Pure & Appl. Chem., Vol. 50, pp. 185-196. Pergamon Press, 1978. Printed in Great Britain.

MASS SPECTROMETRY - A SENSITIVE PROBE OF MOLECULAR GEOMETRY

Mark M. Green

Department of Chemistry, Clarkson College of Technology, Potsdam, New York, 13676, USA

Abstract - The unimolecular rearrangement chemistry of molecular ions encountered in mass spectrometers is ideally suited for stereoanalytical purposes. This follows from the spatial dependency of the bond making stage of the rearrangement. Examples are noted which demonstrate how such rearrangements may be utilized to assign configuration with and without deuterium labelling even when only one stereoisomer is available. These rearrangements may occur from molecular ions generated by electron impact or field ionization. Chemical ionization and ion cyclotron resonance mass spectrometry are known to be sensitive to stereoisomerism but have yet to be developed as stereoanalytical devices. Simple cleavage reactions of molecular ions may reveal stereoisomeric information when two diastereomers give rise to identical product ions. In the latter case appearance potential differences and product intensities differences mirror the enthalpic differences of the precursor neutral diastereomers. These thermochemical effects could be utilized to assign configuration to stereoisomers.

In 1865 a sample of honeystone was brought to the laboratory of Adolph von Baeyer in the Technical High School at Charlottenburg. Baeyer, whose interests in structural chemistry were shaped by his mentor, Friedrich Kekule, began a series of investigations which by the 1880's had brought him to the stereochemical problems associated with the hydrogenated forms of the various carboxyl substituted benzenes derived from the honeystone (ref. 1).

In a paper concerned with the geometrical isomers of hexahydroterephthalic acid (ref. 2), a material derived from his honeystone investigations, Baeyer recognized that the relationship of the two carboxylic acid groupings in the two cyclohexane isomers paralleled that in fumaric and maleic acids, named the isomers accordingly as fumaride and maleide, and utilized the exclusive formation of anhydride with loss of water to assign configuration to the maleide hexahydroterephthalic acid (Fig. 1).

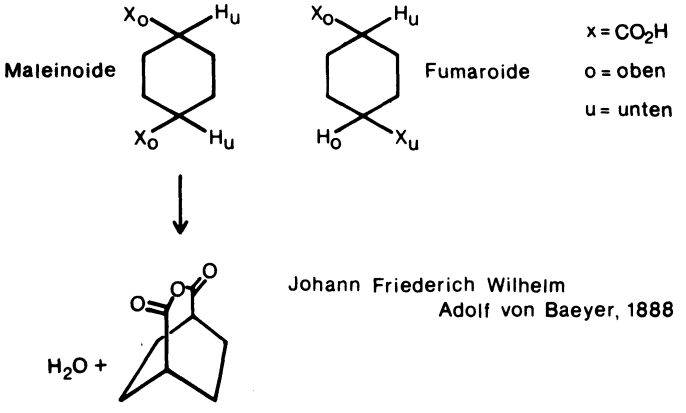


Fig. 1. Assignment of configuration to diasteromeric dicarboxylic acids.

Baeyer later created the cis and trans nomenclature as a broader means of nomenclature for stereoisomers (ref. 1). We now wish to point out that the loss of water on heating of the cis-terephthalic acid to reveal its relative molecular geometry constitutes a broader means of sterochemical analysis (ref. 3) which finds its way into the twentieth century field of mass spectrometry.

Anhydride formation in cis 1,4-cyclohexane dicarboxylic acid (ref. 2) is an example of a spatially dependent rearrangement reaction. Rearrangements are commonly observed in the mass spectra of organic molecules. Indeed, the two most common functional groups in natural product chemistry, hydroxyl and carbonyl, most commonly exhibit rearrangement chemistry on electron impact (ref. 4). This rearrangement chemistry exhibits spatial dependency (ref. 5) and thus could be utilized, following Baeyer (ref. 2), to elucidate molecular geometry. The prerequisite information for such a utilization is a firm mechanistic understanding of the rearrangements of interest. With that latter point in mind, we may ask what we know about the nature of the rearranging molecular cation radicals of hydroxy and carbonyl compounds, the chemically active intermediates of immediate interest.

In the late 1930's, Walker Bleakney and his students at Princeton University began a series of investigations on the energetics associated with the ionization and fragmentation of simple alcohols (ref. 6). Focusing on the difference between the ionization potential of methanol and the appearance potential for loss of a single hydrogen from the methanol cation radical, Bleakney found (ref. 6) that the A.P.-I.P. value was much less than the bond strength of either hydrogen bound to oxygen or to carbon. He discovered that the numbers made sense if a carbon oxygen double bond was formed in partial compensation for the breaking of a carbon hydrogen bond and thus proposed the scheme in Figure 2 (ref. 6). Bleakney in this way foresaw a reasonable formulation of the lowest energy molecular ion of methanol as a protonated methoxy radical, a species formed by ionization of one of the nonbonding electrons on oxygen (ref. 6).

