The First Evidence for Triple Cation Binding by Multi-ring Macrocyclic Polyethers: An Electrospray Ionization Mass Spectral Study

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Unequivocal evidence was obtained for three-cation complexation by three-crown, bola-amphiphilic systems by use of the electrospray ionization mass spectrometric technique.

In previous studies, we have shown that two- and three-ring macrocycles can be constructed and used for the formation of membranes[1] or to form cation-conducting channels in phospholipid vesicles.[2] We have also demonstrated by use of the fast atom bombardment mass spectrometry (FAB-MS) technique that multi-ring macrocycles can cooperate intramolecularly to complex cations.[3] In the latter case, the separation of the simple bola-amphiphilic[4] macrocycles ranged from 10 to 22 carbons but the tris(macrocycle)s studied contained $-(CH_2)_6-$ spacers. In both the two- or three-ring cases, FAB-MS revealed that two cations could be bound by the macrocyles. In the present study, we demonstrate simultaneous, three-cation complexation by a tris(macrocyclic) system.

Two macrocycles were prepared for the present study. The tris(macrocycle), **1**, was previously reported.[2] Compound **2** is a bis(macrocycle) in which the aza-18-crown-6 rings are connected by a relatively rigid, xylene-1,4-diyl spacer.[5]

The electrospray ionization (ESI) mass spectra[6] were acquired on a triple quadrupole tandem mass spectrometer equipped with an electrospray interface.† Details of the electrospray ion source have been reported previously.[7] Two sets of experiments were done with each substrate. In the first, shown in the Table as set 'A', a solution of either **1** or **2** (0.05 mmol dm^{-3}), KCl (0.05 mmol dm^{-3}) and NaCl (0.5 mmol dm^{-3}) was prepared in 10% *v/v* $CHCl_3$–MeOH. In the second set 'B', **1** (0.05 mmol dm^{-3}), KCl (0.15 mmol dm^{-3}) and NaCl (0.5 mmol dm^{-3}) were dissolved in 10% *v/v* $CHCl_3$–MeOH. The masses and assignments of the observed peaks (compounds **1** and **2**) along with relative intensity data are recorded in Table 1.

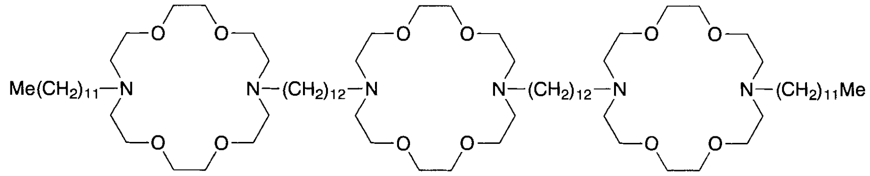
The concentration differences used for conditions 'A' and 'B' reflect an effort to assess differences in sodium or potassium selectivities. When the Na^+ : ligand concentration ratio was 10 : 1, the base peak in the spectra of **1** or **2** (see column 'A', Table 1) was the complex between the ligand and a number of sodium cations equal to the number of macrorings present. Potassium complexation was certainly observed but the spectrum was dominated by sodium cation complexes. When the K^+ concentration was increased to equimolar with the number of cation-binding rings present in **1**, an ion having the composition $[\mathbf{1}{\cdot}3K]^+$ was prominent, although the base peak was due to $[\mathbf{1}{\cdot}2Na{\cdot}K]^+$. This result is expected because of the differential in cation concentrations.

To our knowledge, the data presented herein constitute the first definitive evidence for triple cation complexation. In a previous study, we found that FAB-MS could detect the presence of certain dication complexes of tris(macrocycles). In the latter case, the three macrocyclic rings were separated by $-(CH_2)_6-$ spacers. Thus, in the extended conformation, these rings were closer together than they are in the present systems and electrostatic repulsions could have been greater. The observation of species $[\mathbf{2}{\cdot}2Na]^{2+}$ is particularly notable since the macrorings of **2** are separated by a fairly rigid spacer which is expected to prevent the sort of ring–ring cooperation that might be possible in **1**.

