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SPECIAL FEATURE: PERSPECTIVE

Structurally diagnostic ion/molecule reactions: class and functional-group identification by mass spectrometry

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This article discusses the application of gas-phase ion/molecule reactions for fine structural elucidation in mass spectrometry. This approach is illustrated via a representative collection of class- and functional group–selective reactions, a few of historical relevance as well as by more recent and instructive examples, and their applications. The focus is on reactions performed under well-controlled conditions of sequential mass spectrometry, discussing key mechanistic details and potential applications. Recent and innovative strategies that allow these reactions to be performed under ambient conditions, making this fast, selective and sensitive approach for structural investigation much more generally applicable, are also discussed.

KEYWORDS: ion/molecule reactions; gas-phase reactivity; ion chemistry; mass spectrometry; structure elucidation

INTRODUCTION

In chemistry there is often a direct, rational and very elegant relationship between structure and reactivity. Many types of structural information can therefore be obtained by screening for the occurrence or lack of a specific reaction. In mass spectrometry, for historical and practical reasons, structure/reactivity relationships have been explored mainly for unimolecular reactions that lead to ion dissociation.[1,2] Many diagnostic fragment ions of fragmentation processes have been found and extensively studied throughout the development of MS for structural investigation. Today, ion dissociation occurring as the result of ionization or induced by extra energy provided by collisions or radiation is by far the most frequent process used in MS to access structural information including atom connectivity and spatial arrangements.

Thomson,[3] as early as 1913, noticed that ion/molecule reactions occur efficiently inside mass spectrometers, but these reactions began to be explored in MS only after 1966 when Munson and Field introduced chemical ionization (CI).[4] Many CI gases and reactions have since been developed and used to screen for classes of compounds and to identify specific functional groups.[5,6] These structurally diagnostic reactions are therefore usually referred to as either *class-selective* or *functional group–selective* ion/molecule reactions. In CI, however, ion formation and its further reaction are performed in the same region (ion source) without previous ion isolation; hence the desired reaction may occur concurrently with other reactions involving cogenerated ions. With the development of MS instrumentation, particularly after the introduction of tandem-in-space and tandem-in-time mass spectrometers, reactions of ions with neutral molecules can now be performed with much greater refinement than is possible under CI environments. Very efficient multiple-stage mass spectrometers, most particularly the ICR,[7] quadrupole ion traps,[8] flowing afterglows[9] and triple- and penta-quadrupoles,[10] and the many scan modes they offer, make possible the isolation of the desired reacting ion with minimized internal energy, its reaction under low-enough energies to allow for efficient bimolecular reactions accompanied by minimal or no ion dissociation, as well as under controlled and well-defined reaction conditions such as pressure, temperature and time. Using MS^n techniques, reactions can be monitored as a function of time with instant structural investigation of product ions employing either CID or a second structurally diagnostic ion/molecule reaction, or a combination of both.

The mass spectrometrist, with great refinement and exploiting the outstanding speed, sensitivity and selectivity as well the diverse environments that only MS offers, can therefore use the rational relationship between structure and chemical reactivity to investigate the structure of

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gaseous neutral molecules and ions by using an arsenal of class- and functional group–selective ion/molecule reactions. These investigations, which have been reviewed from other perspectives,[7–16] also uncover the intrinsic mechanistic details of these structurally diagnostic reactions that occur under the unique solvent- and counter ion–free environments of mass spectrometers.

This article illustrates the effectiveness of ion/molecule reactions for structural investigation by collecting a representative set of class- and functional group–selective ion/molecule reactions. This set includes a few examples of historical relevance as well as more recent and instructive examples, with a focus on those reactions performed under the well-defined conditions of sequential mass spectrometry using mass-selected ions. Key mechanistic details and potential applications are also discussed. Recently, ion/molecule reactions have been shown to occur efficiently under atmospheric pressure conditions and even under ambient conditions for molecules present on surfaces, and these innovative strategies that greatly widened the scope of applications of such reactions will also be presented.

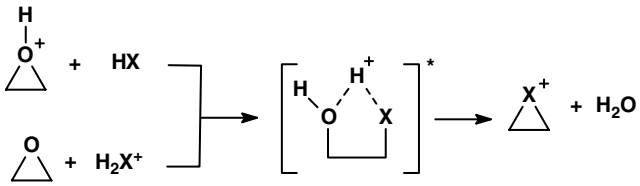
PIONEERING WORK INSPIRING FURTHER DEVELOPMENTS

Epoxides

The applicability for structural elucidation of gas-phase ion/molecule reactions performed via MS^n experiments was already evident from the very earliest experiments. Many illustrative examples of these pioneering works could be mentioned, such as those of Gross, Beauchamp, Jennings and Ausloos and their coworkers. In 1972, Gross and coworkers[17] studied by ion cyclotron resonance-mass spectrometry (ICR-MS) the reactions of the isolated radical cation of 1,3-butadiene with isomeric olefins of C_5H_{10} composition, which display similar EI-MS. They observed distinct sets of product ions, lack of reactivity or significant differences in product distribution, which allowed them to differentiate all eight isomers tested. In 1974, Beauchamp and coworkers[18] showed that nucleophilic attack on protonated epoxides in the gas phase, promoted for instance by PH_3, as well as of protonated PH_3 on the neutral epoxide, differentiates the epoxide from isomeric ketones and carbonyl ylides. They observed that protonated ethylene oxide reacts selectively with PH_3 (Scheme 1, X = PH_2) via an interesting ring-opening/re-forming mechanism leading to the cyclic phosphonium ion of m/z 61.

Later, Cooks and coworkers[19] reported another class-selective reaction for protonated epoxides. They found that ethyl vinyl ether adds promptly to protonated cyclohexene epoxide but not to protonated molecules bearing other oxygen-containing functional groups, i.e. protonated ketones, esters, ethers and alcohols (Fig. 1). Note that selective reactions of protonated molecules are attractive considering that such gas-phase species, even from heavy and polar analytes, are today easily obtained using atmospheric pressure ionization (API) techniques (ESI, APCI, APPI) or matrix assisted laser desorption ionization (MALDI).

Epoxides are common reagents, and key intermediates in a variety of synthetically important reactions, and therefore strategies to identify such important class of compounds on the basis of selective gas-phase reactions are likely to find broad applications. More recently, we[20] reported another



Scheme 1

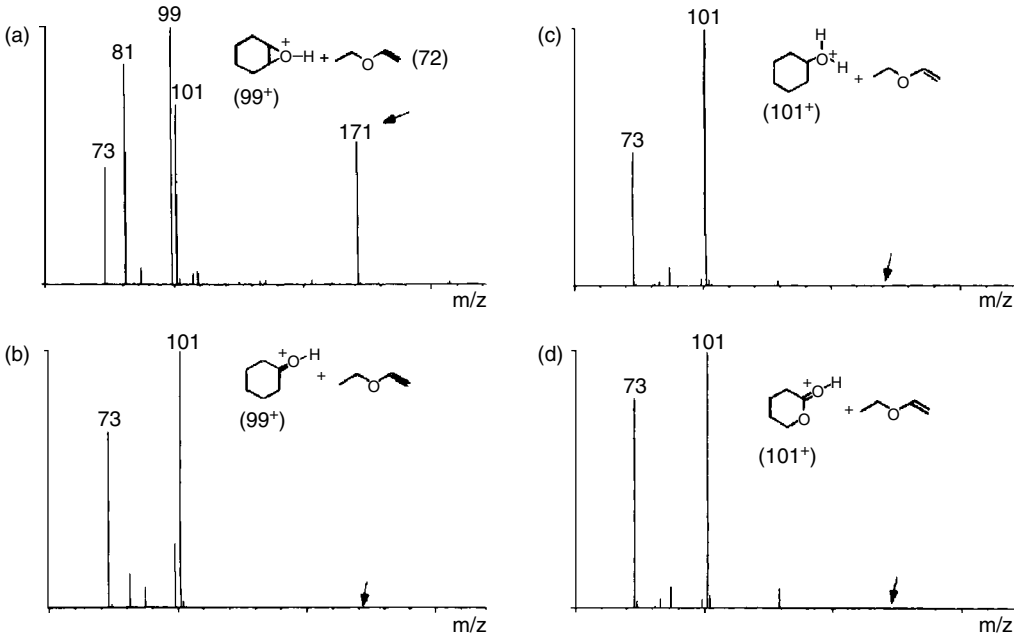


Figure 1. Product ion mass spectra for the reactions with ethyl vinyl ether of protonated molecules of (a) cyclohexene oxide (b) cyclohexanone (c) cyclohexanol and (d) γ-valerolactone. Adapted from Ref. 19.

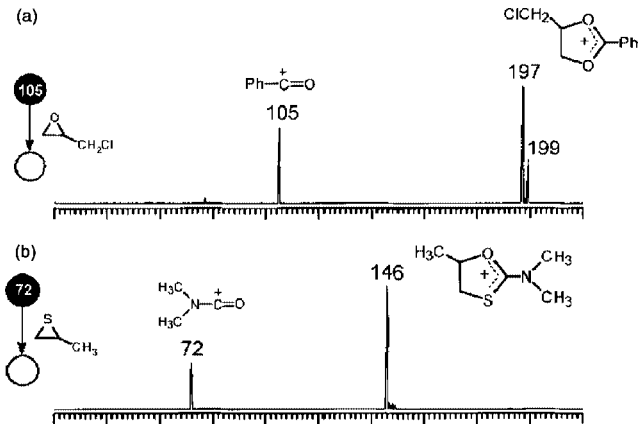
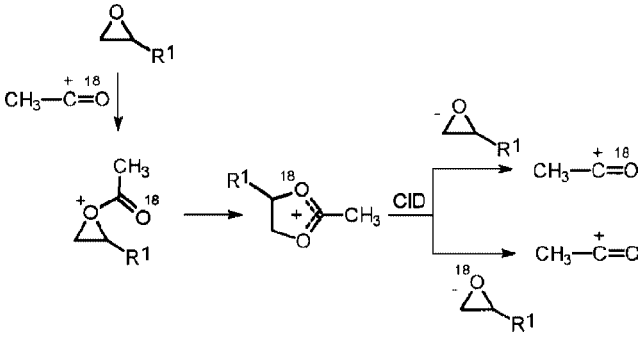


Figure 2. Product ion mass spectra for the reactions of (a) $Ph-C^+=O$ with epichlorydrin and (b) of $(CH_3)_2N-C^+=O$ with propylene sulfide. Adapted from Ref. 20.