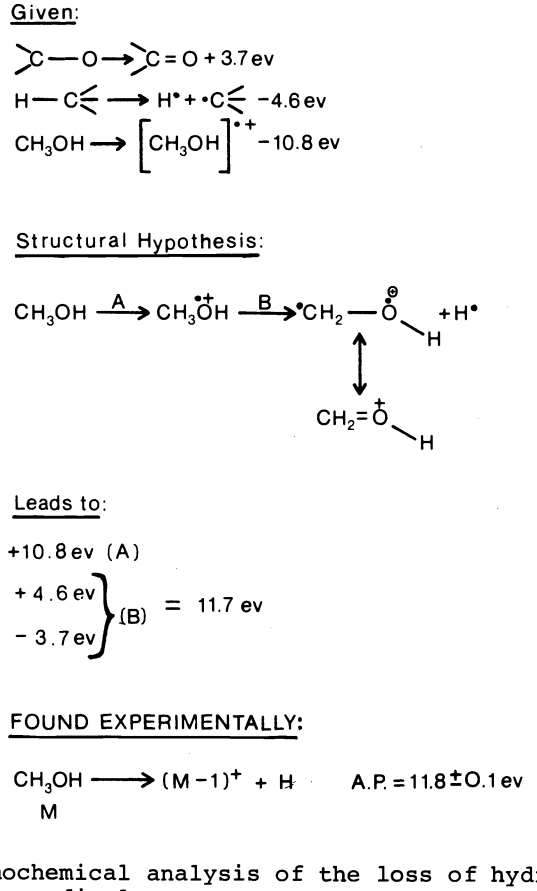


Fig. 2. Thermochemical analysis of the loss of hydrogen atom from methanol cation radical.

The formulation of the lowest energy molecular ions of hydroxyl and carbonyl compounds as arising by promotion of nonbonding electrons is an exceptionally useful concept because we might therefore expect that the chemistry of these ions would resemble that of the comparable free radicals. Photoelectron spectroscopy supports this view since alcohols and ketones and related functional groups of varied structure always exhibit discrete low energy bonds associated with promotion of nonbonding electrons on the heteroatom (ref. 7). In addition, considerations based on the quasi-equilibrium theory of mass spectrometry predict, with experimental support (ref. 4), that the rearrangement reactions of interest here (see above) are most competitive from molecular ions of the lowest internal energy (ref. 8).

This key idea that the rearrangement chemistry of hydroxy and carbonyl compound cation radicals in mass spectrometers will parallel the behavior of free radicals in solution suggests that the geometrically dependent hydrogen abstraction reactions found for example in the free radical chemistry of alkoxy radicals and triplet carbonyl compounds will carry over to mass spectrometry. The stereoanalogous diastereotopic hydrogen abstractions pictured in Figure 3 provide strong evidence for this idea.

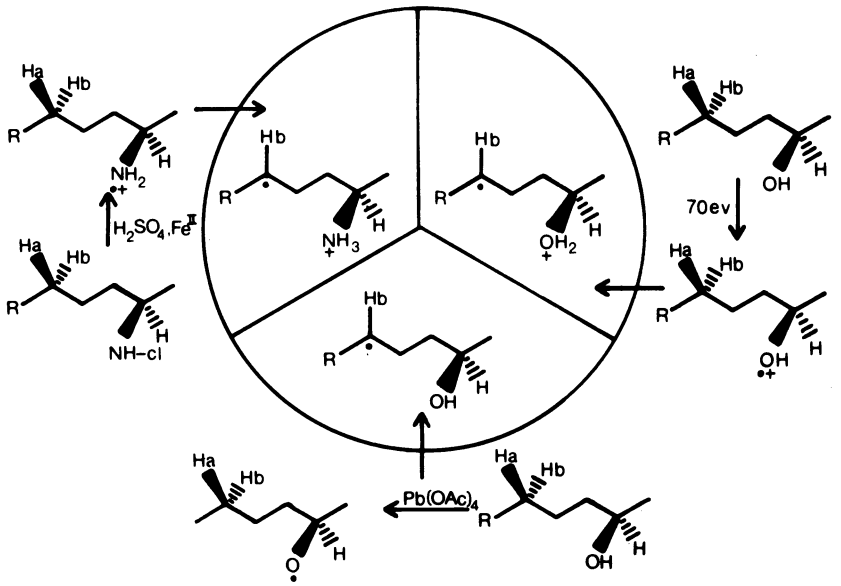


Fig. 3. Comparison of free radical chemistry and mass spectrometry. Transfer of Ha is always favored over Hb but not exclusively (ref. 9).

This rearrangement chemistry occuring in mass spectrometers and involving spatially dependent hydrogen transfer manifests itself effectively in the behavior of the cis and trans isomers of 4-tertiarybutyl and 4-isopropyl cyclohexanol (Fig. 4). As seen here only hydrogens accessible in the structurally intact molecules are available for intramolecular transfer in these model systems (ref. 10).

This phenomenon arises again in the behavior of 5-β steroids (ref. 11) where the high stereoselectivity for water loss from 3α-alcohols has been shown (ref. 12) to arise via regiospecific abstraction of the 9α-hydrogen. Figure 5 outlines these results (ref. 11, 12).

