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# Synthesis and NMR Studies on a $C_3$-Symmetrical Triquinolina Triscationic Bicyclophane

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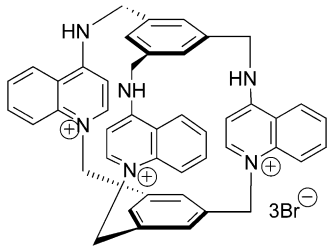


Two-step syntheses of triple-bridged triscationic bicyclophanes are presented. Molecular modeling has been undertaken by means of ab initio calculations (6-31G** level) on the triquinolina triscationic bicyclophane. This compound exists as two diastereomeric sets of enantiomers, one with $C_3$ symmetry and the other with $C_1$ symmetry. The $C_3$-symmetrical derivative is 1.94 kcal mol$^{-1}$ more stable than its $C_1$-symmetrical one. This energy difference is sufficient to consider the former and its enantiomer the only two conformations existing in solution at room temperature.

Cyclophanes represent a field of research of increasing significance. Their NMR studies have been recently recorded.[1] The synthesis and structures of several neutral cyclophanes, including tetrapeptides incorporating 14-membered-cycloisodityrosine moieties,[2] and [6.6]- and [8.8]cyclophanes with 1,4-dioxabut-2-yne and 1,6-dioxahexa-2,4-diyne bridges have been reported.[3] The synthesis and structural studies of some cationic cyclophanes were previously reported by Cabildo et al.[4] Other tetracationic species containing furan, thiophene, and benzo[*b*]thiophene-bridged macrocycles of 4,4′-bipyridine were reported by Scheytza and Reissig.[5] We designed and synthesized the most simple model of macrocyclic compounds that have only one benzene ring as a linker (**3**−**6**).[6] These compounds are dissimilar in the substitutionpattern shown by the upper (first prefix) and lower (second prefix) benzene rings (Figure 1). We have recently studied the conformational dynamics of cyclophane **3**.[5]

Bicyclophanes are compounds seldom referred to in the literature. In bicyclophanes, two aromatic rings are joined by three bridges, all containing aromatic rings, and structures of this type have been only previously described by Högberg and Wennerström[8] and by Olsson et al.[9] more than 20 years ago. As part of a program leading to the synthesis and biological evaluation of choline kinase (ChoK) inhibitors, we were interested in finding bicyclophane analogues in this unexplored area, and also in new triscationic ChoK inhibitors. This paper describes the synthesis of the reference compounds **5**−**7** (Figure 1) and the NMR and theoretical studies necessary to elucidate the conformational behavior of **7**.

Bicyclophanes **5**−**7** were synthesized according to the Scheme 1. The tripyridine **11** and the triquinoline **12** are novel and were prepared from the triamine **8**[10] and 4-bromopyridine **9** or 4-chloroquinoline **10**, respectively, in the presence of phenol, which is known to catalyze the reaction in the case of 2- and 4-haloquinolines.[11] Phenol reduces both the reaction time and temperature of halogen substitution reaction as a reaction medium.

Tripyridine **11** and triquinoline **12** were characterized as the tris-hydrobromide compounds. The conversion of **11** and **12** to the desired bicyclophanes was carried out in acetonitrile under high-dilution conditions (0.001 M) at the reflux temperature of the mixture (Scheme 1). A solution of the tribromide **13**[12] or **14** was added drop by drop to the trispyridine or trisquinoline structures. In this way, the preparation of the target molecules was favored and polymer formation kept to a minimum. The products **5** (87%) and **6** (56%) were isolated by vacuum filtration and purified by washing with ethyl acetate and diethyl ether. When the same procedure was applied for the preparation of **7**, the product obtained proved to be impure and was purified by repeated recrystallizations from ethanol, which resulted in a low yield (8%). The structures of all the compounds were characterized by elemental analyses (C, H, N), HRMS, and $^1$H and $^{13}$C NMR spectroscopies.

