Stable and Selective Guanidinium and Imidazolium Complexes of a Macrocyclic Receptor Molecule

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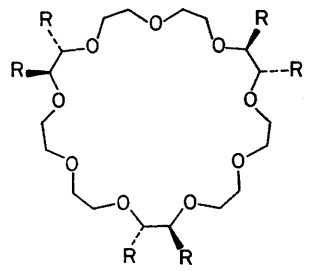
Summary The hexacarboxylate [27]-O_9 macrocyclic polyether (**1**) is a suitable receptor molecule forming stable and selective inclusion complexes with guanidinium and imidazolium as well as ammonium salts in aqueous solution.

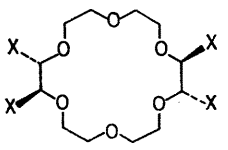
GUANIDINIUM groups of arginine residues play a most important biological role as binding sites for anionic substrates in enzyme active sites,[1] in receptor sites of antibodies,[2] and in nucleic acid interaction with the highly charged protamines and histones.[3] They are also present in hormones (like bradykinin) and in toxins (tetrodotoxin and saxitoxin).[4] A low molecular weight receptor molecule capable of strongly complexing guanidinium groups would thus be of great interest. The design of such a system also represents an interesting problem in molecular recognition. It has been shown that [18]-crown-6[5] and [27]-crown-9[6] macrocyclic polyethers are able to complex guanidinium chloride in the solid state or in an organic solvent. The latter ring provides a suitable arrangement of oxygen binding sites for guanidinium complexation.[6]

We now report that the chiral hexacarboxylate [27]-crown-9 derivative (**1**) forms complexes with guanidinium and imidazolium groups, which are *stable and selective even in aqueous solution.* We had shown earlier that the tetracarboxylate 18-crown-6 derivative (**2**) forms much more stable complexes than the parent system (**2** with X = H).[7]

The [27]-crown-9 hexacarboxamide (**3**) is obtained as a glassy colourless solid in about 10% yield, by careful chromatography of the reaction mixture resulting from the condensation of $[ICH_2CH_2OCH_2CH_2O\text{-}(R)CHX]_2$ with $[HO\text{-}(R)CHX\text{-}(R)CHX\text{-}OCH_2CH_2]_2O$ [X = $CONMe_2$; both derived from the bis-dimethylamide of (*R,R*)-(+)-tartaric acid, see ref. 8] using a procedure similar to that described



(1) R = CO_2^- $^+HN(CH_2CH_2OH)_3$
(3) R = $CONMe_2$



(2) R = CO_2^- $^+NMe_4$
(4) R = $CONMe_2$

for the preparation of the [18]-crown-6 tetracarboxamide (**4**);[8] it may also be isolated in about 1% yield from the mother liquors of the preparation of compound (**4**)[8]. It has been characterized by microanalysis, mass spectroscopy, ^{13}C- and ^1H-n.m.r. spectroscopy, and specific optical rotation $[\alpha]_D = 110°$ (*c* 1·07 in $CHCl_3$). The hexacarboxylic acid of (**1**) (colourless glassy solid; $[\alpha]_D = 40°$, *c* 0·445 in tetramethylammonium phosphate buffer, 0·1 M, at pH 6·0) is obtained by acid hydrolysis of (**3**) under conditions similar to those in the preparation of (**2**) from (**4**).[7] The stabilities of the complexes formed by the ligand (**1**) have been determined using a competition method. The 1 : 1 stoicheiometry of the [(**1**)–NH_4^+] complex was shown by n.m.r. studies and its stability was measured by titration of (**1**) with NH_4Cl using a NH_4^+-selective electrode. The stability constants of the other complexes were obtained by performing the same titration in the presence of the other substrates which displace the bound NH_4^+ ions; they are listed in the Table. The following conclusions may be drawn.

(i) The presence of anionic carboxylate groups markedly increases the stability of the complexes of (**1**) with respect to the uncharged macrocycle (**3**) ($K_S < 5$ for NH_4^+). The present guanidinium (G^+) and ammonium (N^+) complexes are by far the most stable ones known to date. As in the case of (**2**),[7] electrostatic interactions are the prime factors leading to *high stability*.

(ii) *Selectivity* of complexation arises from *central discrimination* by the macrocyclic ring. The guanidinium complex of (**1**) is more stable than its NH_4^+ complex while the reverse holds for (**2**).† This agrees with a recognition pattern of ionic N^+–H...O hydrogen bonds as depicted in (**5**) and confirmed by the steric effects due to the introduction of substituents. The stability of the (G^+) complexes of (**1**) decreases markedly as the number and bulk of substituents (Me, Et) on (G^+) increases; this strong steric discrimination is similar to that found for the (N^+) complexes of (**2**).[7]† The stability of the (N^+) complexes of (**1**) also decreases on substitution, but less markedly, indicating less steric interference than with substitution on (G^+); *e.g.*, $Me_2NH_2^+$ forms a stable complex with the larger macrocycle (**1**), where inclusion is still possible, but not with (**2**)† where central discrimination between primary and secondary ammonium salts is much more efficient.[7,9] These selectivity patterns confirm the formation of cryptate complexes by inclusion of the (G^+) groups into the macrocyclic cavity, making compound (**1**) a *guanidinium receptor* molecule.

