Chemical Ionization Mass Spectrometry of Complex Molecules. IV.[1] Amino Acids

G. W. A. Milne, T. Axenrod,[2] and H. M. Fales

Contribution from the Molecular Disease Branch, National Heart and Lung Institute, National Institutes of Health, Bethesda, Maryland 20014. *Received November 21, 1969*

Abstract: The chemical ionization mass spectra of 21 of the 23 free "natural" amino acids have been measured. All give quasimolecular ions and simple fragmentation patterns which can be rationalized as arising from decomposition of the molecule protonated at several sites.

We have recently reported[3] the chemical ionization (CI) mass spectra of a series of natural products, in particular alkaloids[4] whose high proton affinity makes them especially susceptible to protonation by the strong acids CH_5^+ and $C_2H_5^+$ formed by electron bombardment of methane at pressures of the order of 1 Torr.[5]

This paper deals with the results obtained when this technique is applied to free amino acids. Earlier work in the area of electron impact (EI) mass spectrometry of free amino acids,[6-8] revealed that satisfactory spectra could be obtained with no contribution from diketopiperazines if either a direct insertion probe[6,8] or a crucible ion source[7] is used. In general however, the high ionizing energies led to mass spectra in which the molecular ion (M^+) was often of very low abundance (<1% except for methionine, phenylalanine, and tryptophan) and in which many rearrangements and much homolysis were occurring. Even when recourse is made to ester derivatives,[9-11] the molecular ion intensities are only 0.05–0.2% of the base peak except in the cases of methionine and the aromatic amino acids.

As an adjunct to our work on the problem of peptide sequencing by CI mass spectrometry[12] we have studied the CI mass spectra, with methane as the reactant gas, of all the common naturally occurring amino acids. These spectra have been measured on an MS-902 mass spectrometer equipped with the source described previously[3] using a direct insertion probe. Thus the high temperatures (~200–250°) required to vaporize the free amino acids have not been avoided but since ion formation is now carried out by protonation rather than the removal of an electron, the resulting even-electron species is relatively stable. The quasimolecular (QM^+) ion at *m/e* (M + 1) is, in fact, among the most intense ions except in those cases (glutamine, glutamic acid, ornithine) where cyclization with loss of water or ammonia might be expected to be particularly facile. The unambiguous assignment of this ion[13] probably constitutes the most important aspect of the technique from an analytical point of view.

Fragmentation Processes

(a) Loss of Water and $COOH_2$. Two types of cleavage common to all α-amino acids are loss of 18 (H_2O) and 46 amu ($COOH_2$) from the QM^+ ion. Loss of water from the QM^+ ion might be expected to be more important generally than the loss of HO· or H_2O from the EI molecular ion since it involves a transition between even-electron species (QM^+ and $QM^+ - H_2O$) and elimination of a stable molecule (ΔH_{H_2O} = *ca.* −60 *vs.* $\Delta H_{HO\cdot}$ = *ca.* +10 kcal/mol).[14] On the other hand, loss of the carboxyl or carbalkoxyl group has ample precedent in the EI mass spectrometry of both free amino acids[6-8] esters[9-11] where the resulting ion ($M^+ -$ COOH or $M^+ -$ COOR) is often the most abundant in the spectrum.

As seen in Tables I and II, the ions resulting from such cleavages are always present, loss of $COOH_2$ usually being particularly marked. In simple aliphatic, amino acids, such as glycine, alanine, valine, leucine, isoleucine, and proline, this is the total extent of the fragmentation, giving rise to very simple mass spectra such as that shown in Figure 1 for proline. The mechanism by which these fragmentations take place may involve protonation by the reagent gas (CH_5^+, $C_2H_5^+$, etc.) at one or another of the carboxyl oxygens (in addition to the nitrogen) followed by two-electron shifts. Thus all three ions in a CI mass spectrum such as that in Figure 1 can be rationalized as follows. This general scheme is supported by the observation that proline gives identical CI mass spectra with either CH_4 or H_2O as the reagent gas, but that with D_2O as the reagent gas, the QM^+ ion (*m/e* 119) loses 20 (D_2O) or 48 amu ($COOD_2$) to give the other major ions in the spectrum. The fragmentation reactions shown in the above scheme are all supported by metastable ion evidence.

(1) Part III: H. M. Fales, H. Ziffer, G. W. A. Milne, and F. H. Field, *J. Amer. Chem. Soc.*, **92**, 1597 (1970).

(2) National Institutes of Health Special Fellow, 1969; on leave from the City College of the City University of New York.

(3) H. M. Fales, G. W. A. Milne, and M. L. Vestal, *J. Amer. Chem. Soc.*, **91**, 3682 (1969).

(4) H. M. Fales, H. A. Lloyd, and G. W. A. Milne, *ibid.*, **92**, 1590 (1970).