The first thing that drew our attention was the high symmetry of **5**−**7**, expressed by the number of proton and carbon signals corresponding to one-third of the molecules. In the $^1$H NMR spectra of **5**−**7**, H-3 shows the largest heterocycle shift (upfield), and H-2 behaves in a manner similar to the α-proton of pyridinium ions[13]

(1) Ernst, L. *Prog. Nucl. Magn. Reson. Spectrosc.* **2000**, *37*, 47−190.
(2) Cristau, P.; Martin, M.-T.; Dau, M.-E. T. H.; Vors, J.-P.; Zhu, J. *Org. Lett.* **2004**, *6*, 3183−3186.
(3) Srinivasan, M.; Sankararaman, S.; Hopf, H.; Dix, I.; Jones, P. G. *J. Org. Chem.* **2001**, *66*, 4299−4303.
(4) Cabildo, P.; Sanz, D.; Claramunt, R. M.; Bourne, S. A.; Alkorta, I.; Elguero, J. *Tetrahedron* **1999**, *55*, 2327−2340.
(5) Scheytza, H.; Reissig, H.-U. *Tetrahedron* **1999**, *55*, 4709−4720.
(6) Conejo-García, A.; Campos, J. M.; Sánchez-Martín, R. M.; Gallo, M. A.; Espinosa, A. *J. Med. Chem.* **2003**, *46*, 3754−3757.
(7) Conejo-García, A.; Campos, J. M.; Entrena, A.; Sánchez-Martín, R. M.; Gallo, M. A.; Espinosa, A. *J. Org. Chem.* **2003**, *68*, 8697−8699.
(8) Högberg, H.-E.; Wennerström, O. *Acta Chim. Scand. B* **1982**, *36*, 661−667.
(9) Olsson, T.; Tanner, D.; Thulin, B.; Wennerstroem, O. *Tetrahedron* **1981**, *37*, 3485−3490.
(10) Garrett, T. M.; McMurry, T. J.; Hosseini, M. W.; Reyes, Z. E.; Hahn, F. E.; Raymond, K. N. *J. Am. Chem. Soc.* **1991**, *113*, 2965−2977.
(11) Surrey, A. R.; Cutler, R. K. *J. Am. Chem. Soc.* **1951**, *73*, 2623−2626.
(12) Vögtle, F.; Zuber, M.; Lichtenthaler, R. G. *Chem. Ber.* **1973**, *106*, 717−718.
(13) Katritzky, A. R.; Dega-Szafran, Z. *Magn. Reson. Chem.* **1989**, *27*, 1090−1093.

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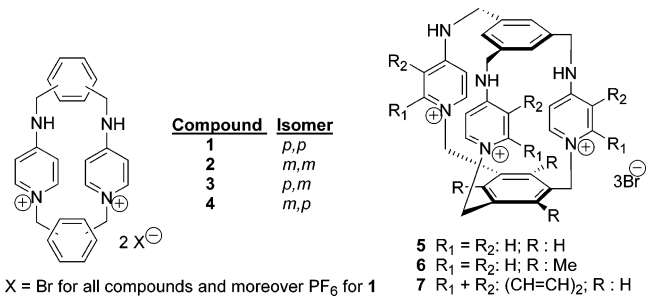
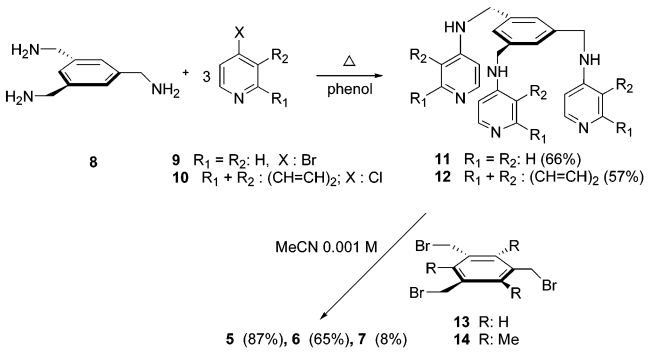


