Tetrahedron Letters,Vol.29,No.48,pp 6231-6234,1988 0040-4039/88 $3.00 + .00
Printed in Great Britain Pergamon Press plc

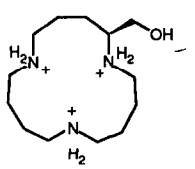
Complexation of ATP to a Synthetic [15]-N_3 Macrocyclic Polyammonium Receptor

James F. Marecek, Peter A. Fischer and Cynthia J. Burrows*

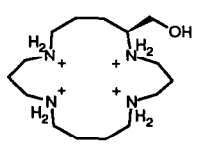
Department of Chemistry
State University of New York at Stony Brook
Stony Brook, NY 11794-3400, U.S.A.

Summary. *A new macrocyclic triamine, (S)-2-hydroxymethyl-1,6,11-triazacyclopentadecane, was synthesized from ornithine, and complexation of its conjugate acid with ATP was studied by ^{31}P-NMR.*

Anion complexation to synthetic molecular receptors is of interest in both organic and biological chemistry as researchers attempt to understand the molecular interactions involved in anion binding.[1] In particular, macrocyclic polyammonium compounds have been investigated as anion complexones since they mimic the biological polyamines, especially spermine and spermidine, in their association to inorganic phosphate and phosphate ester or anhydride anions.[2,3] In addition, certain of them have been studied for their complexation to halides and trigonal anions such as NO_3^-.[4,5] Additional functional groups in the macrocyclic receptor which are not involved in binding may act as reagents or catalysts for reactions of anionic substrates. This concept has been elegantly demonstrated in the case of an artificial ATPase.[6] One of our goals in this area is the design of a phosphate-binding unit which may ultimately be elaborated to include additional reactive or binding functional groups. Toward this goal, a new [15]-ane-N_3 macrocycle, **1**, was synthesized from L-ornithine, and its complexation to ATP^{4-} was compared with a macrocyclic spermine analog, **2**.[7]



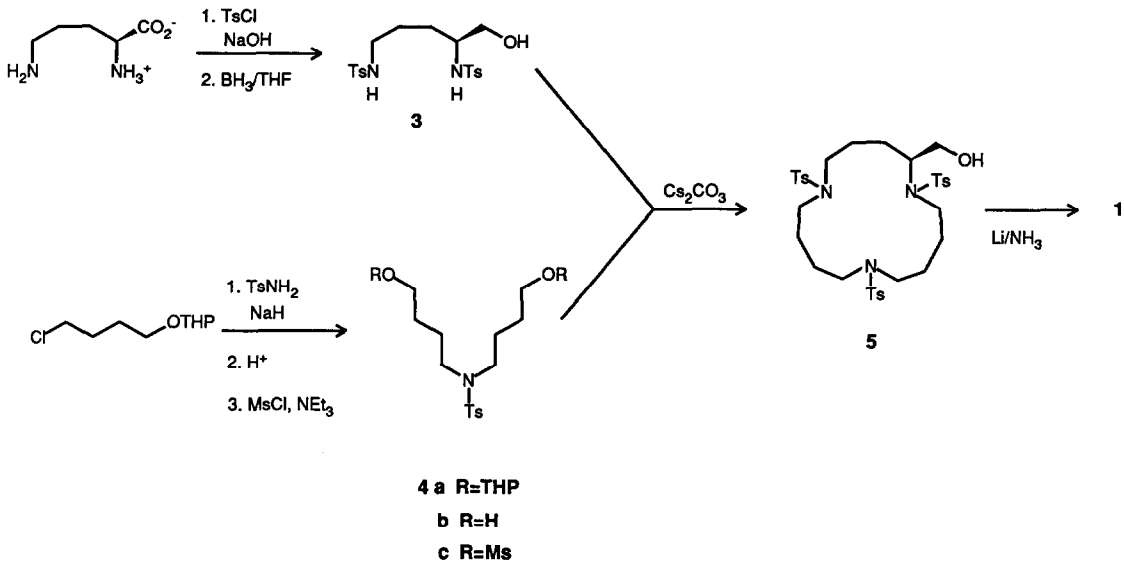
1



2

Previous work[8,9] has demonstrated that the [18]-ane-N_4 ring system results in association constants with ATP^{4-} in excess of 10^5. This strong association appears to stem from the charge-charge interactions of a polycation with high positive charge density with a polyanion (ATP^{4-}) of high negative charge density. The spacing of ammonium groups in **2** is ideal. A distance of 3-4 methylene groups prevents the lowering of pK_a's seen in $(\text{-NHCH}_2\text{CH}_2\text{NH-})_n$ systems, and the polyamine is therefore fully protonated near pH 7. At the same time, the ring is as small as possible resulting in a high charge density on the macrocycle. With these factors in mind, it was of interest to study anion complexation using a [15]-ane-N_3 compound with a tripodal, pseudo-C_3

Scheme I



Scheme I: Synthesis of Macrocycle **1**. Ts = *p*-$CH_3C_6H_4SO_2$-; THP = tetrahydropyranyl; Ms = CH_3SO_2-.

arrangement of hydrogen bond donors. The $(CH_2)_4$ spacing between ammonium groups of **1** should allow triprotonation near pH 7 in a relatively small macrocycle.