(5) F. H. Field and M. S. B. Munson, *ibid.*, **87**, 3289 (1965). For a review of this field see F. H. Field, *Accounts Chem. Res.*, 1, 42 (1968).

(6) K. Biemann and J. A. McCloskey, *J. Amer. Chem. Soc.*, **84**, 3192 (1962).

(7) G. Junk and H. Svec, *ibid.*, **85**, 839 (1963).

(8) K. Heyns and H. Grützmacher, *Ann.*, **667**, 194 (1963).

(9) C.-O. Andersson, *Acta Chem. Scand.*, **12**, 1353 (1958); C.-O. Andersson, R. Ryhage, S. Stalberg, F. Stenhagen, and E. Stenhagen, *Ark. Kemi*, **19**, 405 (1961).

(10) K. Biemann, J. Seibl, and F. Gapp, *Biochem. Biophys. Res. Commun.*, **1**, 307 (1959).

(11) K. Biemann, J. Seibl, and F. Gapp, *J. Amer. Chem. Soc.*, **83**, 3795 (1961).

(12) A. A. Kiryushkin, H. M. Fales, G. W. A. Milne, T. Axenrod, and E. Gilbert, submitted for publication.

(13) In fact, low-intensity ions at *m/e* $(M + CH_3)^+$, $(M^+ + C_2H_5)^+$, and $(M^+ + C_3H_3)^+$ are also observed but are easily identified by the mass and intensity ratio to the QM^+ ion.

(14) F. H. Field and J. L. Franklin, "Electron Impact Phenomena," Academic Press, New York, N. Y., 1958, p 129.

Table I. Methane CI Mass Spectra of Aliphatic and Hydroxy Aliphatic Amino Acids[a]

Amino acid (mol wt)	Nature of ion: QM^+	$QM^+ - H_2O$	$QM^+ - COOH_2$	$QM^+ - H_2O - COOH_2$	$QM^+ - NH_3$	$QM^+ - 2H_2O$	% Σ_I[b]
Gly (75)	36	9	100				98.7
Ala (89)	50	5	100				94.7
Val (117)	58	5	100				95.6
Leu (131)	43	7	100				98.2
Ile (131)	100	2	70				90.9
Pro (115)	100	4	39				98.7
Ser (105)	27	21	100			5	94.8
Thr (119)	100	25	57	23		9	97.1
Hypro (131)	35	10	100	73		7	96.8
β-Ala (89)	100	52	7		4		91.2

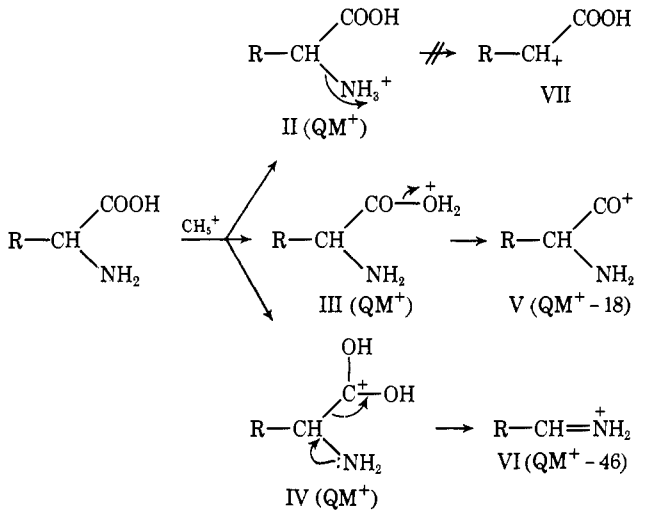
[a] Relative abundances of the ions are normalized to the base peak of each spectrum. [b] Percentage of the total ion current (ignoring methane ions) given by the ions reported plus the contribution from their satellites containing one ^{13}C atom.

Table II. Methane CI Mass Spectra of Substituted and Aromatic Amino Acids[a]

Amino acid (mol wt)	QM^+	$QM^+ - H_2O$	$QM^+ - COOH_2$	$QM^+ - NH_3$[b]	$QM^+ - NH_3 - H_2O$	$QM^+ - H_2O - COOH_2$	Other	% i[c]
Cys (121)	100	6	80	9	19		5[d]	91.6
Met (149)	100	3	20	40			5;[e] 5[f]	89.3
Asp (133)	26	27	100	3	4	10	79[g]	92.8
Asn (132)	60	50	100	35	18	4	26;[h] 27[i]	87.4
Glu (147)	9	100	17	4		33		85.8
Glun (146)	5	7	3	100	2	6	5;[h] 72[j]	92.5
Orn (132)	9	13	2	14	5		100;[j] 7[k]	94.5
Lys (146)	14	3	2	29	5		100[j]	89.9
Phe (165)	100	10	83	2	4		9;[l] 7[m]	97.5
Tyr (181)	28	10	100	55	11		30[l]	86.7
Trp (204)	13	3	48	100			70[l]	82.2
His (155)	26	7	100	4			11[l]	72.7