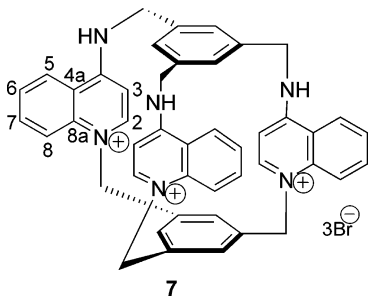

**FIGURE 1.** Structures of compounds **1**−**7**.

**SCHEME 1. Synthesis of Target Molecules 5−7**




(largest downfield). In the case of **5** and **6**, the assignment of all the proton and carbon resonances is straightforward. In compounds **5** and **6**, the symmetrical hydrogen atoms of the pyridinium rings show an important difference in chemical shift. Protons H-2 and H-3 give rise to two sets of signals ($\delta$ 8.34 ppm, d, $J$ = 7.4 Hz and $\delta$ 7.81 ppm, d, $J$ = 7.7 Hz for **7**, and $\delta$ 8.42 ppm, d, $J$ = 7.1 Hz and $\delta$ 6.97 ppm, d, $J$ = 7.1 Hz for **6**, in DMSO-$d_6$ in both cases). Similarly, two sets of resonances are observed for protons H-3 and H-5 ($\delta$ 6.85 ppm, dd, $J$ = 2.7 and 7.4 Hz and $\delta$ 6.41 ppm, dd, $J$ = 2.7 and 7.7 Hz for **5**, and $\delta$ 6.81 ppm, dd, $J$ = 2.9 and 7.1 Hz and $\delta$ 6.33 ppm, dd, $J$ = 2.7 and Hz for **6**, in DMSO-$d_6$ in both cases). Such a process was studied in depth in **1**·$PF_6^-$, and the process observed by which the different chemical shifts of the pyridinium protons showed coalescence at a high-temperature $^1$H NMR was the rotation around the C-4$_{pyridinium}$−NH$_{exocyclid}$ bond.[5] We did not consider it necessary to undertake a similar study on **5** and **6**, which would have provided only a different free energy activation, and decided to focus our efforts on the triquinolina triscationic bicyclophane **7**.

Unequivocal $^1$H and $^{13}$C NMR assignments are more complicated for bicyclophane **7**. Together with the $^1$H and $^{13}$C NMR spectra (in $CD_3OD$), a variety of two-dimensional NMR experiments were performed to allow the assignment of proton and carbon chemical shifts and to elucidate unambiguously the structure of compound **7** at room temperature. The strategy followed provided for the concerted application of several gradient-enhanced experiments such as $^1$H/$^{13}$C two-dimensional HSQC[14] and HMBC.[15] A diagram is provided to show the numbering of both proton and carbon atoms for compound **7**.




The unequivocal assignment of the benzene hydrogen atoms of the quinolinium moiety of **7** was the following step. The coupling pattern for H-5 and H-8 was the same (doublets), as was the coupling pattern for H-6 and H-7 (double doublets). The chemical shifts attributed to C-4, C-4a, and C-8a were distinguished on the basis of their HMBC connectivities. Cross correlations were observed between H-3 ($\delta$ 6.57 ppm, d, $J$ = 7.6 Hz) and C-4a ($\delta$ 119.03 ppm), between H-2 ($\delta$ 7.92 ppm, d, $J$ = 7.6 Hz), C-4 ($\delta$ 155.70 ppm), and C-8a ($\delta$ 138.62 ppm), and between C-4a ($\delta$ 119.03 ppm), H-6 ($\delta$ 7.75 ppm, dd, $J$ = 8.8 and 1.4 Hz), and H-8 ($\delta$ 8.46 ppm, d, $J$ = 8.8 Hz). Additionally, H-7 ($\delta$ 8.05 ppm, dd, $J$ = 8.8 and 1.4 Hz) exhibited a two-bond correlation to C-8a (138.62 ppm). Finally, to complete the structural assignment, the protonated carbons were unmasked by the HMQC spectra. Complete $^1$H and $^{13}$C NMR assignments are reported in the Experimental Section

To determine the conformational behavior of **7**, the structures of the conformers and the interconversion pathway between them warrant consideration. We have tried to record the $^1$H NMR spectra at low temperatures in order to analyze the conformational behavior of **7** in a $CD_3OD$ solution, but all the attempts were fruitless because no coalescence phenomenon was observed. We have considered the methylene protons and tried to exchange environments via a dynamic process over a range of temperatures. No coalescence of these two proton signals was observed in the temperature range from 25 to −60 °C. Moreover, we were unable to obtain a crystalline sample suitable for molecular structure determination by X-ray crystallography. To overcome all these problems, molecular modeling was carried out as reported previously,[7] but optimizing the different conformations by means of ab initio calculations at the 6-31G** level.

