Available online at www.sciencedirect.com
SCIENCE DIRECT®
Carbohydrate Research 340 (2005) 1567–1572
Carbohydrate RESEARCH

Note

Mass spectrometric decompositions of cationized β-cyclodextrin

Rafał Frański,[a,*] Błażej Gierczyk,[a] Grzegorz Schroeder,[a] Sebastian Beck,[b] Andreas Springer[b] and Michael Linscheid[b]

[a]*Adam Mickiewicz University, Faculty of Chemistry, Grunwaldzka 6, 60-780 Poznań, Poland*
[b]*Humboldt University Berlin, Department of Chemistry, Brook-Taylor-Str. 2, 12489 Berlin, Germany*

Received 18 November 2004; received in revised form 30 March 2005; accepted 31 March 2005
Available online 5 May 2005

Abstract—The mass spectrometric decompositions of β-cyclodextrin (β-CD) complexed with a number of common divalent metal cations (Mg, Ca, Cd, Cu, Co and Pb), obtained under electrospray ionization conditions, are reported. The main fragmentation pathways of $[\beta\text{-CD} + \text{Cat}]^{2+}$ ions studied (Cat stands for divalent cation) consist of consecutive losses of sugar units. The rupture of C–C bond in sugar units, which occurs via hydrogen atom transfer from the fragment ion formed to the eliminated species, was also observed. Isotope labelling consisting of the exchange of all hydroxyl hydrogens for deuteriums, has been applied in order to understand better the formation of fragment ions. It was found that C–H hydrogen transfer proceeds only during fragmentation across C–C bonds.

Keywords: β-Cyclodextrin; Metal cation complexes; Fragmentation pathways

Mass spectrometric fragmentation pathways, especially under electrospray ionization (ESI) conditions, of mono- and oligosaccharides ionized by metal cations are of importance to researchers involved in carbohydrate studies, mainly from the point of view of saccharide structure analysis and the interesting gas-phase ion chemistry of these processes. Saccharides cationized by alkali and alkali earth metals,[1–8] transition metals[8–14] and even by lead[15] have been extensively studied. Gas-phase chemistry of metal cationized saccharides has been shown to be a powerful tool for linkage position determination. On the other hand, there are few data on mass spectrometry of metal cationized cyclic saccharides (for example cyclodextrins). Madhusudanan has studied fragmentation pathways of lithiated α-, β- and γ-cyclodextrin.[16] Cai et al. have shown that iron(II) or magnesium cations play an important role in holding the toluene molecule inside β-cyclodextrin cavity.[17] Mass spectrometric fragmentation pathways of protonated, deprotonated and sodiated cyclodextrins (generated by different ionization methods) or their derivatives have been already investigated.[18–23] Many papers were also devoted to the mass spectrometry of cyclodextrin complexes with organic compounds.[24–34] However, to the best of our knowledge, the mass spectrometric fragmentation pathways of cyclodextrins cationized by divalent metals have not been reported yet. Recently, Kurokawa et al. have obtained a β-cyclodextrin–$CuCl_2$ complex in condensed phase.[35] This paper reports the mass spectrometric decompositions of β-cyclodextrin (β-CD) complexed with a number of common metal cations, obtained under electrospray ionization conditions (so in the gas-phase).

The results obtained by performing the MS/MS experiments for cationized β-CD are summarized in Table 1 and MS/MS spectra of $[\beta\text{-CD} + \text{Mg}]^{2+}$ (*m*/*z* 579) and $[\beta\text{-CD} + \text{Cd}]^{2+}$ (*m*/*z* 624) ions are shown in Figure 1 as representative examples. Decomposition of $[\beta\text{-CD} + \text{Na}]^{+}$ ion consists of consecutive losses of mass 162. Analogous fragmentation pathway has been observed for $[\alpha\text{-, }\beta\text{-, }\gamma\text{-CD} + \text{Li}]^{+}$ ions.[16] This process can occur either according to the formation of Y- or Z-type ions

* Corresponding author. Tel.: +48 61 8291245; fax: +48 61 8658008; e-mail: franski@main.amu.edu.pl

0008-6215/$ - see front matter © 2005 Elsevier Ltd. All rights reserved.
doi:10.1016/j.carres.2005.03.014

Table 1. Results of MS/MS experiments performed for β-cyclodextrin ionized by metal cations