[a] Relative abundances of the ions are normalized to the base peak of each spectrum (100%). [b] Corrected for abundance of the $^{13}C_1$ satellite of the $QM^+ - H_2O$ ion. [c] Percentage of the total ion current (ignoring methane ions) given by the ions reported, plus the contributions from their $^{13}C_1$ satellites. [d] $QM^+ - H_2S$. [e] $QM^+ - CH_3SH$. [f] $QM^+ - CH_3SH - COOH_2$. [g] $QM^+ - CH_3COOH$. [h] $QM^+ - CONH_3$. [i] $QM^+ - CH_3CONH_2$. [j] $QM^+ - COOH_2 - NH_3$. [k] $QM^+ - 2NH_3$. [l] $QM^+ - C_2H_4NO_2$. [m] $QM^+ - C_7H_7$.

A metastable ion is not observed for the collapse of V to VI however, and in fact, we have never observed the loss



of CO under CI conditions.[15] Except in the case of glutamic acid, loss of $COOH_2$ from QM^+ ion is more facile than loss of water. This might be anticipated from the relative stabilities of ions V and VI and by analogy with the corresponding ions in EI mass spectrometry.[6-11] Loss of NH_3 is not observed in the CI mass spectra of these unsubstituted amino acids due presumably to the lack of stability of the ion VII in which a charge would be located next to the polarized carbonyl group. A similar factor has been invoked to explain

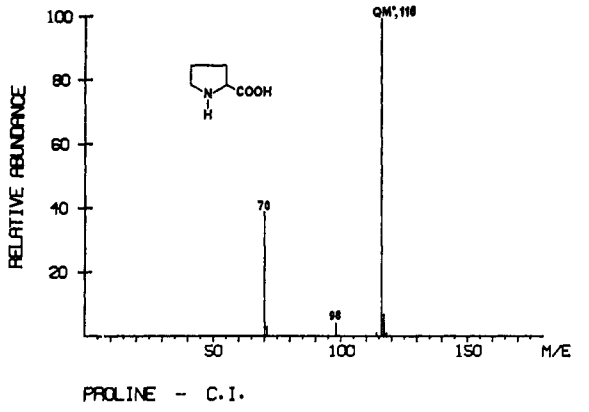


Figure 1.

the low abundance of the "ester fragment"[11] (H_2N^+=CH—COOR) from amino acid esters or the fragment H_2N^+=CH—COOH from amino acids by electron impact, and it is interesting that this fragment (*m/e* 74 in the case of the free acids) is absent or has very low

(15) This fact recalls that loss of CO is not observed in the field ionization process; *cf.* J. N. Damico, R. P. Barron, and J. A. Sphon, *J. Mass. Spect. Ion Phys.*, 2, 161 (1969).

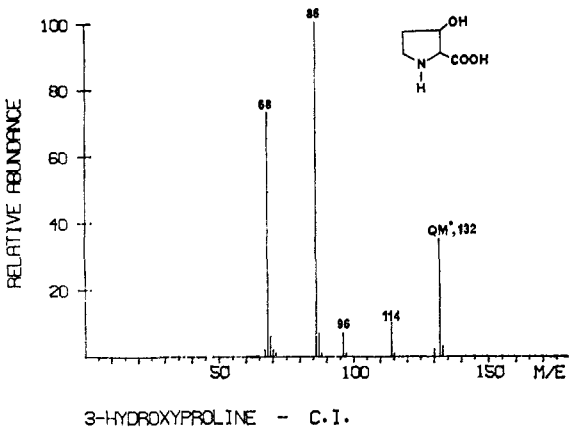


Figure 2.

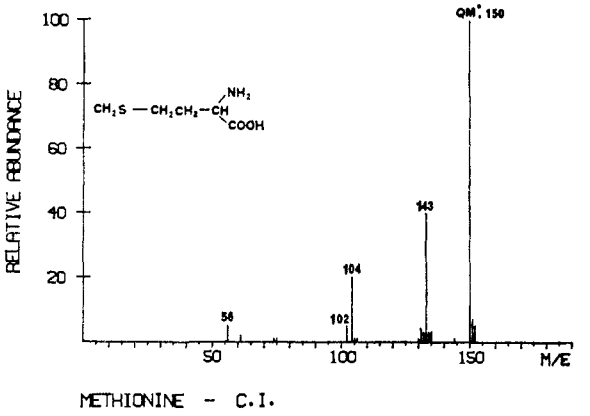
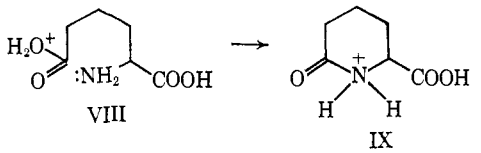


Figure 3.