The synthesis of macrocycle **1** is shown in Scheme I. It makes use of the well-established N-alkylation of *p*-toluenesulfonamides in the presence of Cs_2CO_3.[10] The bis-tosylamide **3** was prepared from L-ornithine hydrochloride by tosylation and subsequent reduction of the carboxyl group as previously described.[7] Compound **4c** was prepared starting with 4-chloro-1-tetrahydropyranylbutanol[11] and *p*-toluenesulfonamide ($TsNH_2$). Equimolar amounts of $TsNH_2$, NaH and the alkyl chloride were allowed to react (70°C, DMF) for 4.5 h. This was followed by treatment with a second equiv. of NaH and alkyl chloride with continued heating overnight. Crude **4a** was not purified but was treated directly with TsOH in EtOH to remove the THP groups. Compound **4b** was purified by column chromatography (silica, EtOAc) and then treated with methanesulfonyl chloride and triethylamine (CH_2Cl_2, overnight) to give **4c** in 43% overall yield. Macrocyclization was carried out by slow addition (6 h) of equimolar amounts of **3** and **4c** to a suspension of 5 equiv. Cs_2CO_3 in DMF at 80-85°C and continued heating for another 16 h. Column chromatography (3:1 EtOAc:hexanes) provided the tritosylamide **5** in 45% yield. Detosylation was effected by reaction with Li in NH_3/THF/MeOH (125:25:1). The desired macrocycle was isolated as its trihydrochloride salt in 45% yield. Physical and chemical data are consistent with the structure of **1**.[12]

Preliminary titration studies of triamine **1** indicated pK_a's of approximately 7, 8 and >11; for tetraamine **2**, pK_a's were determined to be 6.8, 7.0, 10.5, and 10.8. In order to have a qualitative indication of anion binding, ^{31}P-NMR studies were carried out with compound **1** and ATP under conditions identical to those previously reported for receptor **2**.[7] In these experiments, the chemical shift of the γ phosphorus of ATP was monitored

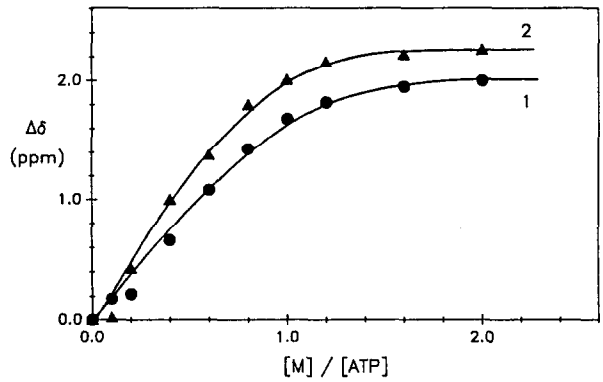


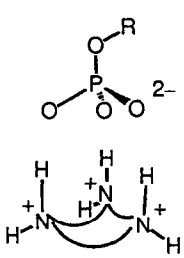
Figure 1. Change in chemical shift of ^{31}P-NMR resonances of ATP upon complexation to a polyammonium macrocycle, **M** (o=**1**, ▲=**2**), at pH 6.80.

as a function of added polyamine. The pH was held constant at 6.80±0.02 in 20% D_2O. Samples of 0.01 M $(NMe_4)_2ATP{\cdot}2H_2O$[13] displayed a γ-P chemical shift of -8.56 ppm (upfield of external 85% H_3PO_4 reference). In the presence of a complexing polyamine, the resonance shifted to lower field. Consistent with other studies,[2] only the γ-P showed a significant chemical shift, presumably due to a conformational change in the triphosphate group upon association to a polyammonium. The results for complexation of both **1** and **2** are shown in Figure 1.

The data in Figure 1 show a somewhat larger overall shift upon complexation to tetraamine **2** than to triamine **1**, although the magnitude of the shift is not an indication of strength of binding. In the limit of infinitely strong binding, a straight line with a change to zero slope at 1:1 stoichiometry would be expected. For **2**, the observed binding curve is slightly steeper and lies closer to the theoretical one suggesting, qualitatively, that the tetraammonium receptor may be a modestly better binding unit than its triammonium analog. More accurate binding constants can be determined by potentiometric titration, and these results are currently under analysis. For both complexes the stoichiometry is approximately 1:1, although there is a possibility that higher order complexes, such as 2 ATP:1 macrocycle may also be present.