At molecular level, helical chirality[16] is usually a consequence of strong conformational preferences around covalent bonds. This is the case in molecular propellers, chiral molecules possessing two or more subunits which can be considered as "blades" (e.g., aryl or spirocyclic rings), which radiate from an axis of rotation (propeller axis).[17] Triarylmethanes are the most extensively studied structures of this class.[16a,18]

(14) Bax, A.; Pochapsky, S. *J. Magn. Reson*. **1992**, *99*, 638−643.
(15) Bax, A.; Summers, M. F. *J. Am. Chem. Soc.* **1986**, *108*, 2093−2094.

(16) (a) Meurer, K. P.; Vögtle, F. *Top. Curr. Chem.* **1985**, *127*, 1−76. (b) Prins, L. J.; Huskens, J.; De Jong, F.; Timmerman, P.; Reinhoudt, D. N. *Nature* **1999**, *398*, 498−502.
(17) Eliel, E. L.; Wilen, S. H.; Mander, L. N. *Stereochemistry of Organic Compounds*; Wiley: New York, 1994, p 1156.
(18) (a) Mislow, K. *Acc. Chem. Res.* **1976**, *9*, 26−33. (b) Mislow, K.; Gust, D.; Finocchiaro, P.; Boettcher, R. J. *Top. Curr. Chem.* **1974**, *47*, 1−28.

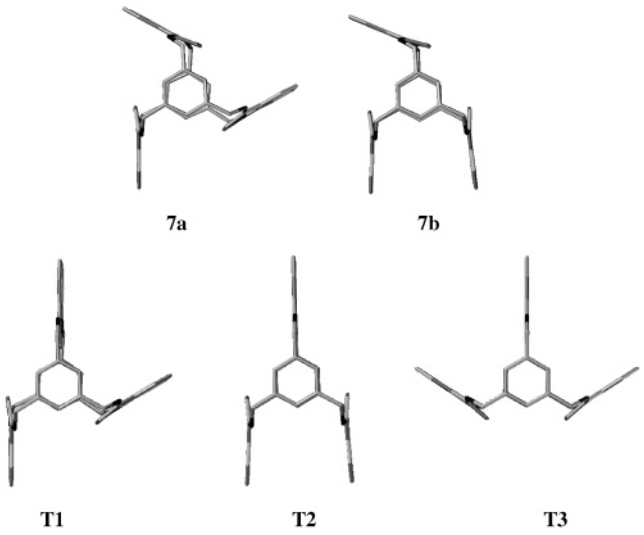



**FIGURE 2.** Top view of conformers (**7a** and **7b**) and transition states (**T1**, **T2**, and **T3**) (ab initio) for **7**.

Figure 2 shows the two energetically different conformations for compound **7**. A careful study of the conformational behavior of this molecule based on symmetry considerations allowed us to identify a total of eight conformations. These conformations make up two diastereomeric sets of enantiomers. One set of enantiomers is of $C_3$ symmetry, and consequently each enantiomer has three equivalent (homotopic) quinolinium rings (**7a** in Figure 2). The other six have $C_1$ symmetry, and each enantiomer has three nonequivalent quinolinium rings. They differ from each other in the orientation sense (clockwise or counterclockwise) of the quinolinium rings, showing two quinolinium rings in almost parallel dispositions as is shown in Figure 2 for the **7b** conformation.

The computational method indicates that conformation **7a** is the most stable conformer of the molecule. The $C_3$-symmetrical derivative **7** was found to be 1.94 kcal mol$^{-1}$ more stable than its $C_1$-symmetrical conformation. This energy difference suffices to consider **7a** and its enantiomeric one the only two conformations existing in solution at 25 °C, the conformational population being calculated as 92.2% (46.1% for each one), considering a Boltzman distribution. The other six conformations account for only 7.8 (1.3% for each one) of the conformational distribution.

