

Improved procedure for the synthesis of calix[4]arenes with four amino groups on the wide rim

Xiao-Qiang Sun^{1*}, Hao Zheng¹, Zheng-Yi Li¹, Myroslav O. Vysotsky^{1*}

¹ School of Petrochemical Engineering, Changzhou University, Changzhou, 213164, Jiangsu, P. R. China

ABSTRACT

Calix[4]arenes fixed in the cone conformation and bearing four amino functionalities on the wide rim, are well known and often used starting materials in calixarene chemistry. During our studies in this field we have noticed, that the safe procedures known (which use palladium on carbon as catalyst and hydrazine hydrate as reducing agent in methanol at reflux) not always fit well for the synthesis of all possible even rather simple derivatives of this type and undesired mixtures are usual outcome in these cases. High resolution MS and ¹H NMR indicate, that the main impurity (nearly 30%) is a partially reduced calix[4]arene containing three amino- and one nitro groups. We have modified the synthetic procedure by changing the solvent to *iso*-propanol, which allowed to shorten the reaction times and to obtain the desired products in high purity and yields.

Keywords: Aromatic amines; calix[4]arenes; nitro compounds; palladium; reduction.

1. INTRODUCTION

Calix[4]arenes [1] fixed in the *cone* conformation and bearing four amino groups at the wide rim **1** [2] are used often as the starting materials in the synthesis of many interesting and even useful compounds (Fig. 1) such as tetra-ureas [3] (or similar [4]), calixsugars [5], carbamoyl methyl phosphine oxide (CMPO) derivatives [6] and some water soluble calix[4]arenes [2a,b,7]. Calix[4]arene ureas are well known to self-assemble into dimeric molecular capsules [3,8], which can be also used in effective synthesis of mechanically interlocked structures [9]. This self-assembly motif had been also successfully used in assembly of hydrogen bonded polymers [10], dendrimers of defined structures [11], and elucidation of self-sorting phenomena between self- assembled capsules of similar structure [12]. Calixsugars can have plenty of different applications in bio-relevant area including inhibition of lectin binding to cell-surface glycoconjugates [5a]. CMPO derivatives are known for their very high extraction abilities of lanthanides and actinides [6], which potentially can be used in treatment of highly radioactive wastes obtained during exploitation of nuclear reactors. Some of water soluble calix[4]arenes (like guanidinium derivatives shown on Fig. 1) proved to be quite effective in DNA cell transfection [7].

* Tel.: +86 0519-86330003;

E-mail addresses: Xqsun@cczu.edu.cn, m.o.vysotsky@gmail.com.

