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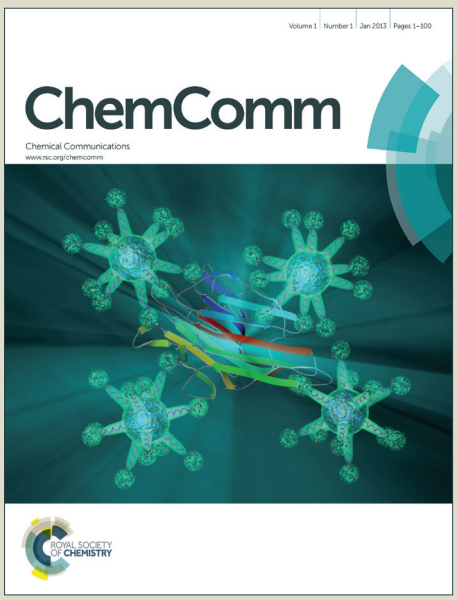

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Journal Name

COMMUNICATION

Template-controlled synthesis of chiral cyclohexylhemi-cucurbit[8]uril

Received 00th January 20xx,
Accepted 00th January 20xx

DOI: 10.1039/x0xx00000x

www.rsc.org/

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Enantiomerically pure cyclohexylhemicucurbit[8]uril (cycHC[8]), possessing a barrel-shaped cavity, has been prepared in high yield on a gram scale from either (*R,R,N,N'*)-cyclohex-1,2-diylurea and formaldehyde or cycHC[6]. In either case, a dynamic covalent library is first generated from which the desired cycHC can be amplified using a suitable anion template.

Research on new and selective host-guest systems and their applications is currently progressing very quickly[1]. Along with the search for new selective host-guest pairs, new and more efficient synthesis methods for hosts are being developed. Based on the recent success in the field of reversible non-covalent interactions in supramolecular chemistry[2], the concept of dynamic covalent chemistry (DCC) has been established.[3] Controlling covalent bond formation by non-covalent interactions can serve as an excellent tool for developing efficient adaptive systems, where the formation of the host molecule is based on the structure of the guest.

Cucurbit[*n*]urils[4] (CB) are non-toxic host molecules[5] with a wide range of applications.[1a,1d,6] Mechanistic studies have shown that the formation of oligomers and larger CBs proceeds reversibly, indicating that the principles of DCC are applicable in CB chemistry.[7] Hemicucurbiturils[8] (HC) are a sub-group of the cucurbituril family (Figure 1). HCs are known to form complexes with anions[9] and unsubstituted HCs have been applied as catalysts in organic reactions[10]. It has been shown that biotin[6]uril esters can be applied as transmembrane anion carriers.[9g] Miyahara *et al.*[8a] were the first to describe an efficient synthesis of HC[6] and HC[12]. High selectivity towards the HC[6] was explained by the template effect of the chloride anion, which was recently confirmed in a biotin[6]uril

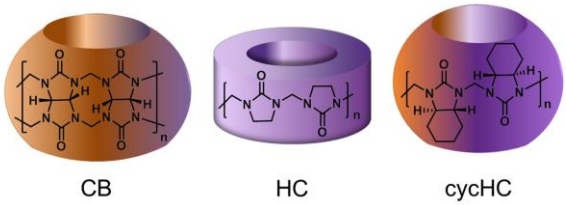

Figure 1. Generalized shapes of normal CB, HC and chiral cycHC.

synthesis.[8f] The halogen anion is also the necessary templat in the synthesis of bambus[6]urils (BU),[11] which can b classified as substituted HCs. Presently, besides HC[12], only 6 membered HCs[8] and 4- and 6- membered BUs[11] have bee isolated as main products. Until now, there has not been a efficient synthetic method available for the synthesis of 8 membered HCs. The existence of norbornahemicucurbit[8]uril[8d] has been detected only by mass-spectrometry and (*all-R*)-cyclohexylhemicucurbit[8]uril (cycHC[8]) has only been isolated as a by-product in low yield.[8e]

Herein we report an efficient synthesis of enantiomerical pure cycHC[8], starting either from its homologue cycHC[6] o (*R,R,N,N'*)-cyclohex-1,2-diylurea **1a** and paraformaldehyde. A mechanism of the transformation of cycHC[6] to cycHC[8] proposed and proof of complexation with carboxylic acids presented.