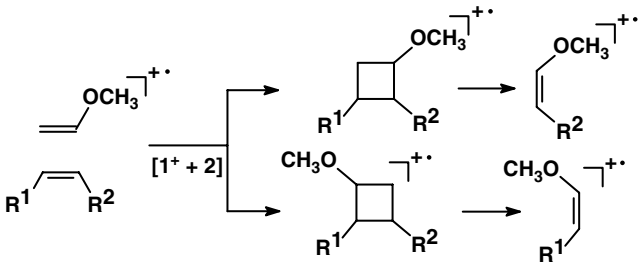
Scheme 2

selective ion/molecule reaction for epoxides (Fig. 2). Neutral molecules of epoxides and thioepoxides were found to react distinctively with acylium (and phosphonium ions) via a three-to-five membered ring expansion mechanism (probed by ^{18}O-labelling) that led to 1,3-dioxolanylium ions and analogues (Scheme 2). To honor Meerwein, who in 1955 reported an analogous reaction in solution, we termed this as *the gas-phase Meerwein reaction*. Cooks and coworkers[21] also reported gas-phase Meerwein reactions with phosphonium ions, and proposed its use as a method of recognizing compounds that yield these ions, which include a number of chemical warfare agents.

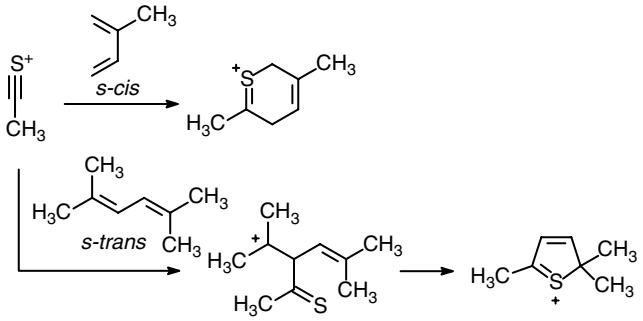
Dienes and dienophiles

Because of their high selectivity for molecular class (dienes with dienophiles) and geometry (*s-cis* conformation), cycloadditions are prime candidates for class-selective reactions, and many examples of their use for gas-phase polar cycloadditions have been described and recently reviewed.[13] Such applications have been demonstrated for several systems, and we will present here a few of the most illustrative cases. In 1978, Jennings and coworkers[22] performed one of the first gas-phase, structurally diagnostic cycloadditions, i.e. of ionized methyl vinyl ether with olefins and dienes. Upon dissociation mono- and di-unsaturated compounds were found to form structurally diagnostic product ions, which indicate the location of the initial double bond (Scheme 3).

We,[23,24] also taking advantage of the high specificity of cycloaddition reactions, showed later that *s-cis* conjugated



Scheme 3



Scheme 4

dienes react selectively with acylium and thioacylium ions via polar $[4+2^+]$ cycloadditions, as Scheme 4 illustrates for the thioacetyl cation, whereas Caserio and coworkers[25] showed that an *s-trans* locked diene reacts distinctively.

We[26] also showed that $[4+2^+]$ cycloadditions with isoprene are able to distinguish protonated nitriles from isomeric protonated isonitriles, and used the proof-of-principle pair of methyl isomers to demonstrate this possibility. The protonated isonitrile reacts mainly by cycloaddition to give the product ion of m/z 110, whereas the protonated nitrile reacts predominantly by proton transfer

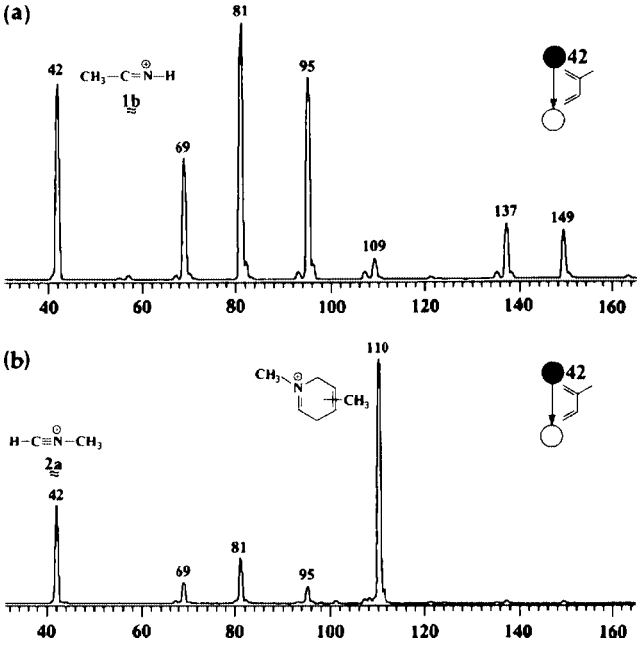


Figure 3. Product ion mass spectra for the reactions of the isomeric ions and (a) $CH_3-C\equiv N^+-H$ and (b) $H-C\equiv N^+-CH_3$ of *m/z* 42 with isoprene. Adapted from Ref. 26.

to give the ion of m/z 69 as well as a number of secondary hydrocarbon product ions (Fig. 3).

In a series of reports, we[27–29] showed that polar $[4^+ + 2]$ cycloadditions of cationic 2-azabutadienes are selective for enol ethers and analogues and that CID of the cycloaddition product ions can provide refined structural information. Figure 4 illustrates such an application for two members of the homologue series of enol ethers, i.e. methyl and *n*-butyl vinyl ether. Both molecules react extensively with MAB^+ (Scheme 5) to form the respective cycloadducts of m/z 128 and 170. Upon CID, the cycloadduct of m/z 128 loses a methanol molecule of 32 Da, whereas that of m/z 170 loses *n*-butanol of 84 Da. These dissociations disclose the mass of the alkyl group, i.e. methyl or *n*-butyl respectively. This principle is further illustrated (Fig. 5) for vinyloxy trimethylsilane since it reacts promptly with MAB^+, whereas its cycloadduct loses trimethylsilol of 90 Da.

The presence and the nature of substituents at the double bond of enol ethers can also be investigated via this combined ion/molecule reaction plus CID approach. As Fig. 6 shows, the two methyl ether cycloadducts lose a methanol molecule,

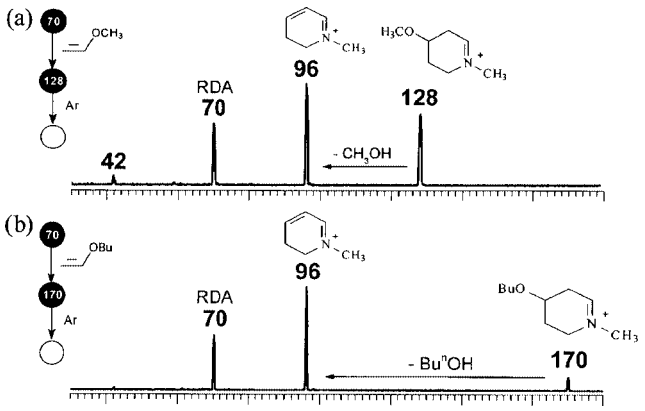
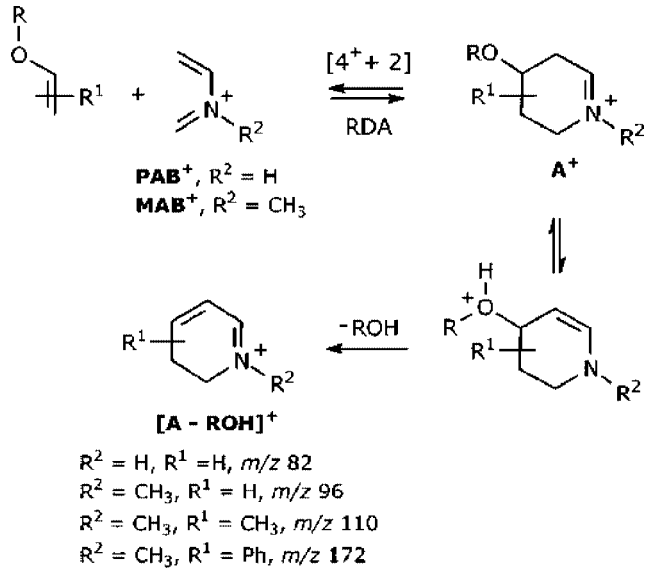


Figure 4. Sequential product ion mass spectrum for the $[4^+ + 2]$ cycloadduct of MAB^+ with (a) methyl vinyl ether of m/z 128 and (b) *n*-butyl vinyl ether of m/z 170. Adapted from Ref. 28.



Scheme 5

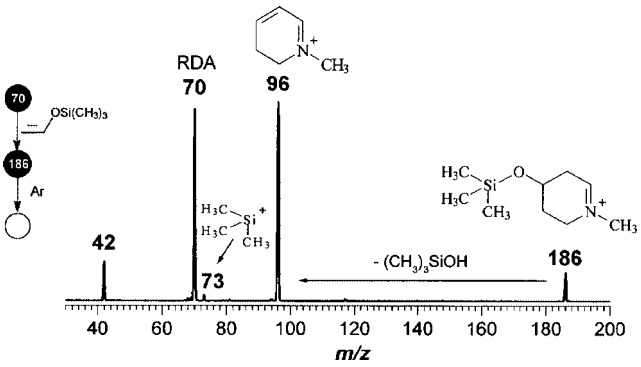


Figure 5. Sequential product ion mass spectrum for the $[4^+ + 2]$ cycloadduct of m/z 186 formed via reactions of MAB^+ with vinyloxy trimethylsilane. Adapted from Ref. 28.