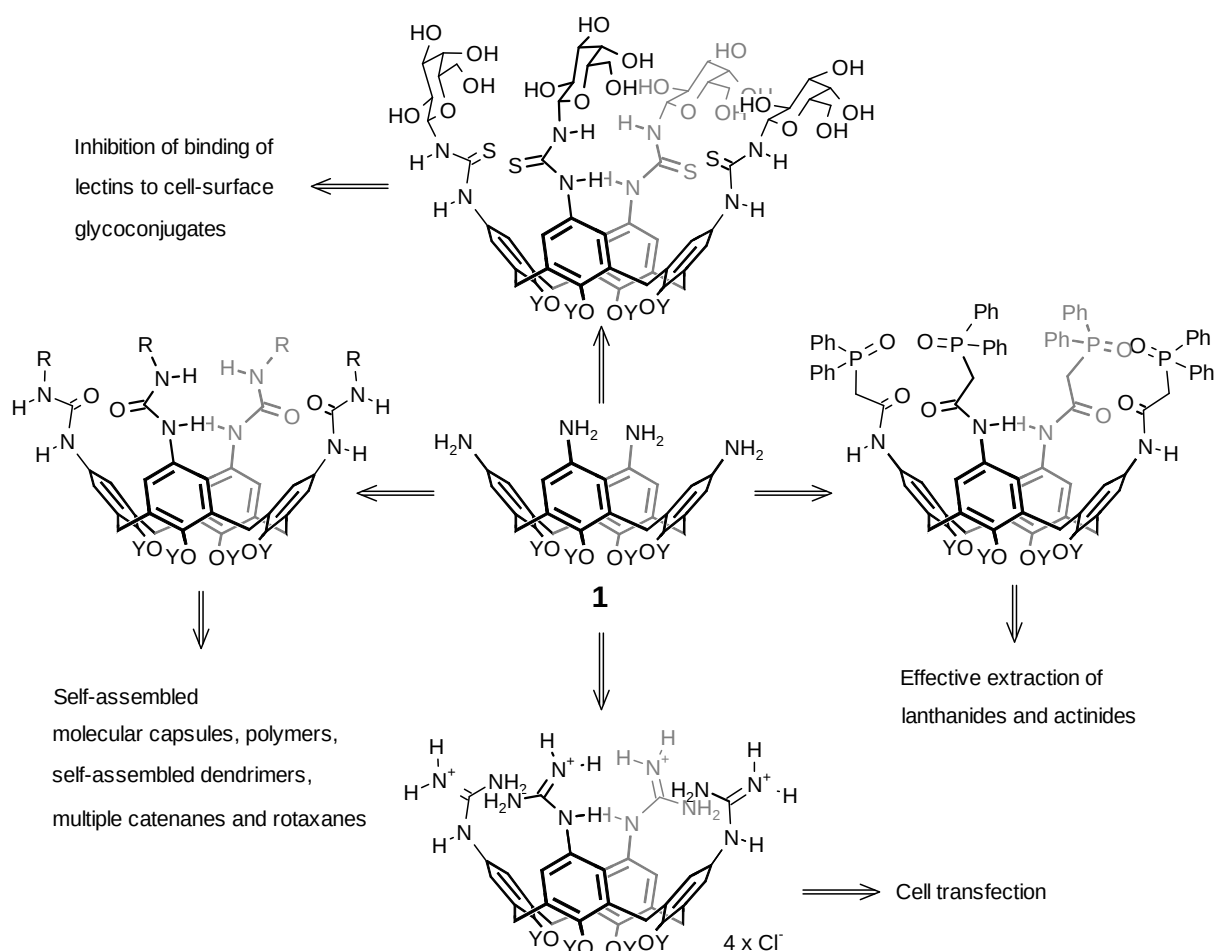
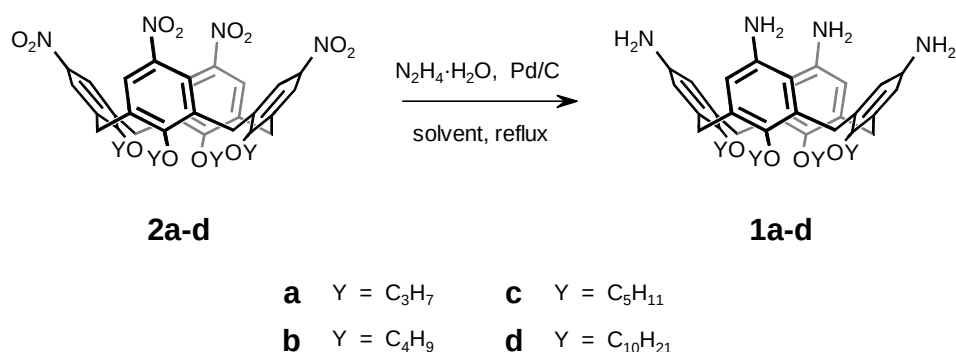


Fig. 1. Derivatives obtained from tetra-amines of calix[4]arenes, and their applications.



Scheme 1. Synthesis of tetra-amino derivatives of calix[4]arenes **1a-d**.

The very first method of the synthesis of calix[4]arene with four amino functions on the wide rim **1** (Scheme 1, large variation of alkyl chains Y) included exhaustive hydrogenation of corresponding tetra-nitro derivatives **2** using iron on active charcoal (obtained *in situ*) and hydrazine hydrate as reducing agent (Table 1, entry 1) [2a,b]. Interestingly, this method had never been used afterwards, to the best of our knowledge. Instead, another and very often used procedure had been developed, which uses Raney nickel as catalyst and hydrogen as reducing agent [2c], at room temperature (usually, overnight). This is a general procedure for

Table 1. Summary of the conditions known from the literature, which are used in reduction of tetra-nitro derivatives of the type of 2 (many different alkyl chains Y).

