

Inclusion of Quaternary Ammonium Compounds by Calixarenes^{*}

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Abstract. In weakly polar solvents, strong association occurs between calixarene anions and tetraalkylammonium cations, with the magnitude of the observed equilibrium constants depending upon the charge on the anion, the solvent, the ring size of the calixarene and the nature of the alkyl group of the cation. Large upfield shifts of the methyl resonances of the $[(\text{CH}_3)_4\text{N}]^+$ cation in solutions of $[(\text{CH}_3)_4\text{N}]_2[p\text{-}t\text{-butylcalix[6]arene} - 2\text{H}]$ indicate cation inclusion in a structure which is possibly identical with that found for the solid 'salt' by X-ray crystallography. This shows one of the cations to be included within a partial cone structure of a 'hinged 3-up, 3-down' conformation of the calixarene. The functionalised tetramethylammonium ions, choline and acetyl choline, are also strongly included by various calixarene anions but attempts to detect significant modification of the reactivity of acetyl choline resulting from inclusion have not been successful.

Key words: Calixarene anions, tetra-alkylammonium cations, association equilibria, inclusion, acetyl choline, catalysis.

1. Introduction

Thermodynamic [1], kinetic [2–4], spectroscopic [5, 6] and synthetic [7–9] studies have shown that calixarenes may be readily and sometimes extensively deprotonated by strong bases in nonaqueous solvents. Although solids containing six- and even seven-fold deprotonated calixarenes have been isolated as rare earth [7] or transition metal [8] ion complexes, solid salts of weakly or formally noncoordination cations are known only for degrees of deprotonation up to a maximum of three [9]. Obviously, these differences must in some way be associated with the nature of the cation–anion interactions occurring, and we have recently succeeded in characterising some aspects of these phenomena through structural studies of some alkali metal and tetraalkylammonium ion complexes [9–12]. It is apparent, as also suggested by theoretical calculations [13] and thermodynamic [1] measurements, that the negative charge on deprotonated calixarenes is delocalised and that cation

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binding in both solution and solid phases can involve inclusion provided there is appropriate dimensional matching [2–4, 9–12]. The occurrence of inclusion raises the prospect of conducting reactions with the calixarene as catalyst, especially if charge delocalisation is such that the nucleophilicity of the calixarene itself may be ignored, so that inclusion of an electrophile does not simply result in attack on this negative charge. A relatively simple electrophile of interest because, in its natural role, it undergoes binding of its cationic centre into a hydrophobic cavity, is acetylcholine [14, 15]. We describe here measurements of equilibrium binding of some tetraalkylammonium ions and both choline and acetylcholine by calixarene anions, and our attempts to characterise the reactivity of included acetylcholine. The structure of the *bis*(tetramethylammonium) salt of *p-t*-butylcalix[6]arene is also reported as a model for the binding of acetylcholine by such a calixarene.

2. Experimental

2.1. SYNTHESIS

The salts $[\text{Pr}_4\text{N}][\text{calix}[4]\text{arene} - \text{H}]$, $[\text{Et}_4\text{N}][p\text{-}t\text{-butyldihomooxalix}[4]\text{arene} - \text{H}]$, $[(\text{CH}_3)_4\text{N}]_2[p\text{-}t\text{-butylcalix}[6]\text{arene} - 2\text{H}]$, $[\text{Et}_4\text{N}]_2[p\text{-}t\text{-butylcalix}[6]\text{arene} - 2\text{H}]$, $[\text{Pr}_4\text{N}][p\text{-}t\text{-butylcalix}[6]\text{arene} - \text{H}]$ and $[\text{Pr}_4\text{N}]_2[\text{calix}[6]\text{arene} - 2\text{H}]$ ($\text{Et} = \text{CH}_3\text{CH}_2$, $\text{Pr} = \text{CH}_3\text{CH}_2\text{CH}_2$) were prepared as described previously [9]. Choline and acetylcholine perchlorates were precipitated by addition of NaClO_4 to aqueous solutions of the chlorides (Sigma Chemical Co.), and were recrystallised from methanol and from acetonitrile by the addition of ether, respectively. The previously unknown *bis*- and *tris*-(tetrapropylammonium) salts of *p-t*-butylcalix[6]arene, $[\text{Pr}_4\text{N}]_2[p\text{-}t\text{-butylcalix}[6]\text{arene} - 2\text{H}]$ and $[\text{Pr}_4\text{N}]_3[p\text{-}t\text{-butylcalix}[6]\text{arene} - 3\text{H}]$, were prepared by a slight variation of methods established for the other species, as follows.

[Pr₄N]₂[p-t-butylcalix[6]arene - 2H] Trihydrate

Approximately 1 mol L^{-1} methanolic $\text{Pr}_4\text{N}^+\text{OH}^-$ (4.0 mL) was added to a slurry of *p-t*-butylcalix[6]arene (1.93 g) in methanol (40 mL). The mixture was stirred vigorously for 60 min (without dissolution of the solid occurring) and the solvent was then evaporated under reduced pressure. The white residue was dissolved in CH_2Cl_2 (40 mL), the solution filtered and the product (1.75 g) precipitated as a white powder by the addition of hexane. Note that the attempts to recrystallise this material from hot methanol resulted in the deposition of $[\text{Pr}_4\text{N}][p\text{-}t\text{-butylcalix}[6]\text{arene} - \text{H}]$. *Analysis Calcd.* for $\text{C}_{90}\text{H}_{138}\text{N}_2\text{O}_6 \cdot 3\text{H}_2\text{O}$: C, 77.26; H, 10.37; N, 2.00. *Found*: C, 77.24; H, 9.88; N, 1.10%. Nitrogen analyses on these compounds were typically erratic and cation to anion ratios were more reliably established through integration of $^1\text{H-NMR}$ spectra.

[Pr₄N]₃[p-t-butylcalix[6]arene – 3H] Hexahydrate

Approximately 1 mol L⁻¹ methanolic Pr₄N⁺OH⁻ (6.5 mL) was added to a slurry of *p-t*-butylcalix[6]arene (1.93 g) in methanol (40 mL). The mixture was stirred vigorously for 10 min and the nearly clear solution produced was then filtered and the solvent evaporated under reduced pressure. The white residue was dissolved in CH₂Cl₂ (40 mL) and the product was precipitated as clusters of fine, white needles by the addition of hexane. These were collected, washed with hexane and dried in air, efflorescing in this process to a white powder (2.45 g). *Analysis Calcd.* for C₁₀₂H₁₆₅N₃O₆·6H₂O: C, 74.81; H, 10.89; N, 2.57. *Found:* C, 74.73; H, 10.68; N, 2.02%.

Choline Trifluoromethanesulphonate (Choline Triflate)

Choline chloride (5 g) and trifluoromethane sulphonic acid (CF₃SO₃H, 4 mL) were mixed together and heated on a steam bath for 5 min, giving a clear, viscous solution. This was cooled to room temperature and added, with stirring, to ether (200 mL). A white, crystalline precipitate formed immediately. It was recrystallised from ethanol by the addition of ether to the point of permanent turbidity and storage at 4°C for 24 h. *Analysis Calcd.* for C₆H₁₄F₃NO₄S: C, 28.46; H, 5.57; N, 5.53. *Found:* C, 28.39; H, 5.79; N, 5.38%.

2.2. EQUILIBRIUM MEASUREMENTS

2.2.1. *Solution Spectrophotometry*

Acetonitrile (Ajax Chemicals HPLC Grade) solutions of constant calixarene anion (host) concentration and varying tetramethylammonium ion, choline or acetylcholine (guest) concentrations were prepared by appropriate dilutions of 10⁻² mol L⁻¹ stock solutions of the tetrapropylammonium salts of the calixarenes, tetramethylammonium triflate, choline triflate and acetylcholine perchlorate. Spectra were recorded in the range 200–350 nm on a Hewlett-Packard 8452 Diode Array spectrophotometer. Nonlinear least-squares fitting of absorbance vs. composition data to models of the reaction system was conducted using a program written locally by Dr. E. S. Kucharski.