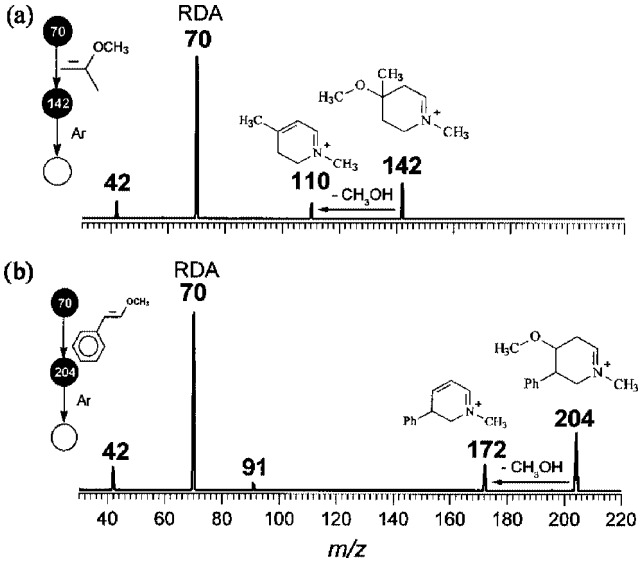


Figure 6. Sequential product ion mass spectrum for the $[4^+ + 2]$ cycloadduct of MAB^+ with (a) 2-propenyl vinyl ether (m/z 142) and (b) β-methoxy styrene (m/z 204). Adapted from Ref. 28.

but the methyl substituent at the vinyl group causes an m/z shift for the fragment ion of 14 units (m/z 110 as compared to the unsubstituted ion of m/z 96, Fig. 4) whereas a phenyl substituent causes the corresponding m/z shift of m/z units (m/z 172).

The direct relationship between structure and reactivity so clearly perceived in these structurally diagnostic ion/molecule reactions is also evident for cyclic enol ethers. Their cycloadducts dissociate exclusively via the retroaddition pathway (Fig. 7) owing to the cyclic nature of the neutral reactant; the 'alcohol-loss' pathway in this case is precluded causing just isomerization via opening of the THF ring.

Recently, Cooks and coworkers[30] applied structurally selective cycloadditions with ethyl vinyl ether to diagnostic fragment ions for the secure detection of the explosives trinitrotoluene (TNT) and hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX). For instance, a major fragment ion of RDX was selected for the reaction, and a series of diagnostic product ions were formed (Scheme 6).

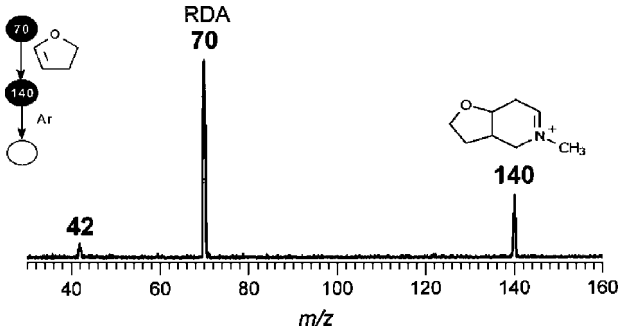
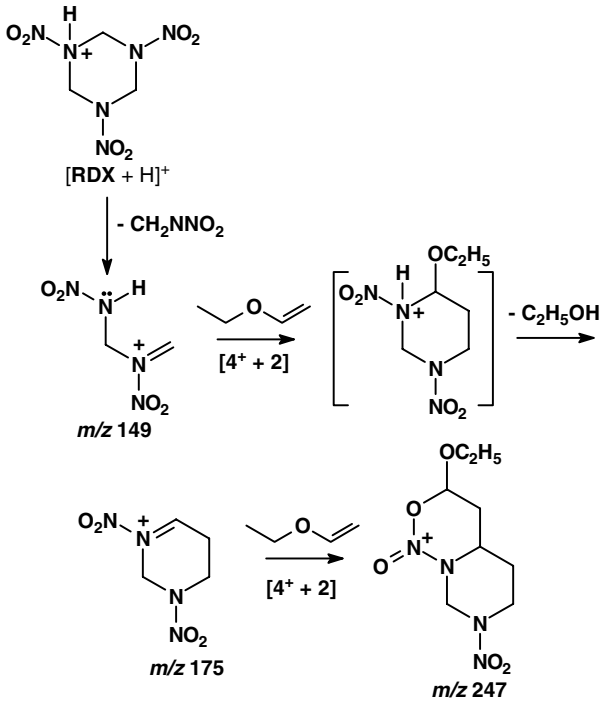


Figure 7. Sequential product ion mass spectrum for the $[4^+ + 2]$ cycloadduct of *m/z* 140 formed by reactions of MAB^+ with a cyclic vinyl ether. Adapted from Ref. 28.



Scheme 6

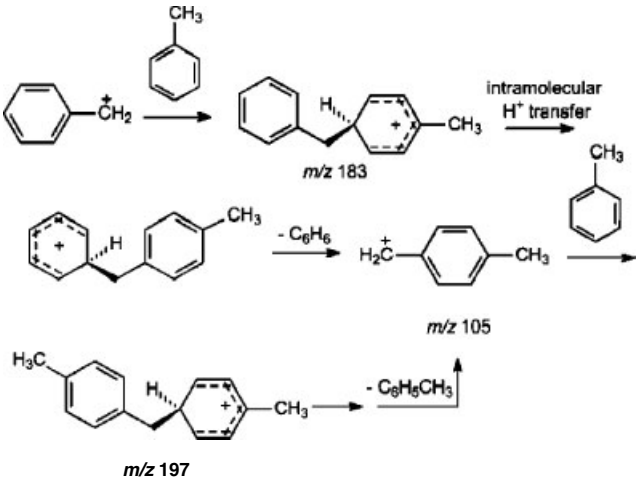
Methylene transfer reaction

In 1982, Ausloos,[31] using an ICR cell, reacted CD_2F^+ with benzene to form $[C_7H_5D_2]^+$ ions and found that 90% of such ions (benzylium ions) react further with toluene to form $[C_8H_7D_2]^+$ via a structurally diagnostic methylene transfer reaction. This reaction characterizes both the benzylium ion (since it is the only stable $C_7H_7^+$ isomer bearing a methylene unit) as well as the benzene derivative molecule, whereas the isomeric tropylium ion was found to be unreactive. Later, we[32] performed this classical structurally diagnostic reaction (Scheme 7) to probe the structure of a $C_7H_7^+$ product ion under penta-quadrupole reaction conditions obtaining very clear and similar results (Fig. 8), i.e. the methylene transfer product of *m/z* 105 was formed to a great extent together with minor products from a similar secondary reaction.

Hydroxy ketones

In 1989, Kenttamaa and Cooks[33] reported the first class-selective reaction applied with efficiency to considerably complex molecules. They found that protonated β-hydroxycarbonyl compounds react selectively with ethyl vinyl ether to form characteristic cyclic products (Scheme 8), and demonstrated its use to distinguish a pair of isomeric diterpenoid dilactones, i.e. the reactive β- and the unreactive γ-hydroxy ketones shown in Fig. 9.

Similarly, we[34] reported recently that ion/molecule reactions with acylium ions are class-selective for α-, β- and γ-hydroxy ketones. The relative positions of the carbonyl and hydroxy groups along the carbon chain favors specific structurally diagnostic reactions, which Scheme 9 illustrates for a β-hydroxy ketone, and a characteristic product or set of product ions are formed for each isomeric molecule (Fig. 10).



Scheme 7

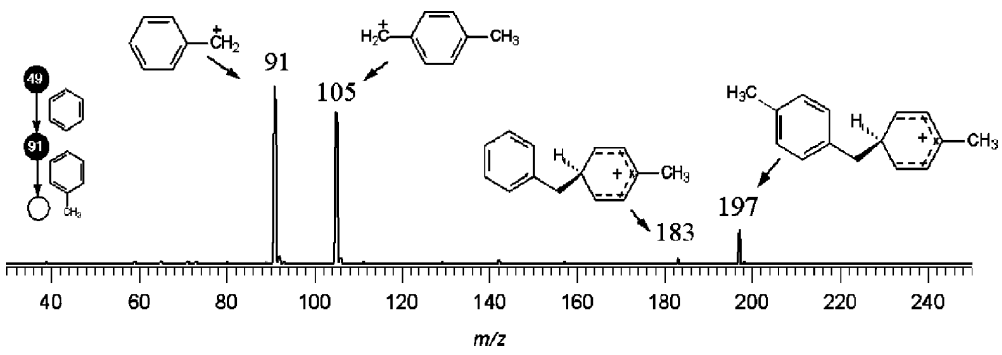


Figure 8. Sequential product ion mass spectrum for the reactions of the benzyl cation of *m/z* 91 (formed via reactions of $CH_2{}^{35}Cl^+$ of *m/z* 49 with benzene) with toluene. Adapted from Ref. 32.