N	(Pre)catalyst	Reducing agent	Solvent	Temperature	Time	Reference
1	FeCl ₃ ·6H ₂ O, active charcoal	N ₂ H ₄ ·H ₂ O	2-methoxy-ethanol	Reflux	12 h	2a, 2b
2	Raney-Ni	H ₂	Toluene or THF	Room temp.	14 h	2c
3	Raney-Ni	N ₂ H ₄ ·H ₂ O	MeOH	Reflux	4 h, 2h	2d, 2e
4	Raney-Ni	N ₂ H ₄ ·H ₂ O	EtOH	Reflux	6 h	2f
5	Raney-Ni	N ₂ H ₄ ·H ₂ O	THF/MeOH	Reflux	2 h	2g
6	Pd/C	H ₂ (2 bar)	Ethyl acetate	Room temp.	24 h	2h
7	Pd/C	H ₂	EtOH/toluene/1M HCl solution	Room temp.	Not specified	2i
8	Pd/C	N ₂ H ₄ ·H ₂ O	EtOH	Reflux	5 h	2h
9	PtO ₂	H ₂	THF	Room temp.	18 h	2k
10	-	SnCl ₂ ·2H ₂ O	EtOH	70 °C	48 h	2l
11	-	SnCl ₂ ·2H ₂ O	EtOH	Reflux	12 h	2m

reduction of nitro-compounds, of course. These conditions are known to be very mild ones, which could be applied to many different calix[4]arenes bearing also some sensitive functionalities (like ester groups) on the narrow rim [11]. However, the obvious drawback of this method, is the additional safety issues. The first issue is the active Raney nickel, which is flammable in the dry state. That is why one should be always very careful in handling it and especially after the reaction, when the reaction mixture should be filtered. In the last case see sand should be used to prevent the drying out of the catalyst. Then it should be destroyed by treatment with hydrochloric acid. This produces, obviously, additional wastes, that are also not desirable. The second obvious safety issue is the use of gaseous hydrogen, which easily forms explosive mixtures with air and thus is not always welcome in industrial processes in developed countries.

The further development of the reduction conditions includes the use of other reducing reagents like hydrazine hydrate (Table 1, entries 3-5) [2f,g]. This allows already to lower one of the hazards discussed. The usual solvent used in thus modified method is methanol (or ethanol) and times applied, are 2-6 hours. Some other modifications include the change of heterogeneous catalyst from Raney nickel to palladium on carbon (Table 1, entries 6,7) [2h], which also reduces the hazards discussed. Under these hydrogenation conditions the times applied are up to 24 hours (at room temperature). Finally, the most safe method (from the view point of these hazards) includes the use of palladium on carbon as heterogeneous catalyst and hydrazine hydrate (Table 1, entry 8, under reflux, 5 hours). Some other modifications include the use of Adams catalyst (which is also pyrophoric, Table 1, entry 9) or the use of tin dichloride (Table 1, entries 10,11). The reaction mixtures in the last cases usually are difficult to work-up (due to possible problems with precipitation of tin complexes). In this paper we disclose our studies on the reduction of tetra nitro calix[4]arenes having alkyl chains of different length on the narrow rim, which show, however, that the desirable conditions (Pd/C and N₂H₄·H₂O, in alcohols) not always give the desired results.

2. MATERIALS AND METHODS / EXPERIMENTAL DETAILS

The reagents of analytic grade were purchased from Aladdin Chemical Company (China) and were used without further purifications. Compounds **2a-d** had been obtained in accordance with the typical methods described in the literature [2c]. ¹H NMR spectra were recorded in chloroform-d₁ on a Burkcr Avance III 300 MHz spectrometer. All reactions were carried out under nitrogen atmosphere.

Typical procedure for synthesis of 1a-d: *Iso*-propyl alcohol (600 mL) was poured into the one-neck 1 L round-bottom flask containing tetra-nitro compound **2** (9.16mmol), Pd/C (0.75 g) and hydrazine hydrate (70 mL or 1.44 mol). The flask was equipped with condenser ending on the top with a balloon. The reaction mixture was brought to reflux at stirring (as the reaction proceeds, the balloon becomes larger and the system should be opened from time to time for a very short period to let the excess of gases to leave the system). After four hours of reflux, the reaction mixture was cooled to the room temperature, then the solvent was removed at the reduced pressure and the residue was thoroughly dried in vacuum. Dichloromethane (250 mL) was added. After swirling for several minutes, the suspension (due to the presence of Pd/C), was filtered through a paper filter, the solution thus obtained, was evaporated and dried yielding the desired compound in 80 - 95% yield (see Table 2). ¹H NMR of the products obtained, showed the expected pattern of signals [2].