2.2.2. *NMR Spectroscopy*

Solutions in acetone-*d*₆ of known concentration close to 3 × 10⁻² mol L⁻¹ in choline or acetylcholine perchlorates were prepared and used as solvents for calixarene salts so as to obtain series of solutions in which the host : guest (calixarene : quaternary ammonium group) ratio varied between 0 and 2.00. Proton chemical shifts were measured for the trimethylammonium resonances of all solutions using a Bruker WP80 instrument. Inclusion equilibrium constants were determined from nonlinear least-squares fitting of the chemical shift vs. host : guest ratio curves.

2.3. CRYSTALLOGRAPHY

Recrystallisation of $[(\text{CH}_3)_4\text{N}]_2[p\text{-}t\text{-butylcalix[6]arene - 2H}]$ from acetonitrile provided a mixture of relatively large rhombs and some very fine needles. The structure determination was performed on a capillary-mounted specimen of the larger crystals.

2.3.1. Structure Determination

A unique diffractometer data set (Siemens P4; $2\theta_{\text{max}} 50^\circ$, $2\theta/\theta$ scan mode, filtered MoK_α radiation, $\lambda = 0.71073 \text{ \AA}$) was measured at 293(2) K, yielding 22347 independent reflections, 12637 of these with $I > 3\sigma(I)$ being considered 'observed' and used in the full-matrix least-squares refinement on F^2 . The structure solution was obtained using SHELXS-86 [16], refinement being carried out with SHELXL-93 [17]. Anisotropic thermal parameters were refined for the ordered nonhydrogen atoms. The positions of the hydrogen atoms on carbon were calculated from geometrical considerations and were refined in constraint with the bonded carbon atoms. Phenolic hydrogen atoms were located by difference Fourier syntheses and were refined in constraint with the bonded oxygen atoms. Positional parameters for the nonhydrogen atoms are given in Table I and some selected torsional angles are given in Table II. Tables of bond lengths, bond angles, hydrogen positional parameters, least-squares planes, and structure factor amplitudes are available from the authors. Views of one calixarene dianion/tetramethylammonium cations unit, showing the inclusion of one of the cations by the calixarene (and the atom numbering scheme used), are given in Figure 1. There is extensive disorder within the structure; three of the six *t*-butyl groups on each independent calixarene anion are disordered, as are three of four cations associated with these calixarene moieties, and the lattice solvent molecules (water and acetonitrile). Thermal motion is high, especially for the solvent molecules, so that the crystal appears to be bordering on the liquid state. The formulation (below) based on the structure solution shows that preparation for elemental analysis [9] results in considerable loss of solvent. The two independent calixarene species are both identified as a dianion on the basis of the detection of two protons in association with each group of three commonly oriented phenyl rings. The final indices of fit were $R_1 = 0.139$, $wR_2 = 0.356$ with weight = $[\sigma^2 F_0^2 + (0.1551P)^2 + 28.1194P]^{-1}$.

2.3.2. Crystal Data

$\text{C}_{76}\text{H}_{115}\text{N}_3\text{O}_9$ ($[(\text{CH}_3)_4\text{N}]_2[p\text{-}t\text{-butylcalix[6]arene - 2H}] \cdot \text{CH}_3\text{CN} \cdot 3\text{H}_2\text{O}$), $M = 1214.7$, triclinic, space group $P\bar{1}$, $a = 17.424(3)$, $b = 18.447(4)$, $c = 27.229(6) \text{ \AA}$, $\alpha = 82.10(2)$, $\beta = 98.27(3)$, $\gamma = 107.83(2)^\circ$, $V = 8204(3) \text{ \AA}^3$, D_c ($Z = 4$) = 0.983 g cm^{-3} , $F(000) = 2656$, $\mu_{\text{Mo}} = 0.63 \text{ cm}^{-1}$, specimen $0.65 \times 0.60 \times 0.20 \text{ mm}^3$.

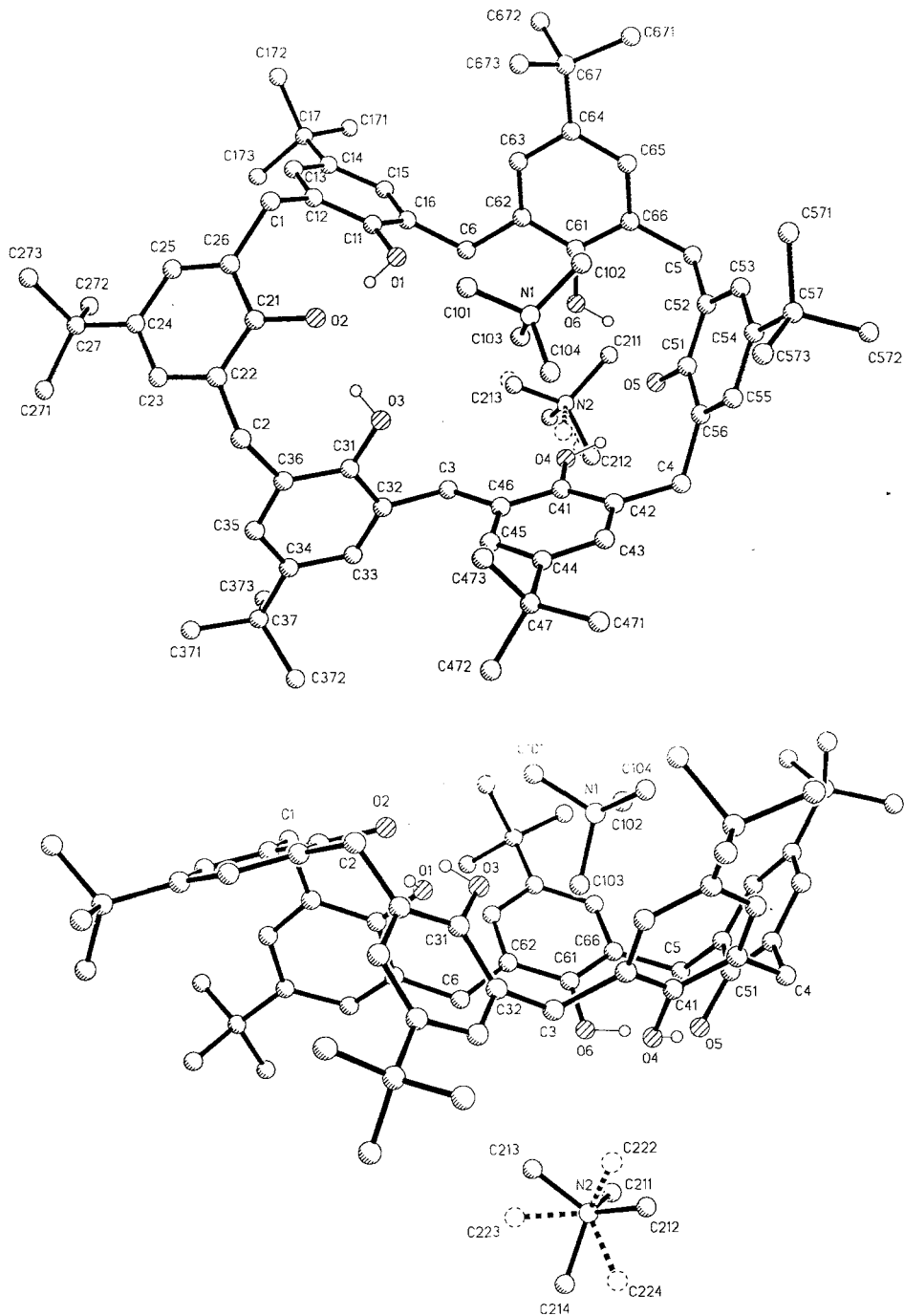


Fig. 1. Views of one of the two independent calixarene moieties in the unit cell of the salt $[(\text{CH}_3)_4\text{N}]_2[p\text{-}t\text{-butylcalix[6]arene} - 2\text{H}]$, in which an included cation is ordered and another associated but nonincluded cation is disordered over two configurations. Only the phenolic hydrogen atoms are shown.