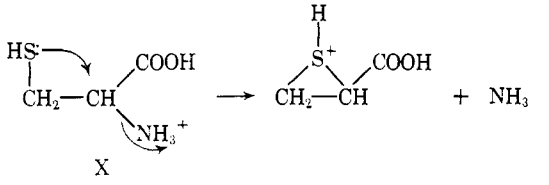
abundance in most of the CI spectra reported here (but see below). On the other hand, the CI mass spectrum of β-alanine, which cannot form an ion of type VI, shows loss from the QM^+ ion of both NH_3 (4%) and H_2O (52%), but unusually little loss of $COOH_2$ (7%).

In the spectra of the hydroxyamino acids serine, threonine, and 3-hydroxyproline (Table I) there is superimposed upon this simple fragmentation scheme, loss of a second molecule of water from both ($QM^+ - H_2O$) and ($QM^+ - COOH_2$). The first of these has no analogy in the EI mass spectra since it represents loss of ($H_2O + HO\cdot$) from the molecular ion. Loss of H_2O from the ($QM^+ - COOH_2$) ions of hydroxyamino acids is, however, directly parallel to the processes seen in the EI mass spectra which lead to ions such as ($M^+ - COOH - H_2O$)[6-8] or ($M^+ - COOR - H_2O$)[9-11] in the case of esters. Loss of $COOH_2$ and H_2O from the QM^+ ion of serine gives an ion of very low abundance but the corresponding ion is very prominent in the spectra of threonine and 3-hydroxyproline (Figure 2). Metastable ions at 45.6 (120 → 74), 42.3 (74 → 56), and 86.7 (120 → 102) in the spectrum of threonine indicate that H_2O may be lost both before and after loss of $COOH_2$ from the QM^+ ion. Aspartic acid and asparagine both lost H_2O from the QM^+ in relatively large amounts (27 and 50%, respectively) perhaps due to some assistance from neighboring functions, and in the case of glutamic acid (VIII), where loss of H_2O from the δ-carboxyl group of the QM^+ ion may be especially effectively assisted by the α-amino nitrogen, the ($QM^+ - H_2O$) ion gives the base peak of the spectrum. This may be due in part to prior thermal dehydration of glutamic acid to pyroglutamic acid whose CI mass spectrum shows a QM^+

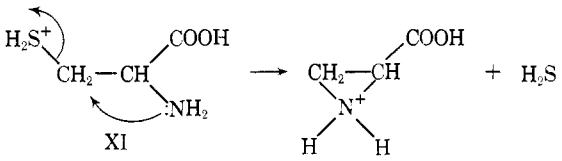


ion at *m/e* 130 (100%) and $QM^+ - COOH_2$ at *m/e* 84 (60%). In a closely related process, the QM^+ ion from glutamine loses ammonia, presumably from the amido group to form the same species (**IX**). Both of these processes are very similar to the loss of ammonia and ethanol from the ($M^+ - COOR$) ions in the EI mass spectra of amino acids (R = H)[6-8] and their esters ($R = C_2H_5$).[9-11]

(b) Loss of Ammonia and RSH. Loss of H_2O from the QM^+ ion of cysteine gives the ion at *m/e* 104 (6%). Loss of 17 amu from the QM^+ ion, however, gives the ion at *m/e* 105 (9%) and is ascribed to the expulsion from the QM^+ ion of NH_3. The absence of such a reaction in the CI mass spectra of the simple unsubstituted amino acids prompts the suggestion that in the cysteine case, participation of the sulfhydryl group, as in **X**, is involved. Similar participation of the carboxyl group may be responsible for the loss of ammonia



(4%) from the QM^+ ion of β-alanine. Expulsion of H_2S from the QM^+ ion of cysteine on the other hand, gives an ion at *m/e* 88 (5%) and might be a result of nitrogen participation after protonation of the sulfhydryl group as in **XI**. The CI mass spectrum of methionine, shown in Figure 3, has abundant ions corresponding to the loss from the QM^+ ion (*m/e* 150, 100%) of H_2O, $COOH_2$, and NH_3. Methyl mercaptan loss (5%) is quite analogous to the loss of H_2S from the QM^+ ion of cysteine (5%). An ion at *m/e* 56 (5%) in the methionine spectrum results from the loss of both CH_3SH and $COOH_2$ from the QM^+ ion, a process also seen in the EI mass spectrum.[6,7] In contrast to this, the important ions in the EI mass spectrum at *m/e* 61 ($CH_3SCH_2^+$) and 75 ($CH_3SCH_2CH_2^+$) are effectively absent from the CI mass spectrum.