3. RESULTS AND DISCUSSION

We have applied the modified conditions (Table 1, entry 8, in methanol) to the synthesis of tetra-amino calix[4]arenes **1** fixed in the *cone* conformation, which bear four equal alkyl chains having three, four, five and ten carbons on the narrow rim. In contrast to the reactions on derivatives with butyl and pentyl chains, which went without any problems (Table 2, **1b**, **c**), the reactions on derivatives with propyl and decyl chains, however, led to the corresponding products only with ~70% and ~86% purity, respectively, to our big surprise (Fig. 2). This result was *constantly* reproducible. Increasing the reaction times also did not change much the outcome.

A rough look at ¹H NMR spectrum of such mixtures gives a hint, that there is, perhaps, predominantly only one impurity present in each case, since only two additional pairs of doublets for methylene bridge protons Ar-CH₂-Ar are observed (Fig. 2, on example of **1a**). If this assumption would be true, this impurity should be of AAAB (or ABAC) substitution type of calix[4]arene skeleton, which should give a) two pairs of doublets for Ar-CH₂-Ar protons and b) two singlets and one pair of *meta*-coupled doublets in the aromatic region, nearly exactly the picture observed in ¹H NMR (Fig. 2, two *meta*-coupled doublets are represented

Table 2. Different conditions applied in our studies for the synthesis of 1a-d.

Product	Solvent	Time	Yield	Purity ^a
1a	methanol	overnight	58%	~70%
1a	<i>iso</i> -propanol	4 h	80%	>98%
1b	methanol	overnight	57%	>98%
1b	<i>iso</i> -propanol	4 h	83%	>98%
1c	methanol	overnight	65%	>98%
1c	<i>iso</i> -propanol	4 h	95%	>98%
1d	methanol	overnight	58%	~86%
1d	<i>iso</i> -propanol	4 h	82%	>98%

^a Estimated by the integration of signals in ¹H NMR spectra.

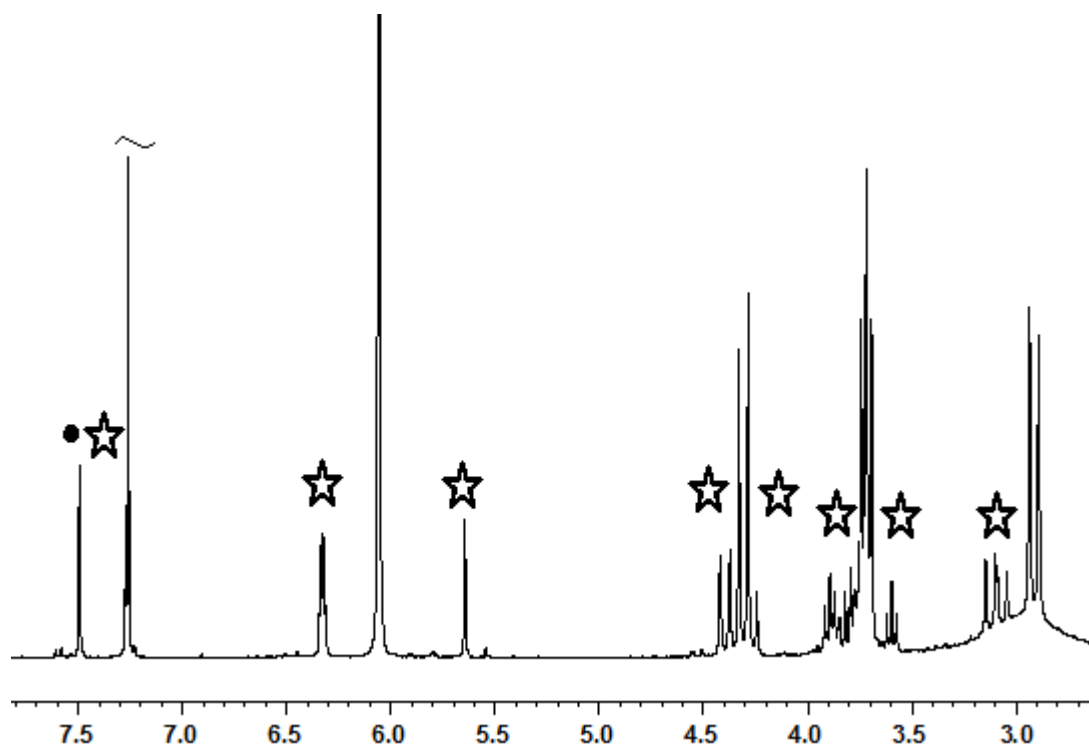


Fig. 2. A part of ^1H NMR spectrum (chloroform- d_1 , 300 MHz) of product **1a** obtained by applying the conditions indicated in Table 2 (methanol, overnight). The signals of main by-product are indicated with a star, another impurity is indicated with a filled black circle, the residual signal of solvent is cutted (indicated with a waved line), while the signals of tetra-amino derivative **1a** are *not* indicated.