TABLE I. Atomic coordinates/Å, equivalent isotropic displacement parameters/Å² and site occupancies (population parameters) for [N(CH₃)₄]₂[*p*-*t*-butylcalix[6]arene – 2H]. $U_{(eq)}$ is defined as one-third of the trace of the orthogonalised U_{ij} tensor.

Atom	<i>x</i>	<i>y</i>	<i>z</i>	$U_{(eq)}$	Occupancy
C(1)A	0.5990(6)	0.5377(5)	0.2965(4)	0.060(3)	1.0
O(1)A	0.4800(5)	0.4101(4)	0.3440(3)	0.082(2)	1.0
C(11)A	0.4988(6)	0.4042(6)	0.2981(4)	0.061(3)	1.0
C(12)A	0.5557(5)	0.4646(5)	0.2738(4)	0.053(2)	1.0
C(13)A	0.5700(6)	0.4542(6)	0.2263(4)	0.058(3)	1.0
C(14)A	0.5329(6)	0.3882(6)	0.2035(4)	0.063(3)	1.0
C(15)A	0.4786(6)	0.3296(6)	0.2304(4)	0.061(3)	1.0
C(16)A	0.4595(6)	0.3367(6)	0.2762(4)	0.063(3)	1.0
C(17)A	0.5495(6)	0.3812(7)	0.1503(4)	0.071(3)	1.0
C(171)A	0.5144(8)	0.3009(8)	0.1351(5)	0.109(5)	1.0
C(172)A	0.6389(8)	0.404(1)	0.1460(6)	0.142(7)	1.0
C(173)A	0.508(1)	0.432(1)	0.1127(5)	0.131(6)	1.0
C(2)A	0.4006(6)	0.6547(5)	0.3515(4)	0.057(3)	1.0
O(2)A	0.4890(4)	0.5443(4)	0.3634(2)	0.067(2)	1.0
C(21)A	0.4976(6)	0.5961(5)	0.3223(4)	0.056(2)	1.0
C(22)A	0.4543	0.6506(5)	0.3139(4)	0.057(3)	1.0
C(23)A	0.4627(6)	0.7012(5)	0.2707(4)	0.058(3)	1.0
C(24)A	0.5136(6)	0.7010(5)	0.2356(4)	0.057(3)	1.0
C(25)A	0.5560(6)	0.6476(5)	0.2465(4)	0.061(3)	1.0
C(26)A	0.5500(5)	0.5958(5)	0.2873(4)	0.054(2)	1.0
C(27)A	0.5221(8)	0.7571(7)	0.1890(4)	0.078(3)	1.0
C(271)A	0.468(1)	0.810(1)	0.183(1)	0.15(1)	0.67
C(272)A	0.502(1)	0.713(1)	0.1427(8)	0.14(1)	0.67
C(273)A	0.611(1)	0.808(1)	0.1893(7)	0.109(7)	0.67
C(274)A	0.442(2)	0.759(2)	0.166(2)	0.15(1)	0.33
C(275)A	0.567(3)	0.743(3)	0.155(2)	0.14(1)	0.33
C(276)A	0.1899(7)	0.3981(6)	0.3691(3)	0.065(3)	1.0
O(3)A	0.3472(4)	0.4947(4)	0.3886(3)	0.076(2)	1.0
C(31)A	0.2931(6)	0.5278(6)	0.3631(3)	0.057(3)	1.0
C(32)A	0.2116(6)	0.4840(5)	0.3545(3)	0.055(3)	1.0
C(33)A	0.1542(7)	0.5160(6)	0.3284(4)	0.071(3)	1.0
C(34)A	0.1750(7)	0.5937(6)	0.3100(4)	0.070(3)	1.0
C(35)A	0.2554(7)	0.6356(6)	0.3184(4)	0.066(3)	1.0
C(36)A	0.3150(6)	0.6060(5)	0.3452(3)	0.054(2)	1.0
C(37)A	0.1076(8)	0.6254(7)	0.2795(6)	0.098(4)	1.0
C(371)A	0.139(1)	0.7096(9)	0.2665(9)	0.22(1)	1.0
C(372)A	0.035(1)	0.606(1)	0.3088(8)	0.167(8)	1.0
C(373)A	0.078(1)	0.586(1)	0.2318(7)	0.20(1)	1.0
C(4)A	0.1398(7)	0.1796(6)	0.5042(4)	0.072(3)	1.0
O(4)A	0.1569(5)	0.2479(4)	0.4016(2)	0.072(2)	1.0

TABLE I. (Continued)

Atom	x	y	z	$U_{(eq)}$	Occupancy
C(41)A	0.1651(6)	0.2932(5)	0.4389(4)	0.056(3)	1.0
C(42)A	0.1581(6)	0.2648(6)	0.4883(4)	0.058(3)	1.0
C(43)A	0.1657(3)	0.3161(6)	0.5226(4)	0.068(3)	1.0
C(44)A	0.1806(7)	0.3941(6)	0.5100(4)	0.072(3)	1.0
C(45)A	0.1900(7)	0.4209(6)	0.4598(4)	0.071(3)	1.0
C(46)A	0.1822(6)	0.3712(6)	0.4244(4)	0.058(3)	1.0
C(47)A	0.188(1)	0.4504(8)	0.5486(5)	0.107(5)	1.0
C(471)A	0.176(1)	0.410(1)	0.6028(7)	0.16(1)	0.67
C(472)A	0.130(2)	0.500(2)	0.533(1)	0.18(1)	0.67
C(473)A	0.280(1)	0.507(1)	0.5541(8)	0.135(9)	0.67
C(474)A	0.096(2)	0.411(2)	0.571(2)	0.16(1)	0.33
C(475)A	0.186(4)	0.517(3)	0.528(2)	0.18(1)	0.33
C(476)A	0.239(3)	0.433(2)	0.589(2)	0.135(9)	0.33
C(5)A	0.3295(7)	0.0616(5)	0.4407(4)	0.072(3)	1.0
O(5)A	0.1961(4)	0.1237(4)	0.4271(2)	0.064(2)	1.0
C(51)A	0.2369(6)	0.1213(5)	0.4735(4)	0.058(3)	1.0
C(52)A	0.3033(6)	0.0918(5)	0.4827(4)	0.058(3)	1.0
C(53)A	0.3436(6)	0.0906(5)	0.5305(4)	0.068(3)	1.0
C(54)A	0.3231(7)	0.1201(6)	0.5695(4)	0.065(3)	1.0
C(55)A	0.2560(7)	0.1483(5)	0.5586(4)	0.063(3)	1.0
C(56)A	0.2129(6)	0.1488(5)	0.5124(4)	0.056(2)	1.0
C(571)A	0.456(1)	0.113(1)	0.6176(9)	0.16(1)	0.67
C(572)A	0.320(1)	0.047(1)	0.6536(8)	0.16(1)	0.67
C(573)A	0.375(1)	0.194(1)	0.6438(7)	0.119(7)	0.67
C(574)A	0.385(2)	0.028(2)	0.637(2)	0.16(1)	0.33
C(575)A	0.333(3)	0.128(3)	0.662(2)	0.16(1)	0.33
C(576)A	0.461(3)	0.166(2)	0.621(2)	0.119(7)	0.33
C(6)A	0.3968(6)	0.2739(5)	0.3020(4)	0.068(3)	1.0
O(6)A	0.2901(5)	0.1654(4)	0.3575(3)	0.076(2)	1.0
C(61)A	0.3691(7)	0.1705(6)	0.3720(4)	0.062(3)	1.0
C(62)A	0.4279(7)	0.2236(5)	0.3457(4)	0.063(3)	1.0
C(63)A	0.5087(7)	0.2312(6)	0.3590(4)	0.068(3)	1.0
C(64)A	0.5350(7)	0.1834(7)	0.3938(4)	0.072(3)	1.0
C(65)A	0.4754(8)	0.1301(6)	0.4236(4)	0.068(3)	1.0
C(66)A	0.3931(8)	0.1218(6)	0.4108(4)	0.068(3)	1.0
C(67)A	0.6240(8)	0.1923(8)	0.4100(5)	0.089(4)	1.0
C(671)A	0.641(1)	0.135(1)	0.4525(6)	0.140(6)	1.0
C(672)A	0.658(1)	0.179(1)	0.3632(6)	0.140(6)	1.0
C(673)A	0.6694(9)	0.2725(9)	0.4241(7)	0.137(6)	1.0
C(1)B	0.0964(6)	0.8994(5)	0.1678(4)	0.061(3)	1.0
O(1)B	0.1303(4)	1.0577(4)	0.1221(3)	0.068(2)	1.0
C(11)B	0.1842(6)	1.0381(5)	0.1594(3)	0.049(2)	1.0
C(12)B	0.1719(6)	0.9637(5)	0.1818(3)	0.54(2)	1.0