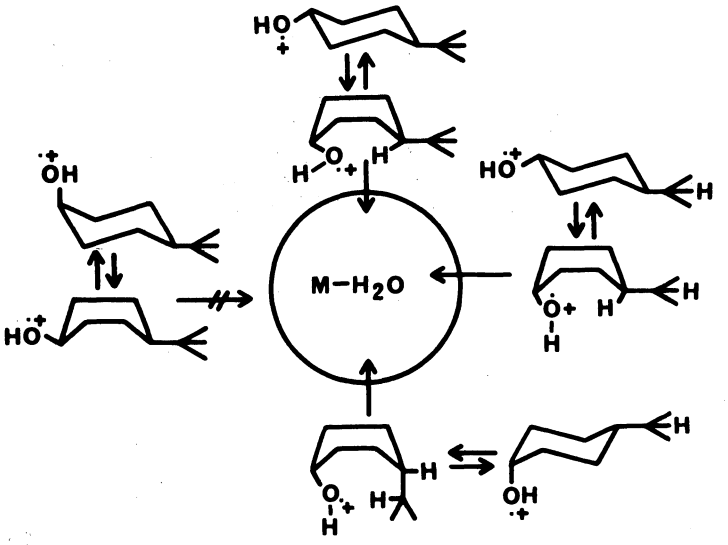


Fig. 4. Stereoselective reactions of cyclic alcohol molecular ions (ref. 10).

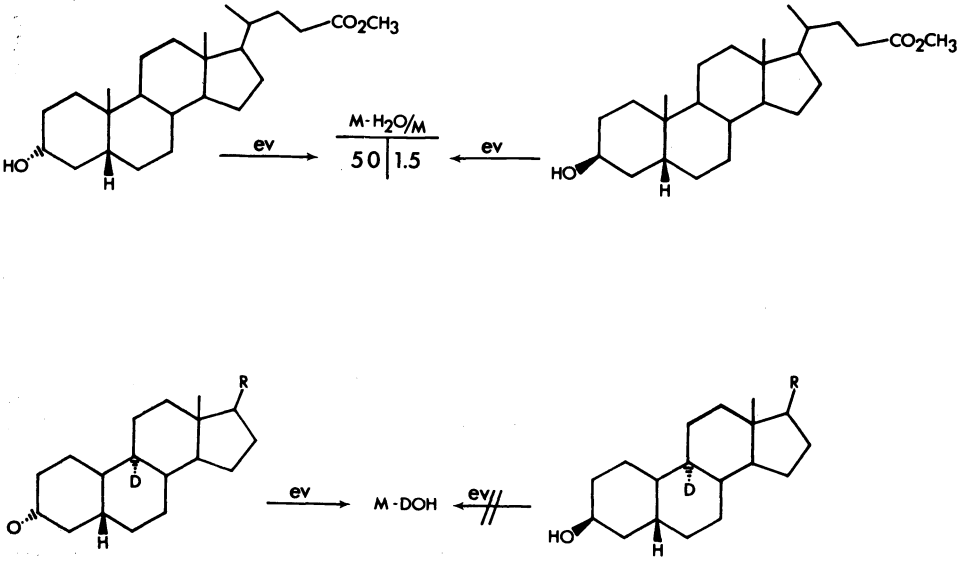


Fig. 5. Regiospecific hydrogen transfer in 5β-sterols.

The results presented in Figures 4 and 5 (ref. 10, 11, 12) suggest that these intramolecular hydrogen-abstracting rearrangement reactions could be used to assign stereochemistry in molecules of uncertain geometry. This could be carried out by connecting the rearrangement to the spatial accessibility of the groups, e.g. OH and H in Figure 4, and thereby to assign configuration. Such an approach would not be limited to elimination of water or other hydrogen transfers, but could rest on the rearrangement interaction of other functional groups. The work of Grützmacher and his students on the mass spectrometry of diols and dimethylethers of various cyclohexanes and decalins may be widely applicable in this regard (ref. 13). One of their seminal studies (ref. 14) now shown to model numerous more complex molecules (ref. 15) is shown in Figure 6.

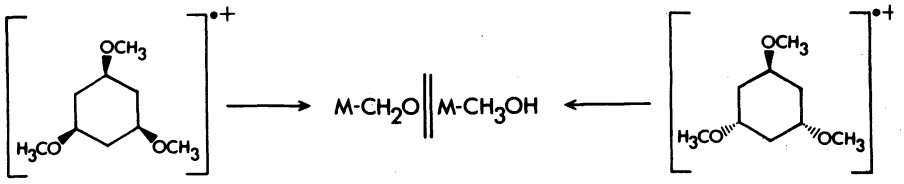


Fig. 6. Rearrangement interaction of functional groups (ref. 14).

The three examples to follow have been chosen to demonstrate the utilization of spatially dependent rearrangements to elucidate diastereomeric configuration by mass spectrometry.

MacLeod and Wells (ref. 16) prepared 2-bicyclo-[4.3.1.]-decen-10-one which upon reduction with a variety of reagents yielded only a single alcohol with configuration of the hydroxyl group either syn or anti to the double bond (Fig. 7).

