Supramolecular stabilization of hydroxylamine TEMPOH by complexation with an amphiphilic calixarene†‡

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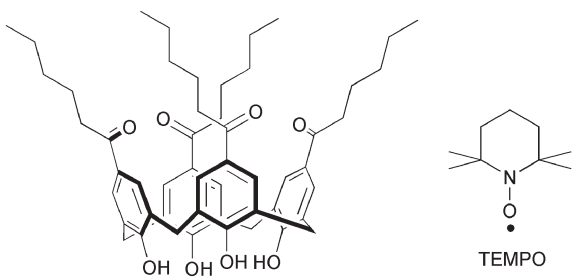
In an ethanol/water mixture, the nitroxyl radical TEMPO abstracts a hydrogen atom from a phenolic OH group of the amphiphilic *para*-hexanoylcalix[4]arene, and the hydroxylamine TEMPOH formed yields a stable inclusion complex with another molecule of the calixarene.

The amphiphilic calixarenes, particularly *para*-hexanoyl calix[4]-arene (Scheme 1),[1] belong to one of the major groups of synthetic macrocyclic hosts in supramolecular chemistry. It was shown previously that the conversion of ordinary calixarenes into their amphiphilic (lipidic) derivatives is straightforward[1b,d] and that they can be obtained as solvent-free solids.[1c] Interaction with a guest in an appropriate solvent leads to inclusion complexes of different morphologies.[2,3] Aiming to analyse the main peculiarities in the formation of various complexes, we attempted to trap the stable nitroxyl radical TEMPO (Scheme 1) into the *para*-hexanoyl calix[4]arene cavity. However, crystallization of the expected complex from an ethanol solution containing the components was not immediate and required the presence of some water and two weeks of waiting.§ A complex precipitated as long colourless needles, and proved to be stable in air for at least several days.

A single crystal X-ray diffraction study¶ revealed that the complex crystallizes in the monoclinic $P2_1/c$ crystal system with unit cell parameters: 10.46, 22.04 and 24.90 Å, and 95.87°, and consists of calixarene, hydroxylamine TEMPOH (*i.e.* the reduced form of TEMPO), EtOH and water (Fig. 1) in equimolar ratios. All components of the complex are ordered, except for EtOH, which is outside the cavity and is disordered over three positions,



Scheme 1

providing hydrogen bonding with two neighbouring C=O groups of the calixarene (*ca.* 72, 14 and 14% occupancy, respectively). The acyl groups of the calixarene chains are turned towards the EtOH (the left hand side in Fig. 1), opening the container from the right hand side (of Fig. 1). All protons shown in Fig. 1 (white) were solved by direct methods during the refinement, and hence, this confirms the formation of stable hydrogen bonds between the host and guests. The system of hydrogen bonds includes: phenolic-O···H–O–N (1.70 Å), N–H···O–H_2 (1.88 Å), H–O–H···O=C (1.93 Å), as well as H–O–H···OH-phenolic (1.98 Å). In addition, the calixarene is stabilized in a cone conformation by hydrogen bonding between the phenolic-OH groups, but in this case only 3 of the possible 4 hydrogen bonds are present. The fourth hydrogen atom has shifted completely to the oxygen atom of the hydroxylamine. The nitrogen in the hydroxylamine is protonated and has a pyramidal structure (O–N–H angle is 107.6°), which agrees with literature data.[4]

The total number of protons located during the refinement confirmed the expected electroneutrality of the complex. The diamagnetic nature of the complex was confirmed by NMR spectroscopy. In particular, the white solid was dissolved in a N_2 atmosphere in deuterated acetone purged with N_2. The 400 MHz

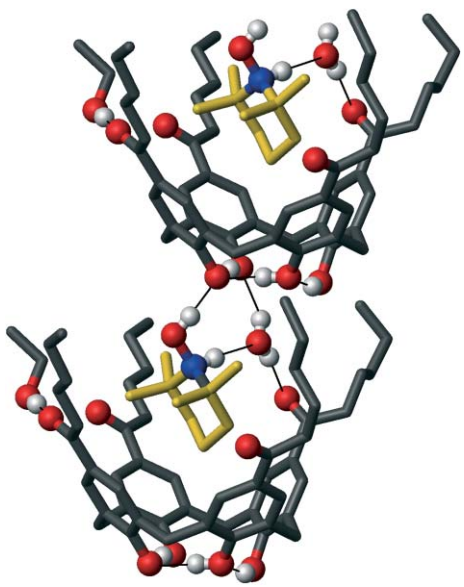


Fig. 1 X-Ray structure of the complex of *para*-hexanoylcalix[4] arene with TEMPOH, EtOH and water. Only the ethanol with the largest occupation number is shown for simplicity. Thin solid lines denote the hydrogen bonds.