with a signal of AB-system with close chemical shifts at ~ 6.35 ppm). To find out the elemental composition, a high resolution MS spectrum was measured for this case. It shows only one unexpected peak with mass of 683.3813 (the other peaks of mono-charged species of sufficient intensities correspond to $\mathbf{1a}^+$, $[\mathbf{1a}+\text{H}]^+$, $[\mathbf{1a}+\text{Na}]^+$ and $[\mathbf{1a}+\text{K}]^+$). This mass corresponds well with the formula $\text{C}_{40}\text{H}_{51}\text{N}_4\text{O}_6$ (calc. mass 683.3809). This is protonated species of calix[4]arene (Fig. 3) having either 1) three amino and one nitro functions on the wide rim (**3a**), or 2) two amino, one hydroxylamino and one nitroso functions on the wide rim (**4a**). If these assumptions in the last case should stay in accordance with the ^1H NMR spectra, this impurity must have ABAC substitution pattern (**4a**), in accordance with C_s symmetry (two amino functions on *distal* aromatic units and -NO and -N(OH)H groups on the remaining two ones, Fig. 3). It would be very surprising to have such di-amine without having observed the formation of *proximal* regio-isomer, which should have even much more

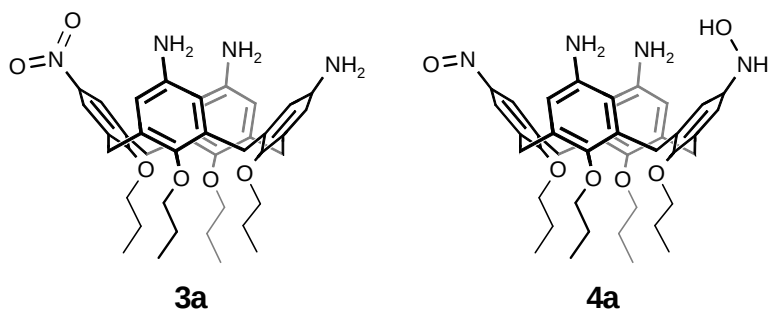


Fig. 3. Possible structures of the main side product observed in the synthesis of **1a**.

complicated spectrum due to its C_1 symmetry. Thus, taking this fact and the usual chemical liability of nitroso and hydroxylamino functions into account, we suppose to deal rather with tri-amino mono-nitro derivative **3a**. This is the first observation in chemistry of calixarenes, which describes a *partial* reduction of nitro-derivatives of calixarenes, to the best of our knowledge. In searching for new conditions for reductions of **2**, we have changed the solvent to *iso*-propanol and kept the reaction mixture under stirring at reflux for four hours only. These conditions appear already quite good ones, which lead to the desired compounds of high purities (Fig. 4, Table 2) and in high yields. These facts can be rationalized, most probably, by higher lipophilicity and higher boiling point of *iso*-propanol in comparison to methanol, since the starting materials are not soluble under usual conditions in either of the solvent mixtures used. The intermediates (partially reduced calix[4]arenes) should be better soluble in *iso*-propanol than in methanol, thus increasing their concentrations and the rate of the whole process.