One motivation for studying triamine **1** was to test whether a tripodal arrangement of ammonium groups might more closely match the trigonal oxyanion array of a phosphate substrate (see Figure 2). If hydrogen-bonding to the terminal phosphate group of ATP contributes highly to the stability of the complex, a fully protonated triammonium macrocycle with approximate C_3 symmetry would show enhanced binding. Since this effect was not borne out in the experiment, we conclude that the major contribution to the stability of anion complexation in H_2O is the sum of the charge-charge interactions. Tetraprotonation of receptor **2** in the presence of the tetraanion ATP^{4-} leads to a stronger interaction than with that of **1**. These conclusions assume that no unusual conformational effects in **1** prevent its strong association with ATP. Future studies will address the quantitative aspects of association of **1** with a family of anionic substrates, including polyphosphates and polycarboxylates and explore functionalized side chains on the polyammonium macrocycles.

Figure 2



Acknowledgement. Support of this work by a grant from the National Science Foundation (CHE-8706616) is gratefully acknowledged.

References and Notes.

1. Reviews: (a) F. Voegtle, H. Sieger and W. M. Mueller *Topics Curr. Chem.* **1981**, *98*, 109-161. (b) J.-L. Pierre and P. Baret *Bull. Soc. Chim. Fr.* **1983**, II-367-380. (c) B. Dietrich in *Inclusion Compounds, Vol.2*, J. L. Atwood, J. E. D. Davies and D. D. MacNicol, Eds.; Academic: London, 1984, chap. 10.
2. B. Dietrich, M. W. Hosseini, J.-M. Lehn and R. B. Sessions *J. Am. Chem. Soc.* **1981**, *103*, 1282-1283. (b) M. W. Hosseini and J.-M. Lehn *Helv. Chim. Acta* **1986**, *69*, 587-603. (c) M. W. Hosseini and J.-M. Lehn *Helv. Chim. Acta* **1987**, *70*, 1312-1319. (d) M. W. Hosseini and J. M. Lehn *Helv. Chim. Acta* **1988**, *71*, 749-756.
3. (a) E. Kimura *Topics Curr. Chem.* **1985**, *128*, 113-141. (b) E. Kimura, A. Sakonaka, T. Yatsunami, M. Kodama *J. Am. Chem. Soc.* **1981**, *103*, 3041-3045. (c) E. Kimura, A. Watanabe and M. Kodama *J. Am. Chem. Soc.* **1983**, *105*, 2063-2066.
4. (a) J. Cullinane, R. I. Gelb, R. N. Margulis and L. J. Zompa *J. Am. Chem. Soc.* **1982**, *104*, 3048-3053. (b) R. I. Gelb, B. T. Lee and L. J. Zompa *J. Am. Chem. Soc.* **1985**, *107*, 909-916.
5. (a) E. Graf and J.-M. Lehn *J. Am. Chem. Soc.* **1976**, *98*, 6403-6405. (b) B. Dietrich, J. Guilhem, J.-M. Lehn, C. Pascard and E. Sonveaux *Helv. Chim. Acta* **1984**, *67*, 91-104. (c) D. Heyer and J.-M. Lehn *Tetrahedron Lett.* **1986**, *27*, 5869-5872.
6. (a) M. W. Hosseini, J.-M. Lehn, L. Maggiora, K. B. Mertes and M. P. Mertes *J. Am. Chem. Soc.* **1987**, *109*, 537-544. (b) M. W. Hosseini, A. J. Blacker and J.-M. Lehn *J. Chem. Soc., Chem. Commun.* **1988**, 596-598.
7. J. F. Marecek and C. J. Burrows *Tetrahedron Lett.* **1986**, *49*, 5943-5946.
8. E. Kimura, M. Kodama and T. Yatsunami *J. Am. Chem. Soc.* **1982**, *104*, 3182-3187.
9. J. F. Marecek and C. J. Burrows, unpublished results with **2**.
10. (a) J. E. Richman, T. J. Atkins and W. F. Oettle *Org. Synth.* **1979**, *58*, 86-97. (b) B. K. Vriesema, J. Buter and R. M. Kellogg *J. Org. Chem.* **1984**, *49*, 110-113.
11. E. S. Ferinandi and G. Just *Can. J. Chem.* **1971**, *49*, 1070-1084.
12. Compound **1** was isolated as a white solid, mp 283-285°C (dec.), $[\alpha]_D$=+5.3° (C=1.3, H_2O), ^{1}H-NMR (300 MHz, D_2O) δ 1.6-1.95 (m, 12H), 2.9-3.3 (m, 10H), 3.5-3.66 (m, 1H), 3.63 (dd, 1H, J=6.9, 12.5 Hz), 3.79 (dd, 1H, J=3.8, 12.5 Hz), ^{13}C-NMR (75.4 MHz, D_2O) δ 26.22, 26.77, 26.85, 26.94, 27.18, 28.92, 48.72, 49.47, 49.60, 49.69, 49.79, 63.12, 65.28. Anal. Calcd for $C_{13}H_{32}Cl_3N_3O$: C, 44.25, H, 9.14, N, 11.91. Found: C, 43.91, H, 9.34, N, 11.61.
13. F. Ramirez and J. F. Marecek *Biochim. Biophys. Acta* **1980**, *589*, 21-29.

(Received in USA 22 August 1988)