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† Electronic Supplementary Information (ESI) available: Crystal structure parameters and data tables. See DOI: 10.1039/b511810g
‡ In memoriam Professor Hanns Fischer

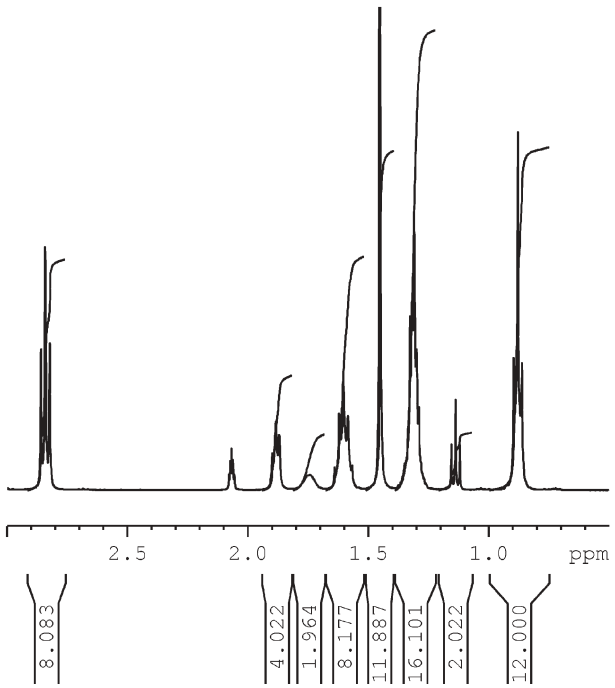

Fig. 2 A fragment of the 400 MHz ^{1}H-NMR spectrum of a solution, in deuterated acetone, of the complex of *para*-hexanoylcalix[4]arene with TEMPOH, EtOH and water.

^{1}H-NMR spectrum shows the presence of all components of the complex (Fig. 2). The signals of hydroxylamine TEMPOH can be easily seen in the spectrum: 1.45 ppm from the 4 × CH_3 groups, and 1.88 and 1.74 ppm from the protons of the piperidine ring. The ratio of the integrals of the host (4 × CH_3 at 0.88 ppm, 4 × $2CH_2$ at 1.31 ppm, and 4 × CH_2 and 4 × CH_2 at 1.60 and 2.84 ppm, respectively) to those of TEMPOH (4 × CH_3 at 1.45 ppm or 2 × CH_2 at 1.88 ppm) is close to 1, confirming the composition of the complex. The molar ratio of EtOH (CH_3 at 1.14 ppm) in the complex is smaller than 1, probably because of a partial loss of the guest during storage of the complex. Upon keeping the solution in the open air, the signals of TEMPOH slowly disappeared, the colour of the solution turning reddish and the lines in the spectrum becoming broader, unambiguously confirming the oxidation of TEMPOH by atmospheric oxygen back to the free nitroxide.

TGA and DSC traces show that the complex is relatively stable at room temperature, then slowly ejects water and ethanol between 30 and 100 °C, and melts with decomposition at *ca.* 105 °C (Fig. 3). The presence of a water molecule is essential for complex formation and stability as it provides an additional hydrogen bond linkage between the host and the guests, and hence stabilizes the entire system.