A major point of difference between the spectrum of cysteine and that of methionine is found in the abundance of the ions resulting from the loss of NH_3 from the QM^+ ion. A somewhat minor process in the case of the former leads to the major fragment ion in the spectrum of methionine. Sulfur participation, such as

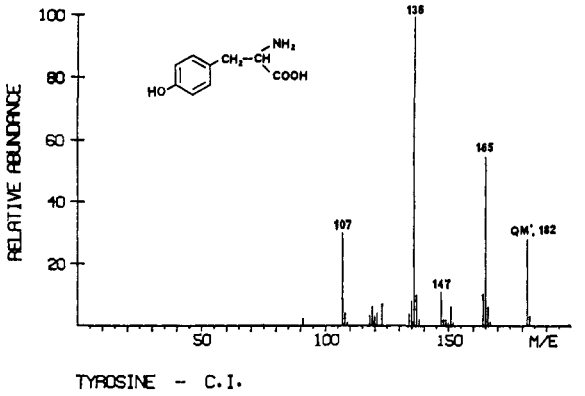


Figure 4.

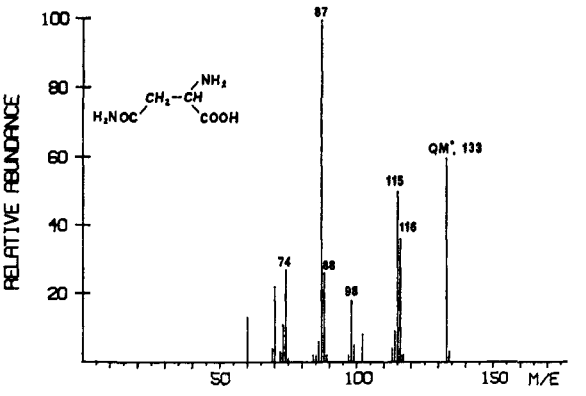
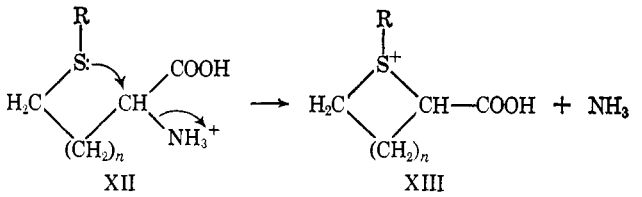


Figure 5.

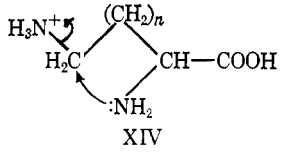
in XII, must be invoked to explain this difference in behavior and while it is tempting to assume that the value of n, and the size of the ring in XIII, will be a controlling factor, it transpires that such is not the



XII XIII

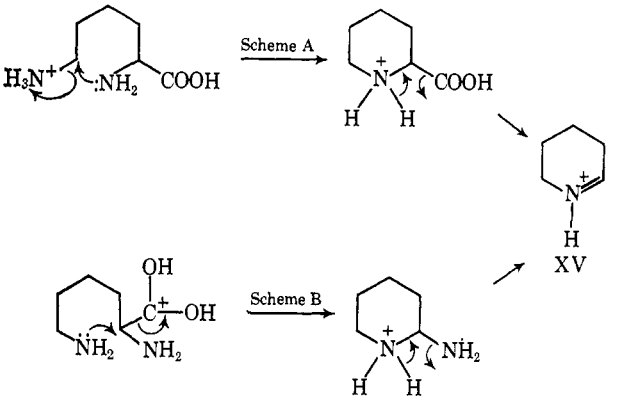
case; the QM^+ ions of methionine (R = CH_3, n = 1) and S-methylcysteine (R = CH_3, n = 0) both exhibit this facile loss of NH_3 but the QM^+ ions of cysteine (R = H, n = 0) and homocysteine (R = H, n = 1) both fail to do so. The reason clearly lies in the superior nucleophilicity of $-SCH_3$ groups, compared to un-ionized –SH groups. This in turn is considered to be due to electron release by the methyl group. This fact permits the differentiation between methionine and cysteine in proteins, because the former reacts readily with iodoacetamide at pH 2, conditions which result in no reaction at cysteine sites.[16]

The CI mass spectra of ornithine and lysine both have only minor ions resulting from the loss of $COOH_2$ or H_2O. Loss of NH_3 however, is relatively important in both spectra, being twice as facile in the lysine case (29%) as the ornithine spectrum (14%). The CI mass spectrum of lysine labeled with ^{15}N in the ϵ-amino group[17] shows loss of only $^{15}NH_3$ from the QM^+ ion and it must be concluded that participation as in XIV (n = 3) is occurring. This conclusion was confirmed by the absence in the CI mass spectrum of α-$^{15}NH_2$-lysine[17] of an ion corresponding to $QM^+ - {}^{15}NH_3$.



