above ratios becoming 1.34, 2.52, 2.78, 2.25, 3.21, and 2.19, respectively.
(8) J. D. Baty, G. W. A. Milne, and H. M. Fales, unpublished work.
(9) F. H. Field and M. S. B. Munson, *J. Amer. Chem. Soc.*, **87**, 3289 (1965).