The mechanism for formation of the complex is not yet fully clear, but some assumptions based on its structure can be made. We observed that the solubility of the calixarene in an ethanol–TEMPO mixture is much higher than in ethanol alone. This is probably due to the formation of hydrogen bonds between the nitroxide and phenolic-OH group of the calixarene.[5,6] It is also known[7–8] that TEMPO can reversibly abstract a hydrogen atom from phenolic-OH groups to form the corresponding hydroxylamine TEMPOH. Since the complex has been obtained

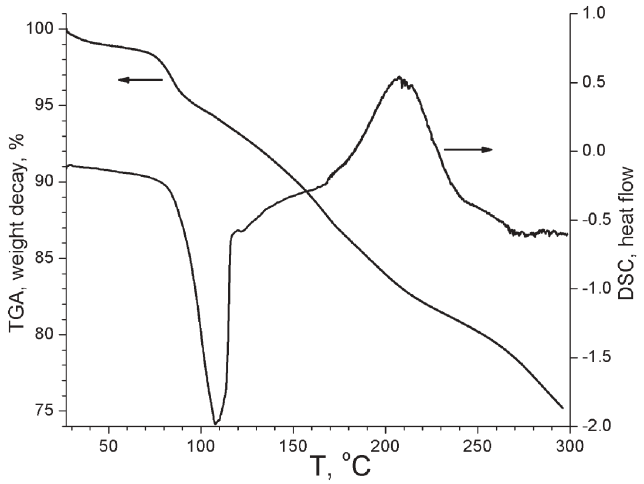

Fig. 3 TGA and DSC traces of the complex of *para*-hexanoylcalix[4]-arene with TEMPOH, EtOH and water.

from ethanol, the most likely mechanism is that of sequential proton loss electron transfer (SPLET).[9] The weak base formed, TEMPOH, abstracts a proton from another calixarene molecule,[10] yielding the corresponding salt $[TEMPO(H)H]^+$ $[calixarene(-H)]^-$. The latter forms the inclusion complex involving water, and precipitates. It is not necessary that the hydrogen atom connected to the O-atom of TEMPOH comes from the neighbouring calixarene, although the structure of the complex allows one to make such a conclusion.

Of course, other reaction mechanisms cannot be completely ruled out, such as (i) conventional H-atom abstraction[7,8,11] or (ii) protonation of the free nitroxide to form a radical cation, followed by electron transfer from the phenolate.[8] The first mechanism can be competitive with SPLET but the second one seems to be less probable because of the extremely low concentration of protonated TEMPO in weakly acidic solutions.[12] We are currently investigating the mechanisms of formation of such inclusion complexes by the variation of nitroxide, calixarene and reaction conditions.

In summary, reduction of nitroxyl radicals to their corresponding hydroxylamines, followed by the trapping and stabilizing of the reaction products, allows control over the composition of the reaction mixture and the reactivity of its components. Taking into account that (i) systems involving nitroxide–phenol pairs[5,7–9] are often considered as models to study the antioxidant activity of phenolic compounds,[13] (ii) nitroxides can be employed in magnetic resonance imaging experiments,[14] and (iii) amphiphilic calixarenes can easily form solid lipid nanoparticles[15] that can serve as prospective delivery systems, one can conclude that the combination of these factors gives the opportunity for fine tuning of the properties of encapsulated species in order to monitor their transport and to match biological targets.

Notes and references

§ 200 mg (0.24 mmol) of calixarene was placed in a solution of 1 g (6.4 mmol) of TEMPO in 8 ml of 96% EtOH and stirred with boiling until the solution became homogeneous. The solution was left at room temperature for crystallization to occur. The solvent was allowed to slowly evaporate for 5 days. The first crystals appeared after 10 days, and 42 mg (17%) of the complex was isolated after 2 weeks.

¶ The single crystal structure studies were performed on a Bruker SMART diffractometer with Mo-Kα radiation. Crystal data: $C_{63}H_{91}NO_{11}$, M =

1038.42, monoclinic, space group $P2_1/c$ (no. 14), $Z = 4$, $a = 10.457(2)$, $b = 22.041(4)$, $c = 24.899(4)$ Å, $\beta = 95.870°$, $V = 5708.8(17)$ Å^3, $T = 125(2)$ K, μ(Mo-Kα) = 0.081 mm^{-1}, 70995 reflections measured, 15946 unique (R_{int} = 0.043) which were all used in calculations. The final $wR(F2)$ was 0.080. The structure was solved by direct methods and refined by full matrix least-squares analysis (SHELXL-97). All non-hydrogen atoms were refined anisotropically. Hydrogen atoms were refined isotropically in the observed positions (shown on Fig. 1), except where disordered (in which case fixed, calculated positions were used). CCDC 279736. For crystallographic data in CIF or other electronic format see DOI: 10.1039/b511810g

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