XIV

Interestingly, these same compounds show, by virtue of the intensities of the ions at m/e 84 (100% in α-$^{15}NH_2$-lysine, 45% in ϵ-$^{15}NH_2$-lysine), and m/e 85 (46% in α-$^{15}NH_2$-lysine, 100% in ϵ-$^{15}NH_2$-lysine), that the ion at m/e 84 (XV) in unlabeled lysine arises from two processes: loss from the QM^+ ion of the ϵ-amino group as discussed above, followed by loss of $COOH_2$ (Scheme A), and in addition, loss from the QM^+ ion of $COOH_2$



and subsequent loss as ammonia of the α-amino group (Scheme B). The driving force in both cases is presumably the stability of the pyrrolinium ion (XV) as in EI mass spectrometry.[11] The fact that a five-membered ring would be involved in the corresponding intermediates (*e.g.*, XIV, n = 2) explains the greater difficulty of the elimination of ammonia in the ornithine case. In an analogous study of lysine and lysopine ethyl esters[11] the conclusion was reached that ammonia was being lost from both amino groups during the formation of XV. In the CI mass spectra, although the method of formation of ions is different, the same results pertain. In addition, it is clear that the first loss of NH_3 from the QM^+ ion (a process not observed in the EI mass spectra) is selective with respect to the amino groups of lysine.

The participation reactions that are observed in the CI mass spectra of the aromatic amino acids tyrosine, tryptophan, phenylalanine, and histidine are of particular interest. The spectrum of tyrosine (Figure 4) shows loss from the QM^+ ion of H_2O (10%), presumably from the carboxyl group, loss of $COOH_2$ giving the base peak, and ammonia (55%). The facile loss of ammonia must be due to Ar_1^--3 participation of the p-hydroxyphenyl ring in the manner first postulated by Baird and Winstein.[18] Such participation (XVI)

(16) R. Cecil and J. R. McPhee, *Advan. Protein Chem.*, **14**, 225 (1959).

(17) J. A. Grove, T. J. Gilbertson, R. H. Hammerstedt, and L. M. Henderson, *Biochim. Biophys. Acta*, **184**, 329 (1969). We should like to thank Professor Henderson for a generous gift of samples of α-$^{15}NH_2$-lysine and ϵ-$^{15}NH_2$-lysine.

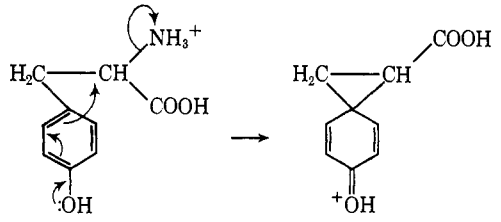
(18) R. Baird and S. Winstein, *J. Amer. Chem. Soc.*, **85**, 567 (1963); *cf.* H.-F. Grützmacher, *Org. Mass. Spectrom.*, **3**, 131 (1970).

Table III. Relative Abundances of Selected Ions in the CI and EI Mass Spectra of Aromatic Amino Acids and Esters

	Esters EI[b]			Acids EI[c]			Acids CI			
	Phe	Tyr	Trp	Phe	Tyr	Trp	Phe	Tyr	Trp	His
Amine type ion	100	51	8.5	74	3.5	3.4	83	100	48	100
Ester type ion	83	52	4	100	6.3	2	5	0	0	2
$(ArCH_2)^+$ [a]	23	100	100	55	100	100	9	30	70	11

[a] $(ArCH_3)^+$ in the EI mass spectra of the free acids Trp and Tyr. [b] Reference 10. [c] Reference 7.

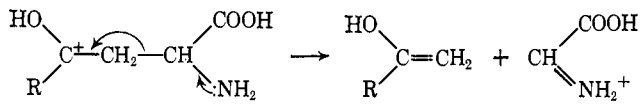
is present in all the "aromatic" amino acids, and the order of its effectiveness, *i.e.*, the abundance of the ion



XIV

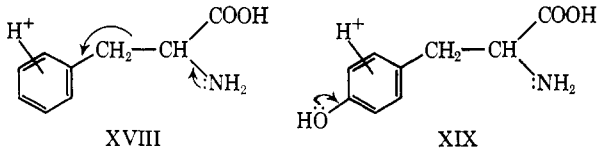
formed in each case by the loss of ammonia is, predictably, tryptophan, tyrosine, histidine, and phenylalanine.