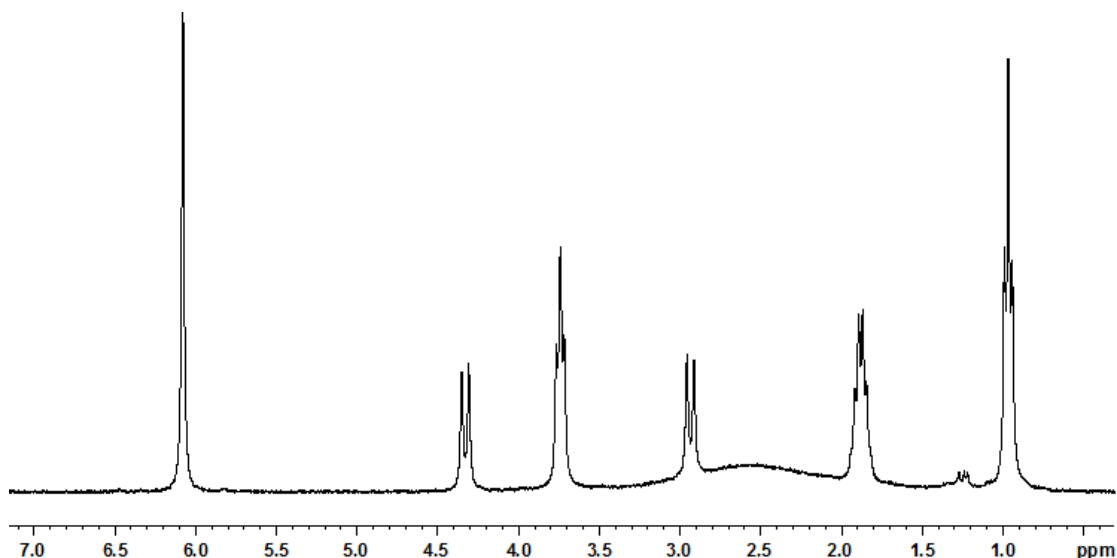


Fig. 4. ^1H NMR spectra (chloroform- d_1 , 300 MHz) of products **1a** obtained by applying new conditions (Table 2, *iso*-propanol, reflux 4h).

4. CONCLUSION

The reduction conditions including Pd/C as heterogeneous catalyst, hydrazine hydrate as reducing agent and methanol as solvent are typical most safe conditions used in exhaustive reduction of calix[4]arenes with four nitro groups on the wide rim. They work well, however,

in 50% of cases only. The main impurity identified by high resolution MS and ^1H NMR, is the tri-amino mono-nitro derivative (formed up to nearly 30%). This is the first report on *partial* hydrogenation reaction of nitro compounds in calixarene chemistry. The use of *iso*-propanol instead of methanol, leads already to the reliable method giving the desired tetra-amino derivatives in high yields (80-95%) and high purities (>98%).

ACKNOWLEDGEMENTS

We gratefully acknowledge the financial support from the Natural Science Foundation of China (No. 21002009), Major Program for the Natural Science Research of Jiangsu Colleges and Universities (12KJA150002), and Jiangsu Key Laboratory of Advanced Catalytic Materials and Technology (BM2012110). This work was also funded by the Priority Academic Program Development (PAPD) of Jiangsu Higher Education Institutions and the Qing-Lan Project of Jiangsu Province.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

AUTHORS' CONTRIBUTIONS

This work was done in collaboration between all authors. XQS and MOV designed this work, while ZH carried out all experiments. The experimental results were analyzed by ZH and MOV and discussed by all authors. MOV wrote the manuscript, while all authors read and approved the final version.

REFERENCES

- (a) Böhmer V. Calixarenes, macrocycles with (almost) unlimited possibilities. *Angew Chem Int Ed Engl.* 1995;34:713-745.
(b) Gutsche CD. Calixarenes revisited. Cambridge: Royal Society of Chemistry; 1998.
(c) Calixarenes 2001. Eds.: Asfari MZ, Böhmer V, Harrowfield J, Vicens J. Dordrecht: Springer; 2001.
- (a) Shinkai S, Shirahama Y, Tsubaki T, Manabe O. Calixarene-catalysed basic hydrolysis of p-nitrophenyl dodecanoate: a possible change in the mechanism from "deshielding" to "host-guest". *J Am Chem Soc.* 1989;111:5477-5478.
(b) Shinkai S, Arimura T, Araki K, Kawabata H, Satoh H, Tsubaki T, Manabe O, Sunamoto J. Syntheses and aggregation properties of new water-soluble calixarenes. *J Chem Soc Perkin Trans 1.* 1989;2039-2045.
(c) Jakobi RA, Böhmer V, Grüttner C, Kraft D, Vogt W. *New J Chem.* 1996;20:493-501.
(d) Matthews SE, Saadioui M, Böhmer V, Barbosa S, Arnaud-Neu F, Schwing-Weill MJ, Carrera MG, Dozol JF. Conformationally mobile wide rim carbamoylmethylphosphine oxide (CMPO)-calixarenes. *J Prakt Chem.* 1999;341:264-273.
(e) Klimentová J, Vojtišek P. New receptors for anions in water: synthesis, characterization, X-ray structures of new derivatives of 5,11,17,23-tetraamino-25,26,27,28-tetra-propyloxycalix[4]arene. *J Mol Str.* 2007;826:48-63.

