Dear Sir

The Use of On-line H/D Exchange for the Investigation of Alcohols and Carbonyl Compounds by Reaction Gas Chromatography/Mass Spectrometry

Deuterium labelling is well known and is one of the most powerful tools for the interpretation of mass spectral fragmentation. At the same time, deuteriated analogues appear to assist in solving the structural problems. This is particularly true when simple procedures are used for the synthesis of labelled products or deuteriation is accomplished directly in the inlet system of a mass spectrometer. For instance, the determination of the number of active hydrogens such as OH, COOH, NH or SH by simple exchange of the sample in the presence of heavy water is widely used in mass spectrometric practice. The determination of the number of enolizable hydrogens in carbonyl compounds can also give a characteristic, structurally informative parameter regarding the substitution pattern at carbon atoms neighbouring to the carbonyl group. In this respect, the use of gas phase exchange of enolizable hydrogens for deuterium seems to be the most promising since the corresponding reactor may be coupled to the mass spectrometer, which avoids the necessity to isolate the labelled products. For this purpose, the reaction column devised by Burlingame and coworkers[1] for preparative gas phase synthesis of carbonyl compounds may be useful.

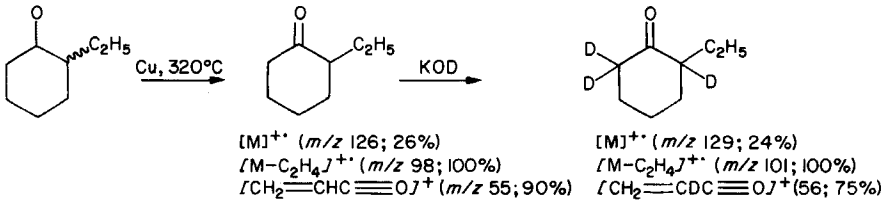
The necessity for such a column arose from our intention to improve the method of reaction gas chromatography/mass spectrometry (GC/MS) for the investigation of alcohols within mixtures, which involves the use of the gas phase dehydrogenation microreactor (Cu, 320 °C) located between the GC column and the mass spectrometer.[2] The mass spectra of carbonyl compounds registered after passage of alcohols through the catalyst allow the detection of the ions due to McLafferty rearrangement and, hence, branching at carbon atoms α to the functional group. The mass numbers of such ions, however, do not always indicate the character of the branching, e.g. *gem*-dimethyl v. ethyl groups. This uncertainty might be resolved by the determination of the number of enolizable hydrogen atoms.

A stainless steel 1.5 m × 3 mm column packed with 5% SE-30 and 10% KOH on Chromaton W was pretreated with deuterium oxide by injection of 10 μl portions several times during 4 h. The column thus prepared achieved quantitative exchange of enolizable hydrogens for deuterium in 0.05–0.1 μl (50–100 μg) of individual aldehydes or ketones at 40–300 °C. This reaction column was included in the systems for the investigation of alcohols within mixtures (GC column → dehydrogenation microreactor → reaction column), of individual alcohols (dehydrogenation microreactor → reaction column), of carbonyl compounds within mixtures (GC column → reaction column) and of individual carbonyl compounds (reaction column). All the systems may be coupled to the mass spectrometer (LKB-2091).

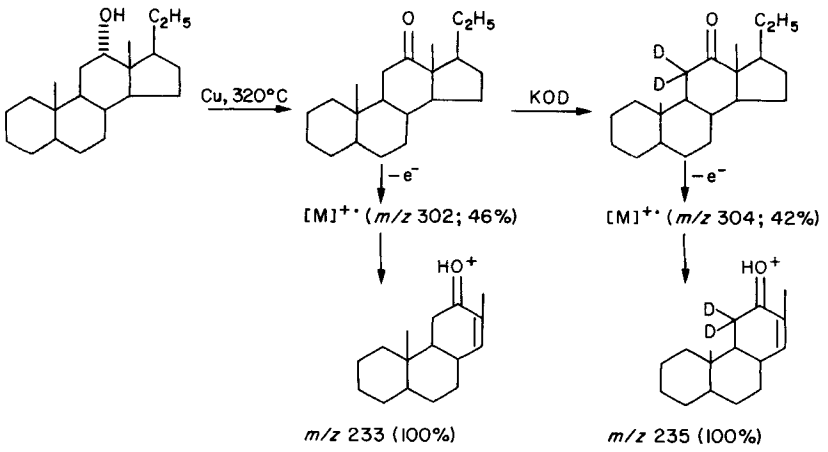
Table 1. Principal ions in the mass spectra of isomeric octanols, the corresponding carbonyl compounds and their labelled analogues (*m/z*; relative intensity in %)