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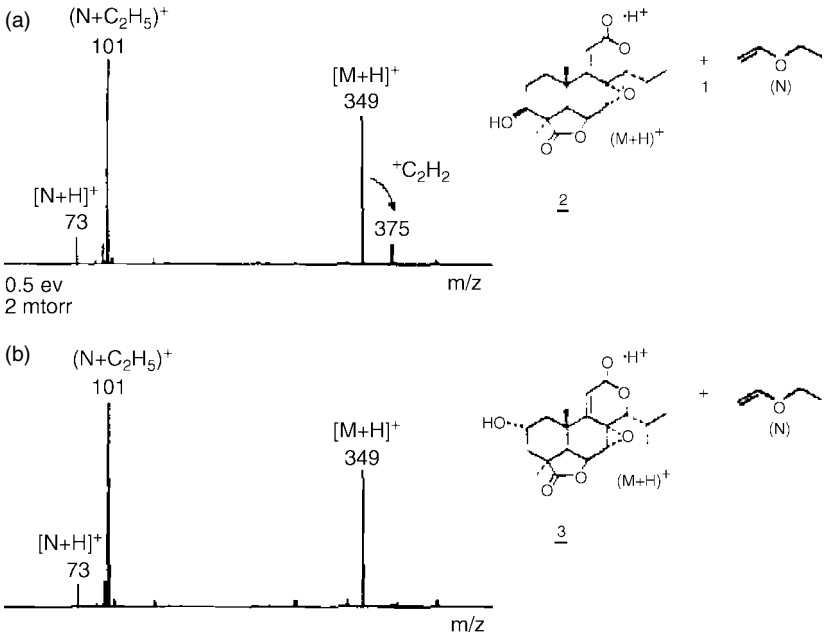
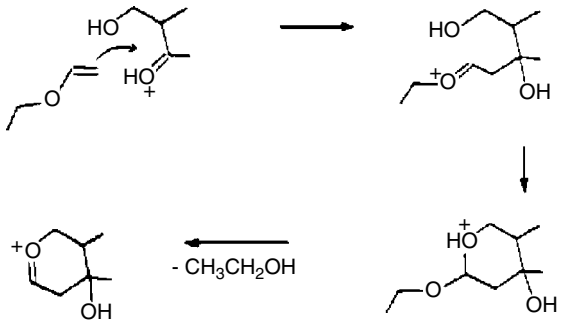
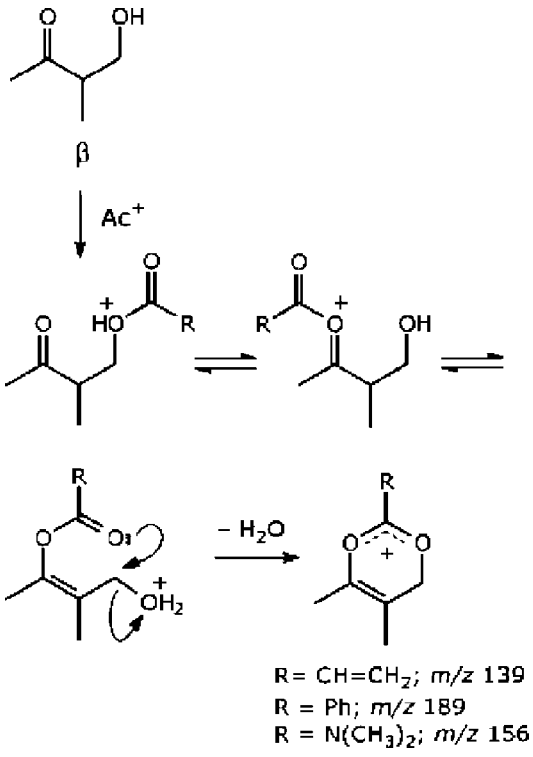


Figure 9. Product ion mass spectra for the reactions of an isomeric pair of protonated (a) β- and (b) γ-hydroxycarbonyl compounds with ethyl vinyl ether (N). The class-selective product is that of *m/z* 375. The ion of *m/z* 101 arises from ethylation of N by $[N + H]^+$. Adapted from Ref. 33.



Scheme 8



Scheme 9

Acetals, diols and analogues

In solution, acetalization reactions are known to be highly class-selective, i.e. they occur most favorably for α- and β-diols in reaction with aldehydes and ketones. We[35–37] therefore tested gas-phase acetalizations with acylium ion as class-selective for, and to reveal structural details of, α- and β-diols and amino, thiol, ether and thioether analogues. These reactions were often found to occur to great extents and to proceed in a very functional group–selective manner. For instance, loss of H_2O occurs for diols, loss of H_2S for dithiols, loss of NH_3 for alcoholamines and loss of CH_3OH for monoalkyl ethers of diols (Fig. 11), whereas some bifunctionalized molecules react accordingly losing both neutrals (although to contrasting extents), as Scheme 10 exemplifies for ethanolamine.

Stereoisomers

A challenging tasks for structural elucidation is certainly the differentiation of stereoisomers, those that differ only by different spatial arrangements. Bimolecular reactions, which are greatly affected by spatial arrangement, are therefore most likely to succeed for stereoisomer assignment. Polar acetalization involving ring-closure is, in this sense, a candidate to differentiate geometric stereoisomers, and indeed it was shown to be stereoselective for *trans* 1,2-diamino cyclohexane (Fig. 12).[36] The *cis* isomer is inert toward acetalization, whereas the *trans* isomer (for which backside nucleophilic attack is favored, Scheme 11) reacts selectively to form the diagnostic product ion of *m/z* 169. Similar stereoselective reactions for diamines and

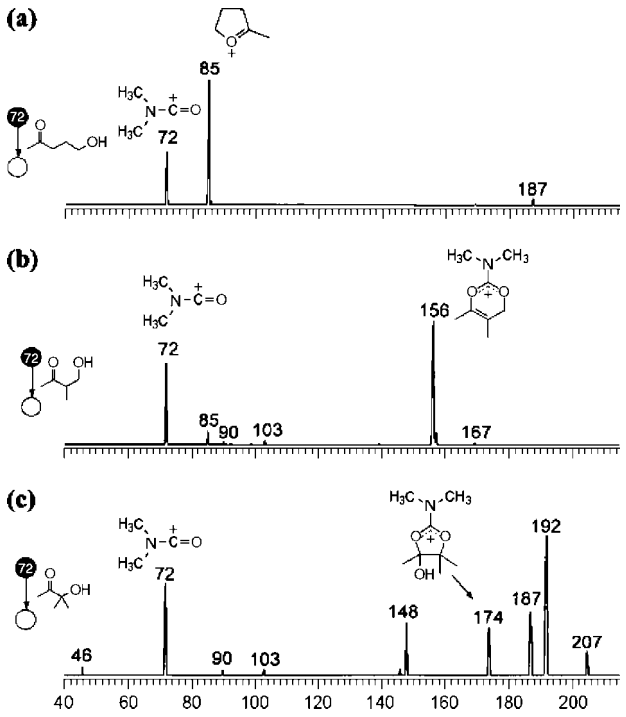


Figure 10. Product ion mass spectra for the reactions of the acylium ion $(CH_3)_2N{-}C^+{=}O$ with each member of an isomeric set of (a) α-, (b) β- and (c) γ-hydroxyketones. Adapted from Ref. 34.

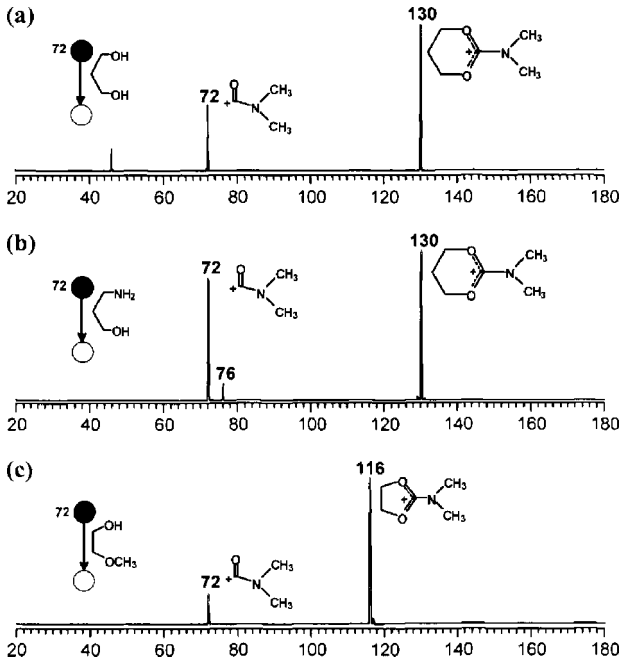
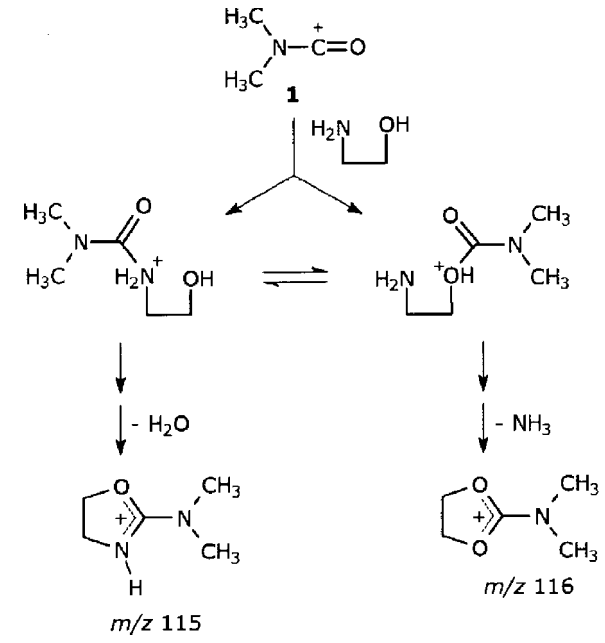


Figure 11. Product ion mass spectra for reactions of the acylium ion $(CH_3)_2NCO^+$ of *m/z* 72 with (a) 1,3-propanediol, (b) 3-amino-1-propanol and (c) 2-methoxyethanol. Adapted from Ref. 28.

diols have been reported by Kenttamaa and coworkers using phosphenium[38] and borinium ions.[39] Meyerhoffer and Bursey[40] also have shown that $(CH_3)_3Si^+$ reacts stereoselectively with *cis*-1,2-cyclopentenediol by water abstraction to form $(CH_3)_3SiOH_2^+$, whereas the *trans* isomer is much less effective, requiring a higher activation energy.