Parent ion *m/z* (% ri)	Detected daughter ions *m/z* (% ri)
$[\beta\text{-CD} + Na]^+$ 1157 (100)	995 (15), 833 (30), 671 (40), 509 (50) 347 (20)
$[\beta\text{-CD} + 2Na]^{2+}$ 590 (25)	1157 (100), 995 (3), 833 (4), 671 (5), 509 (6), 347 (2)
$[\beta\text{-CD} + Mg]^{2+}$ 579 (30)	Series A 498 (25), 417 (70), 336 (100), 255 (70) Series B 995 (2), 833 (4), 671 (15), 509 (60), 347 (10) Series C 731 (3), 569 (4), 407 (20) Series D 815 (0.5), 653 (1), 491 (15), 329 (10)
$[\beta\text{-CD} + Ca]^{2+}$ 587 (25)	Series A 506 (20), 425 (45), 344 (100), 263 (100) Series B 1011 (2), 849 (4), 687 (6), 525 (8) 363 (9) Series C 747 (5), 585 (10), 423 (10)
$[\beta\text{-CD} + Cd]^{2+}$ 624 (30)	Series A 543 (35), 462 (80), 381 (80), 300 (100) Series B 1085 (15), 923 (25), 761 (40), 599 (80), 437 (40) Series C 821 (10), 659 (20), 497 (25), 335 (10) Series D 905 (0.5), 743 (2) 581 (10), 419 (10)
$[\beta\text{-CD(D)} + Cd]^{2+}$ 634.5 (50)	Series A 552 (50), 469.5 (95), 387 (100), 304.5 (90) Series B 1102 (15), 937 (20), 772 (35), 607 (70), 442 (40) Series C 833 (10), 668 (20), 503 (20), 338 (10) Series D 917 (1), 752 (2), 587 (10), 422 (10)
$[\beta\text{-CD} + Co]^{2+}$ 596.5 (20)	Series A 515.5 (15), 434.5 (25), 353.5 (15), 272.5 (5) Series B 1030 (10), 868 (20), 706 (40), 544 (100), 382 (5) Series C 766 (0.5), 604 (1), 442 (2), 280 (3)
$[\beta\text{-CD} + Cu]^{2+}$ 598.5 (70)	Series A 517.5 (45), 436.5 (40), 355.5 (15) Series B 1034 (10), 872 (40), 710 (55), 548 (100), 386 (5) 649 (5), 487 (15), 325 (85)
$[\beta\text{-CD} + Pb]^{2+}$ 671 (30)	Series A 590 (20), 509 (25), 428 (10) Series B 1179 (25), 1017 (55), 855 (65), 693 (100), 531 (75), 369 (40) Series C 915 (1.5) 753 (2), 591 (6), 429 (3)

(Scheme 1).[36] The former ions are considerably more common than the latter ones, thus we believe that the loss of mass 162 proceeds according to the Y-type ion formation. It is worth adding that internal monosaccharide residue loss can occur for protonated linear oligosaccharides but it does not proceed for sodium cationized ones.[37,38] β-Cyclodextrin cationized by two sodium cations ($[\beta\text{-CD} + 2Na]^{2+}$ ion at *m/z* 590) loses Na^+ which leads to the $[\beta\text{-CD} + Na]^+$ ion (*m/z* 1157) and then consecutive losses of mass 162 are observed (Table 1).

For linear oligosaccharides, besides doubly charged $[M + Cat]^{2+}$ ions, singly charged $[M-H + Cat]^+$ or $[M + CatX]^+$ ions (Cat stands for divalent cation, X stands for counter ion) usually form under ESI conditions.[7–14] On the other hand, the dominant species formed from chloride salts are $[M + Cat]^{2+}$ ions.[7,8] We found the same dominant species for cyclodextrin and we studied these ions here, since we were interested in the determination of mass spectrometric fragmentation pathways of cationized cyclodextrin unaffected by counter ion or negatively charged sites formed by deprotonation.