TABLE. Stability constants, K_S of guanidinium, ammonium, and imidazolium complexes of the macrocyclic receptor molecule (**1**).[a]

Substrate	K_S/l mol^{-1}
Guanidinium	9,000 ± 700
Methylguanidinium	450 ± 40
Ethylguanidinium	150 ± 30
NN'-Diethylguanidinium	<10
NNN'N'-Tetramethylguanidinium	<10
G^+–$[CH_2]_n$–G^+	
$n = 2$	14,000 ± 1,500
$n = 3$	4,300 ± 400
$n = 4$ (Arcaïne)	2,100 ± 200
$n = 5$ (Andouine)	1,500 ± 250
$n = 6$	1,000 ± 150
G^+–$[CH_2]_4$–NH_3^+ (Agmatine)	4,700 ± 400
L-Arginine	250 ± 30
Streptidine	1,100
Creatine	<10
NH_4^+	6,000
$MeNH_3^+$	1,200
$EtNH_3^+$	160
$Me_2NH_2^+$	110
Me_3NH^+	<10
Me_4N^+	<10
^+H_3N–$[CH_2]_n$–NH_3^+	
$n = 2$	$2 \pm 0.5 \times 10^5$
$n = 3$	$6 \pm 1.5 \times 10^4$
$n = 4$	7,200
L-Ornithine	700
D,L-Lysine	165
Imidazolium	350
N-Methylimidazolium	<10
2-Methylimidazolium	*ca.* 30
N-Acetylhistamine–H^+	120

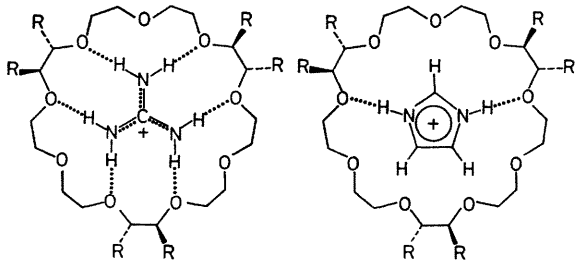
[a] In aqueous solution at 25 °C: 0·1 M tris-(2-hydroxyethyl)-ammonium hydrochloride buffer, pH 7·3. The concentrations of (**1**) and substrate were usually *ca.* 4·3 mM and 6 mM respectively; a higher concentration, 12 mM, was used for the weakly complexed substrates. The error in K_S is *ca.* ±10% as estimated from accuracy and reproducibility of the measurements. The data agree with 1 : 1 stoicheiometry for all complexes; however the formation of weak complexes containing more than one substrate molecule cannot be excluded at higher concentrations. $G^+ = -NH-C(NH_2)=NH_2^+$.

(iii) Both types of doubly charged substrates G^+–$[CH_2]_n$–G^+ and ^+H_3N–$[CH_2]_n$–NH_3^+ form much more stable complexes than the mono (G^+) and (N^+) compounds even at large separations (*e.g.* G^+–$[CH_2]_6$–G^+); however the bis-(N^+) complexes now become appreciably more stable than the bis-(G^+) ones, owing both to more localized charges and to weaker steric effects (as already discussed). The biological substances arcaïne, andouine, and agmatine are strongly complexed. This is also the case for the amino-acids arginine, ornithine, and lysine despite the presence of a carboxylate group on the substrate.

(iv) The *imidazolium ion*, which presents some structural analogy with guanidinium, is also appreciably complexed by the receptor (**1**), although much less well than guanidinium (Table). The introduction of substituents decreases markedly the association constant in the order: N–CH_3 < C(2)–CH_3 < C(4)–R, no complexation being detected for *N*-methylimidazolium (Table). These substituent effects, as well as consideration of space-filling molecular models, suggest that the imidazolium ion binds to the macrocyclic receptor (**1**) by forming two N–H^+....O hydrogen bonds with oxygen sites in 1,5 (or perhaps 1,4) positions in the ring; its molecular plane may be tilted to

† The stability constants for the complexes of (**2**) are 45 (G^+),[9] 3200 (NH_4^+),[7] 750 ($MeNH_3^+$),[7] 260 ($EtNH_3^+$),[9] and <5 ($Me_2NH_2^+$)[9] (in aqueous solution at pH 7·0).

some extent with respect to the average plane of the macrocycle, since a substituent at C(4) is less detrimental to complexation than one at C(2). The structure of the complex is probably close to that represented schematically by (6).



(5) R = CO_2^- $^+HN(CH_2CH_2OH)_3$ (6) R = CO_2^- $^+HN(CH_2CH_2OH)_3$

One may envisage the formation of complexes of other amidinium salts (2-aminopyridinium, protonated adenine *etc.*) with (1) or larger polyanionic macrocycles. The receptor molecules (1) and (2) should also form strong complexes with highly charged, biologically important molecules and macromolecules like polyamines, protamines, and arginine-rich or lysine-rich nucleohistones.

It is clear that the combined use of a macrocyclic cavity for selection and charged sites for stability may lead to complexing agents for a wide variety of molecular substrates. Extension to macropolycyclic cavities may increase stabilities and selectivities by allowing a better design of the recognition site of the receptor molecule.

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