(c) Fission of the α,β Bond. As mentioned earlier, the ion at *m/e* 74 (H_2N^+=CH—COOH) which is analogous to the "ester fragment" in the EI mass spectra of the esters of amino acids, is usually absent in the CI mass spectra even when branching is present such as in valine. It is, however, an important ion (*m/e* 74) in the CI mass spectrum of aspartic acid (XVII, R = OH) and asparagine (XVII, R = NH_2, Figure 5) with abundances of 79 and 22%, respectively, and we propose that it is formed by a retro-Aldol type of reaction triggered by protonation of the β-carbonyl group. In this connection, it is of interest to note that



XVII

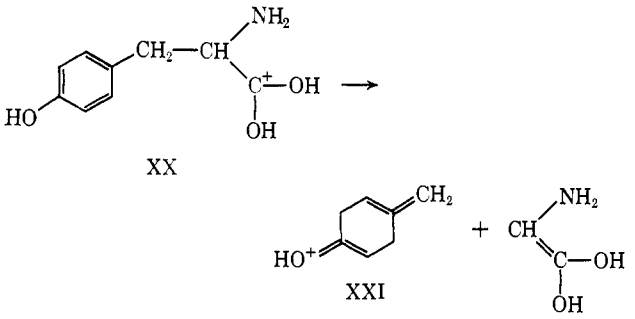
this same ion of *m/e* 74 is present in the CI spectrum (*vide infra*) of phenylalanine where it may be considered to arise by protonation of the aromatic ring and subsequent cleavage of the α,β bond as in XVIII. In tyrosine and tryptophan by contrast, the electron-releasing nature of the aromatic substituent successfully competes with this reaction (*e.g.*, XX) and the ion of *m/e* 74 is not formed. On the other hand, protonation



XVIII XIX

of the carboxyl or amino functions of these aromatic amino acids leads to α,β fission (XX → XXI) with charge retention on the benzylic fragment whose abundance is proportional to the degree of resonance stabilization. This effect was first described by Biemann, *et al.*,[11] and later supplemented by Junk and Svec[7] who reported the data in Table III to which we now add the CI abundances.

The CI mass spectrum of histidine is of further interest in that the ion at *m/e* (M + 15) arising from



XX XXI

methylation of the molecule by the ionized reaction gas has a high relative abundance (8 *vs.* 26% for the QM^+ ion). In the histidine case, this ion has an abundance which is temperature and pressure dependent but in all other amino acids, the ion at *m/e* (M + 15) has a negligible intensity and this seems therefore another example of the nucleophilic tendencies of the imidazole ring of histidine which in the EI mass spectra of peptide methyl esters is responsible for intermolecular methylation prior to ionization.[19]

Conclusions

While similarities do exist between the EI and CI mass spectra of the amino acids and their derivatives, there are also some important differences. Many of the fragmentation processes, particularly those of diagnostic value, in the EI mass spectra arise from decomposition of the "amine fragment" (R—CH=NH_2^+) of even-electron character. In the CI mass spectra, the QM^+ ion is itself an even-electron ion and most of the important processes are rationalized as arising from the prior formation of this species. The overall result is to focus attention on ions such as QM^+, $QM^+ - H_2O$, $QM^+ - NH_3$, and $QM^+ - COOH_2$ instead of the "amine fragment" and the ions derived from it. In the case of an unknown compound, the CI mass spectrum would be highly diagnostic of an amino acid while the EI mass spectrum might be of additional aid in the determination of the structure of the amino acid side chain.

Since fragmentations in methane CI mass spectrometry all result from protonation of the molecule rather than electron abstraction as in EI mass spectrometry, it is tempting to draw analogies between decompositions of the QM^+ ion and mechanisms encountered in proton-catalyzed reactions in solution chemistry. This should be valid as long as it is recognized that the time scale is much shorter (*ca.* 10^{-6} sec), the energies higher ($\Delta H_{CH_5^+}$ = 229 kcal/mol) and solvation is absent. Thus important solution reactions such as acid-catalyzed decarboxylation and dehydration of

(19) G. W. A. Milne, A. A. Kiryushikin, Yu. A. Alakhov, V. M. Lipkin, and Y. A. Ovchinnikov, *Tetrahedron*, **26**, 299 (1970); M. Senn, R. Venkataraghavan, and F. W. McLafferty, *J. Amer. Chem. Soc.*, **88**, 5593 (1966).

amino acids may be expected to have analogies in CI mass spectrometry (*viz.*, $QM^+ - COOH_2$ and $QM^+ - H_2O$) but important CI processes (*e.g.*, α,β fission, etc.) may be unknown or obscure in solution chemistry.

Rearrangement processes so prevalent in the EI mass spectra of amino acids[6-8] and their derivatives[9-11] are generally absent from the CI mass spectra probably because of the even-electron character of the ions as well as the possibility of their dissipating excess energy through collision processes. While rearrangement ions in EI mass spectra may provide considerable structural information when the mechanism of their formation is well understood, they may cause considerable confusion when their origin is unclear. It is therefore both a strength and a weakness of CI mass spectra that few rearrangements are encountered.

It seems highly artificial to consider the CI technique as an alternative rather than as a complement to EI mass spectrometry since merely stopping the reagent gas flow into the source permits a rapid return to EI conditions. It is clear that both spectra have advantages and disadvantages and in application to a genuine problem the combination of both methods will be most advantageous.