Decomposition of each of the $[\beta\text{-CD} + Cat]^{2+}$ ions studied proceeds in two directions. The first one consists of consecutive losses of mass 81 (Table 1, series A). This fragmentation pathway is analogous to that described above for singly charged $[\beta\text{-CD} + Na]^+$ ions, that is, it consists of consecutive losses of mass 162 since 162/2 = 81. The second direction (series B) can be regarded as the so-called charge separation reaction.[39–41] The loss of protonated 162 species (*m/z* 163) leads to singly charged ions, for example, *m/z* 995 for $[\beta\text{-CD} + Mg]^{2+}$ or *m/z* 1030 for $[\beta\text{-CD} + Co]^{2+}$ ion (Table 1), and this is followed by consecutive losses of mass 162. There are also series C and D consisting of rather low abundant ions, not observed for each of the $[\beta\text{-CD} + Cat]^{2+}$ ions studied. The formation of ions of series C can be rationalized assuming that together with the loss of protonated 162 species (*m/z* 163) the loss of mass 264 (e.g., for $[\beta\text{-CD} + Mg]^{2+}$ ion 995 − 264 = 731; for $[\beta\text{-CD} + Cd]^{2+}$ ion 1085 − 264 = 821) takes place, followed by consecutive losses of mass 162 (Fig. 1). The loss of mass 264 occurs via hydrogen atom transfer from the fragment ion formed to the eliminated species (Scheme 1). Analogous fragmentation pathway has been observed for lithiated cyclodextrins.[16] For $[\beta\text{-CD} + Mg]^{2+}$ and $[\beta\text{-CD} + Cd]^{2+}$ ions there are also fragment ions (series D) which can be rationalized as ions of series B deprived of water molecules.

In order to understand better the formation of fragment ions of series A–D, isotope labelling has been applied. Deuterium labelling has been found to be useful in mass spectrometric studies of sugars and their conju-