TABLE I. (Continued)

Atom	x	y	z	$U_{(eq)}$	Occupancy
C(13)B	0.2306(6)	0.9488(6)	0.2182(3)	0.058(3)	1.0
C(14)B	0.3020(6)	1.0037(6)	0.2340(4)	0.060(3)	1.0
C(15)B	0.3091(6)	1.0782(6)	0.2104(4)	0.061(3)	1.0
C(16)B	0.2521(6)	1.0956(5)	0.1739(3)	0.054(2)	1.0
C(17)B	0.3635(7)	0.9833(7)	0.2734(4)	0.082(3)	1.0
C(171)B	0.4391(9)	1.0534(9)	0.2839(6)	0.137(6)	1.0
C(172)B	0.389(1)	0.921(1)	0.2588(9)	0.23(1)	1.0
C(173)B	0.327(1)	0.959(1)	0.3219(5)	0.160(8)	1.0
C(2)B	-0.1715(6)	0.9586(6)	0.1356(4)	0.063(2)	1.0
O(2)B	-0.0156(4)	0.9699(3)	0.1094(2)	0.053(2)	1.0
C(21)B	-0.0350(6)	0.9339(5)	0.1549(3)	0.048(2)	1.0
C(22)B	-0.1097(5)	0.9285(5)	0.1709(3)	0.050(2)	1.0
C(23)B	-0.1260(6)	0.8963(5)	0.2186(4)	0.059(3)	1.0
C(24)B	-0.0708(7)	0.8668(6)	0.2518(4)	0.064(3)	1.0
C(25)B	0.0002(6)	0.8708(5)	0.2339(4)	0.058(3)	1.0
C(26)B	0.0184(6)	0.9017(5)	0.1868(3)	0.053(2)	1.0
C(27)B	-0.0928(7)	0.8344(6)	0.3052(4)	0.075(3)	1.0
C(271)B	-0.173(1)	0.7725(8)	0.3029(5)	0.134(6)	1.0
C(272)B	-0.098(1)	0.8987(7)	0.3325(4)	0.110(5)	1.0
C(273)B	-0.025(1)	0.804(1)	0.3357(5)	0.148(7)	1.0
C(3)B	-0.0426(6)	1.2448(6)	0.1191(4)	0.065(3)	1.0
O(3)B	-0.0489(4)	1.0976(4)	0.1037(3)	0.072(2)	1.0
C(31)B	-0.1099(6)	1.1034(6)	0.1273(3)	0.055(3)	1.0
C(32)B	-0.1099(6)	1.760(5)	0.1384(4)	0.057(3)	1.0
C(33)B	-0.1727(8)	1.1834(6)	0.1574(4)	0.079(3)	1.0
C(34)B	-0.2365(8)	1.1217(8)	0.1719(6)	0.087(4)	1.0
C(35)B	-0.2331(7)	1.0500(7)	0.1650(4)	0.082(4)	1.0
C(36)B	-0.1712(6)	1.0386(6)	0.1424(3)	0.056(3)	1.0
C(37)B	-0.309(1)	1.1313(9)	-0.1938(7)	0.131(6)	1.0
C(371)B	-0.338(2)	1.069(1)	0.2358(9)	0.138(9)	0.67
C(372)B	-0.379(2)	1.128(2)	0.150(1)	0.19(1)	0.67
C(373)B	-0.287(3)	1.212(1)	0.2138(9)	0.147(9)	0.67
C(374)B	-0.385(2)	1.053(2)	0.189(2)	0.138(9)	0.33
C(375)B	-0.329(3)	1.197(4)	0.177(2)	0.19(1)	0.33
C(376)B	-0.277(3)	1.122(3)	0.252(3)	0.147(9)	0.33
C(4)B	0.0744(6)	1.4365(5)	-0.0217(4)	0.074(3)	1.0
O(4)B	0.786(4)	1.365(4)	0.0805(2)	0.070(2)	1.0
C(41)B	0.0120(6)	1.3381(5)	0.0475(4)	0.057(3)	1.0
C(42)B	0.0056(6)	1.3713(5)	-0.0022(4)	0.055(2)	1.0
C(43)B	-0.0628(6)	1.3398(6)	-0.0331(4)	0.061(3)	1.0
C(44)B	-0.1267(6)	1.2779(6)	-0.0184(4)	0.063(3)	1.0
C(45)B	-0.1188(6)	1.2479(6)	0.0317(4)	0.060(3)	1.0
C(46)B	-0.0513(6)	1.2773(5)	0.0648(4)	0.053(2)	1.0

TABLE I. (Continued)

Atom	<i>x</i>	<i>y</i>	<i>z</i>	$U_{(eq)}$	Occupancy
C(47)B	-0.2018(7)	1.2431(8)	-0.0524(5)	0.088(4)	1.0
C(471)B	-0.202(1)	1.285(1)	-0.1051(8)	0.21(2)	0.67
C(472)B	-0.280(1)	1.240(2)	-0.0310(9)	0.16(1)	0.67
C(473)B	-0.207(1)	1.158(1)	-0.0582(9)	0.14(1)	0.67
C(474)B	-0.231(2)	1.311(2)	-0.077(2)	0.21(2)	0.33
C(475)B	-0.262(3)	1.186(3)	-0.026(2)	0.15(1)	0.33
C(476)B	-0.174(3)	1.212(3)	-0.094(2)	0.14(1)	0.33
C(5)B	0.3418(6)	1.3721(6)	0.0040(4)	0.065(3)	1.0
O(5)B	0.2146(4)	1.4132(4)	0.0400(2)	0.063(2)	1.0
C(51)B	0.2070(6)	1.4027(5)	-0.0086(3)	0.054(2)	1.0
C(52)B	0.2666(6)	1.3816(5)	-0.0290(4)	0.055(2)	1.0
C(53)B	0.2556(6)	1.3699(5)	-0.0781(4)	0.055(2)	1.0
C(54)B	0.1884(5)	1.3771(5)	-0.1112(3)	0.050(2)	1.0
C(55)B	0.1319(6)	1.4003(5)	-0.0906(4)	0.057(3)	1.0
C(56)B	0.1397(6)	1.4133(5)	-0.0410(4)	0.053(2)	1.0
C(57)B	0.1752(6)	1.3608(6)	-0.1652(4)	0.064(3)	1.0
C(571)B	0.0964(7)	1.2998(6)	-0.1761(4)	0.084(3)	1.0
C(572)B	0.2424(8)	1.334(1)	-0.1800(5)	0.127(6)	1.0
C(573)B	0.1719(8)	1.4329(7)	-0.2001(4)	0.103(5)	1.0
C(6)B	0.2636(6)	1.1775(6)	0.1505(4)	0.062(3)	1.0
O(6)B	0.2728(4)	1.3157(4)	0.0979(2)	0.066(2)	1.0
C(61)B	0.3019(5)	1.2703(6)	0.755(4)	0.054(2)	1.0
C(62)B	0.2981(5)	1.1976(5)	0.1011(3)	0.051(2)	1.0
C(63)B	0.3292(6)	1.1495(6)	0.0804(4)	0.058(3)	1.0
C(64)B	0.3637(6)	1.1694(6)	0.0350(4)	0.061(3)	1.0
C(65)B	0.3660(6)	1.2426(6)	0.0114(4)	0.065(3)	1.0
C(66)B	0.3363(5)	1.2932(6)	0.0305(3)	0.053(3)	1.0
C(67)B	0.3979(8)	1.1157(7)	0.0138(5)	0.087(4)	1.0
C(671)B	0.487(1)	1.160(1)	0.0020(8)	0.115(7)	0.67
C(672)B	0.346(1)	1.095(1)	-0.0365(8)	0.129(8)	0.67
C(673)B	0.399(1)	1.042(1)	0.0477(8)	0.13(1)	0.67
C(674)B	0.410(2)	0.129(2)	-0.038(2)	0.115(7)	0.33
C(675)B	0.309(2)	1.028(2)	0.010(2)	0.129(8)	0.33
C(676)B	0.449(3)	1.087(2)	0.051(2)	0.13(1)	0.33
N(1)	0.4313(5)	0.3455(4)	0.4797(3)	0.062(2)	1.0
C(101)	0.4966(9)	0.4140(7)	0.4658(5)	0.116(5)	1.0
C(102)	0.465(1)	0.2832(8)	0.4987(6)	0.133(6)	1.0
C(103)	0.3723(7)	0.3240(6)	0.4352(4)	0.081(3)	1.0
C(104)	0.391(1)	0.361(1)	0.5186(5)	0.143(7)	1.0
N(2)	0.0858(4)	0.1195(3)	0.2696(2)	0.080(3)	1.0
C(211)	0.1236(6)	0.0545(5)	0.2856(4)	0.115(5)	1.0
C(212)	0.0348(9)	0.1239(8)	0.3094(5)	0.115(7)	0.5
C(213)	0.1525(7)	0.1947(6)	0.2628(7)	0.143(9)	0.5