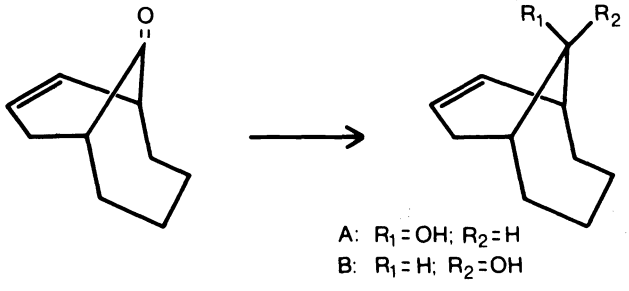


Fig. 7. Problem of configurational assignment to A and B.

Conversion of the alcohol A or B of unknown configuration to the dideutero-saturated derivative by homogenous addition of deuterium to the double bond and subsequent observation of the loss of water on electron impact gave rise to no loss of HDO. The deuterium was known to add to the exo face and following from model studies showing that exo deuterium would form HDO with the hydroxyl group syn to the unsaturated bridge (ref. 16) these workers assigned configuration A (Fig. 7) to the material in question (Fig. 8).

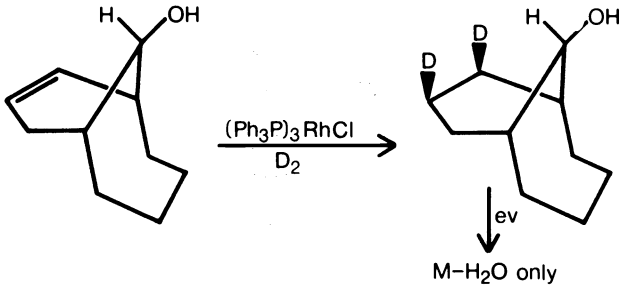


Fig. 8. Assignment of configuration utilizing the loss of water rearrangement (ref. 16).

In the mass spectra of various gibberic acids (ref. 17) the diastereomers show discrete fragmentation elimination which is sensibly related to structure. Figure 9 shows a typical example. While A, Fig. 9 gives rise to a simple mass spectrum with base peak at m/e 238 the diastereomer B exhibits only slight ion intensity at m/e 238. The latter material shows increased fragmentation though to m/e 239.

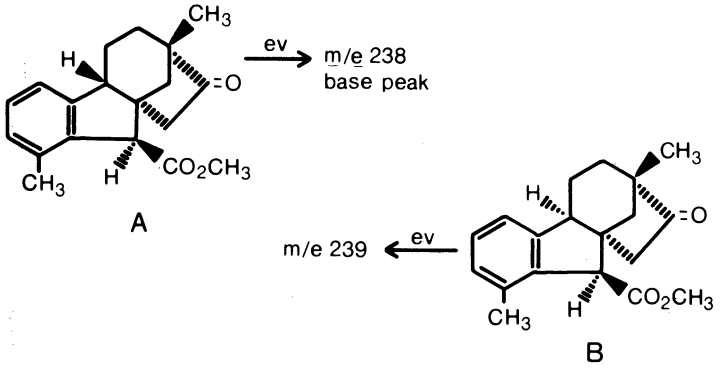


Fig. 9. Stereoselective rearrangements of gibberic acids.

High resolution mass spectrometry confirms that while A is losing the elements of methylformate to form m/e 238, the diastereomer B is simply cleaving the carboxymethyl group to yield m/e 239. Deuterium labelling of the benzylic hydrogen on C-9 (Fig. 9) caused the loss of monodeuteromethyl formate to almost the exclusion of methylformate. This general result of cis-specificity for formation of $[M - HCO_2CH_3]^+$ from the C-9 hydrogen and the carboxymethyl group manifests itself in the related allogibberates as well and was utilized by these workers (ref. 17) to assign configuration to the molecules exhibited in Figure 10.

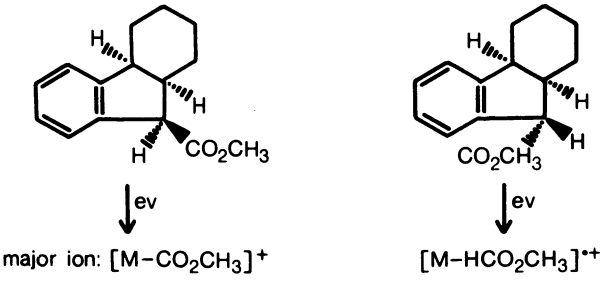
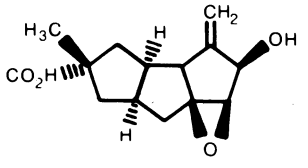


Fig. 10. Assignment of configuration by mass spectrometry utilizing a spatially dependent rearrangement of a carboxyl functional group.

A related stereoanalytical utilization of mass spectrometry arises in the case of the antibiotic material Hirsutic Acid C isolated from the hairy fungus stereum hisutum (ref. 18). In an attempted synthesis of this material the two diastereomers of unassigned configuration shown in Figure 11 were prepared (ref. 19).