Polyethylene glycols [PEGs, $RO(CH_2CH_2O)_nR$, R = H, Me) are capable of binding more than one cation simultaneously and in methanol solution, a plot of log K_S *vs.* log M_w is linear.[8] Nevertheless, the ability of PEGs to catalyse phase-transfer reactions was not enhanced by the use of longer chains. Instead, catalysis was correlated to the molarity of PEG chains available to transfer cations from the polar to the non-polar phase. The present case is remarkable in this context as the macrocycle appears capable of binding three cations in the gas phase in the absence of a neutralizing anion or coordinating solvent; no peak observable above the baseline appears to contain methanol.

The spectrum of **2** shows no peak of relative intensity greater than 25% of that due to $[\mathbf{2}{\cdot}2Na]^{2+}$. Two peaks of approximate intensity 25%, ($[\mathbf{2}{\cdot}Na{\cdot}K]^{2+}$ and $[\mathbf{2}{\cdot}2Na{\cdot}Cl]^+$), both contain two cations.

Compound **1** has three macrorings and a correspondingly larger number of cation complexation possibilities. As noted above, under 'A' conditions, the base peak is due to $[\mathbf{1}{\cdot}3Na]^{3+}$. A peak observed at *m/z* 500.7 has the composition $[\mathbf{1}{\cdot}H{\cdot}2Na]^{3+}$ and a relative intensity of 98%. The corresponding $[\mathbf{1}{\cdot}H{\cdot}2K]^{3+}$ ion is also observed, but its relative intensity is < 10%. Four other prominent ions (relative intensities > 50%) are observed corresponding to $[\mathbf{1}{\cdot}2H{\cdot}Na]^{3+}$, $[\mathbf{1}{\cdot}H{\cdot}Na{\cdot}K]^{3+}$, $[\mathbf{1}{\cdot}2Na{\cdot}K]^{3+}$ and $[\mathbf{1}{\cdot}2Na]^{2+}$. The fact that K^+ is present in several of the major ions observed in the spectra of **1** and **2** affirms its stronger binding despite its lower concentration. When the concentration of K^+ is higher (conditions 'B', $[Na^+]$: $[K^+]$ = 10 : 3; condition



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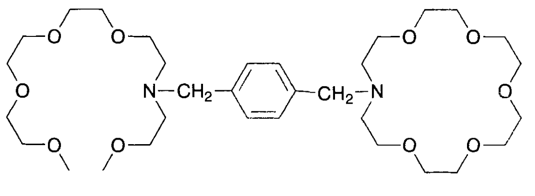

Table 1 Electrospray ionization mass spectra for compounds **1** and **2**[a]