Scheme 10

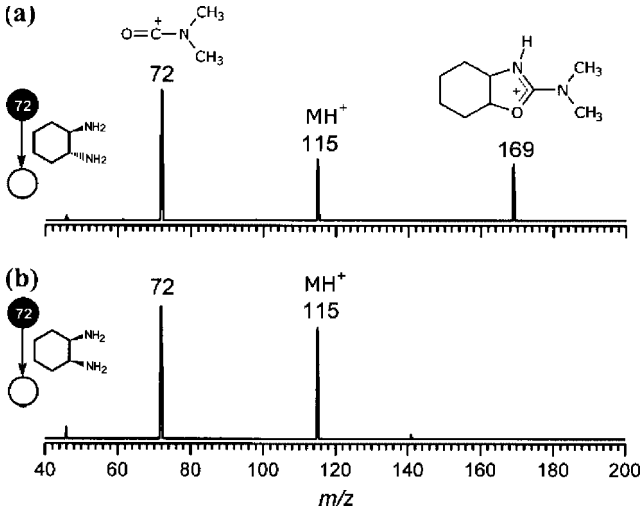


Figure 12. Product ion mass spectra for the reaction of the acylium ion $(CH_3)_2N{-}C^+{=}O$ of *m/z* 72 with the stereoisomeric pair of (a) *trans*- and (b) *cis*-diamine cyclohexane. Adapted from Ref. 36.

Transacetalization

We[41,42] also showed that polar transacetalization with acylium and many related amphoteric ions (Scheme 12) serve as a very selective reaction for cyclic acetals and analogues. This versatile gas-phase reaction has been extensively explored, and a variety of synthetic and analytical applications have been found. An extensive review on this reaction is to appear.[15] Figure 13 illustrates this effective reaction for two cyclic acetals reacting with the $CH_2{=}CH{-}C^+{=}O$ acylium ion of *m/z* 55. Transacetalization is by far the dominant process for both neutral acetals and occurs with the structurally diagnostic release of either cyclohexanone or formaldehyde to form the same ionic acetal of *m/z* 99. This class-selective reaction demonstrates clearly that both neutral reactants are ethylene glycol acetals (from the *m/z* of the product) with

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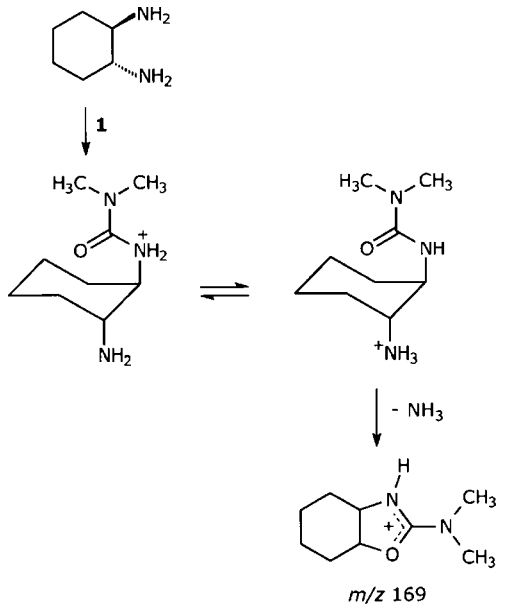
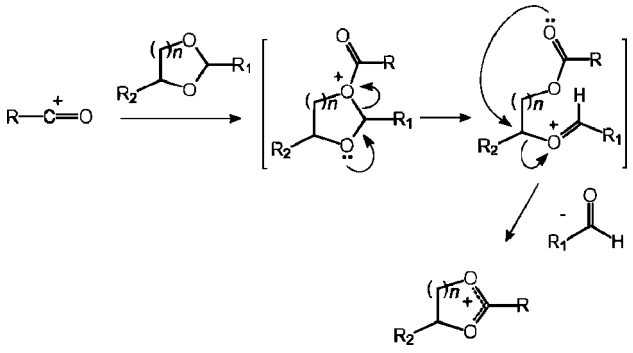


Scheme 11



Scheme 12

no ring substituents at the 4 and 5 positions. If the mass of the neutral is known (or determined via the $[M + H]^+$ or $[M - H]^+$, species that are often formed concurrently) the mass of the released carbonyl compound can easily be determined. If there is a substituent at the 4 or 5 ring positions (or both), the m/z of the ionic acetal is shifted accordingly. Figure 14 illustrates this interesting feature for the differentiation of two isomeric acetals. We also proposed the use of polar transacetalization with dioxolanes of the diagnostic NO_2^+ ion (Scheme 13) as an ion/molecule reaction capable of screening with high selectivity for organonitrate explosives in general.[43]

Cooks and coworkers[44] proposed the use of a similar acetalization-like reaction of phosphonium ions with 1,4-dioxane (Scheme 14) for the selective detection of organophosphorus esters, important warfare-agent simulants (Fig. 15).

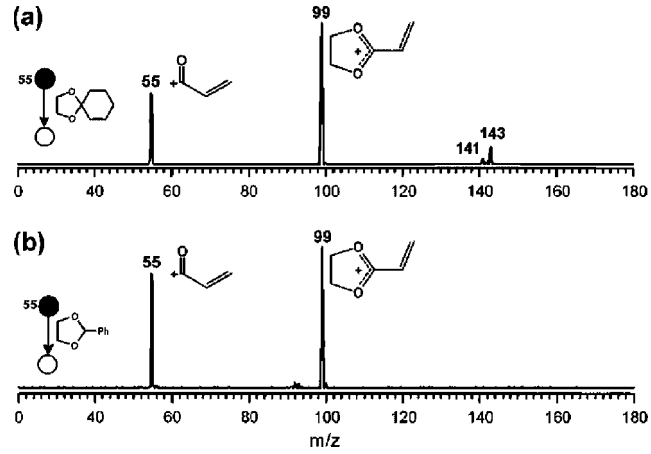
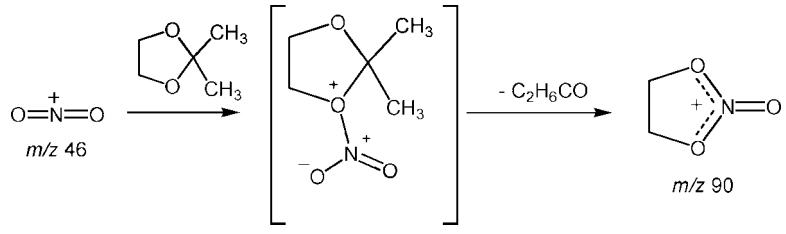


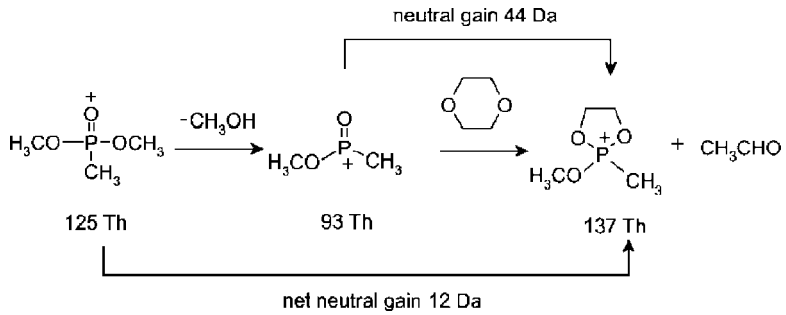
Figure 13. Product ion mass spectra for the reaction of the acylium ion $CH_2{=}CH{-}C^+{=}O$ of m/z 55 with two cyclic acetals. Adapted from Ref. 41.

Gas-phase derivatization

Previous gas-phase derivatization via ion/molecule reactions is also an interesting approach to improve the quality of structural information provided by ion dissociation.



Scheme 13



Scheme 14

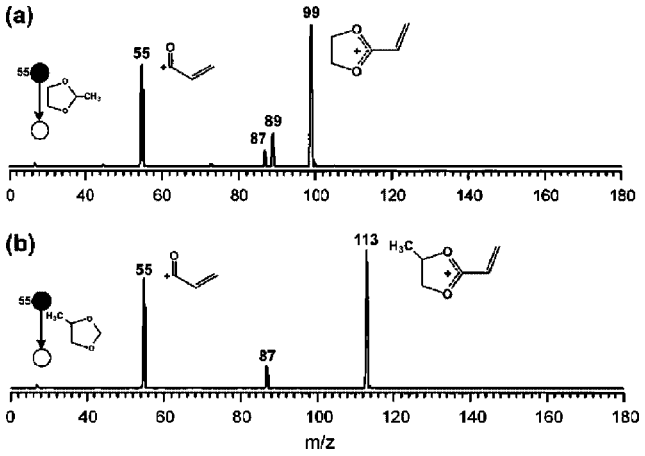


Figure 14. Product ion mass spectra for the reaction of the acylium ion $CH_2{=}CH{-}C^+{=}O$ of *m/z* 55 with two isomeric acetals. Adapted from Ref. 41.

We[45] showed for instance, for PyC_2H_5 isomers as a proof-of-principle set, that ionized methylene transfer occurs promptly upon reactions of the distonic ion $^{\bullet}CH_2{-}O{-}CH_2^+$, and that CID of the products distinguish easily the positional isomers. This distinction is possible owing to the strong structure/reactivity relationships that reveal the position of the ring substituent (Fig. 16). The *ortho* isomer was found to lose preferentially an ethylene molecule of 28 Da after H-shift to the methylene group (an *ortho* effect) to form by an ion of *m/z* 91, the *para* isomer to lose preferentially a methyl radical from the ethyl substituent (a *para* effect) to form by an ion of *m/z* 106, and the *meta* isomer to show a series of nearly equally favored dissociation pathways.

