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Research Article

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Collision Induced Dissociation of Complexes of Isomeric Heptylamines with Crown Ethers

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ABSTRACT

The complexes of protonated isomeric primary heptylamines (C_7H_{15}-NH_2) with crown ethers have been studied by using electrospray ionization-collision induced dissociation-tandem mass spectrometry (ESI-CID-MS/MS). It has been found that besides the gas phase basicity, small structural changes in the amines may also lead to changes in the relative gas phase stabilities of the complexes. For example, 2-aminoheptane and 1,3-dimethylamylamine (the heptylamines that are on the list of the prohibited substances of the World Anti-Doping Agency) form complexes with DB24C8 of remarkably different relative gas phase stabilities. The H/D exchange in the complexes of protonated isomeric heptylamines with crown ethers may also lead to the visible differences in the relative gas phase stabilities.

Keywords: Amine; Crown ether; Electrospray ionization; Tandem mass spectrometry; 1,3-dimethylamylamine; Tuaminoheptane

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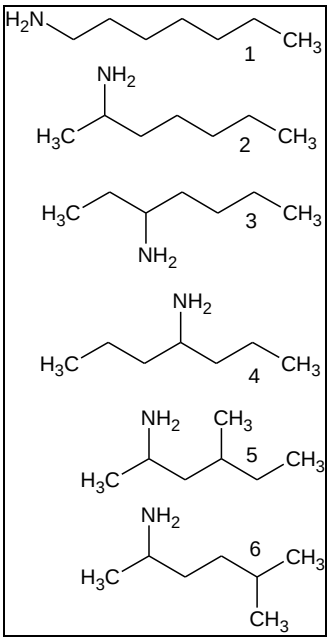
INTRODUCTION

Although crown ethers are the best known for their ability to form complexes with metal cations, their complexes with ammonium cation and its conjugates are also interesting. One of the techniques widely used to the study such complexes is the electrospray ionization mass spectrometry (ESI-MS). There are a number of papers devoted to the ESI-MS complexes of protonated amino acids with chiral crown ether conjugates (chiral recognition) [1-4]. The complexes of crown ethers with protonated peptides have been studied by ESI-MS [5-7]. Obviously, the complexes between crown ether and ammonium cation and the complexes between crown ether and protonated amines have been also studied in details [8-16], and sometimes such complexes are quite large.[17] By using ion mobility spectrometry, the formation of complexes between crown ether and protonated amines may be useful for differentiation of isomeric amines [12,13].

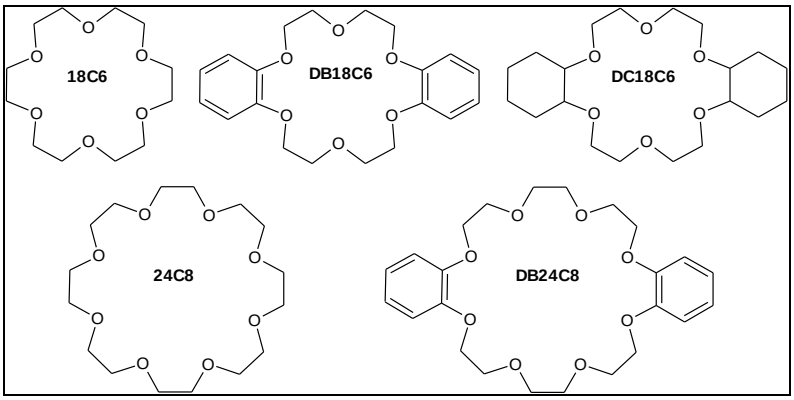
Besides the practical application of the complex formation between crown ether and protonated amines, the factors influencing the relative stabilities of the complexes have been discussed in details (e.g. gas phase basicity of the amines) [10-15]. From the scientific point of view it seems to be the most important topic. Sometimes, subtle structural changes in the amines may lead to the remarkable changes in the complex stabilities [12].