TABLE I. (Continued)

Atom	x	y	z	$U_{(eq)}$	Occupancy
C(214)	0.0321(9)	0.1047(8)	0.2205(5)	0.126(8)	0.5
C(222)	0.097(1)	0.1671(8)	0.3130(5)	0.115(7)	0.5
C(223)	0.126(1)	0.1698(9)	0.2259(6)	0.143(9)	0.5
C(224)	-0.0045(7)	0.0864(7)	0.2539(7)	0.126(8)	0.5
N(3)	-0.0793(3)	0.8441(3)	0.0071(2)	0.048(2)	1.0
C(311)	-0.1026(4)	0.7972(4)	-0.0373(2)	0.066(3)	1.0
C(312)	-0.1513(5)	0.9693(8)	0.0164(5)	0.075(5)	0.3
C(313)	-0.0090(7)	0.9147(6)	-0.0037(4)	0.066(5)	0.3
C(314)	-0.054(1)	0.7962(6)	0.0529(3)	0.056(4)	0.3
C(322)	-0.1300(7)	0.8991(7)	0.0018(4)	0.075(5)	0.3
C(323)	0.0099(5)	0.8891(8)	0.0090(5)	0.066(5)	0.3
C(324)	-0.094(1)	0.7911(5)	0.0548(3)	0.056(4)	0.3
C(332)	-0.079(1)	0.9259(5)	-0.0103(3)	0.075(5)	0.3
C(333)	0.0048(6)	0.8436(9)	0.0309(5)	0.0665(5)	0.3
C(334)	-0.1399(8)	0.8098(7)	0.0449(4)	0.056(4)	0.3
N(4)	0.2952(3)	1.4730(3)	0.2043(2)	0.075(2)	1.0
C(411)	0.2927(6)	1.4289(6)	0.2567(3)	0.169(7)	1.0
C(412)	0.310(1)	1.5587(5)	0.2089(5)	0.17(1)	0.3
C(413)	0.2136(7)	1.4421(9)	0.1727(4)	0.144(9)	0.3
C(414)	0.3640(9)	1.462(1)	0.1789(5)	0.105(7)	0.3
C(422)	0.271(1)	1.5463(6)	0.2051(5)	0.17(1)	0.3
C(423)	0.2356(9)	1.4222(7)	0.1666(4)	0.144(9)	0.3
C(424)	0.3817(6)	1.495(1)	0.1889(5)	0.105(7)	0.3
C(432)	0.240(1)	1.525(1)	0.1986(6)	0.17(1)	0.3
C(433)	0.265(1)	1.4158(7)	0.1646(4)	0.144(9)	0.3
C(434)	0.3838(6)	1.522(1)	0.1974(5)	0.105(7)	0.3
N(5)	0.115(2)	-0.134(2)	0.636(1)	0.167(8)	0.5
C(511)	0.112(3)	-0.096(3)	0.605(2)	0.167(8)	0.5
C(512)	0.116(3)	-0.051(3)	0.571(2)	0.167(8)	0.5
N(6)	-0.146(2)	1.399(1)	0.135(1)	0.094(5)	0.5
C(611)	-0.168(2)	1.393(2)	0.105(1)	0.094(5)	0.5
C(612)	-0.209(2)	1.383(2)	0.067(1)	0.094(5)	0.5
N(7)	0.067(1)	0.342(1)	0.2416(8)	0.093(4)	0.5
C(711)	0.014(2)	0.333(2)	0.255(1)	0.093(4)	0.5
C(712)	-0.055(2)	0.327(2)	0.272(1)	0.093(4)	0.5
N(8)	0.148(2)	0.590(2)	0.658(1)	0.121(6)	0.5
C(811)	0.200(2)	0.597(2)	0.683(1)	0.121(6)	0.5
C(812)	0.263(2)	0.608(2)	0.713(1)	0.121(6)	0.5
O(1W)	0.058(2)	1.483(2)	0.133(1)	0.171(9)	0.5
O(2W)	0.065(1)	0.002(1)	0.4140(8)	0.133(7)	0.5
O(3W)	-0.041(2)	0.315(2)	0.444(1)	0.21(1)	0.5
O(4W)	0.673(2)	0.228(2)	0.573(1)	0.23(1)	0.5
O(5W)	0.688(3)	0.300(3)	0.555(2)	0.25(2)	0.5

TABLE I. (Continued)

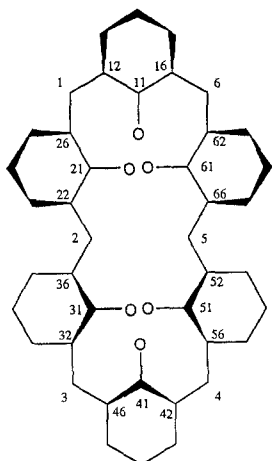
Atom	<i>x</i>	<i>y</i>	<i>z</i>	$U_{(eq)}$	Occupancy
O(6W)	-0.107(2)	1.415(2)	0.144(1)	0.20(1)	0.5
O(7W)	-0.145(2)	1.397(2)	0.161(1)	0.18(1)	0.5
O(8W)	0.302(2)	1.557(1)	0.0584(9)	0.127(9)	0.5
O(9W)	0.338(2)	1.548(1)	0.0660(9)	0.125(9)	0.5
O(10W)	0.385(1)	1.585(1)	0.0302(8)	0.130(7)	0.5
O(11W)	-0.081(3)	0.258(3)	0.486(2)	0.22(2)	0.3
O(12W)	0.091(3)	-0.043(3)	0.426(2)	0.19(2)	0.3
O(13W)	0.772(3)	0.283(3)	0.573(2)	0.19(2)	0.3