We have also demonstrated that ion/molecule reactions could be used to distinguish a set of isomeric ions that in turn would serve individually as diagnostic ions for a whole family of isomers. This interesting principle has been initially explored for isomeric mono-substituted alkyl pyridines using transacetalization-like reactions to locate the

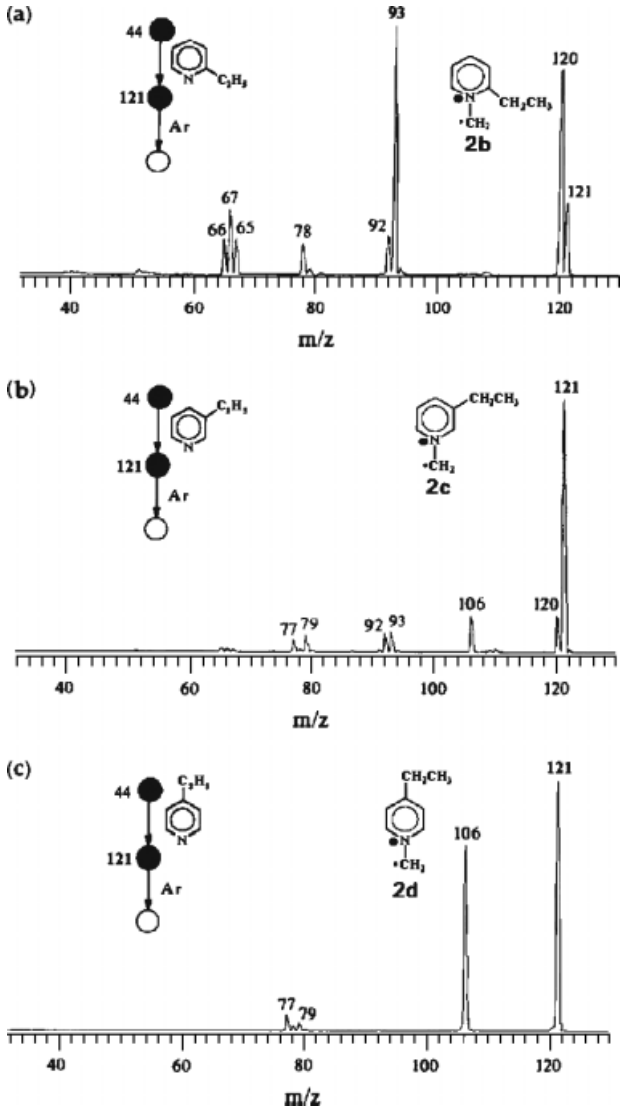


Figure 16. Sequential product ion mass spectra for the product ions of *m/z* 121 formed by $CH_2^{+\bullet}$ transfer from $^{\bullet}CH_2{-}O{-}CH_2^+$ to isomeric ethyl pyridines. Adapted from Ref. 45.

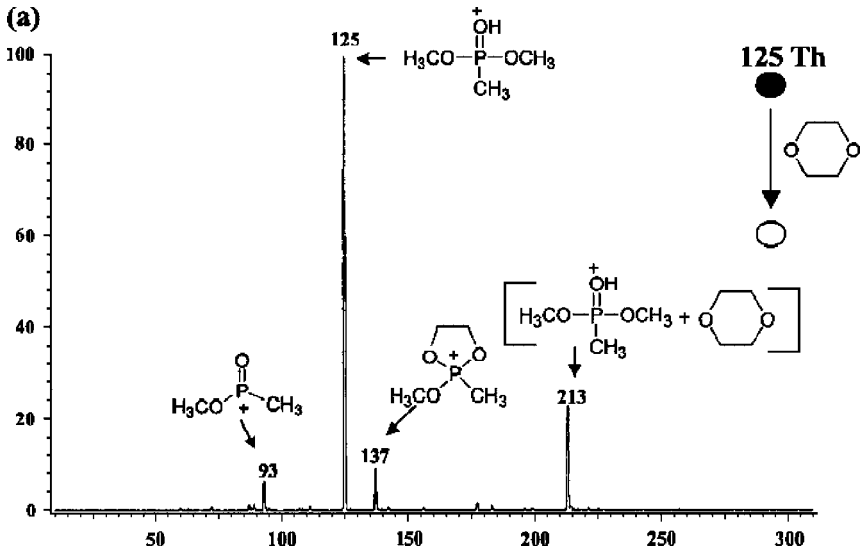
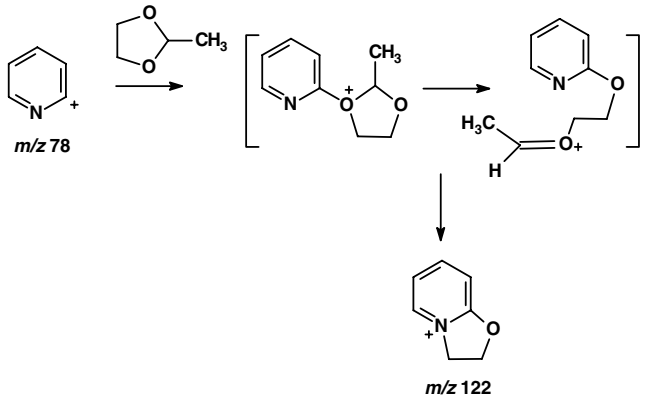


Figure 15. Product ion mass spectra for the reaction of the phosphonium ion $CH_3OP(O)CH_3^+$ of *m/z* 93 formed by methanol loss from protonated dimethyl methylphosphonate (DMMP) of *m/z* 125. Adapted from Ref. 44.

JMS

charge site (hence the position of the original substituent) of pyridyl cations either at the 2- or 3/4-positions (Fig. 17).[46] The *ortho* isomer of *m/z* 78 reacts promptly with 2-methyl dioxolane, likely via the mechanism depicted in Scheme 15, to form the product ion of *m/z* 122 to a great extent, whereas the *meta* and *para* isomers react nearly exclusively by proton transfer (*m/z* 89). This principle, using isomer-diagnostic ions has been expanded to other classes of compounds such as *ortho, meta*, or *para* isomers of acyl nitrobenzenes,[47] acyl and amidyl anilines and phenols[48] and monoalkyl pyrrols.[49]

Brodbelt and coworkers[50] have also demonstrated that ion/molecule reactions applied to a fragment ion rather than the intact protonated molecule can enhance structural characterization of specific target compounds. They used, for instance, a combined CID plus ion/molecule approach to perform the reaction with acetone of the acylium ion formed by dehydration of protonated nalidixic acid. This reaction forms a highly specific cyclic adduct ion favored by the participation of the neighboring carbonyl group (Scheme 16).



Scheme 15

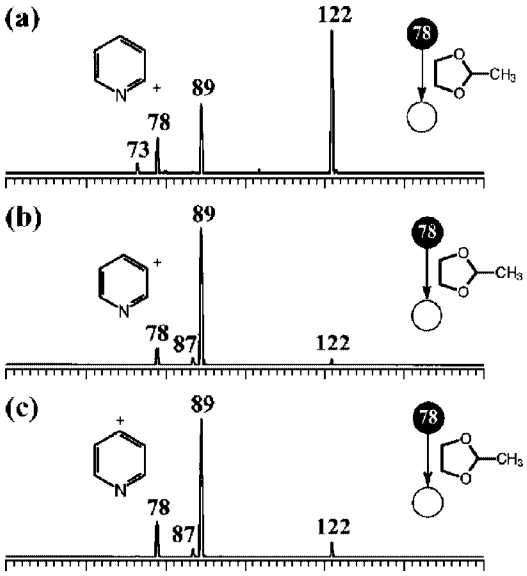


Figure 17. Product ion mass spectra for the reaction of isomeric pyridyl cations with 2-methyl 1,3-dioxolane. Adapted from Ref. 49.

Counting of functional groups

Kenttamaa and coworkers[51] described not only the identification but also the *counting* of hydroxy groups in protonated polyols via structurally selective reactions with neutral diethylmethoxyborane. The reaction occurs by the replacement of a proton with a diethylborenium ion wherein the hydroxy groups are derivatized leading to a characteristic series of product ions separated by 68 *m/z* units. Also, very interestingly, the increasing complexity and characteristic ratio of the ^{10}B and ^{11}B isotopologue ions can be used to confirm the number of B atoms incorporated and hence the number of functional OH groups (Fig. 18).

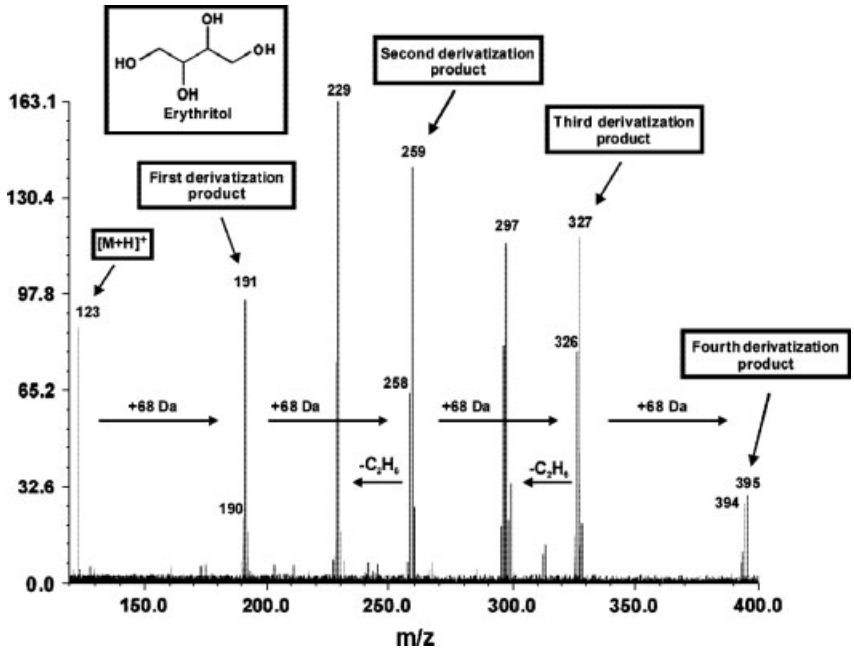
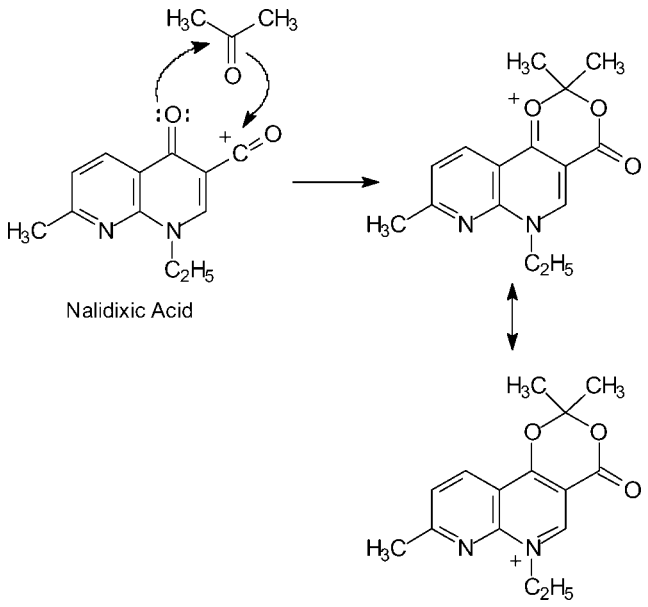


Figure 18. Product ion mass spectrum for the reaction of protonated erythritol (a tetrol) with diethylmethoxyborane. Adapted from Ref. 51.