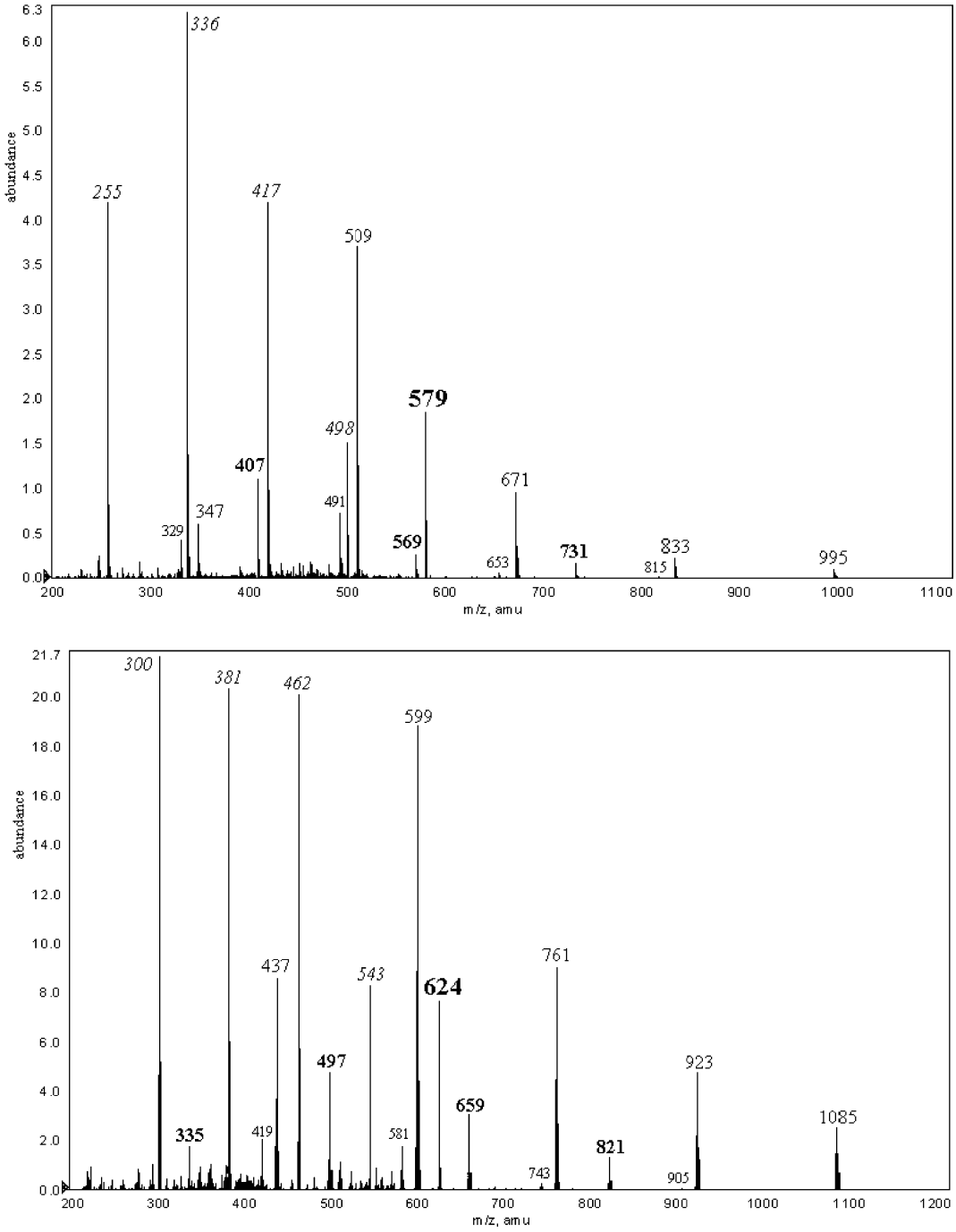
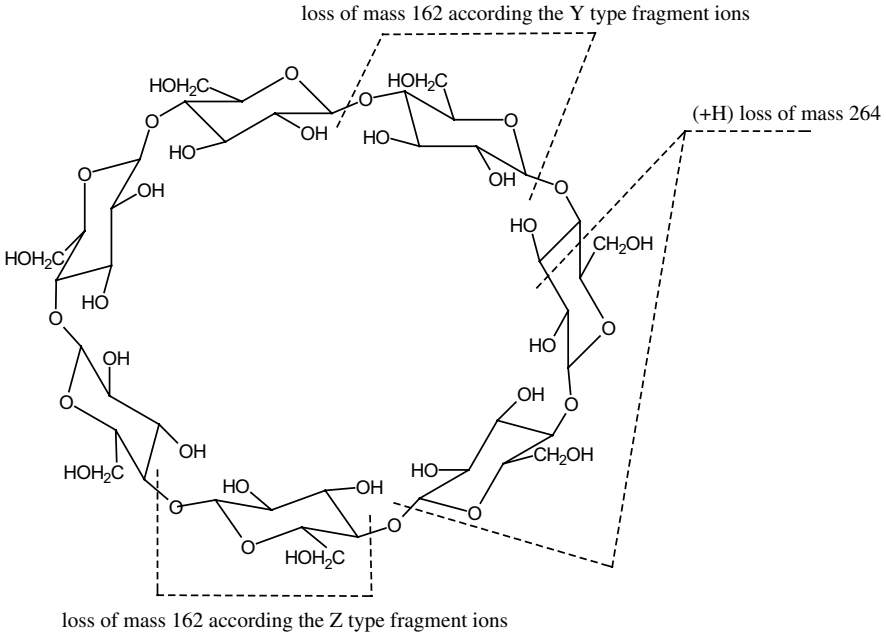


Figure 1. MS/MS spectra of $[\beta\text{-CD} + Mg]^{2+}$ (*m*/*z* 579) and $[\beta\text{-CD} + Cd]^{2+}$ (*m*/*z* 624) ions.

gates.[42–45] For $[\beta\text{-CD} + Cd]^{2+}$ ion all the above series of fragment ions are observed with reasonable abundances, therefore ESI mass spectrum of CH_3OD/D_2O (2/1) solution containing β-cyclodextrin and $CdCl_2$ was recorded (Fig. 2). The $[\beta\text{-CD(D)} + Cd]^{2+}$ ion at *m*/*z* 634.5 has all hydroxyl hydrogens exchanged for deuteriums. Instead of the losses of mass 81 (series A), those of mass 82.5 are observed since all three hydroxyl hydrogens are exchanged for deuteriums (165/2 = 82.5). This means that during this process there is no intramolecular H/D exchange between the hydroxyl and C–H hydrogen atoms. The *m*/*z* values of series B ions indicate that the C–H hydrogen atoms are also not involved in the process of series B ion formation. Consecutive losses of mass 165 occur and the fragment ion of the highest *m*/*z* value at *m*/*z* 1102 is formed by the loss of an ion at *m*/*z* 167. The formation of $[165 + D]^+$ ion takes place in the case of $[\beta\text{-CD(D)} + Cd]^{2+}$ ion at *m*/*z* 634.5, whereas $[162 + H]^+$ ion appears in the case of unlabelled $[\beta\text{-CD} + Cd]^{2+}$ ion. The formation of series C species



Scheme 1.