At this paper we asked the question if the crown ether complexes with isomeric heptylamines (C_7H_{15}-NH_2) have different gas phase stabilities. In order to answer this question we obtained the collision induced dissociation mass spectra (CID-MS/MS spectra, in other words product ion spectra) of the complexes. ESI was used for gas phase ion generation. The structures of heptylamines (1-7) are shown in Scheme 1 and those of crown ethers are shown in Scheme 2. The complexes of 1 with crown ethers have been already studied in details by molecular modeling and

for the complexes the values of $E_{1/2}$ (point at which half of the complexes dissociate) have been determined [10]. Taking into account the size of the crown ether cavity, 18C6 is appropriate to form complexes with protonated primary amines [16]. Therefore we decided to use 18C6 and its two common conjugates (DB18C6 and DC18C6). Investigation of larger crown ethers (24C8 and DB24C8) also seems to be reasonable [17,18]. It has to be stressed that haptylamines 2 and 5 (tuaminoheptane and 1,3-dimethylamylamine - DMAA) are on the list of the prohibited substances of the World Anti-Doping Agency. Obviously, mass spectrometric techniques have been used for their analysis [19-22]. DMAA is especially problematic with respect to its effect on the organism [23,24] and its origin in the food supplements [25-31].



Scheme 1: Structures of the heptylamines (1-7)



Scheme 2: Structures of the crown ethers

EXPERIMENTAL SECTION

Amines and crown ethers were obtained from Sigma-Aldrich (Poznań, Poland) and used without purification. ESI-CID-MS/MS spectra were taken on a Waters/Micromass (Manchester, UK) Q-tof Premier mass spectrometer (software MassLynx V4.1, Manchester, UK) equipped with an electrospray ion source (ESI). The sample solutions were infused into the ESI source by a syringe pump at the flow rate of 10 μl/min. The electrospray voltage was 2.7 kV and the cone voltage - 30 V. The source temperature was 80^{o}C and the desolvation temperature was 250^{o}C. Nitrogen was used as the cone gas and desolvating gas at the flow-rates of 50 and 300 dm^3/h, respectively. Argon was used as a collision gas at the flow-rate 0.5 ml/min in the T-wave collision cell. This flow rate resulted in the

collision cell pressure of 0.3 Pa. The applied collision energy (laboratory frame, E_{lab}), the most important parameter for CID-MS/MS experiments, was 7-25 eV. In order to obtain the respective break down plots for the purpose of this work, the collision energy values 10, 12, 15, 17 and 20 eV were used. Scan time was 0.5 s, inter scan time was 0.1 s and run duration was 6 s. Thus, as can be easy calculated, each mass spectrum (and also each point at the break down plots) was generated by averaging ten mass spectra. The calculated relative standard deviations, for the peak intensities, did not exceeded 5 %.

In this work we analyzed thirty complexes (plus three isotope labeled). For each complex at least five ESI-CID-MS/MS spectra were recorded (it took a few days). It is of key importance that all complexes were analyzed under identical conditions, especially with respect to the pressures inside the mass spectrometer (collision cell 0.3 Pa, quadrupole analyzer 2.8×10^{-3} Pa, tof analyzer 9.7×10^{-5} Pa). Therefore, the pressures were systematically controlled throughout the experiment. Furthermore, each day the ESI-CID-MS/MS spectrum of a selected complex was recorded and confronted with the one recorded on the previous day. The sample solutions were prepared in CH_3CN/H_2O (3/1) with crown ether concentrations and amine concentrations of 10^{-5} mol/dm^3 (the concentrations refer to each ingredient, not to their sum). In order to generate the isotope labeled complexes the mixture CH_3CN/D_2O (3/1) was used as a solvent. In this solvent all hydrogen atoms bonded to the nitrogen atom were immediately exchanged into deuterium. Thus, the masses of the labeled complexes were by 3 units higher than the masses of non-labeled correspondents. By using pure methanol or methanol/water as solvent the abundant complexes of interest were also generated. However, CH_3OD (initially used to generate the isotope labeled complexes) contained relatively high amount of sodium and potassium as contamination. Formation of the abundant sodium and potassium complexes with crown ethers affected the formation of abundant complexes of amines with crown ethers. Therefore, in order to analyze the isotope labeled and non-labeled complexes at identical conditions, acetonitrile/water was used as a solvent, although the solution conditions seemed to be of minor importance for the purpose of this work.

Special care was also taken to make sure if the amine used for the experiments just finished, had been removed from the mass spectrometer (compounds which have high ESI response may be sometimes detected for a relatively long time, even though they are no longer introduced into ESI source). For the purpose of this study it was of crucial importance that the complex subjected to the CID-MS/MS experiment was pure (not a mixture of isomers).