Hirsutic Acid C.

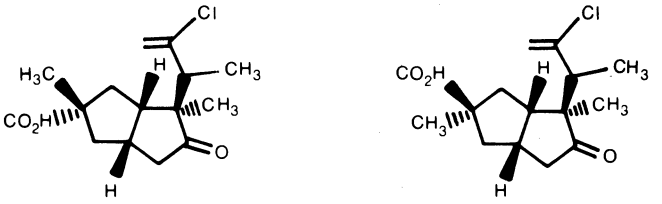


Fig. 11. Synthon diastereomers for the preparation of Hirsutic Acid C.

These two synthons which were known to differ only in the configuration at the carboxyl bearing carbon were usefully distinctive in their mass spectra. Both diastereomers split off the elements of the chlorinated side chain in an apparent McLafferty rearrangement. Only one material then went on to eliminate the elements of the carboxyl group plus one hydrogen. A rational scheme for this is shown in Figure 12 for the endo carboxyl compound. This diastereomer is unique in allowing access of the carboxyl grouping to the presumably allylically activated hydrogens in the enol product of the McLafferty rearrangement. This is well supported by deuteration of the methylene group adjacent to the carbonyl - the lost fragment now bore the elements DOHCO. Both the methyl and ethyl esters of the endo diastereomer showed similar behavior in the McLafferty rearrangement now followed by loss of methanol and ethanol plus carbon monoxide respectively. The combination of these results allowed a firm assignment of endo-carboxylic acid to the diastereomer which eliminates the elements of formic acid after McLafferty rearrangement (ref. 19). This general rearrangement interaction of ester groupings and stereochemistry by mass spectrometry

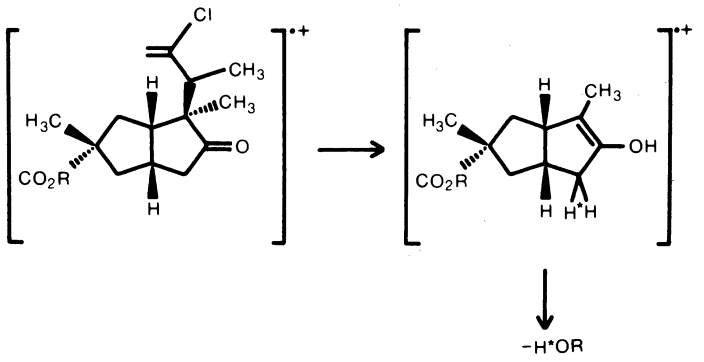


Fig. 12. Assignment of configuration to endo carboxylic acid utilizing a spatially dependent rearrangement reaction.

has been the subject of an in-depth study at the Technion in Israel (ref. 20).

It has not been our purpose here to outline every stereochemically dependent mass spectral process. Any such process has the potential of stereoanalytical use as shown above for the hydrogen rearrangements. The key point is that mechanistic understanding can translate the observed into structural information. The major reviews in this area (ref. 5) may point the way in this regard.

The connecting link of mechanism and structural insight crosses the boundaries of the various fields of mass spectrometry. Indeed, just as one may look to free radical chemistry (ref. 21) as a model for various aspects of electron impact mass spectrometry so solution phase strong acid chemistry models chemical ionization mass spectrometry (CI) (ref. 22). Work worth noting as a model for others and with great potential for stereoanalytical utility is the study of the CI of various steroid amino alcohols by Longevialle, Milne and Fales (ref. 23).

Steroidal amino alcohols functionalized in the A-ring exhibit an interesting pattern of intramolecular hydrogen bonding. This phenomenon which manifests itself in the infra-red as the appearance of a hydrogen bonded hydroxyl stretching mode of reduced frequency compared to free hydroxyl groups shows up in the steroids pictured in Figure 13 (only the A-ring is exhibited) (ref. 23).

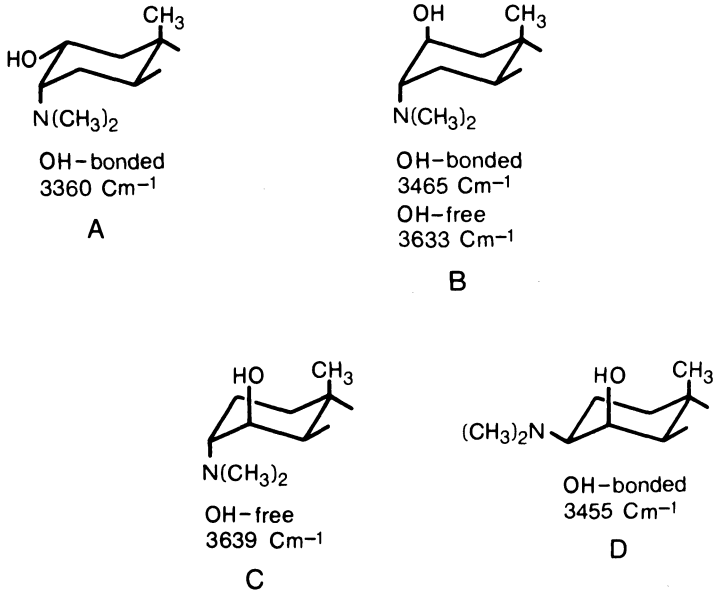


Fig. 13. Intramolecular hydrogen bonding in steroid amino alcohols. Only the A-ring is shown. The wavenumbers (cm^{-1}) are the experimental stretching frequencies found in the infra-red spectra.