3. Results and Discussion

3.1. CRYSTALLOGRAPHY

The structural information available for calix[6]arene and its derivatives is relatively limited compared to that for both calix[4]- and calix[8]arene compounds [18–20]. Nonetheless, conformers which may be described as ‘pinched cone’ and ‘hinged 3-up, 3-down’ species have been well characterised [18, 19]. In both of these, the molecule may be regarded as providing two cavities of similar dimensions to the cup of cone-form calix[4]arene with, however, one of the four walls of the cup missing in each. The present derivative, $[(\text{CH}_3)_4\text{N}]_2[p\text{-}t\text{-butylcalix[6]arene} - 2\text{H}]$, provides another example of the hinged, 3-up, 3-down form, and inclusion of one of the two $[(\text{CH}_3)_4\text{N}]^+$ ions occurs in a manner which is similar to that observed in the simpler of the two inclusion moieties found in $[(\text{CH}_3)_4\text{N}]_2[\text{calix[4]arene} - \text{H}]_2 \cdot \text{calix[4]arene} \cdot \text{H}_2\text{O}$ [12] and is essentially as suggested by us previously on the basis of solution NMR measurements [9]. The two independent calixarene species in the unit cell differ principally in that the included cation is ordered in one (molecule A, Figure 1) and disordered in the other (molecule B); the nonincluded cations are disordered over two equally populated orientations. As in the structures determined for other tetra-alkylammonium ion complexes of calixarenes [9, 12], it is possible to ascribe the cation inclusion to $\text{CH}_3\text{-(phenyl)}\pi$ electron interactions assisted by cation–dipole attractions. Thus, although the included cation can be seen as embedded within a cone-like structure formed by three phenyl rings, placing the hydrogen atoms of at least one of the methyl groups in close proximity to the π electrons, the ‘open’ side of the cone does provide access to the presumably negative array of three oxygen atoms of the other half of the calixarene. The importance of optimising both factors may be the reason why the calixarene moiety does not have C_2 symmetry (with respect to the C(3)–C(6) axis) and show inclusion of both cations. The nitrogen atom of the included cation is significantly closer to the oxygen atoms which are not part of its inclusion cone than to the three which are (in molecule A, for example, $\text{N}(1) \cdots \text{O}(1)$, $\text{N}(1) \cdots \text{O}(2)$, $\text{N}(1) \cdots \text{O}(3)$ are 3.86, 4.47,

TABLE II. Torsion angles/ $^{\circ}$ defining calix[6]arene inner rings. Atom numbering follows the figure below; O(1) is attached to C(11), O(2) to C(21), etc.



Torsion	Molecule A	Molecule B	Eu ligand {21}
C(26)-C(1)-C(12)-C(11)	-84.8(11)	-73.6(11)	-84.8
O(1)-C(11)-C(12)-C(1)	0.6(14)	-2.1(14)	-2.8
C(16)-C(11)-C(12)-C(1)	-179.9(9)	178.2(8)	-177.4
O(1)-C(11)-C(16)-C(6)	1.7(14)	2.7(13)	0.0
C(12)-C(11)-C(16)-C(6)	-177.8(9)	-177.7(8)	-177.4
C(36)-C(2)-C(22)-C(21)	-89.2(11)	-96.6(10)	-96.7
O(2)-C(21)-C(22)-C(2)	2.9(14)	3.9(12)	4.8
C(26)-C(21)-C(22)-C(2)	-177.4(8)	-176.2(8)	178.3
C(12)-C(1)-C(26)-C(21)	91.1(11)	97.3(10)	93.9
O(2)-C(21)-C(26)-C(1)	-1.5(13)	-6.7(12)	-3.3
C(22)-C(21)-C(26)-C(1)	178.8(8)	173.4(8)	-176.5
C(46)-C(3)-C(32)-C(31)	79.2(12)	84.5(12)	70.0
O(3)-C(31)-C(32)-C(3)	-7.3(13)	-2.7(14)	1.9
C(36)-C(31)-C(32)-C(3)	173.8(8)	178.8(8)	178.0
C(22)-C(2)-C(36)-C(31)	82.1(12)	73.4(12)	82.5
O(3)-C(31)-C(36)-C(2)	3.2(14)	2.8(14)	0.9
C(32)-C(31)-C(36)-C(2)	-177.8(8)	-178.8(9)	-175.0
C(56)-C(4)-C(42)-C(41)	82.8(12)	85.6(12)	86.6
O(4)-C(41)-C(42)-C(4)	-0.4(15)	3.0(14)	1.4
C(46)-C(41)-C(42)-C(4)	-179.7(9)	-179.1(8)	-179.3
C(32)-C(3)-C(46)-C(41)	-177.9(9)	-175.8(8)	-177.3
O(4)-C(41)-C(46)-C(3)	0.0(13)	-0.3(13)	0.5
C(42)-C(41)-C(46)-C(3)	179.3(9)	-178.3(8)	-178.9
C(66)-C(5)-C(52)-C(51)	90.4(12)	89.6(11)	94.1
O(5)-C(51)-C(52)-C(5)	-0.5(13)	-1.2(13)	5.9
C(56)-C(51)-C(52)-C(5)	179.8(8)	177.9(8)	-173.5
C(42)-C(4)-C(56)-C(51)	-90.4(12)	-89.0(11)	-96.3
O(5)-C(51)-C(56)-C(4)	1.8(13)	-0.3(13)	-4.4
C(52)-C(51)-C(56)-C(4)	-178.5(8)	-179.4(8)	175.1
C(11)-C(16)-C(6)-C(62)	-76.8(13)	-82.8(13)	-72.1
C(16)-C(6)-C(62)-C(61)	174.3(9)	173.4(8)	172.7
O(6)-C(61)-C(62)-C(6)	1.1(14)	-0.6(12)	4.9
C(66)-C(61)-C(62)-C(6)	178.0(9)	177.4(8)	179.6
C(52)-C(5)-C(66)-C(61)	-83.6(13)	-83.3(12)	-79.0
O(6)-C(61)-C(66)-C(5)	-2.8(15)	-0.6(13)	-3.0
C(62)-C(61)-C(66)-C(5)	-179.6(9)	-178.5(8)	-177.5

3.93 Å, respectively, whereas N(1)··O(4), N(1)··O(5), N(1)··O(6) are 4.85, 5.03, 4.88 Å, respectively) and, interestingly, the nitrogen atom of the nonincluded cation approaches the inclusion-cone array of oxygens in a very similar way to that in which the included cation nitrogen approaches the noninclusion-cone oxygens (in molecule A again, N(2)··O(5), N(2)··O(6), N(2)··O(6) are 4.40, 4.44, 3.92 Å, respectively).

Detailed comparison of the calixarene conformations observed in the present compound and in a 'true' metal complex of Eu(III) [21] where the cation appears to bind through a single coordinate bond can be made in terms of the macrocycle torsion angles given in Table II. It is apparent that the calixarene, which is formally dianionic in all three situations, also has very closely similar conformations in all. Flexibility and the presence of numerous energy minima associated with closely similar configurations are properties expected of the larger calixarenes in particular [13, 20, 22], so that it is somewhat remarkable that greater differences are not observed, although it is true that in the present structure phenoxide-oxygen separation indicate significantly higher symmetry than in the europium complex. Thus within the separate 'tripods' of the calixarene entities in the tetramethylammonium salt, the separation of adjacent oxygen atoms are O(2)··O(1) 2.57 Å (molecule A), 2.57 Å (molecule B), O(2)··O(3) 2.52 Å (molecule A), 2.57 Å (molecule B), and O(5)··O(4) 2.57 Å (molecule A), 2.61 Å (molecule B), O(5)··O(6) 2.57 Å (molecule A), 2.57 Å (molecule B), while the corresponding separations in the europium complex are 2.77, 2.76 Å (Eu end) and 2.51, 2.65 Å. Nonetheless, it has also been observed in calix[8]arene systems that the macrocycle conformation is seemingly more sensitive to the formal charge on the ligand than to the particular nature of the bound cation [8, 23, 24], and in this regard it is interesting to note that in $[\text{Cl}_3\text{TiOTiCl}_2]_2[p\text{-}t\text{-butylcalix[6]arenetetramethylether} - 2\text{H}]$ [25] the calixarene again has the hinged conformation, whereas in $[\text{Ti}_4\text{O}_2][p\text{-}t\text{-butylcalix[6]arene} - 6\text{H}]_2$ [26] both calixarene moieties are in the cone form. Further, it has recently been shown that the substituent conformation in a calix[4]arene derivative is remarkably sensitive to the ionisation of one phenolic group [27].