RESULTS AND DISCUSSIONS

As expected upon CID conditions, each of the complexes was dissociated into a neutral crown ether molecule and a protonated amine. Figure 1 shows the CID-MS/MS spectrum (product ion spectrum) of $[5+H+24C8]^+$ as a representative example.

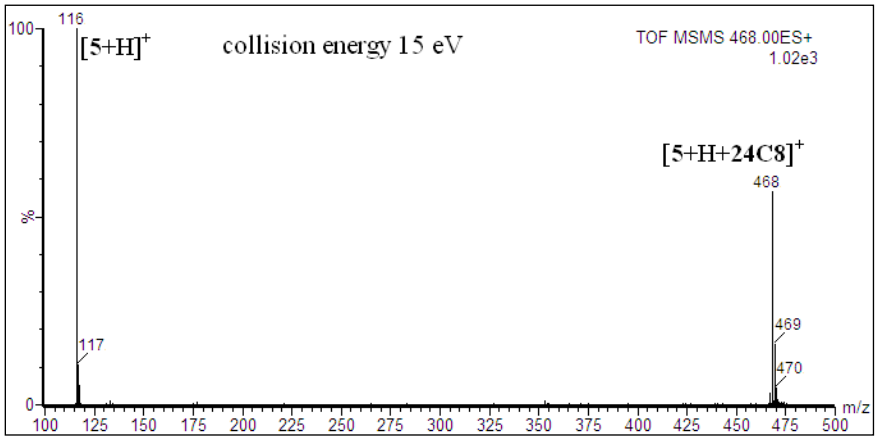


Figure 1: Product ion spectrum of $[5+H+24C8]^+$.

In order to compare the relative gas phase stabilities of the complexes we made the respective break down plots of the ratios $[amine+H]^+/[amine+H]^+ + [complex]^+$ against collision energy. Collision energy in the laboratory frame can be converted into the center-of-mass frame (E_{cm}= 40/40+M_{ion} × E_{lab}, 40 - mass of argon) [32]. However, in this work we compared the isomeric ions of identical masses; therefore such a conversion seemed to be not necessary.

For clarity, the heptylamines were divided into two groups. The first group contained compounds 1-4. Thus, the amine group was substituted to the first carbon atom of the heptane chain - 1, to the second - 2, to the third - 3 and fourth - 4 (Scheme 1). The second group contained compounds 2, 5 and 6. The general chemical formula of these amines is $CH3(NH_2)CHCH_2$-C_4H_9 (2 belongs to both groups). The amines belonging to the first group can be easy differentiated by the EI mass spectra; however, the amines belonging to the second group have identical EI mass spectra (http://webbook.nist.gov/chemistry/).

Figure 2 shows the break down plots obtained for the first group. It is well known that the stabilities of the amine-crown ether complexes increase as the gas-phase basicity of the amine decreases. Thus, it is expected that the complexes of 1 will be more stable than the other ones. It was confirmed by the break down plots obtained for the complexes of large crown ethers.

By comparing the known gas-phase basicities of the primary aliphatic amines it is clear that the more branched the aliphatic chain, the higher the gas-phase basicity (http://webbook.nist.gov/chemistry/). Obviously the most important is the number of alkyl groups attached to the carbon atom bonded with the nitrogen atom (well-known inductive effect). In the first group only 1 has one alkyl group attached to the carbon atom (2, 3 and 4 have two alkyl groups attached to this carbon atom, Scheme 1). Thus, it can be taken for granted that 1 has the lowest the gas-phase basicity so, as consequence 1 should form the most stable complexes with crown ethers. As shown in Figure 2, it was observed for the complexes of large crown ethers (24C8 and DB24C8). For the complexes of 18C6 and DB18C6 it was less pronounced. The complexes of DC18C6 have similar stabilities. The low stability of $[2+H+DB24C8]^+$ is worth noting, however difficult to explain (Figure 2).

Figure 3 shows the break down plots obtained for the second group. The differences in relative stabilities of the complexes are observed for the complexes of DC18C6 and DB24C8. The complex $[6+H+DC18C6]^+$ is definitely less stable than $[2+H+DC18C6]^+$ and $[5+H+DC18C6]^+$, while $[5+H+DB24C8]^+$ is more stable than $[2+H+DB24C8]^+$ and $[6+H+DC18C6]^+$.

