Carbon-14 as a Label in Mass Spectrometry

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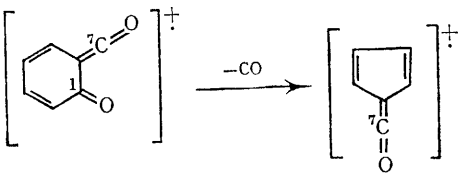
ISOTOPE LABELLING has been a useful means of establishing the mechanisms of ionic reactions in the mass spectrometer. Although deuterium labelling is usually used there are instances where it is necessary, or preferable, to label the skeletal atoms of a molecule, *e.g.* in aromatic compounds where hydrogen scrambling can precede fission.[1] In these instances labelling with the heavy isotopes of carbon, nitrogen, and oxygen may be used.

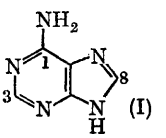
In a recent paper[2] Knöppel and Beyrich have demonstrated the use of ^{14}C-labelling for producing autoradiographic mass spectrograms in order to follow the course of ionic reactions, and have shown that this approach has advantages over the usual labelling with ^{13}C. In addition, we have noted that the level of ^{14}C-incorporation normally used for radiochemical studies (about 5%) is suitable for studying, by isotopic abundance determination, ionic species which occur at relatively isolated regions of a mass spectrum. In these cases interference to abundance determination is from naturally occurring isotopes only, and this interference is about ten times less for ^{14}C- than for ^{13}C-labelled ions.

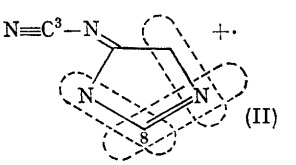
Some experimental results for ^{14}C-labelled salicylic acid and adenine are presented in the Table.

fragment from salicylic acid shows the CO elimination to be exclusively from C-1, assuming no oxygen migration. The scheme which results in a highly conjugated fragment is a possible, though not unique, rationalization of the labelling data.



The complete retention of C-8 in the $[M - HCN]$ fragment from adenine (I) shows that the initial loss of HCN from the molecular ion is more specific than was evident[3] from a study of the spectra of $[^2H_3]$- and $[^2H_4]$-adenine. In the context of the carbon-labelling results, quantitative deuterium-labelling studies made in these





TABLE

Compound	Ion	^{14}C-Content
[7-^{14}C]Salicylic acid	$M^{+\cdot}$	3·9 ± 0·2%†
	$[M - H_2O]^{+\cdot}$	3·9 ± 0·2
(2·31 mc/mM = 3·70% ^{14}C)† ..	$[M - H_2O - CO]^{+\cdot}$ ..	4·1 ± 0·5
[8-^{14}C]Adenine	$M^{+\cdot}$	6·4 ± 0·1
(3·7 mc/mM = 6·0% ^{14}C) ..	$[M - HCN]^{+\cdot}$	6·4 ± 0·1
	$[M - 2HCN]^{+\cdot}$	2·1 ± 0·2
	$[M - HCN - H_2CN]^{+}$ ..	2·5 — 0·2

† Standard deviation, from six or more scans over region of interest. Peak heights corrected for presence of naturally occurring isotopes.
‡ From scintillation counting.

The ^{14}C-compositions of those ions which should fully retain the label are in good agreement with scintillation-counting results obtained for the compounds themselves. The data in the Table provides new information on the modes of fragmentation of salicylic acid and adenine. Complete retention of C-7 in the $[M - H_2O - CO]^{+\cdot}$ laboratories indicate a 55% loss of HCN from C-1 and its attached amine group and 45% HCN loss from C-3 and its attached nitrogen atoms. In contrast only about 35% of the carbon label is retained in those ions resulting from the elimination of two molecules of HCN, and of a molecule of HCN and an H_2CN radical† from the molecular

† The defocussing method for locating metastables[4] showed the transition $[M - HCN]^{+\cdot} \rightarrow [M - HCN - H_2CN]^{+}$.

ion. The postulation of an intermediate such as (II) which may eliminate HCN or $H_2CN\cdot$, with hydrogen migration in some cases, in the manner shown, is in agreement with the ^{14}C retention found for the fragment ions.

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[2] H. Knöppel and W. Beyrich, *Tetrahedron Letters*, 1968, 291.
[3] J. M. Rice and G. O. Dudek, *J. Amer. Chem. Soc.*, 1967, **84**, 2719.
[4] M. Barber and R. M. Elliot, ASTM E14 Conference on Mass Spectrometry, Montreal, June, 1964.