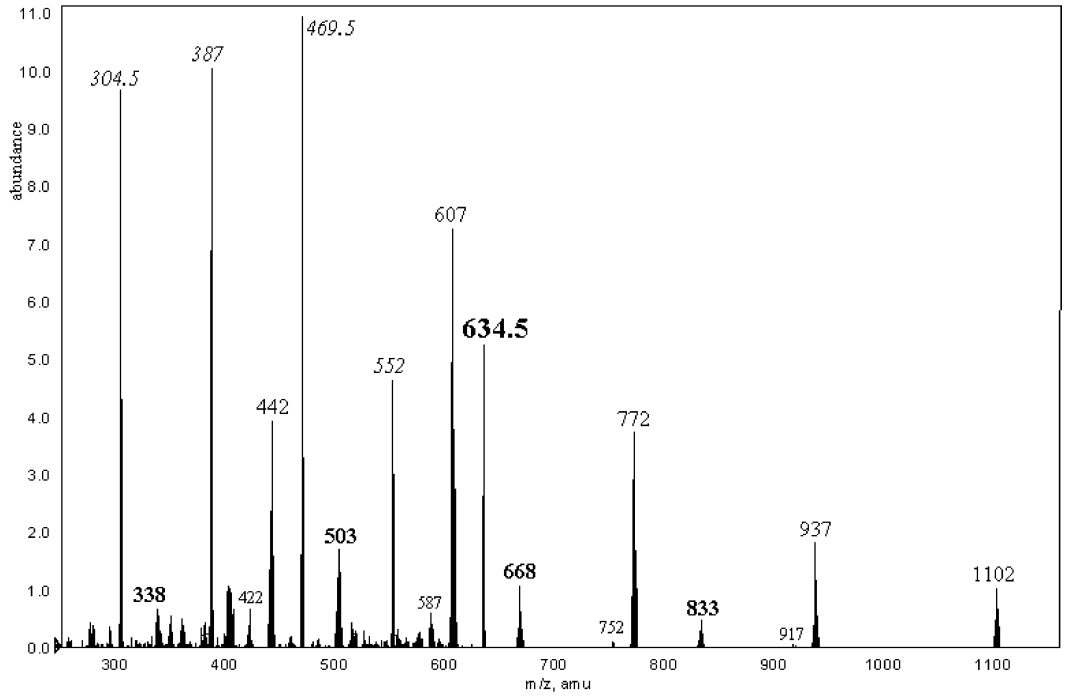


Figure 2. MS/MS spectrum of $[\beta\text{-CD(D)} + Cd]^{2+}$ ion at *m*/*z* 634.5.

from $[\beta\text{-CD(D)} + Cd]^{2+}$ ion proceeds via the loss of mass 269 instead of 264 as it is for unlabelled $[\beta\text{-CD} + Cd]^{2+}$ ion. This means that the eliminated species has five hydrogen atoms exchanged for deuterium ones, so all these hydrogen atoms exchanged originate from hydroxyl groups (Scheme 1). However, as mentioned above, the loss of mass 264 proceeds via hydrogen atom transfer from the fragment ion formed to the eliminated species (Scheme 1). Therefore, the conclusion can be drawn that in the above process we deal with C–H hydrogen transfer. Complexation of metal cation can activate the C–H bond to form organometallic species.[46] Series D can be rationalized as ions of series B deprived of water molecules. For $[\beta\text{-CD(D)} + Cd]^{2+}$ ion the D

series ions can be interpreted as the B series ones deprived of D_2O, thus also in this process C–H hydrogen atoms are not involved.

Only in the MS/MS spectrum of $[\beta\text{-CD} + Cu]^{2+}$ ion fragment ions which do not contain metal cation are observed (*m*/*z* 649, 487 and 325) and it is difficult to explain the reason for this. These ions can be rationalized as $[(162)_n + H]^+$ ions and can be regarded as complementary partners of series B ions. In other words, by charge separation reaction of $[\beta\text{-CD} + Cu]^{2+}$ ion (*m*/*z* 598.5) the following pairs of fragment ions are formed: *m*/*z* 649 and 548, 487 and 710 and 325 and 872.