The structural differences between the isomeric amines 2, 5 and 6 are relatively far from the nitrogen atom (third and fourth carbon atom counting from the carbon atom bonded with nitrogen atom, Scheme 1). Therefore, the observed differences in the stabilities of their crown ether complexes can be regarded as a good example illustrating the remarkable changes in the complex stabilities in response to subtle structural changes in the amine structure.

The complexes between protonated amines and crown ethers exist due to the formation of hydrogen bonds. For primary amines we deal with three hydrogen bonds. The exchange of the hydrogen atoms involved in the hydrogen bond into deuterium may affect the stabilities of the complexes. Furthermore, the question is if the changes in the complex stabilities can be different for different complexes. The stabilities of the complexes of 2, 5 and 6 with 18C8 are practically identical (Figure 3). Therefore, if the changes in the complex stabilities can be different for different complexes, they should be clearly seen for the complexes of 2, 5 and 6 with 18C8 (more clearly than for other complexes). Figure 4 shows the break down plots obtained for the deuterium labeled complexes of 2, 5 and 6 with 18C8 and an exemplary representative CID-MS/MS spectrum. As evidenced by Figure 4, deuterium labeled complex of 2 with 18C6 is more stable than the deuterium labeled complexes of 5 and 6 with 18C6. At low collision energy, the deuterium labeled complex of 5 with 18C6 is more stable than the deuterium labeled complexes of 6 with 18C6. As mentioned above, the stabilities of the normal (non-deuterium labeled) complexes of 2, 5 and 6 with 18C8 are practically identical (Figure 3). The observed isotope effect illustrates the unexpectedly strong effect on the complex stabilities of subtle structural changes in the amines.