Scheme 16

Multi-task reactions

Kenttamaa and coworkers[52] also demonstrated recently the use of selective ion/molecule reactions with diethylmethoxyborane to identify functional groups in protonated oxygen-containing monofunctional compounds including protonated alcohols, ethers, aldehydes and ketones, esters and carboxylic acids. Class-selective reactivity was observed for each class of compound; for instance, protonated carbonylic acids undergo two consecutive reactions of nucleophilic addition followed by a fast methanol loss (Fig. 19).

The reactions of the methoxymethylene cation $CH_3–O{=}CH_2^+$ have also been extensively explored by Brodbelt and coworkers[8] as functional-group selective for many classes of compounds including ethylene glycols and their monomethyl and dimethyl ethers and amino alcohols, including several biologically active molecules. Reactivity trends have been observed as a function of formal interfunctional distance, and two reactions normally compete: methylation $[M + 15]^+$ and methylene substitution $[M + 13]^+$. The analytical applications of these reactions have been extensively reviewed recently.[8] Flammang and coworkers[53] reported an interesting structurally diagnostic ion/molecule reaction with *t*-butyl nitrite capable of distinguishing isomeric pairs of ionized carbonyl compounds and enol ethers. The most relevant difference is that enol ions undergo a formal and structurally selective substitution of a hydrogen atom by nitric oxide.

Kenttamaa and coworkers[54] have extensively used the reaction with disulfides and diselenides to perform the challenging task of distinguishing distonic ions from conventional radical cations. This charged radical-selective reaction could be applied as diagnostic for dialkyl disulfides and diselenides, since in the course of reaction the S–S or Se–Se bonds break homolitically providing therefore information on both S- and Se-substituents (Fig. 20). Recently, they demonstrated that a similar functional group–selective reaction with dimethyl disulfide could be applied to screen for N-oxides via SCH_3 derivatization of the protonated molecule (Fig. 21).[55]

Nitroaromatics

In their search for efficient ways to detect all major explosives, Cooks and coworkers[56] observed an interesting class-selective de-nitration reaction of ionized benzonitrile with nitroaromatic compounds including TNT (Scheme 17). Note that the elimination of NO_2 (46 Da) is structurally diagnostic for the presence of an organic nitrate.

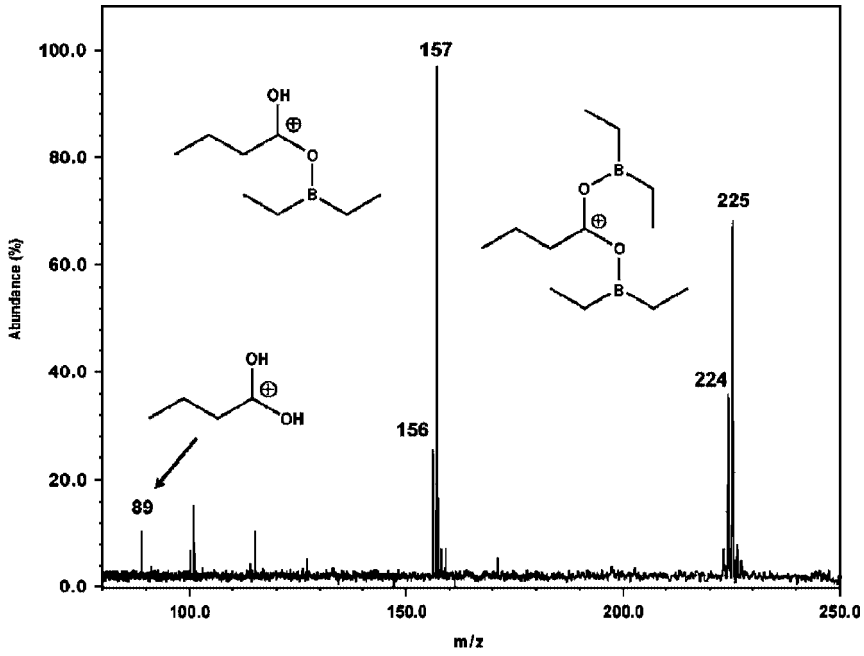


Figure 19. Product ion (MS^2) mass spectrum for the reaction of protonated butanoic acid with diethyl methoxyborane. Adapted from Ref. 39.

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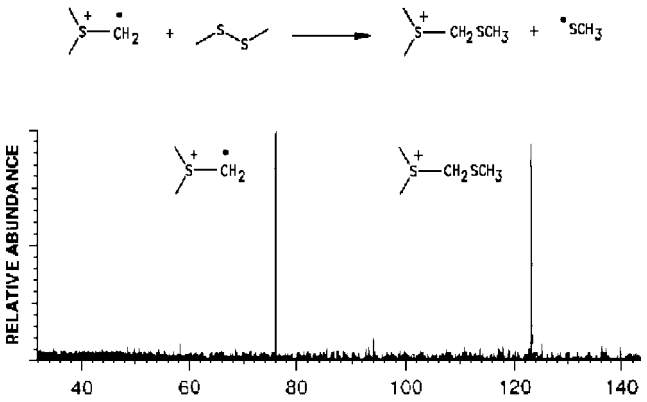


Figure 20. Product ion mass spectra for the reaction of the distonic ion $(CH_3)_2S^+-CH_2^\bullet$ with dimethyl disulfide. Adapted from Ref. 54.

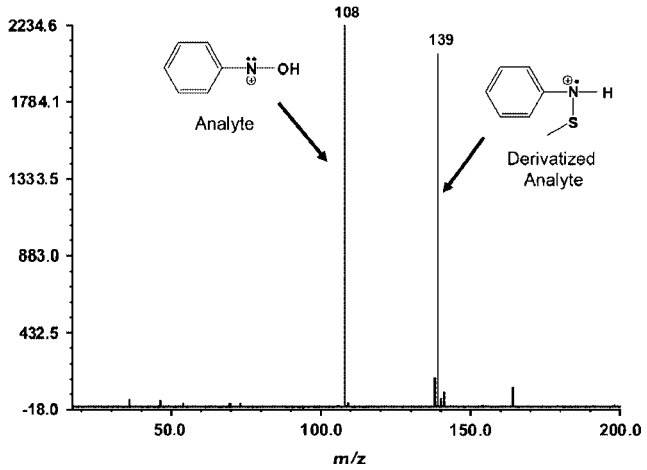


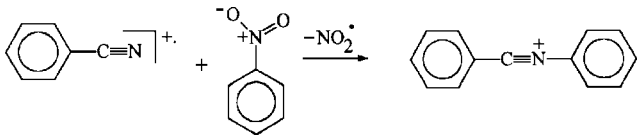
Figure 21. Product ion (MS^2) mass spectra for the reaction of protonated nitrosobenzene with dimethyl disulfide. Adapted from Ref. 55.

Borinium ions

Borinium ions such as $B(OCH_3)_2^+$ also have been used extensively in several structurally diagnostic ion/molecule reactions. For instance, Nibbering and coworkers[57] observed distinct behavior for several types of amines toward reactions with $B(OCH_3)_2^+$, whereas Brodbelt and coworkers[58] used this reaction to characterize several drugs.

Metal ions

Under CI conditions, gaseous metal ions have been extensively used in class-selective ion/molecule reactions,[5] but the use of mass-selected metal ions in tandem mass spectrometry for structural investigation is less common. Ni and Harrison,[59] for instance, performed reactions of gaseous Ti^+ with a set of C_5H_8 isomers that were hard to distinguish



Scheme 17

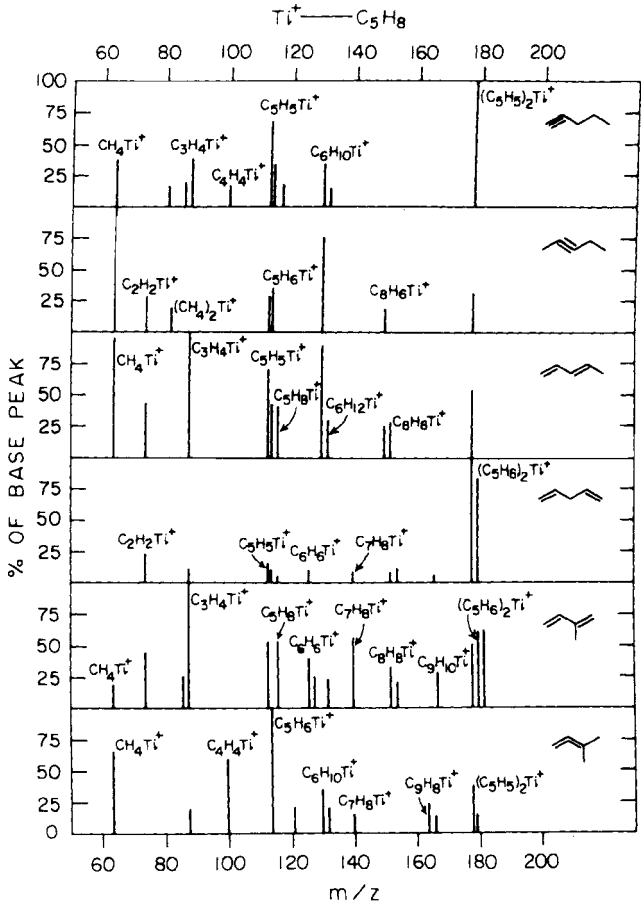


Figure 22. Product ion (MS^2) mass spectra for the reaction of Ti^+ with several C_5H_8 isomeric molecules. Adapted from Ref. 59.

by EI-MS, and observed very characteristic distribution of product ions (Fig. 22).