CycHC[6] was synthesized earlier in our group.[5] Subsequently, a small amount of its homologue cycHC[8][8e] was isolated from the crude product of cycHC[6]. Moreover, we noticed that in the chromatographic sample of cycHC[6] containing formic acid the amount of cycHC[8] gradually increased over time. The screening of reaction conditions for this conversion showed that cycHC[6] was transformed to cycHC[8] in the presence of sulphuric, formic and trifuoroacetic acid, but not acetic acid (see SI S4). The conversion of cycHC[6] to cycHC[8] by trifluoroacetic acid catalysis is approximately ten times faster than by formic acid

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Electronic Supplementary Information (ESI) available: A detailed description of synthesis, MS, NMR, crystallographic and computational details. See DOI: 10.1039/x0xx00000x

Table 1. Selected reaction conditions and the list of templates for cycHC synthesis.

No.	Starting comp.	(Additive)/Acid/Solvent[a]	Template	Time (h), *T*	Ratio[b] of cycHC[8] to cycHC[6]	Product	Isolated yield of product
1.	cycHC[6]	HCOOH/CH_3CN	HCO_2^-	24, rt	92:8	cycHC[8]	71%
2.	cycHC[6]	CF_3COOH/CH_3CN	$CF_3CO_2^-$	1.5, rt	95:5	cycHC[8]	71%
3.	cycHC[6]	$NaPF_6$ (50 eq)/CH_3COOH/CH_3CN	PF_6^-	24, rt	99:1	cycHC[8]	90%
4.	**1a**	HCOOH/CH_3CN	HCO_2^-	24, rt	92:8	cycHC[8]	7%
5.	**1a**	$NaPF_6$ (50 eq)/CH_3COOH/CH_3CN	PF_6^-	24, rt	95:5	cycHC[8]	55%
6.	**1a**	CF_3COOH/CH_3CN	$CF_3CO_2^-$	2, rt	96:4	cycHC[8]	73%
7.[c]	**1a**	HCl/H_2O	Cl^-	24, 70 °C	0:100	cycHC[6]+HCl	85%
8.	cycHC[8]	HCl/H_2O	Cl^-	24, 70 °C	5:95	cycHC[6]+HCl	71%
9.	cycHC[8]	NaCl (50 eq)/CH_3COOH	Cl^-	24, 70 °C	40:60	cycHC[6]+HCl	21%

[a] Generally 300 eq of organic acid and 4M HCl were used; [b]Determined by HPLC; [c]Described previously in ref 8c.

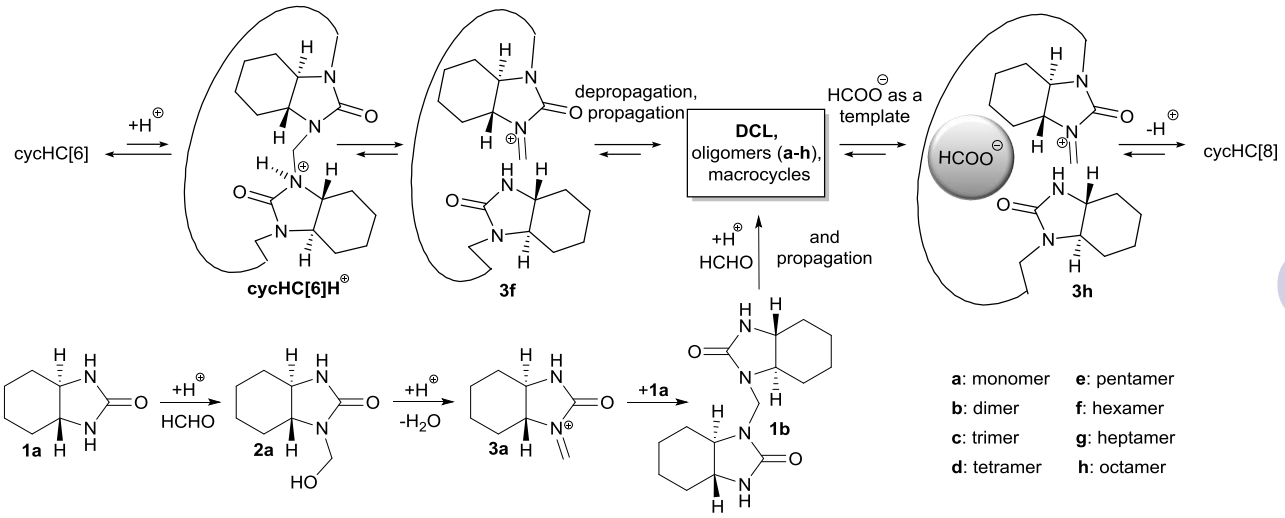
(Table 1, lines 1, 2). Nevertheless, the isolated yield of cycHC[8] was in both cases 71% in gram scale.