1. Experimental

Electrospray ionization (ESI) mass spectrometric experiments were carried out on a Qstar mass spectrometer (Applied Biosystems, Darmstadt, Germany, hybrid QqTOF instrument). The β-cyclodextrin and a respective salt (NaCl, $MgCl_2$, $CaCl_2$, $CdCl_2$, $CoCl_2$, $CuCl_2$, $Pb(CH_3COO)_2$) were dissolved in methanol/water (2/1) at a concentration of about 10^{-5} M and the solutions were directly infused into the ESI source with flow rate of 10 μl min^{-1}. The MS/MS experiments were performed with collision energies 25–35 V to achieve in reasonable abundance both parent and daughter ions. The used collision gas was nitrogen of >99.5% purity. The pressure parameter of the collision gas was set at 4 which resulted in the pressure of about 3.3×10^{-5} Torr in the collision cell and was maintained at this value for all fragmentation experiments.

References

1. Quéméner, B.; Désiré, C.; Debrauwer, L.; Rathahao, E. *J. Chromatogr. A* **2003**, *984*, 185–194.
2. An, J.; Zhang, L.; O'Neill, M. A.; Albersheim, P.; Darwill, A. G. *Carbohydr. Res.* **1994**, *264*, 83–96.
3. Lee, S.; Wyttenbach, T.; Bowers, M. T. *Int. J. Mass Spectrom. Ion Process.* **1997**, *167/168*, 605–614.
4. Penn, S. G.; Cancilla, M. T.; Lebrilla, C. B. *Int. J. Mass Spectrom.* **2000**, *195/196*, 259–269.
5. Striegel, A. M.; Timpa, J. D.; Piotrowiak, P.; Cole, R. B. *Int. J. Mass Spectrom. Ion Process.* **1997**, *162*, 45–53.
6. Xie, Y.; Lebrilla, C. B. *Anal. Chem.* **2003**, *75*, 1590–1598.
7. Zaia, J. *Mass Spectrom. Rev.* **2004**, *23*, 161–227.
8. Harvey, D. J. *J. Am. Soc. Mass Spectrom.* **2001**, *12*, 926–931.
9. König, S.; Leary, J. A. *J. Am. Soc. Mass Spectrom.* **1998**, *9*, 1125–1134.
10. Sible, E. M.; Brimmer, S. P.; Leary, J. A. *J. Am. Soc. Mass Spectrom.* **1997**, *8*, 32–42.
11. Gaucher, S. P.; Leary, J. A. *Int. J. Mass Spectrom.* **2000**, *197*, 139–148.
12. Shahgholi, M.; Callahan, J. H.; Rappoli, B. J.; Rowley, D. A. *J. Mass Spectrom.* **1997**, *32*, 1080–1093.
13. Carlesso, V.; Fournier, F.; Tabet, J. C. *Eur. J. Mass Spectrom* . **2000**, *6*, 421–428.
14. Carlesso, V.; Fournier, F.; Afonso, C.; Tabet, J. C. *Eur. J. Mass Spectrom* . **2001**, *7*, 331–341.
15. Salpin, J. Y.; Tortajada, J. *J. Mass Spectrom.* **2002**, *37*, 379–388.
16. Madhusudanan, K. P. *J. Mass Spectrom.* **2003**, *38*, 409–416.
17. Cai, Y.; Tarr, M. A.; Xu, G.; Yalcin, T.; Cole, R. B. *J. Am. Soc. Mass Spectrom.* **2003**, *14*, 449–459.
18. Sforza, S.; Galaverna, G.; Corradini, R.; Dossena, A.; Marchelli, R. *J. Am. Soc. Mass Spectrom.* **2003**, *14*, 124–135.
19. Carroll, J. A.; Ngoka, L.; Beggs, C. G.; Lebrilla, C. B. *Anal. Chem.* **1993**, *65*, 1582–1587.
20. Metzger, J. W.; Jung, M.; Schmalzing, D.; Bayer, E.; Schurig, V. *Carbohydr. Res.* **1991**, *222*, 23–35.