The CI mass spectra, produced using isobutane reagent gas, of the steroids shown in Figure 13 exhibit loss of water from the protonated molecular ions $(MH^+) \rightarrow (MH-H_2O)^+$ only when the i.r. spectrum indicates the absence of intramolecular H-bonding to some degree. Thus in Figure 13 B and C yield substantial ions for $(MH-H_2O)^+$ while A and D do not. This pattern repeats itself for various other steroid amino alcohols studied (ref. 23). Thus, the CI presence of $(MH-H_2O)^+$ in these systems finds a basis in the conformational factors attendant to the hydrogen bonding in the neutral (ref. 24). The authors (ref. 23) reasonably ascribed the impedence of water loss from the protonated molecular ion as arising from the intervention of cyclic hydrogen bonded structures resembling those revealed by infra-red spectrophotometry on the related neutrals (Fig. 14).

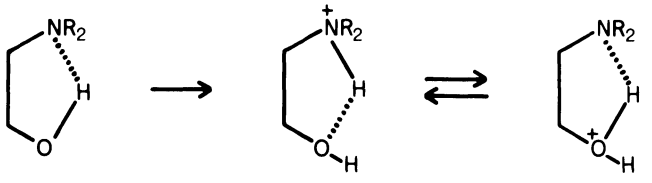


Fig. 14. Proposed analogous hydrogen bonded structures in the protonated and unprotonated steroid amino alcohols of Fig. 13.

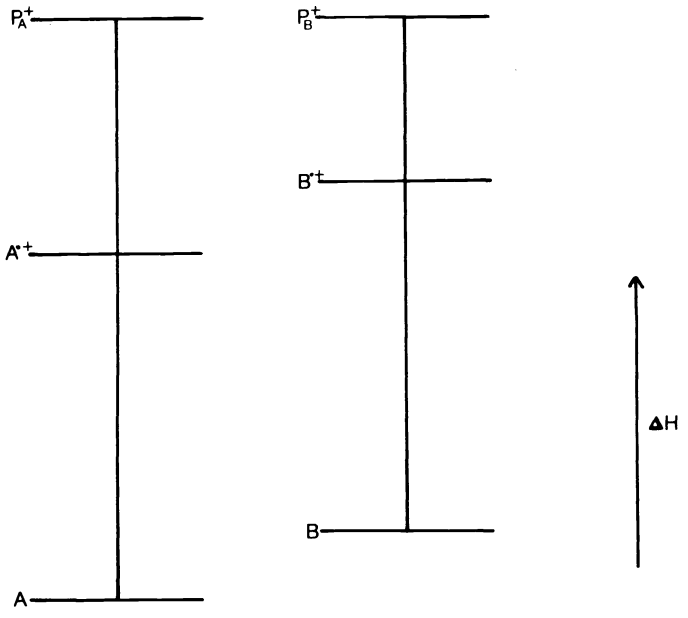
It seems that there is great potential for the study of diastereomers by CI and other intermolecular mass spectral procedures such as ion cyclotron resonancy (I.C.R.). In the latter regard Bursey has worked out methods by acetylation in the I.C.R. to distinguish certain stereoisomeric alcohols (ref. 25). We may reasonably expect excellent return on investments in stereochemical work in C.I. and I.C.R. mass spectrometry, areas in which the surface is hardly scratched (ref. 26).

All of the work discussed up to now shares the feature of a discriminating chemical reactivity as the probe of molecular three dimensionality. Another approach with considerable apparent potential involves the energetics of ionization and fragmentation. Although there are clear drawbacks of an experimental and theoretical nature concerning the accuracy of appearance potential data there is no question that such information constitutes a sensitive structural probe (ref. 27, 28).

The utility of energetic measurements in mass spectrometry (ref. 29) for the elucidation of the relative configuration of diastereomers has received impetus from an approach emphasized recently in Finland (ref. 29, 30) and Israel (ref. 29, 31). The idea is simple: if one were to study two diastereomers of differing heats of formation and measure the energy necessary to ionize and carry out a simple cleavage process which leads each diastereomer directly to a common product ion then it would follow that the difference in the appearance potentials for the two diastereomers will be in proportion to the difference in their heats of formation (ref. 29, 30). Such a scheme is presented in Figure 15.

Before going further with the appearance potential implications ((2), Fig. 15) there is an interesting kinetic consequence of this idea which has been explored by S. Meyerson and A.W. Weitkamp (ref. 32). These workers reasoned that the further decomposition of P_B^+ in Fig. 15 should be faster if, as is reasonable, both diastereomers incorporated similar packets of energy in the ionization process. In this case similar means the difference in P(E) of the two diastereomers would be small compared to the $\Delta\Delta H$ between them. Figure 16 shows some of the data they have gathered to support this idea (ref. 32).