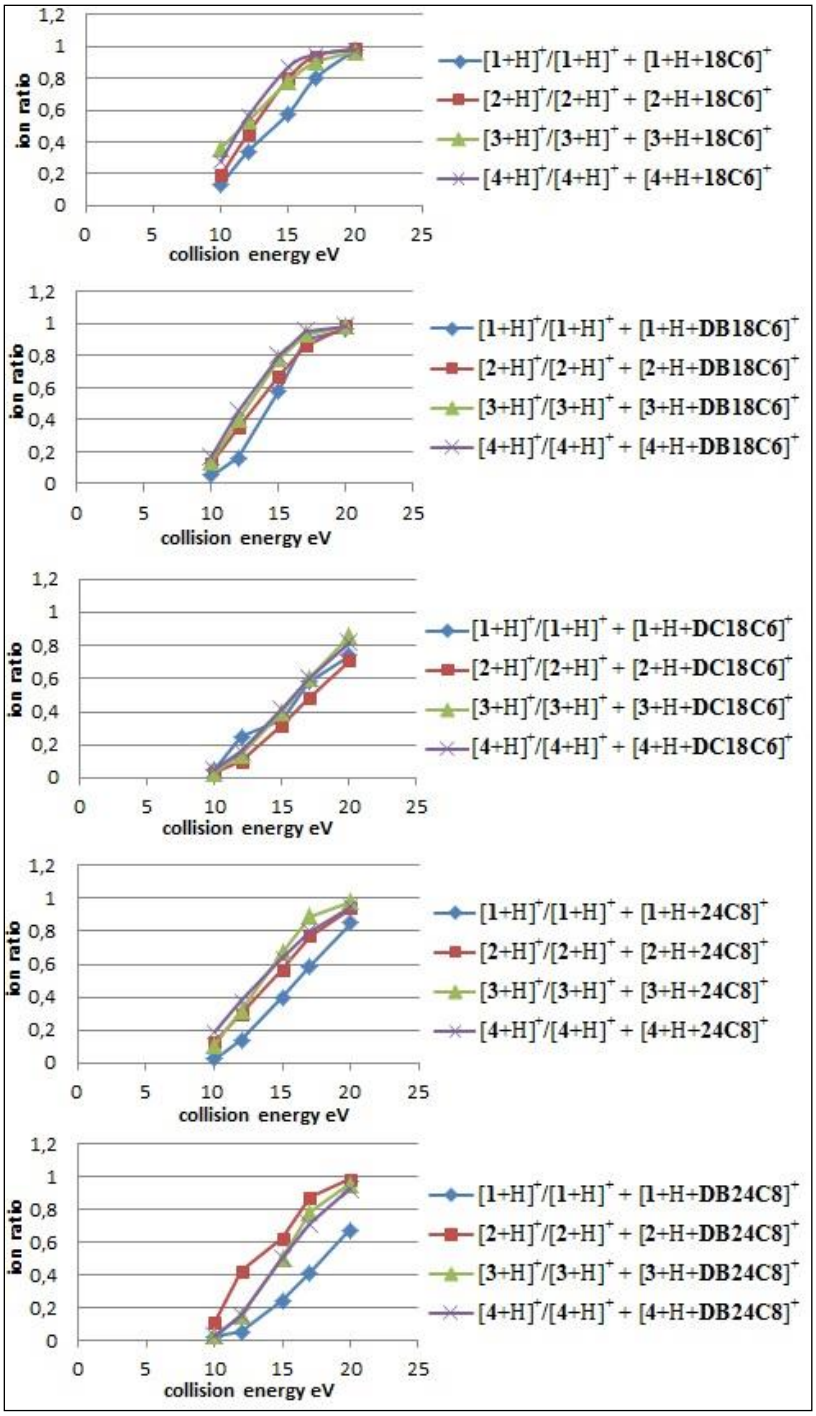


Figure 2: The break down plots obtained for the first group

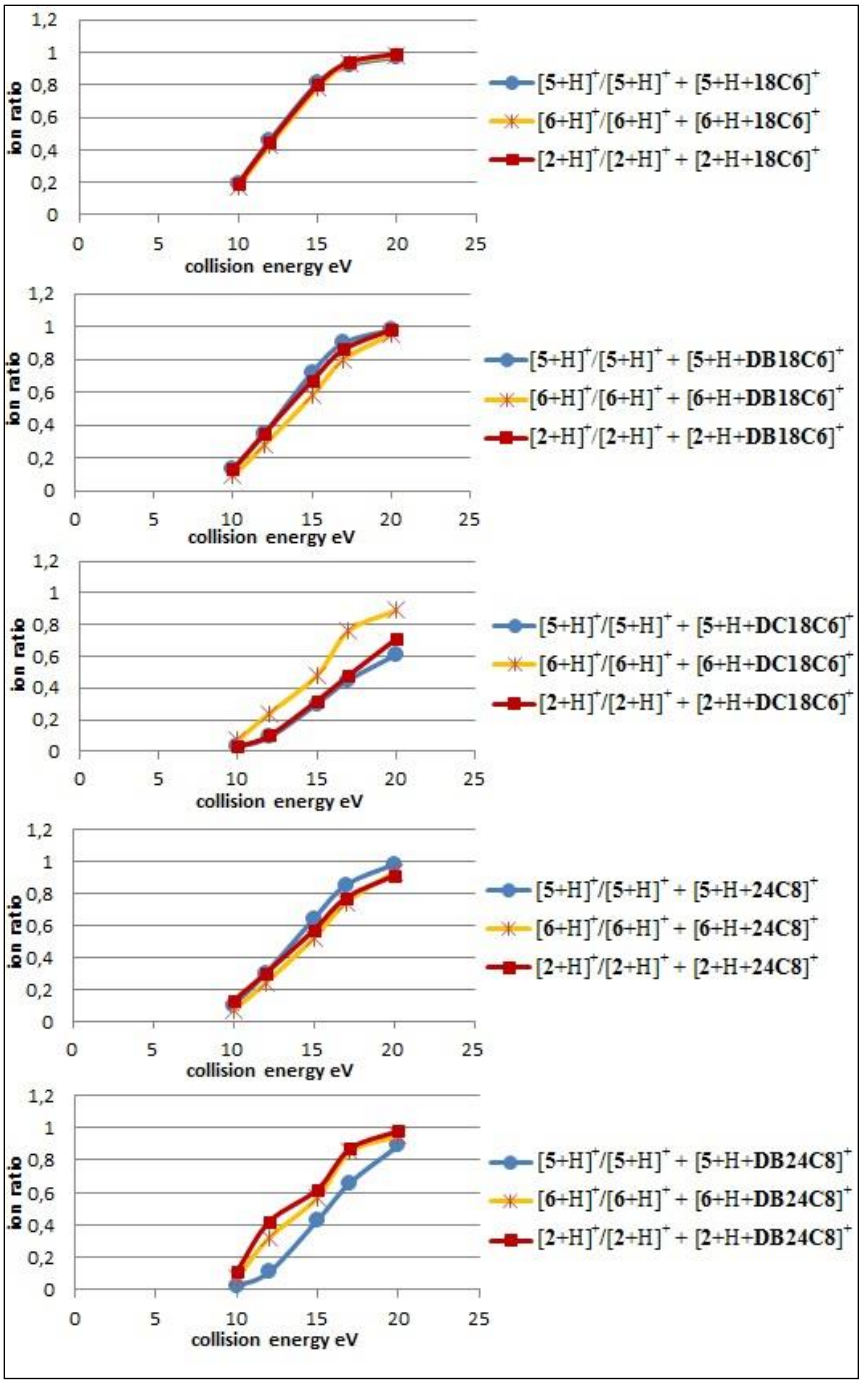


Figure 3: The break down plots obtained for the second group

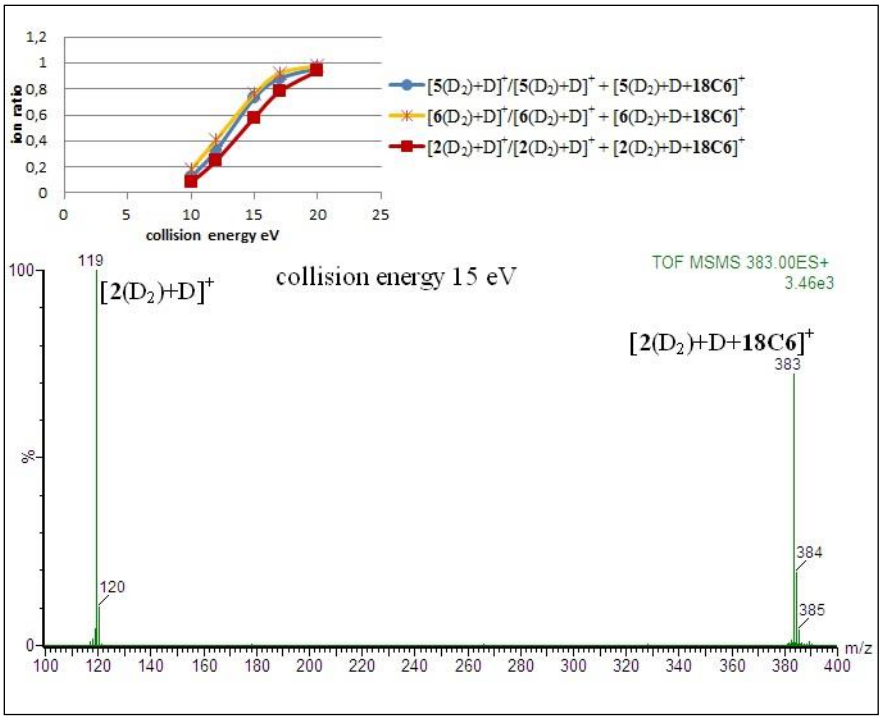


Figure 4: The break down plots obtained for the deuterium labeled complexes of 2, 5 and 6 with 18C8 and exemplary representative product ion spectrum

CONCLUSIONS

The complexes of protonated isomeric heptylamines with crown ethers may have very similar or different gas phase relative stabilities. For example, the complexes of 3-aminoheptane and 4-aminoheptane (heptylamines 3 and 4, Scheme 1) have similar stabilities for all crown ethers used in this work. 1-Aminoheptane (1), which surely has lower gas phase basicity than the other heptylamines, forms more stable complexes with large crown ethers than the complexes formed by the other heptylamines with large crown ethers. The most important in this work are heptylamines 2 and 5 as their complexes with DB24C8 have remarkably different gas phase stabilities. The H/D exchange in the complexes of protonated isomeric heptylamines with crown ethers may also lead to the visible differences in the relative gas phase stabilities.