Experimental Section

The amino acids used in this work were all Mann assayed amino acids.[20] Mass spectra were measured at low resolving power on an MS-902 using the dual EI/CI source described previously.[3] All spectra were measured at a source temperature of 250 ± 5°.

(20) Mann Research Laboratories, Inc., New York, N. Y. 10006.

Photolysis of Peresters. Reactions of Alkoxy–Alkyl Radical Pairs in Solution

Roger A. Sheldon and Jay K. Kochi

Contribution No. 1851 from the Department of Chemistry, Indiana University, Bloomington, Indiana 47401. *Received November 8, 1969*

Abstract: A series of peresters $RCO_2OC(CH_3)_2R'$ cleanly liberate 1 mol of carbon dioxide with a quantum yield of unity when irradiated in solution at 2537 Å. An alkyl radical R· and an alkoxy radical $\cdot OC(CH_3)_2R'$ are the first discernible intermediates. The electron spin resonance spectrum of the alkyl radical R· is obtainable when photolyses are carried out directly in the cavity of the spectrometer. The esr spectrum of R'· from the secondary fragmentation of the alkoxy radical is readily observed with isopropyl, *t*-butyl, and benzyl simultaneously with its partner R· at −115°, but is not seen when R' is methyl or ethyl. Reactions of the alkyl–alkoxy radical pair are considered to proceed from a solvent cage. Combination to form ether $ROC(CH_3)_2R'$ and disproportionation to alcohol $R'(CH_3)_2COH$ and alkene R(–H) are exclusively cage processes. The relative rates of disproportionation and combination k_d/k_c for alkyl–alkoxy and alkyl–alkyl radical pairs are compared and discussed in terms of the transition state for disproportionation. The yield of ether from *t*-butyl peracetate can be quantitatively related to the efficiency of the cage process in a series of solvents. Cage reactions in the thermal and photochemical decomposition of peresters are qualitatively compared. Examination of the rearrangement of cyclopropylmethyl radical leads to an estimate of 10^9 sec^{-1} for the combination of an alkyl and an alkoxy radical pair.

Peresters are useful as free radical precursors because the thermal decomposition[1] represented in eq 1 is

$$R{-}\overset{\overset{O}{\|}}{C}O{-}OC(CH_3)_2R' \longrightarrow R\cdot + CO_2 + \cdot OC(CH_3)_2R' \quad (1)$$

less complicated by competing heterolytic processes such as carboxy inversion[2] than it is with diacyl peroxides. The photochemical decomposition of *t*-butyl percaprate and perlaurate as pure liquids produced carbon dioxide with a quantum yield of unity (at 2537 Å) and other products derived from free radical intermediates.[3] The high quantum yield for decomposition (Φ = 1.5–1.8) and the formation of isobutylene oxide and carboxylic acid, however, indicated that induced decomposition was also occurring under these conditions. The photosensitized decomposition of *t*-butyl peracetate has also been reported.[4]

Direct photolysis of aliphatic peresters in solution was recently shown to be a useful method of generating alkyl radicals for electron spin resonance (esr) studies.[5,6] In conjunction with these esr results we now wish to report a product study from the photolysis of a series of aliphatic peresters with the following objectives: (a) correlation of the esr results with the products of photolysis; (b) determination of the efficiency of the photoinduced homolytic decomposition; (c) assessment of the competition from heterolytic processes and the comparison with thermolytic decomposition; (d) delineation of possible reactions of alkyl–alkoxy

(1) See for example: (a) P. D. Bartlett and R. R. Hiatt, *J. Amer. Chem. Soc.*, **80**, 1398 (1958), and subsequent papers in this series; (b) N. A. Milas and A. Golubovic, *ibid.*, **80**, 5994 (1958); (c) M. Trachtman and J. G. Miller, *ibid.*, **84**, 4828 (1962); (d) Y. K. Syrkin and I. I. Moiseev, *Russ. Chem. Rev., Engl. Trans.*, **29**, 163 (1960).

(2) (a) P. D. Bartlett and T. G. Traylor, *J. Amer. Chem. Soc.*, **83**, 856 (1961); (b) R. Criegee and R. Kaspar, *Ann.*, **560**, 127 (1948); (c) E. Hedaya and S. Winstein, *J. Amer. Chem. Soc.*, **89**, 1661 (1967).

(3) W. Simpson and J. Miller, *ibid.*, **90**, 4093 (1968); also *cf.* L. A. Singer and N. P. Kong, *ibid.*, **88**, 5213 (1966), and E. N. Cain, R. Vukov, and S. Masamune, *Chem. Commun.*, 98 (1969).

(4) C. Walling and M. Gibian, *J. Amer. Chem. Soc.*, **87**, 3413 (1965).

(5) J. Kochi and P. Krusic, *ibid.*, **91**, 3940 (1969).

(6) P. Bakuzis, J. Kochi, and P. Krusic, *ibid.*, **92**, 1434 (1970).