The data in Figure 16 are consistent with the correlation predicted from the model outlined in Figure 15. The ΔH_f in these stereoisomers is a reliable predictor of the relative percent of total ionization for loss of a methyl-group. This is an ideal case study in that the diastereomeric difference centers on the methyl group which is lost, thereby making it highly likely that identical ions are produced from each diastereomer. Such is precisely the situation which Jalonen and Pihlaja (ref. 30) predict will allow determination of $\Delta\Delta H_f$ values between diastereomers by direct measurement of the $\Delta\Delta$ Appearance Potential values. They have in fact demonstrated this correspondence in a number of heterocycles and in other model systems including certain constitutional isomers (ref. 30). These workers (ref. 30) point out that the prerequisite for such information from appearance potential measurements is not accuracy, which has always been a problem in these measurements (ref. 27), but precision.



Simple model predicts:

(1) $\% \Sigma P_B^+ < \% \Sigma P_A^+$

(2) A.P. $(P_A^+) > (P_B^+)$

Where A & B are diastereomers and P_A^+ & P_B^+ are structurally identical.

Fig. 15. Scheme for relating appearance potential differences and product ion intensities to the relative heats of formation of certain diastereomers and constitutional isomers.

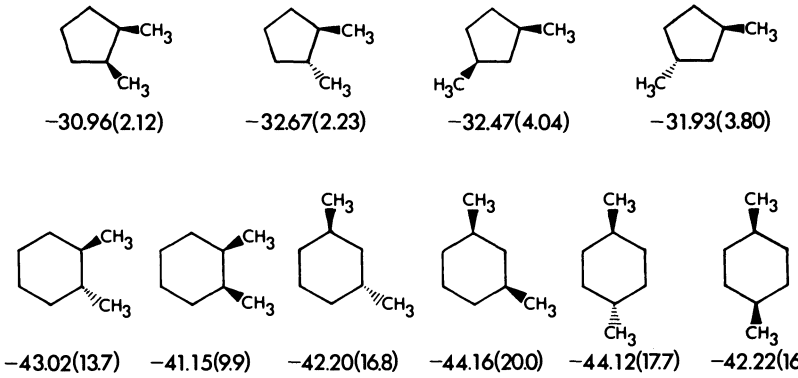


Fig. 16. Relationship between thermochemical stability and ion intensity for loss of methyl radical. Data presented as $\Delta H_f(\% \Sigma [M-CH_3]^+)$.

Zaretskii and Kelner (ref. 31) in further investigations in this general area have carefully measured the ionization potentials and appearance potentials for loss of methyl from various stereoisomers of 5α and β androstanes and pregnanes. In a very pretty confirmation of the simple idea outlined in Figure 15 the stereoisomers of the 5α series (trans AB ring juncture) exhibit higher appearance potentials for loss of methyl over the 5β series while the ionization potentials are unaffected by the epimeric difference. This is certainly an area worthy of much further exploration.

Acknowledgement - The author's investigations in this area have been generously supported by the National Institute of General Medical Sciences.