3.2. SOLUTION CHEMISTRY

A species depositing from a given solution may not necessarily be that which is most abundant in solution, and in the case of tetraalkylammonium salts of calixarenes it is apparent that large solubility increases (seemingly regardless of solvent over a large range of polarity) associated with increasing degrees of deprotonation can cause salts of the singly-charged anions to deposit even when those anions are minor components of the solution mixtures. Thus, in our first syntheses [9, 12], calixarene monoanion salts were frequently obtained from solutions containing quite large excesses of tetraalkylammonium hydroxide. Nonetheless, the nature of the isolated species is obviously determined by a variety of factors other than mere relative solubilities. We have, for example, been able to readily isolate salts

presumed to contain the *p-t*-butylcalix[6]arene trianion even though equilibrium measurements [1] in benzonitrile indicate that it is difficult to remove more than two protons from this calixarene, and in the case of calix[4]arene [12], the isolated solid containing the tetramethylammonium salt of the monoanion also contains the unionised calixarene. The full complexity of this chemistry remains to be established but from the present and earlier work, a useful variety in stoichiometry is available.

The different solubility characteristics of the various calixarene salts presently studied made comparison of their properties in a common solvent difficult. A mixture of one volume (CD₃)₂CO to ten volumes of CDCl₃, however, does provide a solvent suitable for at least qualitative study of the inclusion characteristics of most by ¹H-NMR spectroscopy. For the tetrapropylammonium salts of calix[4]-, *p-t*-butylcalix[4]-, *p-t*-butyldihomooxalix[4]-, *p-t*-butylcalix[6]- and *p-t*-butylcalix[8]arene monoanions and of *p-t*-butylcalix[6]- and *p-t*-butylcalix[8]arene di- and trianions, the various propyl group resonances were little shifted, if at all (but see below), from those of [Pr₄N]Br in this solvent, indicating that the [Pr₄N]⁺ cation is not included to a marked degree in any case. When choline triflate was added to these solutions, near-quantitative precipitation of the choline cation/calixarene anion salt occurred with calix[4]arene and *p-t*-butylcalix[4]arene, but, in all the other systems a solution was obtained for which the quaternary ammonium methyl resonance was shifted ~1.5–2 ppm upfield from the position in the absence of the calixarene anion. Thus, choline inclusion was presumed to be strongly favoured in all these cases. (Similar observations were made when tetramethylammonium triflate was used in place of the choline salt, but greater problems were experienced with product precipitation. Interestingly, close comparison of the *N*-propyl group resonances before and after addition of choline triflate revealed some subtle differences between the various systems, including those which formed precipitates. Small shifts were in fact detectable for both the NCH₂ and terminal methyl resonances, and these varied in both magnitude and direction. For all the anions derived from *p-t*-butylcalix[6]arene and *p-t*-butylcalix[8]arene, small (~0.1–0.2 ppm) downfield shifts appeared to result from choline inclusion, with the shift being greater for the methylene than the methyl protons except for the trianion of *p-t*-butylcalix[6]arene, where the methylene shift was barely detectable. In any case, these shifts are possibly indicative of a rather unfavourable inclusion equilibrium involving the simple [Pr₄N]⁺ salts, an equilibrium which may involve two distinct forms, one where the N⁺ centre can be regarded as inserted (so placing NCH₂ protons close to aromatic π electrons) and one where the terminal methyl group is simply inserted. For the tetrapropylammonium salts of *p-t*-butyldihomooxalix[4]arene and calix[4]arene, addition of choline triflate resulted in very small (~0.05 ppm) downfield shifts of the methyl proton resonances only, perhaps indicating a very unfavourable inclusion equilibrium involving these methyl groups in the original solution. For the tetrapropylammonium (and the tetraethylammonium) salt of *p-t*-butylcalix[4]arene monoanion,

a unique situation appears to prevail in that addition of choline triflate resulted in small (~ 0.1 ppm) upfield shifts of all the propyl resonances. Since there is evidence (see below) that the *t*-butyl groups inhibit inclusion by this calixarene, this observation may indicate that ion association in the original solution placed the cation in close proximity to the electronegative phenol/phenolate oxygen atoms.

For the salts of the smaller calixarenes, acetone alone was a far superior solvent to acetone/chloroform mixtures, though it was generally apparent that cation inclusion was much weaker in acetone, perhaps as a result of competition for inclusion by the solvent itself [19, 28]. The solubilities of the monoanions of both *p-t*-butylcalix[4]arene and calix[4]arene in the presence of choline and acetylcholine cations were sufficiently high for NMR measurements to be readily made without interference from precipitation and so enabled demonstration of very marked selectivity in the interactions of these two calixarenes with either cation. Thus there was in fact no evidence at all of inclusion of either cation by *p-t*-butylcalix[4]arene, whereas the equilibrium constants (Table III) determined by fitting the marked upfield shifting of $N(^+)CH_3$ resonances in the presence of various amounts of calix[4]arene monoanion to a model of 1 : 1 complexation were so large as to indicate that under the given experimental conditions, effectively all cation that could be included was. The two calixarene anions appeared to be equally discriminatory towards the tetramethylammonium cation, though the solubilities of their salts with this cation were too low to allow extensive measurements, and it is worth noting that when the isolated salts were dissolved in $(CD_3)_2SO$, the $N(^+)CH_3$ resonances for both appeared slightly upfield (~ 0.2 ppm) of that for $[N(CH_3)_4][CF_3SO_3]$ in the same solvent. Nonetheless, unless acetone solvent inclusion is very much more favourable for *p-t*-butylcalix[4]arene than for calix[4]arene monoanion, it would appear that this is an instance of where inclusion by a calix[4]arene is inhibited rather than facilitated [19] by the addition of *t*-butyl substituents.

The binding of choline by calix[4]arene monoanion in acetone is appreciably stronger than that of acetylcholine, and the same relative affinity is seen for *p-t*-butyldihomooxalix[4]arene monoanion (Table III). Note that the absolute values of the equilibrium constants for these two calixarene anions are not directly comparable due to the use of the tetraethylammonium salt of the latter species, in which some inclusion actually occurs initially. Since *p-t*-butyldihomooxalix[4]arene is known to be sufficiently capacious to include two small molecules [28], it was considered that its monoanion might act as a catalyst for acetylation reactions involving acetylcholine, despite the fact that favourable binding of the choline product might be expected to ultimately inhibit reaction otherwise accelerated by the placement of reactants in close proximity [29]. No reaction in which such catalysis or even stoichiometric acetyl transfer can be observed has yet been found, however. The acetylation of benzylamine by acetylcholine, for example, is, if anything, slightly inhibited in the presence of *p-t*-butyldihomooxalix[4]arene.

The apparently quite large values for inclusion equilibrium constants determined by NMR spectroscopy indicated that electronic spectroscopy might well be a more

TABLE III. Association equilibrium constants for calixarene anions and quaternary ammonium cations.

Cation	Anion	Calixarene ⁿ⁻ + Cation ⁺ \rightleftharpoons [Calixarene·Cation] ⁽ⁿ⁻¹⁾⁻	Solvent	Method	K/M ⁻¹
Choline	[<i>p</i> - <i>t</i> -butyl/dihomooxacalix[4]arene - H] ⁻	[<i>p</i> - <i>t</i> -butyl/dihomooxacalix[4]arene - H] ⁻	Acetone	¹ H-NMR titration	(2.50 ± 0.37) × 10 ^{3a,c}
Choline	[calix[4]arene - H] ⁻	[calix[4]arene - H] ⁻	Acetone	¹ H-NMR titration	(5.1 ± 2.7) × 10 ^{2a,c}
Acetylcholine	[<i>p</i> - <i>t</i> -butyl/dihomooxacalix[4]arene - H] ⁻	[<i>p</i> - <i>t</i> -butyl/dihomooxacalix[4]arene - H] ⁻	Acetone	¹ H-NMR titration	(8.5 ± 2.2) × 10 ^{2a,c}
Acetylcholine	[calix[4]arene - H] ⁻	[calix[4]arene - H] ⁻	Acetone	¹ H-NMR titration	(2.55 ± 10 ²) ^{a,c}
Acetylcholine	[<i>p</i> - <i>t</i> -butyl/calix[6]arene - 3H] ³⁻	[<i>p</i> - <i>t</i> -butyl/calix[6]arene - 3H] ³⁻	Acetonitrile	Spectrophotometric titration	(3.3 ± 2.1) × 10 ^{5b,c}
[N(CH ₃) ₄] ⁺	[<i>p</i> - <i>t</i> -butyl/calix[6]arene - 3H] ³⁻	[<i>p</i> - <i>t</i> -butyl/calix[6]arene - 3H] ³⁻	Acetonitrile	Spectrophotometric titration	(3.22 ± 0.71) × 10 ^{5b,c}
[N(CH ₃) ₄] ⁺	[<i>p</i> - <i>t</i> -butyl/calix[6]arene - 2H] ²⁻	[<i>p</i> - <i>t</i> -butyl/calix[6]arene - 2H] ²⁻	Acetonitrile	Spectrophotometric titration	(1.72 ± 0.39) × 10 ^{4b,c}