Initial alcohol	Carbonyl compound	Labelled carbonyl compound
1-Octanol $[M]^{+\cdot}$ (130; 0), $[M-H_2O]^{+\cdot}$ (112; 4)	$CH_3(CH_2)_6CHO$ $[M]^{+\cdot}$ (128; 1), $[CH_2{=}CHOH]^{+\cdot}$ (44; 77)	$CH_3(CH_2)_5CD_2CHO$ $[M]^{+\cdot}$ (130; 1), $[CD_2{=}CHOH]^{+\cdot}$ (46; 79)
2-Ethyl-1-hexanol $[M]^{+\cdot}$ (130; 0), $[M-H_2O]^{+\cdot}$ (112; 4)	$CH_3(CH_2)_3CHEtCHO$ $[M]^{+\cdot}$ (128; 1), $[CHEtCHOH]^{+\cdot}$ (72; 95)	$CH_3(CH_2)_3CDEtCHO$ $[M]^{+\cdot}$ (129; 1), $[CDEtCHOH]^{+\cdot}$ (73; 98)
2-Octanol $[M]^{+\cdot}$ (130; 0), $[M-CH_3]^{+}$ (115; 2), $[M-H_2O]^{+\cdot}$ (112; 4), $[CH_3CHOH]^{+}$ (45; 100)	$CH_3CO(CH_2)_5CH_3$ $[M]^{+\cdot}$ (128; 4), $[CH_3CO]^{+}$ (43; 100), $[CH_2C(OH)CH_3]^{+\cdot}$ (58; 68)	$CD_3COCD_2(CH_2)_4CH_3$ $[M]^{+\cdot}$ (133; 7), $[CD_3CO]^{+}$ (46; 100), $[CD_2C(OH)CD_3]^{+\cdot}$ (63; 75)
4-Octanol $[M]^{+\cdot}$ (130; 0), $[M-H_2O]^{+\cdot}$ (112; 4), $[CH_3(CH_2)_2CHOH]^{+}$ (73; 75), $[CH_3(CH_2)_3CHOH]^{+}$ (87; 52)	$CH_3(CH_2)_2CO(CH_2)_3CH_3$ $[M]^{+\cdot}$ (128; 26), $[CH_3(CH_2)_2CO]^{+}$ (71; 75), $[CH_3(CH_2)_3CO]^{+}$ (85; 55), $[CH_3(CH_2)_2C(OH)CH_2]^{+\cdot}$ (86; 18), $[CH_2(OH_2)CH_2]^{+\cdot}$ (58; 48)	$CH_3CH_2CD_2COCD_2(CH_2)_2CH_3$ $[M]^{+\cdot}$ (132; 22), $[CH_3CH_2CD_2CO]^{+}$ (73; 78), $[CH_3(CH_2)_2CD_2CO]^{+}$ (87; 60), $[CH_3CH_2CD_2C(OH)CD_2]^{+\cdot}$ (90; 15), $[CD_2(OH_2)CD_2]^{+\cdot}$ (62; 55)