Ion	m/z	Rel. intensity		Ion	m/z	Rel. intensity	
		A[b]	B[c]			A	B
$[\mathbf{1}\cdot 3H]^{3+}$	486	12	11	$[\mathbf{1}\cdot 3Na\cdot Cl]^{2+}$	780.5	31	20
$[\mathbf{1}\cdot 2H\cdot Na]^{3+}$	493.5	54	26	$[\mathbf{1}\cdot H\cdot 2K\cdot Cl]^{2+}$	785.1	<5	16
$[\mathbf{1}\cdot 2H\cdot K]^{3+}$	498.6	17	28	$[\mathbf{1}\cdot 2Na\cdot K\cdot Cl]^{2+}$	788.2	21	43
$[\mathbf{1}\cdot H\cdot 2Na]^{3+}$	500.7	98	43	$[\mathbf{1}\cdot Na\cdot 2K\cdot Cl]^{2+}$	795.7	8	36
$[\mathbf{1}\cdot H\cdot Na\cdot K]^{3+}$	506.2	58	95	$[\mathbf{1}\cdot 3K\cdot Cl]^{2+}$	804.2	<5	28
$[\mathbf{1}\cdot 3Na]^{3+}$	508.2	100	25	$[\mathbf{1}\cdot Na]^{+}$	1478.6	22	17
$[\mathbf{1}\cdot H\cdot 2K]^{3+}$	511.5	9	52				
$[\mathbf{1}\cdot 2Na\cdot K]^{3+}$	513.5	81	100	$[\mathbf{2}\cdot 2H]^{2+}$	315.1	<5	—
$[\mathbf{1}\cdot Na\cdot 2K]^{3+}$	518.7	17	83	$[\mathbf{2}\cdot H\cdot Na]^{2+}$	326.2	5	—
$[\mathbf{1}\cdot 3K]^{3+}$	524.2	<5	31	$[\mathbf{2}\cdot 2Na]^{2+}$	337.1	100	—
$[\mathbf{1}\cdot 2H]^{2+}$	729	12	10	$[\mathbf{2}\cdot Na\cdot K]^{2+}$	345.1	25	—
$[\mathbf{1}\cdot H\cdot Na]^{2+}$	740.1	35	17	$[\mathbf{2}\cdot 2K]^{2+}$	353.1	<5	—
$[\mathbf{1}\cdot H\cdot K]^{2+}$	747.9	9	16	$[\mathbf{2}\cdot H]^{+}$	629	<5	—
$[\mathbf{1}\cdot 2Na]^{2+}$	750.8	70	31	$[\mathbf{2}\cdot Na]^{+}$	651.6	18	—
$[\mathbf{1}\cdot Na\cdot K]^{2+}$	759	31	62	$[\mathbf{2}\cdot K]^{+}$	667.6	<5	—
$[\mathbf{1}\cdot 2K]^{2+}$	767	8	28	$[\mathbf{2}\cdot 2Na\cdot Cl]^{+}$	709.5	24	—
$[\mathbf{1}\cdot H\cdot 2Na\cdot Cl]^{2+}$	769.2	6	17	$[\mathbf{2}\cdot Na\cdot K\cdot Cl]^{+}$	725.4	9	—
$[\mathbf{1}\cdot H\cdot Na\cdot K\cdot Cl]^{2+}$	777.1	<5	26	$[\mathbf{2}\cdot 2K\cdot Cl]^{+}$	741.6	<5	—

[a] See footnote † for conditions. [b] *Conditions*: [**1**] = [**2**] = K^+ = 0.05 mmol dm^{-3}, [Na^+] = 0.5 mmol dm^{-3}. [c] *Conditions*: [**1**] = 0.05 mmol dm^{-3}, [K^+] = 0.15 mmol dm^{-3}, [Na^+] = 0.5 mmol dm^{-3}.

'A', 10 : 1), five prominent ions are observed, all containing K^+. Two of the three most intense ions contain three metallic cations, with the base peak being due to $[\mathbf{1}\cdot 2Na\cdot K]^{3+}$. Moreover, four of these five most intense ions are triply charged.

It is also interesting that chloride counter ion appears in several ions. It is presumably coordinated to one or more sodium or potassium cations although this technique does not afford us structural information. It should be noted that chloride is observed only when more than two metallic cations are present, ions of the type [(**1** or **2**)·cation·Cl] would be neutral overall and indetectable by this technique.

A persistent question about the application of gas-phase techniques to solution phenomena is whether the information obtained is really relevant. Recent studies that address this problem[9] for the electrospray ionization method answer in the affirmative. Of course, macrorings separated by distances of 6–12 Å, as is the case with the present molecules, are expected to bind multiple cations in solution.

Overall, we show here for the first time that three cations may be bound by a tris(macrocyclic) ligand. No previous study has established this possibility nor is there any obvious solution technique that can be used to determine cation binding constants in such complex structures. The ESI-MS technique holds considerable promise for assessing cation interactions with binders that are too structurally complex to assess by other methods.

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Footnote

† The N_2 drying gas was held at 100 °C and constant pressure (25 psi) as it entered the electrospray chamber. The signal was averaged for a period of 4 min for each spectrum. Sample solutions (*ca.* 25 °C) were infused directly into the ESI chamber by syringe pump (rate = 2 μl min^{-1}).

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