^a 303 ± 1 K^b 298.2 ± 0.1 K^c 1σ errors.

appropriate method for use in more extensive measurements involving more highly charged anions [30]. Again, however, problems arose in the choice of an appropriate solvent and ultimately in the sensitivity of the method. Acetonitrile is a relatively good solvent for all the compounds presently studied and is also convenient for measurements in the UV region but it is, of course, well known for its tendency to form inclusion complexes with calixarenes [19]. This may well be the reason that for the tetrapropylammonium salts of both *p-t*-butylcalix[4]arene and calix[4]arene monoanions, no effect of even a large molar excess of tetramethylammonium triflate on the UV spectra was discernible. For other anions, changes could be detected but were generally only just outside experimental errors unless a 50-fold or greater molar amount of $[(\text{CH}_3)_4\text{N}]^+$ was added. Indeed, only for the trianion of *p-t*-butylcalix[6]arene could reproducible effects of the addition of a similar quantity of tetramethylammonium ion (or acetylcholine) be determined and associated with an equilibrium constant for a 1 : 1 association (Table III), presumed to be an inclusion process. Surprisingly, the effects of choline addition, though giving rise to absorbance changes at least as great as those resulting from tetramethylammonium or acetylcholine ion additions, were not associated with well-defined isosbestic points (as the others were), and indicated that at least two new species were formed as the ratio of cation to anion varied between 0 and 1. Obviously, many complications may arise in these systems if, for example, inclusion results in the alteration of phenolic proton distributions or calixarene conformations, and more detailed measurements are in progress in an attempt to unravel these complexities. Only limited quantitative data on the binding of cations by uncharged calixarene derivatives are to be found in the literature [31], and any comparison with the present results is vitiated by the different solvent systems involved, though it is interesting to note the observation that cation binding by neutral calixarenes is also inhibited by the presence of *t*-butyl substituents.

Inclusion of acetylcholine by at least the di- and trianions of *p-t*-butylcalix[6]arene is clearly indicated by $^1\text{H-NMR}$ spectroscopy for acetone, dimethylsulphoxide and chloroform solvents and is presumed to be the cause of spectroscopic changes in acetonitrile. If the guest is bound in the same way as is the included cation in solid $[\text{N}(\text{CH}_3)_4]_2[\textit{p-t}$ -butylcalix[6]arene - 2H], then the acetyl carbon electrophile would be brought into close proximity to a nucleophilic phenolate group. Further, in the incompletely deprotonated calixarene, adjacent acidic phenolic protons are available to facilitate alkoxide group departure. Figure 2 shows a possible structure for an acetylcholine : *p-t*-butylcalix[6]arene dianion inclusion complex obtained by using the crystal structure coordinates reported here and modifying the cation (to convert it to acetylcholine) in the molecular modelling program 'Chem3D+' [32]. In this configuration, the separation between the acetyl carbon and the closest phenolic oxygen is 1.5 Å. Despite this further indication that inclusion of acetylcholine by a calixarene may provide a means of accelerating reaction, we have been unable to detect formation of the monoacetate of *p-t*-butylcalix[6]arene, even after extended periods of reaction in various

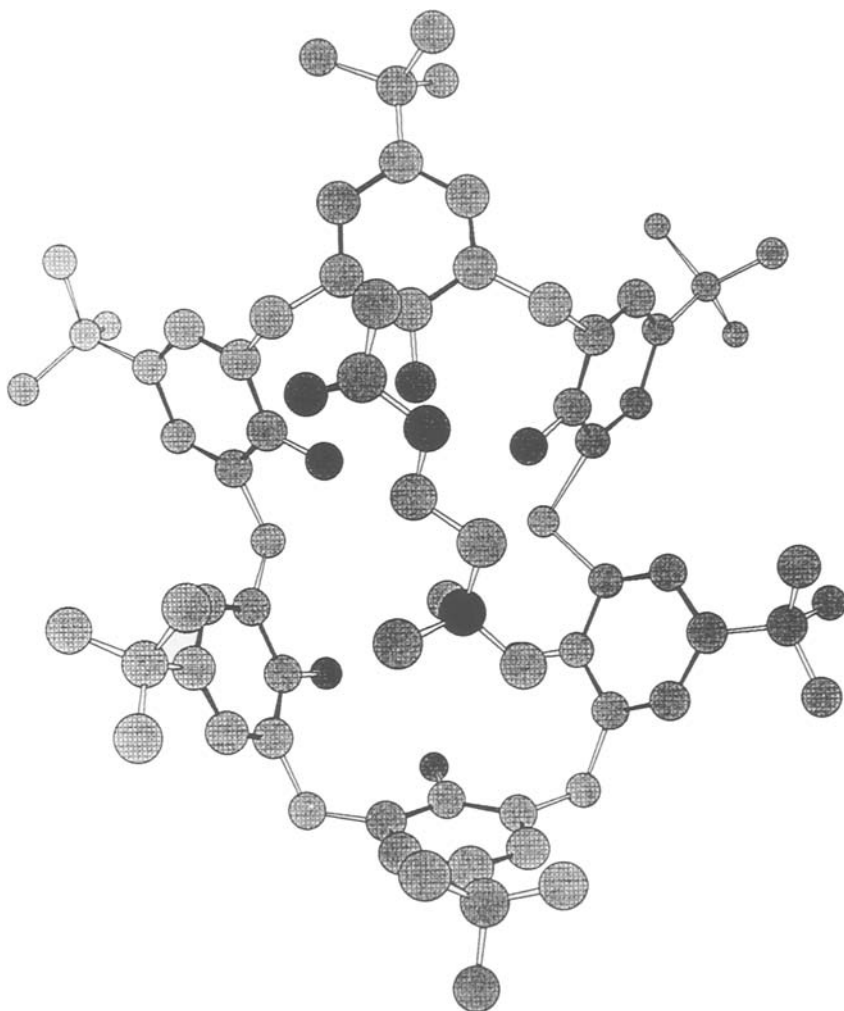


Fig. 2. A possible configuration for the inclusion complex formed between acetylcholine and the dianion of *p*-*t*-butylcalix[6]arene. (Viewed from 'above'.)

solvents. Thus, either the calixarene's oxygen nucleophilicity is too low for reaction to be favourable, or the inclusion complex adopts a conformation, such as the cone, which places the acetyl group remote from any nucleophilic centre. In regard to the latter point, it may be noted that although inclusion of acetylcholine is most easily monitored through the large chemical shift changes occurring for the $N^{+}CH_3$ resonances, the acetyl methyl also shows small upfield shifts, which could be indicative of simultaneous inclusion of this group within the half of a calix[6]arene cone not occupied by a *N*-methyl group. It is also worthy of note that it seems unlikely that proton transfer may be inhibitory, in that an incidental observation during the present work was that dissolution of the di- and trianionic

species in CDCl_3 resulted in the growth of a signal due to CHCl_3 over 10–20 min at room temperature.

Overall, the present observations on acetylcholine support the notion that inclusion by $\text{CH}_3/\text{aromatic-}\pi$ -electron interactions is an effective binding mechanism [14, 15] but they provide little information on how reaction subsequent to binding may be enhanced in an enzyme such as acetylcholine esterase. The calixarene framework does, however, provide a means to bring other nucleophiles into proximity with an included species and to attach activating units such as metal ions, and we are continuing to attempt to develop a more sophisticated model of an acylating enzyme. A recent report [33] of the development of an active acylation catalyst on the basis of principles similar to those described above vividly illustrates the complexities which may be encountered in this task.

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