The described method may be also useful for structure elucidation of cyclic alcohols. This is exemplified, for instance, by the analysis of stereoisomeric 2-ethylcyclohexanols whose electron impact (EI) mass spectra are insufficiently informative. The presence of the $[M]^{+\cdot}$, $[M-H_2O]^{+\cdot}$ and $[CH_2{=}CHCH{=}OH]^{+}$ ions indicates only that the compounds under study are cyclic alcohols which possess no substituents, at least on one of the α-carbon atoms. The mass spectrum of the corresponding ketone registered after passage of the compounds through the dehydrogenation microreactor shows the base peak due to the $[M-C_2H_4]^{+\cdot}$ ion. Since the latter can arise from the McLafferty rearrangement the suggestion is that the ethyl group is situated at the α-carbon atom. This is corroborated by the mass spectrum of the labelled ketone which indicates that only three hydrogen atoms can be replaced by deuterium (Scheme 1).



Scheme 1



Scheme 2

The other example demonstrates that the suggested technique may also be applied in the case of steroid alcohols. No valuable information regarding the position of the hydroxyl group can be extracted from the EI mass spectrum of 12α-hydroxypregnane. However, the mass spectrum of the corresponding ketone shows the base peak at m/z 233 which suggests that the keto group is located at position 12.[3] The shift of the $[M]^{+\cdot}$ and m/z 233 ions by two mass units is in accordance with this suggestion (Scheme 2).

In addition, the described method may be also useful when fragmentation mechanisms of carbonyl compounds are to be investigated. In this case, not only carbonyl compounds but also alcohols may be involved.

Yours

A. I. MIKAYA and V. G. ZAIKIN
Topchiev Institute of Petrochemical Synthesis,
USSR Academy of Sciences,
Moscow,
USSR

A. V. ANTONOVA and N. S. PROSTAKOV
Patris Lumumba Friendship University,
Moscow,
USSR

April 1984

References

1. M. Senn, W. J. Richter and A. L. Burlingame, *J. Am. Chem. Soc.* **87**, 680 (1965).
2. A. I. Mikaya, V. I. Smetanin, V. G. Zaikin, A. V. Antonova and N. S. Prostakov, *Org. Mass Spectrom.* **18**, 99 (1983).
3. H. Budzikiewicz, C. Djerassi and D. H. Williams, *Structure Elucidation of Natural Products by Mass Spectrometry*, Vol. II: *Steroids, Terpenoids, Sugars and Miscellaneous Classes*, p. 67, Holden-Day, San Francisco (1964).

Dear Sir

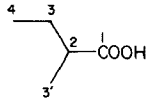
Metastable Fragmentation of 2-Methylbutanoic Acid

Metastable fragmentation of 2-methylbutanoic acid radical cation **1** leads to the loss of a methyl radical only.[1]

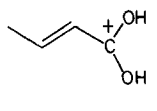
Table 1. Methyl elimination in the MIKE spectra of labelled compounds (VG.ZAB.2F)

	CH_3	CH_2D	CHD_2	CD_3
1	100	—	—	—
1a (*O*-d_1)	99	<1	—	—
1b (2-d_1)	99	<1	—	—
1c (3,3-d_2)	99	<1	—	—
1d (3′,3′,3′-d_3)	52	—	<1	47
1e (3,3,3′,3′,3′-d_5)	64	1	1	34

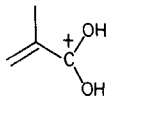
Table 1 shows that H(0), H(2) and H(3) hydrogens remain in the fragment ion and that 2-CD_3-butanoic acid **1d** eliminates CH_3 and CD_3. These data are explained by the mechanism shown in Scheme 1: **1** by β-hydrogen migration leads to **1α**. This reaction is exothermic $\Delta H_f(\mathbf{1}) = 493$ kJ mol^{-1},[2] $\Delta H_f(\mathbf{1\alpha}) = 431$ kJ mol^{-1} [3,4] and irreversible. **1α** isomerizes into **1α′** through the cyclopropanic form **1β** or by 1,2-migration of a CH_2O_2 group.[5] **1α** and **1α′** lead respectively to the enolic ions **1γ** and **1γ′**. Simple cleavages of **1α** and **1α′** lead to ions *a*, while dissociations of **1γ** and **1γ′** lead to ions *b*.