The kinetic data for the conversion of cycHC[6] to cycHC[8] revealed that the overall reaction was pseudo first-order, with a plateau. The fact, that the transformation of cycHC[6] to cycHC[8] proceeds faster in stronger acids (Table 1, compare entries 1 and 2) in combination with the results from DFT computational study of model structures (see SI S29) allows us to state, that the rate-limiting step of this process is protonation of the macrocycle. Occurrence of side reactions was minimal and no intermediates were detected by NMR (see SI S16).

Pittelkow *et al.* have shown that dimers are the main intermediates in the formation of biotin[6]uril.[8f] Also, since cycHC[6] and cycHC[8] differ from each other by a dimer unit, we wanted to examine whether the cycHC[8] formation proceeds via dimer addition. We thus introduced ^{13}C labels to methylene bridges of cycHC[6][8c] and subsequently used a 1:1 mixture of ^{13}C-labelled and non-labelled cycHC[6] in cycHC[8] synthesis. The number of ^{13}C-labelled methylene groups in isolated cycHC[8] varied from 0 to 8, following a normal distribution, thus confirming that beside dimers, other oligomers or monomers are involved in the reaction (see SI S7).

HRMS analysis of the reaction mixture showed the presence of cycHC[6-10][12] and various oligomers (up to an octamer, see SI S14). The large number of observed intermediates pointed to the presence of a dynamic combinatorial library (DCL).[3b]

According to DFT-calculated Gibbs' energies of cycHCs it is not the cycle strain, but the inclusion complex with formate anions that induces a preference towards the formation of cycHC[8] (See SI S27). Based on the experimental observations described above and the energy calculations on a model system (see SI S29), we propose that the transformation of cycHC[6] to cycHC[8] proceeds through the key steps outlined in Scheme 1. First, a reaction rate-limiting protonation of cycHC[6] occurs, then breakage of the first methylene bridge of cycHC[6]H$^+$ takes place, forming the iminium **3f**. The DCL, whose members have been observed by HRMS, is generated through depropagation and propagation reactions. A formate acts as an anionic template and shifts the thermodynamic equilibrium between DCL members towards the formation of cycHC[8].



Scheme 1. Proposed reaction mechanism of the cycHC[8] formation catalysed by formic acid.

To verify that an anionic template is necessary to drive the reaction towards the formation of cycHC, we selected an anion that possessed the size and shape suitable for the cavity of cycHC[8], the hexafluorophosphate, in combination with acetic acid. Acetic acid alone was shown not to facilitate the formation of cycHC[8] (SI S4). As expected, in the presence of $NaPF_6$ in acetic acid/acetonitrile, cycHC[6] was efficiently converted to cycHC[8] (Table 1, entry 3). This observation confirmed that even though reaction rate depends on the acid strength, the macrocycle formation is controlled by the anion, acting as a template. And with catalysis of formic and trifluoroacetic acid, their conjugate anions act as templates (Table 1, entries 1, 2).

Next, based on the proposed mechanism, we envisioned that the DCL members could be generated starting from monomers **1a**. Indeed, using either formic acid, trifluoroacetic acid, or $NaPF_6$/acetic acid as catalysts afforded cycHC[8] (Table 1, entries 4-6). The lower rate of formation of cycHC[8] from **1a** than from cycHC[6], was due to the additional acid-promoted reactions necessary for building methylene bridges. The best yield and selectivity were achieved with trifluoroacetic acid, giving the cycHC[8] from **1a** on a gram scale in 73% yield. This synthetic method allowed for the preparation of enantiopure chiral macrocycle cycHC[8] very efficiently, in only two steps, starting from commercially available 1,2-cyclohexanediamine.[13]

According to the proposed mechanism, the conversion of cycHC[8] to cycHC[6] in the presence of a halide template, should also be possible. Indeed, using the classic conditions of CB formation (see Table 1, entry 8), cycHC[8] was efficiently converted to cycHC[6] with the aid of the chloride anion. Similarly, using NaCl as a templating additive in acetic acid at elevated temperature, cycHC[8] was also converted to cycHC[6] (see Table 1, entry 9), again highlighting the role of the templating anion in the reaction.

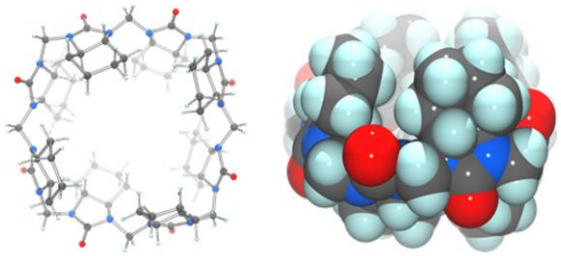


Figure 2. Crystal structure of cycHC[8]: Top view in ball and stick (left) and side view in CPK (right) representations (colour code: C grey, N blue, O red, H turquoise).