REFERENCES

1. J.R. Partington, A History of Chemistry, Vol. IV, p. 775 ff., St. Martin's Press, New York, 1964.
2. J.A. von Baeyer, Ann. Chem., 245, 128-157 (1888).
3. E.L. Eliel, Stereochemistry of Carbon Compounds, p. 185-186, Mc-Graw Hill, New York, 1962.
4. For general discussions of these phenomena and leading references see: R.A.W. Johnstone, Mass Spectrometry for Organic Chemists, Cambridge University Press, Cambridge, 1972; D.H. Williams and I. Howe, Principles of Organic Mass Spectrometry, McGraw-Hill, New York, 1972; F.W. McLafferty, Interpretation of Mass Spectra, 2nd ed., Benjamin, Reading, Mass., 1973.
5. M.M. Green, Topics in Stereochemistry, Vol. 9, (N.L. Allinger and E.L. Eliel, eds.), p. 35-110, Interscience, New York, 1976; A. Mandelbaum, Handbook of Stereochemistry, (H. Kagan, ed.), George Thieme Verlag, Stuttgart, 1977 (in press); S. Meyerson and A.W. Weitkamp, Org. Mass Spectrom. 1, 659-667 (1968).
6. C.S. Cummings, II and W. Bleakney, Phys. Rev., 58, 787-792 (1940).
7. J.B. Peel and G.D. Willett, Austral. J. Chem., 28, 2357-2364 (1975). See reference 6 within reference 8 below; P.F. Bente, III, F.W. McLafferty, D.J. McAdoo and C. Lifshitz, J. Phys. Chem., 79, 713-721 (1975).
8. See: M.M. Green, T.J. Mangner, S.P. Turner, and F.J. Brown, J. Amer. Chem. Soc., 98, 7082-7083 (1976) and references therein for strong support for these ideas.
9. M.M. Green, J.M. Moldowan, M.W. Armstrong, T.L. Thompson, K.J. Sprague, A.J. Hass and J.J. Artus, ibid., 98, 849-851 (1976).
10. This is supported by deuterium labelling. See: C.E. Brion and L.D. Hall, J. Amer. Chem. Soc., 88, 3661-3662 (1966) for the original observation. See: M.M. Green and R.B. Roy, ibid., 92, 6368-6369 (1970) for the deuterium labelling on the isopropylcyclohexanol and leading references to this literature.
11. See: M.M. Green in reference 5 above, p. 53 ff. For a general discussion of the mass spectra of steroids see: Z.V. Zaretskii, Mass Spectrometry of Steroids, Halsted Press, Israel Universities Press, Jerusalem, 1976.
12. H. Klein and C. Djerassi, Chem. Ber., 106, 1897-1904 (1973).
13. H.-F. Grützmacher, Suomen Kemistilehti, 46, 50-69 (1973). See reference 5 above for a discussion of these effects.
14. J. Winkler and H.-F. Grützmacher, Org. Mass Spectrom., 3, 1139-1168 (1970).
15. A recent paper in this series is: H.-F. Grützmacher and G. Tolkien, Tetrahedron, 33, 221-229 (1977). Utilization of these interactions may prove useful in natural product decalin diols. See: W.A. Ayer, L.M. Browne, S. Fung and J.B. Stothers, Can. J. Chem., 54, 3272-3275 (1976).
16. J.K. MacLeod and R.J. Wells, J. Amer. Chem. Soc., 95, 2387-2388 (1973).
17. R.T. Gray and R.J. Pryce, J. Chem. Soc. Perkin II, 955-960 (1974).
18. Leading references and a short review may be found in: Merck Index, ninth edition, p. 618, Merck and Co., 1976.
19. P.T. Lansbury, N.Y. Wang and J.E. Rhodes, Tetrahedron Letters, 1829-1832 (1971); N.Y. Wang, Ph.D. Thesis, State University of New York at Buffalo.
20. See the following leading references as well as reviews of this work in reference 5 above. J. Deutsch and A. Mandelbaum, J. Amer. Chem. Soc., 92, 4288-4291 (1970); S. Weinstein, E. Gil-Av, J.H. Leftin, E.C. Levy and A. Mandelbaum, Org. Mass Spectrom., 9, 774-780 (1974).

21. See references 8 and 9 above and references therein and: M.M. Green, D. Bafus and J.L. Franklin, Organic Mass Spectrom., 10, 679-681 (1975).
22. F.H. Field, Accounts Chem. Res., 1, 42-49 (1968) and subsequent papers in that series. See also: F.H. Field, in MTP International Review of Science, Physical Chemistry Series One, Vol. 5 (A. Maccoll, ed.) Ch. 5, Butterworths, London (1972); G.A. Olah and J. Shen, J. Amer. Chem. Soc., 95, 3582-3584 (1973) and leading references therein.
23. P. Longevialle, G.W.A. Milne and H.M. Fales, J. Amer. Chem. Soc., 95, 6666-6669 (1973).
24. For further conformational effects involving intermolecular hydrogen bonding in CI mass spectrometry see: H.M. Fales and G.J. Wright, J. Amer. Chem. Soc., 99, 2339-2340 (1977).
25. See: M.M. Bursey, J.L. Kao, J.L. Henion, C.E. Parker, and T.I.S. Huang, Anal. Chem., 46, 1709-1712 (1974) for the most recent paper with leading references.
26. W.J. Richter, American Society for Mass Spectrometry meeting May, 1977, Washington, D.C.
27. H.M. Rosenstock, Int. J. Mass Spectrom. and Ion Physics, 20, 139-190 (1976); A.G. Harrison and C.W. Tsang in Biochemical Applications of Mass Spectrometry (G. Waller, ed.), Ch. 4, Wiley-Interscience, New York, 1972; J.D. Morrison in MTP International Review of Science, Physical Chemistry Series One, Vol. 5 (A. Maccoll, ed.), Ch. 2, Butterworths, London, 1972; D.H. Williams and I. Howe, Principles of Organic Mass Spectrometry, Ch. 5, McGraw-Hill, London (1972); J.L. Franklin, J.G. Dillard, H.M. Rosenstock, J.T. Herron, K. Draxl and F.H. Field, Ionization Potentials, Appearance Potentials and the Heats of Formation of Gaseous Positive Ions, National Bureau of Standards, U.S. Government Printing Office, Washington, D.C., (1969); A.G. Harrison in Topics in Organic Mass Spectrometry, (A.L. Burlingame, ed.) p. 121, Wiley-Interscience, New York (1970).
28. Figure 2 (above) and reference 6 constitute a classic case in point.
29. See reference 5 for general discussions and leading references in this area and for references to the earlier pioneering work of P. Natalis.
30. J. Jalonen and K. Pihlaja, Org. Mass Spectrom., 7, 1203-1210 (1973).
31. Z. (V.I.) Zaretskii and L. Kelner, Tetrahedron, 31, 85-87 (1975).
32. S. Meyerson and A.W. Weitkamp, Org. Mass Spectrom., 2, 603-609 (1969).