When the $^1$H NMR chemical shifts of the quinolinium moieties of the open derivatives **15**[19] and bicyclophane **7** are compared (Table 1), it is observed that H-2 and H-3 are more shielded in the latter than in the former ($\delta$ 7.92 and 6.57 ppm for H-2 and H-3 for **7**, against $\delta$ 8.57 and 6.92 ppm, respectively, for **15**), whereas H-8 in **7** ($\delta$ 8.46 ppm) is more deshielded in relation to the same proton in **15** ($\delta$ 7.95 ppm). Obviously, the reason for this behavior lies in the different mobility of the quinolinium fragments, which is more restricted in **7** than in **15**. Both **7a** and **7b** conformations can correctly explain the H-2 shielding observed in relation to compound **15** since this hydrogen atom is located between both benzene rings in compound **7**. Similarly, H-8 suffers deshielding when compared to **15**. This fact is due to the higher rigidity of compound **7** that forces H-8 into a disposition in the shielding zone of the lower benzene ring. Although both **7a** and **7b** conformations may explain these findings, the higher conformational population calculated for **7a** and the simplicity of the NMR spectra in which only one-third of the theoretical signals are observed allow us to consider **7a** as the only conformation existing in solution for compound **7**.

**TABLE 1. $^1$H NMR Chemical Shifts for Compounds 7 and 15 in $CD_3OD$ Solutions**

| proton | **7** | **15**[a] |
|---|---|---|
| H-2 | 7.92 | 8.57 ± 0.01[b] |
| H-3 | 6.57 | 6.92 ± 0.01[b] |
| H-5 | 8.33 | 8.40 ± 0.01[b] |
| H-6 | 7.75 | 7.69 ± 0.01[b] |
| H-7 | 8.05 | 7.89 ± 0.02[b] |
| H-8 | 8.46 | 7.95 ± 0.03[b] |

[a] Taken from ref 19. [b] Standard deviation for six values.

**TABLE 2. Energy and Number of Occurrences for Conformers and Transition States of 7[a]**

| conformation | number of occurrences | Amber | HF 6-31G** | B3LYP 6-31G** |
|---|---|---|---|---|
| **7a** | 2 | 0.00 | 0.00 | 0.00 |
| | | | (48.1) | (46.1) |
| **7b** | 6 | 1.29 | 2.58 | 1.94 |
| | | | (0.62) | (1.29) |
| T1 | 6 | 7.89 | 8.11 | 5.52 |
| T2 | 3 | 9.30 | 9.51 | 6.32 |
| T3 | 3 | 8.03 | 9.51 | 6.63 |

[a] Conformational populations calculated using a Boltzman distribution are shown in parentheses.

Assuming that in solution, the ground state of **7** is a propeller (helical) conformation, the possibilities for its interconversion may be analyzed systematically. The interconversion of diasteromers can be considered in terms of "flip mechanisms", a "flip" being defined as a passage of one quinolinium ring through the plane perpendicular to that of the upper and lower benzene rings, which are parallel each other. To calculate the energy barrier for the interconversion between both types of conformations, transition states have been optimized by means of an ab initio calculation (HF and B3LYP, 6-31G** levels). Three possible types of transition state have been found, all of which show a quinolinium ring oriented in a radial manner in relation to both benzene rings, differing in the orientation of the two remaining heterocycles. The calculated energies for these transition states depend on the theory level employed in the calculation method, and slightly smaller values are found with the B3LYP method: 5.52, 6.32, and 6.63 kcal/mol, respectively.

The smaller values of transition states energy obtained with B3LYP method are consistent with the fact that no decoalescence processes have been observed even at a temperature as low as −60 °C. It is likely that such a dynamic process could be observed at lower temperatures, but the low solubility of this molecule does not permit the use of another solvent (different from $CD_3OD$) able to be cooled to an even lower temperature.

(19) Campos Rosa, J.; Galanakis, D.; Ganellin, C. R. *Magn. Reson. Chem.* **1998**, *36*, 951−955.

To our knowledge, compounds **5**−**7** are the first examples of triscationic tripyridina bicyclophanes, a subclass of the propellerlike cages. Compound **7** exists as two diastereomeric sets of enantiomers, one with $C_3$ symmetry and one with $C_1$ symmetry. The NMR spectrum at room temperature showed the signals corresponding to a single conformer with helical asymmetry. Compounds **5**−**7** proved to be inactive both as ChoK inhibitors (ex vivo assay) and as antiproliferative agents against the HT-29 cell line (in vitro assay).

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**Supporting Information Available:** Experimental Section. This material is available free of charge via the Internet at http://pubs.acs.org.

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