221 (f) Arduini A, Böhmer V, Delmau L, Desreux JP, Dozol JF, Carrera MAG, Lambert B,
 222 Musigmann C, Pochini A, Shivanyuk A, Ugozzoli F. Rigidified calixarenes bearing four
 223 carbamoylmethylphosphineoxide or carbamoylmethylphosphoryl functions at the wide
 224 rim. *Chem Eur J*. 2000;6:2135-2144.
 225 (g) Schühle DT, van Rijn P, Laurent S, Elst LV, Muller RN, Stuart MCA, Schatz J,
 226 Peters JA. Liposomes with conjugates of a calix[4]arene and a Gd-DOTA derivative
 227 on the outside surface; an efficient potential contrast agent for MRI. *Chem Commun*.
 228 2010;46:4399-4401.
 229 (h) Sansone F, Chierici E, Casnati A, Ungaro R. Thiourea-linked upper rim calix[4]-
 230 arene neoglycoconjugates: synthesis, conformations and binding properties. *Org*
 231 *Biomol Chem*. 2003;1:1802-1809.
 232 (i) Zadnarm R, Schrader T. Self-assembly of molecular capsules in polar solvents. *Org*
 233 *Lett*. 2002;4:1687-1690.
 234 (k) Rincón AM, Prados P, de Mendoza J. A Calix[4]arene ureidopeptide dimer self-
 235 assembled through two superposed hydrogen bond arrays. *J Am Chem Soc*. 2001;
 236 123:3493-3498.
 237 (l) Mislin G, Graf E, Hosseini MW. Synthesis of an exo-ditopic receptor based on
 238 calix[4]arene and catechol. *Tetrahedron Lett*. 1996;37:4503-4506.
 239 (m) Stibor I, Budka J, Michlová V, Tkadlecová M, Pojarová M, Cuřínová P, Lhoták P.
 240 Systematic approach to new ligands for anion recognition based on ureido-calix[4]-
 241 arenes. *New J Chem*. 2008;32:1597-1607.
 242 3. (a) Shimizu KD, Rebek J. Synthesis and assembly of self-complementary calix[4]-
 243 arenes. *Proc Natl Acad Sci USA*. 1995;92:12403-12407.
 244 (b) Mogck O, Böhmer V, Vogt W. Hydrogen bonded homo- and heterodimers of tetra
 245 urea derivatives of calix[4]arenes. *Tetrahedron*. 1996;52:8489-8496.
 246 (c) Hamann BC, Shimizu KD, Rebek J. Reversible encapsulation of guest molecules
 247 in a calixarene dimer. *Angew Chem Int Ed Engl*. 1996;35:1326-1329.
 248 4. Shivanyuk A, Saadioui S, Broda F, Thondorf I, Vysotsky MO, Rissanen K,
 249 Kolehmainen E, Böhmer V. Sterically and guest-controlled self-assembly of calix[4]-
 250 arene Derivatives *Chem Eur J*. 2004;10:2138-2148.
 251 5. (a) André S, Sansone F, Kaltner H, Casnati A, Kopitz J, Gabius HJ, Ungaro R.
 252 Calix[n]arene-Based glycoclusters: bioactivity of thiourea-linked galactose/lactose
 253 moieties as inhibitors of binding of medically relevant lectins to a glycoprotein and cell-
 254 surface glycoconjugates and selectivity among human adhesion/growth-regulatory
 255 galectins. *ChemBioChem*. 2008;9:1649-1661.
 256 (b) Galante E, Geraci C, Sciuto S, Campo VL, Carvahlo I, Sesti-Costa R, Guedes
 257 PMM, Silva JS, Hill L, Nepogodiev SA, Field RA. Glycoclusters presenting lactose on
 258 calix[4]arene cores display trypanocidal activity. *Tetrahedron*. 2011;67:5902-5912.
 259 (c) Dondoni A, Marra A, Schermann MC, Casnati A, Sansone F, Ungaro R. Synthesis
 260 and properties of O-glycosyl calix[4]Arenes (calixsugars). *Chem Eur J*. 1997;3:1774-
 261 1782.
 262 (d) Marra A, Schermann MC, Dondoni A, Casnati A, Minari P, Ungaro R. Sugar
 263 calixarenes: preparation of calix[4]arenes substituted at the lower and upper rims with
 264 O-glycosyl groups. *Angew Chem Int Ed Engl*. 1994;33:2479-2481.
 265 6. (a) Dordea C, Brisach F, Haddaoui J, Arnaud-Neu F, Bolte M, Casnati A, Böhmer V.
 266 Tetra-CMPO-derivatives of calix[4]arenes fixed in the 1,3-alternate conformation.
 267 *Supramol Chem*. 2010;22(6):347-357.
 268 (b) Gruner B, Böhmer V, Dordea C, Selucky P, Bubenikova M. Anionic *tert*-butyl-
 269 calix[4]arenes substituted at the narrow and wide rim by cobalt bis(dicarbollide)(1-)
 270 ions and CMPO-groups. Effect of stereochemistry and ratios of the functional groups
 271 on the platform on the extraction efficiency for Ln(III)/An(III). *J Organomet Chem*.
 272 2013;747:155-166.