Proton transfer reactions

Proton transfer and hydrogen/hydride reactions have also been applied with great success for the investigation of several structural aspects, particularly for peptides and proteins,[14] but these reactions are often not useful when the goal is to identify class or functional groups.

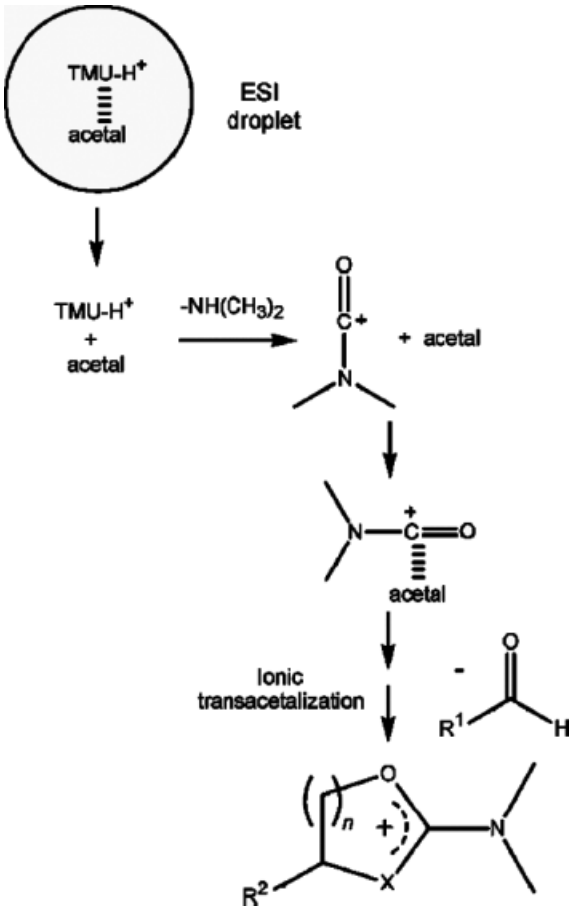
Enantiomers

Optical stereoisomers, the enantiomers, represent by far the greatest challenge for structural elucidation, but even such isomers can also be differentiated using gas-phase enantioselective reactions. This unique application of ion/molecule reactions has been reviewed recently.[16]

Atmospheric pressure ion/molecule reactions

Most gas-phase ion/molecule reactions are limited to neutral compounds that are sufficiently volatile and thermo-stable to be transferred to the gas phase by heating. Recently, we[60,61] showed that ion-molecule reactions can be performed under ESI or APCI conditions and that, under these conditions, the range of neutral molecules participating in structurally diagnostic ion/molecule reactions can be expanded toward those of lower volatility and thermal stability. With API

techniques, considerably more polar, heavier and therefore more complex molecules can be transferred to the gas phase environment in which in-source reactions occur. Scheme 18 illustrates the sequence of events proposed to lead to atmospheric-pressure polar transacetalization reactions under ESI using the ion-evaporation model.



Scheme 18

'On-surface' ion/molecule reactions under ambient pressure and temperature conditions

Very recently, Cooks and coworkers[62] introduced desorption electrospray ionization (DESI), a new method of desorption ionization. DESI is done by directing electrosprayed charged droplets and ions of solvent onto surfaces containing the analyte. The impact of the charged particles on the surface produces gaseous ions of the analyte, from light chemicals to proteins. Owing to these unique and compatible characteristics, DESI has been rapidly used to revolutionize the way ion/molecule reactions are performed. It offers a convenient means of creating a variety of reacting ions and reactions occurring under ambient temperature and pressure conditions and at the interface between the charged microdroplets and the solid surface bearing the condensed-phase analyte (including heavy, polar and nonvolatile molecules). Several structurally diagnostic DESI reactions have already been demonstrated, such as the formation of chloride and trifluoroacetate adducts of RDX and HMX or the Meisenheimer complex of TNT.[63] Similar to solution behavior, ambient stereoselective DESI reactions of the deprotonated molecule of phenylboronic acid (Scheme 19) have been

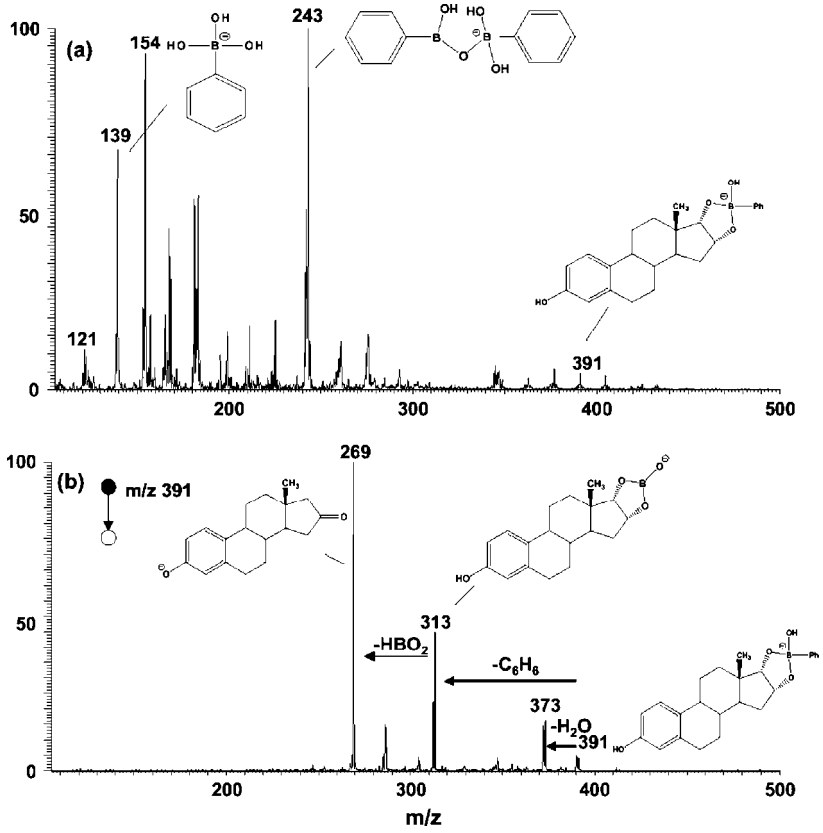
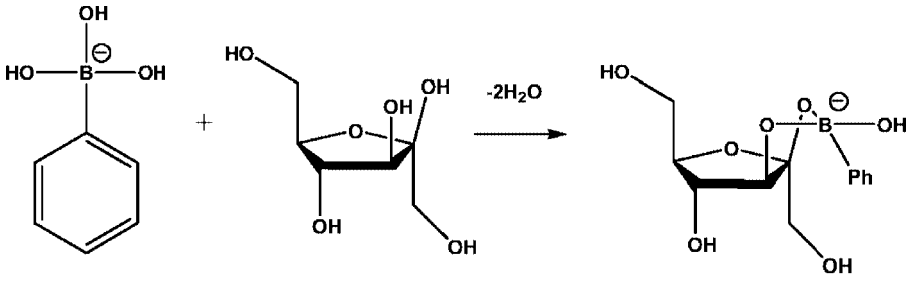


Figure 23. (a) DESI mass spectrum showing the products of on-surface ambient reactions of $PhB(OH)_3^-$ anion with *cis*-estriol; (b) Product ion mass spectrum of the reaction product ion of *cis*-estriol of *m/z* 391. Adapted from Ref. 64.



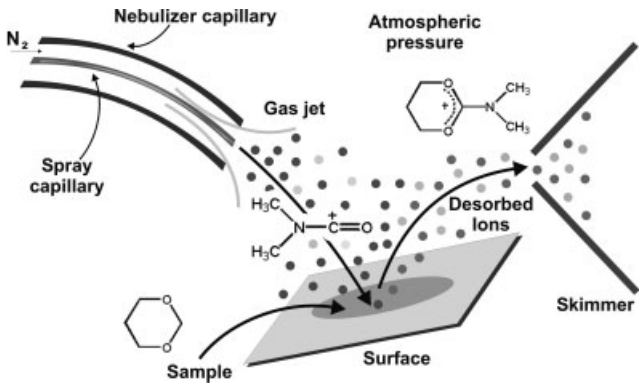
m/z 139 β-D-fructofuranose m/z 283

Scheme 19

recently shown to recognize *cis*-diol functionality for several biologically active molecules including aliphatic diols (carbohydrates and steroids) and aromatic diols (flavonols and catecholamines),[64] as Fig. 23 exemplifies for *cis*-estriol. Very recently, we[65] also showed that transacetalization of acylium ions and cyclic acetals (Scheme 15) occurs under ambient DESI conditions, as Scheme 20 schematically represents for 1,3-dioxane. Through DESI, *in situ* derivatization of the analyte on the surface offers a fast, sensitive and selective method of analyte detection. The possibility of using all API environments, most particularly that of DESI, will certainly greatly expand the scope of structurally diagnostic ion/molecule reactions.

CONCLUDING REMARKS

The combination of a vast arsenal of ionization techniques and multiple-stage MS instruments and scan modes provide access for mass spectrometrists to a world of gaseous ions, reaction environments and ion characterization methods. These versatile tools have allowed us to develop a great variety of structurally diagnostic ion/molecule reactions for class- and functional-group identification thus providing very refined MS information on chemical structures. These reactions, performed with increasing versatility in environments ranging from ultrahigh vacuum to on-surface ambient pressure and temperature conditions and with outstanding speed, selectivity and sensitivity that only mass spectrometry offers, are therefore likely to find increasing applications in many fields, particularly for unknown molecules of increasing complexity.



Scheme 20

Acknowledgements

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