Table 2. Dimensions of cycHC[6,8] and CB[6,8].

Parameters[a]	cycHC[6][b]	cycHC[8]	CB[6][c]	CB[8][c]
Opening diameter (Å)	2.2	4.6	3.9	6.9
Cavity diameter (Å)	5.3	8.5	5.8	8.8
Height (Å)	12.1	12.5	9.1	9.1
Cavity volume (Å^3)	35[d]	123[d]	119±21	356±16

[a]Dimensions account for the van der Waals radii of the various atoms. [b]Opening, cavity and height values are from ref 8c. [c]Opening, cavity and height values are from ref 14a and cavity volume from ref 14b. [c]Cavity volume of cycHC[6] from ref 8c and cycHC[8] calculated by analysing the solvent accessible voids in the respective crystal structures using PLATON[15] with a probe radius of 1.2 Å^3 and grid steps of 0.2 Å.

Table 3. Association constants K_a (M^{-1}) of carboxylic acids with cycHC[6] and cycHC[8] in 1:1 mixtures in $CDCl_3$.

No.	Guest	cycHC[6] K_a	cycHC[8] K_a
1.	CH_3COOH	8.0±0.5 [a]	17±2
2.	HCOOH	97±1	72.6±0.5
3.	CF_3COOH	21(±3)×10^3	29 (±1)×10^3
4.	*R*-MPA	27.2 ± 0.8 [a]	27.0±0.5
5.	*S*-MPA	20.1 ± 0.2 [a]	53±3
6.	*R*-MTPA	n.d.	3.3 (±0.1) ×10^2
7.	*S*-MTPA	n.d.	3.0 (±0.1) ×10^2

[a] Association constants from ref. 8c; n.d. - not determined.

The crystal structure confirmed the barrel-like shape of cycHC[8] (Figure 2). According to the crystal structure, the cavity of cycHC[8], similar in size to that of CB[6], is of sufficient size for the encapsulation of a number of organic and inorganic guests (see Table 2).

Complexation studies of the cycHC[8] with carboxylic acids were performed by diffusion NMR in $CDCl_3$. The comparative results of the complexation of cycHC[6] and cycHC[8] are presented in Table 3. The association constants of simple carboxylic acids – acetic, formic and trifluoroacetic acids - follow the order of their acidity (see Table 3, entries 1–3) for both hosts.

Analogously to small carboxylic acids, complexation with the more acidic α-methoxy-α-trifluoromethylphenylacetic acid (MTPA) was stronger than with α-methoxyphenylacetic acid (MPA) (see Table 3, lines 5, 6). The opposite preference of complexation of *R*-handed cycHC[6] and cycHC[8] toward MPA enantiomers may suggest different geometries of complexes in these cases. Nevertheless *R*- handed cycHC[8] showed nearly double affinity for *S*-MPA, compared to the *R*-MPA. This result confirms that cycHC[8] forms complexes enantioselectively.

In conclusion, we have presented the first highly efficient synthesis of an 8-membered representative of the cucurbituril family, the (*all-R*)-cyclohexylhemicucurbit[8]uril. We have shown that the reversibility of the methylene bridge formation allows the size of the cycHC macrocycles to be controlled by the anionic templates, with halides driving the equilibrium towards the formation of cycHC[6], while carboxylates and PF_6^- promoted the formation of cycHC[8].

Chiral cycHC[8] and cycHC[6] were obtained very efficiently in one step, starting from enantiomerically pure (*R,R,N,N'*)-cyclohex-1,2-diylurea **1a** or either homologue. (*all-R*)-cycHC[8] enantioselectively formed complexes with chiral carboxylic acids, demonstrating chiral discrimination ability. CycHC[8] shows potential for application in host-guest chemistry.[9g,10,16]

In the present study, DCL members were formed from identical monomeric units. It can be envisioned that by utilizing a mixture of different monomeric ureas and suitable templates, a very efficient yet diverse library of useful hemicucurbituril hosts could become accessible *via* dynamic covalent chemistry.

The authors would like to thank Tiina Aid, Marina Kudrjašova and Jasper Adamson for experimental assistance and Aivar Lõokene, Mart Reimund and Omar Parve for help with the manuscript. This research was supported by the Academy of Finland (KR, grants 263256 and 265328), the Estonian Ministry of Education and Research through Grants IUT19-32, IUT19-9, IUT23-7 and PUT692, TUT grant No. B25, the ESF DoRa, and the EU European Regional Development Fund (3.2.0101.08-0017). Computations were performed on the HPC cluster at TUT, which is part of the ETAIS. FT acknowledges support from NGS-NANO through a Ph.D. fellowship.

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