- 273 (c) Peters C, Braekers D, Kroupa J, Kasyan O, Miroshnichenko S, Rudzevich V,
 274 Böhmer V, Desreux JF. CMPO-calix[4]arenes and the influence of structural
 275 modifications on the Eu(III), Am(III), Cm(III) separation. *Radiochim Acta* 2008;96:203-
 276 210.
- 277 7. (a) Sansone F, Dudič M, Donofrio G, Rivetti C, Baldini L, Casnati A, Cellai S, Ungaro
 278 R. DNA condensation and cell transfection properties of guanidinium calixarenes:
 279 dependence on macrocycle lipophilicity, size, and conformation. *J Am Chem Soc.*
 280 2006;128:14528-14536.
- 281 (b) Dudic M., Colombo A, Sansone F, Casnati A, Donofrio G, Ungaro R. A general
 282 synthesis of water soluble upper rim calix[n]arene guanidinium derivatives which bind
 283 to plasmid DNA. *Tetrahedron.* 2004;60:11613.
- 284 (c) Bagnacani V, Franceschi V, Bassi M, Lomazzi M, Donofrio G, Sansone F, Casnati
 285 A, Ungaro R. Arginine clustering on calix[4]arene macrocycles for improved cell
 286 penetration and DNA delivery. *Nature Commun.* 2013;4:1721. DOI: 10.1038/ncomms
 287 2721 available on-line at <http://www.nature.com/ncomms/index.html>.
- 288 8. (a) Rebek J. Reversible encapsulation and its consequences in solution. *Acc Chem*
 289 *Res.* 1999;32:278-286.
- 290 (b) Rebek J. Host-guest chemistry of calixarene capsules. *Chem Commun.* 2000;637-
 291 643.
- 292 9. Bogdan A, Rudzevich Y, Vysotsky MO, Böhmer V. Topologically novel multiple
 293 rotaxanes and catenanes based on tetraurea calix[4]arenes. *Chem Commun.*
 294 2006;2941-2952.
- 295 10. Castellano RK, Rudkevich DM, Rebek J. Polycaps: Reversibly formed polymere-
 296 ric capsules. *Proc Natl Acad Sci USA.* 1997;94:7132-7137.
- 297 11. Rudzevich Y, Rudzevich V, Moon C, Brunklaus G, Böhmer V. Self-assembled
 298 dendrimers with uniform structure. *Org Biomol Chem.* 2008;6:2270-2275.
- 299 12. (a) Rudzevich Yu, Rudzevich V, Klautzsch F, Schalley CA, Böhmer V. A Self-sorting
 300 scheme based on tetra-urea calix[4]arenes. *Angew Chem Int Ed.* 2009;48:3867-3871.
 301 (b) Rudzevich Y, Rudzevich V, Böhmer V. Dimerization and self-sorting of tetraurea
 302 calix[4]arenes. *Synlett.* 2009;1887-1904.
 303 (c) Rudzevich Y, Rudzevich V, Böhmer V. Fine-tuning the dimerization of tetraurea-
 304 calix[4]arenes. *Chem Eur J.* 2010;16:4541-4549.
- 305
 306