REFERENCES

[1] P Gerbaux; J De Winter; D Cornil; K Ravicini; G Pesesse; J Cornil; R Flammang. *Chem Eur J*. **2008**, 14(35), 11039-11049.
[2] CN Stedwell; JF Galindo; K Gulyuz; AE Roitberg; NC Polfer. *J Phys Chem A*. **2013**, 117(6), 1181-1188.
[3] OB Bazanova; ZA Bredikhina; VM Babaev; DR Sharafutdinova; RR Fayzullin; AA Bredikhin. *Rus J Org Chem.* **2015**, 51(11), 1642-1648.
[4] ZA Bredikhina; DR Sharafutdinova; OB Bazanova; VM Babaev; RR Fayzullin; IK Rizvanov; AA Bredikhin. *J Incl Phenom Macrocycl Chem*. **2014**, 80(3-4), 417-426.
[5] BC Bohrer; DE Clemmer. *Anal Chem.* **2011**, 83(13), 5377-5385.
[6] RR Julian; JL Beauchamp. *Int J Mass Spectrom.* **2001**, 210/211(1-3), 613-623.
[7] R Frański;; T G Schroeder; W Kamysz; P Niedzialkowski Ossowski. *J. Mass Spectrom.* **200**7, 42(4), 459-466.
[8] S Malehiat; J Brodbelt. *J Am Chem Soc.* **1993**, 115(7), 2837-2843.
[9] P Hurtado; F Gámez; S Hamad; B Martínez-Haya; JD Steill; J Oomens. *J Phys Chem A*. **2011**, 115(25), 7275-7282.
[10] WM David; JS Brodbelt. *J Am Soc Mass Spectrom.* **2003**, 14(4), 383-392.

[11] MA Zickus; S Koepke; C Hao; K Chong; V Ryzhov. *Int J Mass Spectrom.* **2012**, 312(1), 173-178.
[12] Z Li; SJ Valentin; DE Clemmer. *J Am Soc Mass Spectrom.* **2011**, 22(5), 817-827.
[13] N Alizadeh; P Shahdousti; S Nabavi; M Tabrizchi. *Int J Mass Spectrom.* **2011**, 308(1), 18-25.
[14] BL Williamson; CS Creaser. *Int J Mass Spectrom*. **1999**, 188(1), 53-61.
[15] C-C Liou; H-F Wu; JS Brodbelt. *J Am Soc Mass Spectrom.* **1994**, 5(4), 260-273.
[16] M Schäfer. *Angew Chem Int Ed*. **2003**, 42(17), 1896-1899.
[17] M Lederer; U Hahn; J-M Strub; S Cianférani; A Van Dorsselaer; J-F Nierengarten; T Torres; DM Guldi. *Chem Eur J*. **2016**, 22(6), 2051-2059.
[18] V Rüdiger; H-J Schneider; VP Solov'ev; VP Kazachenko; OA Raevsky. *Eur J Org Chem.* **1999**, 1999(8), 1847-1856.
[19] M Thevis; G Sigmund; A Koch; W Schänzer. *Eur J Mass Spectrom.* **2007**, 13(3), 213-221.
[20] AD Lesiak; KJ Adams; MA Domin; C Hencka; JRE Shepard. *Drug Test Anal.* **2014**, 6(7-8), 788-796.
[21] L Perrenoud; M Saugy; C Saudan. *J Chromatogr A*. **2009**; 877(29), 3767-3770.
[22] V Lopez-Avila; M Zorio. *Forensic Sci Int.* **2013**, 231(1-3), 113-119.
[23] JRH Archer; PI Dargan; AM Lostia; J Van Der Walt; K Henderson; N Drake; S Sharma; DM Wood; CJ Walker; AT Kicman. *Drug Test Anal.* **2015**, 7(5), 433-438.
[24] M Dunn. *Int J Drug Policy*. **2017**; 40(1), 26-34.
[25] C Di Lorenzo; E Moro; AD Santos; F Uberti; P Restani. *Drug Test Anal.* **2012**, 5(2), 116-121.
[26] HL Fleming; PL Ranaivo; PS Simone. *Anal Chem Insights*. **2012**, 7(1), 59-78.
[27] A Lisi; N Hasick; R Kazlauskas; C Goebel. *Drug Test Anal.* **2011**, 3(11-12), 873-876.
[28] B Avula; TJ Smillie; Y-H Wang; J Zweigenbaum; MA ElSohly; IA Khan. *J AOAC Int.* **2015**, 98(3), 757-759.
[29] MA ElSohly; W Gul; C Tolbert; KM ElSohly; TP Murphy; B Avula; AG Chittiboyina; M Wang; IA Khan; M Yang; D Guo; W-D Zhang; J Su. *Drug Test Anal.* **2014**, 7(7), 645-654.
[30] KG Austin; J Travis; G Pacec; HR Lieberman. *Drug Test Anal.* **2013**, 6(7-8), 797-804.
[31] TD Gauthier. *Anal Chem Insights*. **2013**, 8(1), 29-40.
[32] L Sleno; DA Volmer. *J Mass Spectrom.* **2004**, 39(10), 1091-1112.