21. Chankvetadze, B.; Endresz, G.; Blaschke, G.; Juza, M.; Jakubetz, H.; Schurig, V. *Carbohydr. Res.* **1996**, *287*, 139–155.
22. Bashir, S.; Giannakopulos, A. E.; Derrick, P. J.; Critchley, P.; Bottrill, A.; Padley, H. D. *Eur. J. Mass Spectrom.* **2004**, *10*, 109–120.
23. Yamagaki, T.; Nakanishi, H. *Rapid Commun. Mass Spectrom.* **1999**, *13*, 2199–2203.
24. Guo, M.; Song, F.; Liu, Z.; Liu, S. *J. Mass Spectrom.* **2004**, *39*, 594–599.
25. Sun, W.; Cui, M.; Liu, S.; Song, F.; Elkin, Y. N. *Rapid Commun. Mass Spectrom.* **1998**, *12*, 2016–2022.
26. Lamcharfi, E.; Chuilon, S.; Kerbal, A.; Kunesch, G.; Libot, F.; Virelizier, H. *J. Mass Spectrom.* **1996**, *31*, 982–986.
27. Guo, M.; Zhang, S.; Song, F.; Wang, D.; Liu, Z.; Liu, S. *J. Mass Spectrom.* **2003**, *38*, 723–731.
28. Srinivasan, K.; Bartlett, M. G. *Rapid Commun. Mass Spectrom.* **2000**, *14*, 624–632.
29. Cheng, Y.; Hercules, D. M. *J. Mass Spectrom.* **2001**, *36*, 834–836.
30. Dotsikas, Y.; Loukas, Y. L. *J. Am. Soc. Mass Spectrom.* **2003**, *14*, 1123–1129.
31. Kobeti, R.; Jursic, B. S.; Bonnette, S.; Jane, S.-C.; Tsai, J. S.-C.; Salvatore, S. J. *Tetrahedron Lett.* **2001**, *42*, 6077–6082.
32. Gabelica, V.; Galic, N.; De Pauw, E. *J. Am. Soc. Mass Spectrom.* **2002**, *13*, 946–953.
33. Vrkic, A. K.; O'Hair, A. J.; Lebrilla, C. B. *Eur. J. Mass Spectrom.* **2003**, *9*, 563–577.
34. Mele, A.; Panzeri, W.; Selva, A.; Mauri, P. *Eur. J. Mass Spectrom.* **2000**, *6*, 169–174.
35. Kurokawa, G.; Sekii, M.; Ishida, T.; Nogami, T. *Supramol. Chem.* **2004**, *16*, 381–384.
36. Domon, B.; Costello, C. A. *Glycoconjugate J.* **1988**, *5*, 397–409.
37. Kovácik, V.; Pätoprstý, V. *Eur. J. Mass Spectrom.* **1998**, *4*, 417–420.
38. Brüll, L. P.; Kovácik, V.; Thomas-Oates, J. E.; Heerma, W.; Haverkamp, J. *Rapid Commun. Mass Spectrom.* **1998**, *12*, 1520–1532.
39. Lyapchenko, N.; Frański, R.; Schroeder, G.; Kozik, T.; Shvaika, O. P.; Kiselyov, V.; Korotkikh, N. I. *Int. J. Mass Spectrom.* **2003**, *228*, 61–68.
40. Frański, R.; Szymański, A.; Frańska, M.; Gierczyk, B. *Tens. Surf. Det.* **2003**, *40*, 7–9.
41. Kebarle, P. *J. Mass Spectrom.* **2000**, *35*, 804–817.
42. Hofmeister, G. E.; Zhou, Z.; Leary, J. A. *J. Am. Chem. Soc.* **1991**, *113*, 5964–5970.

43. Ma, Y.-L.; Vedernikova, I.; Van den Heuvel, H.; Claeys, M. *J. Am. Soc. Mass Spectrom.* **2000**, *11*, 136–144.
44. Kellersberger, K. A.; Dejsupa, C.; Liang, Y.; Pope, R. M.; Dearden, D. V. *Int. J. Mass Spectrom.* **1999**, *193*, 181–195.
45. Bravo, P.; Viani, F.; Favretto, D.; Traldi, P. *Rapid Commun. Mass Spectrom.* **1996**, *10*, 1103–1106.
46. Wee, S.; O'Hair, R. A. J.; McFadyen, W. D. *Rapid Commun. Mass Spectrom.* **2004**, *18*, 1221–1226.