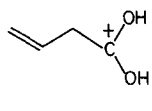
1



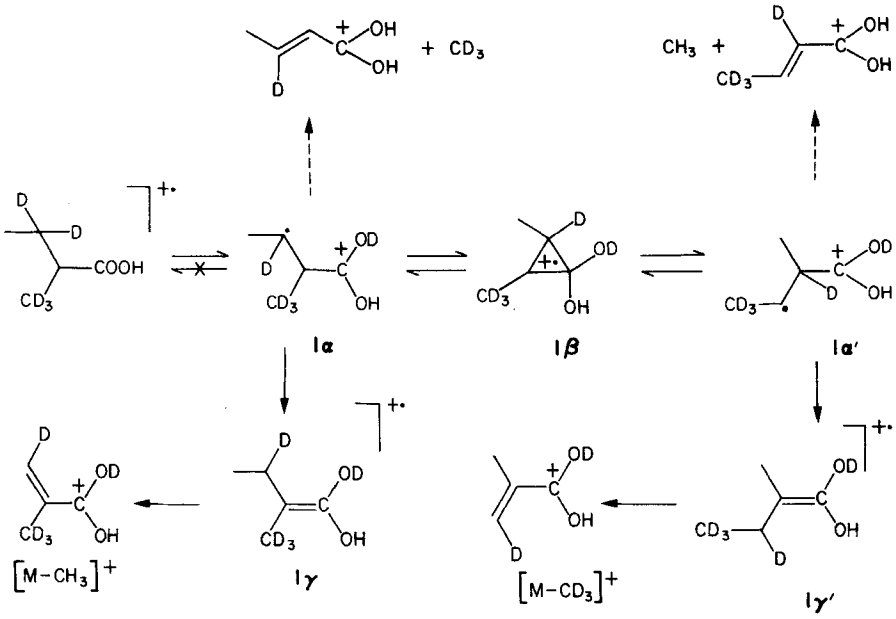
a



b



c



Scheme 1

Several experimental results corroborate this mechanism: (i) $T_{0.5}$ values (23 ± 2 meV) are similar for $[M-CH_3]^+$ and $[M-CD_3]^+$ peaks on the mass analysed ion kinetic energy (MIKE) spectrum of **1d**: these data are in agreement with the occurrence of the symmetrical intermediate **1β** or with a rapid and reversible isomerization, **1α** ⇆ **1α′**. (ii) The MIKE spectrum of the $[M-CH_3]^+$ ion has been compared to those of ions *a*, *b* and *c* generated by protonation of the corresponding acids (chemical ionization). The structure of $[M-CH_3]^+$ is a mixture of *a* and *b*, the latter being more abundant. Furthermore, the difference between $\Delta H_f(a)$ and $\Delta H_f(b)$ is very small (6 kJ mol^{-1} [6]). (iii) $[M-CH_3]^+$ ions eliminate a molecule of water. Table 2 indicates that DHO is mostly eliminated for such ions generated from **1a**, **1c** and **1e**. This result suggests that fragmentation begins with an irreversible β-hydrogen migration.

The observed isotopic effect for **1e** corroborates the proposed 1,2-hydride shift, but can also be explained by a direct formation of **1γ** by 1,3-hydrogen migration. If this latter hypothesis appears less probable (Table 2, compound **1b**), this isotopic effect proves, in any case, that ion *b* comes from the enolic intermediate ions.

This work confirms several mechanisms proposed to explain the fragmentation of butanoic acid and ester radical cations,[7–9] for example, the results of Hemberger *et al.* concerning ionized methyl isobutyrate.[10]

Table 2. Water elimination in the MIKE spectra of $[M-15]^+$ ions generated from labelled compounds

	H_2O	HDO	D_2O
1a	21	78	—
1b	90	10	—
1c	29	71	—
1d	88	12	—
1e	21	75	4