Conformational Analysis of Topiramate and Related Anion in the Solution and Interaction Between the Most Stable Conformer of Topiramate with Active Center of Carbonic Anhydrase Enzyme

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GRAPHICAL ABSTRACT

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Color versions of one or more of the figures in the article can be found online at www.tandfonline.com/lcar.
Density functional theory using the B3LYP/6-311++G** method was employed to calculate the details of the electronic structure and electronic energy of the carbonic anhydrase enzyme active center (CA); topiramate, a sulfamate substituted monosaccharide; and the complex between topiramate and CA. The calculated results indicate that topiramate appears to adopt a twist-boat conformation in the solution. The conformational analysis around the S-N bond (H-N-S-O dihedral angle) in deprotonated topiramate shows that the conformers with a H-N-S-O torsion of 270, 0, and 180 degrees are the minimum, transition state, and maximum energy conformers, respectively. The deprotonated form of topiramate is coordinated to the Zn$^{2+}$ ion.

**Keywords** Carbonic anhydrase; Topiramate; Inhibitor; Thermodynamic functions; Conformational analysis

**INTRODUCTION**

Carbonic anhydrases (CAs) are zinc-containing enzymes, which present in plants, animals, and humans.[1–6] These enzymes catalyze a simple but important physiological reaction, the interconversion of carbon dioxide and carbonic acid. In all known isozymes of CA, the Zn(II) ion is responsible for catalytic activity. The Zn$^{2+}$ ion is located at the bottom of a conical cavity that binds to three histidins from their imidazole nitrogens, with the water molecule as the fourth ligand[7–12] (Sch. 1). The compounds with sulfonamide moiety, R$_1$SO$_2$-NR$_2$R$_3$, can replace this hydroxyl in the anionic form and inhibit the CA enzyme.[13,14]

Epilepsy is a neurological disorder that ranges from a brief cessation of responsiveness to severe spasms with a lack of consciousness. In recent years, some drugs, called anticonvulsants, have been tailored with the objective of controlling epileptic symptoms.[15–18] Among different classes of pharmacological agents, topiramate (TPM),[19–24] a sulfamate substituted monosaccharide (Sch. 2), has antiepileptic effects. One main class of CA inhibitors (CAIs) is known as the metal complexing anions, which bind to the Zn(II) ion of the enzyme by substituting the nonprotein zinc ligand (Eq. 1), generating tetrahedral geometry of the Zn(II) ion. Thus, the CA-inhibitor interaction constitutes the initial stage of the mechanism of inhibitory action:

\[
[E - \text{Zn} - \text{OH}_2]^+ + I^- \overset{\text{inhibitor anion}}{\rightleftharpoons} [E - \text{Zn} - I]^+ + \text{H}_2\text{O} \quad (1)
\]

The results of experimental studies indicate that topiramate has inhibitory activity against the CA enzyme.[25,26] In addition, X-ray crystallographic studies show the binding mode of deprotonated topiramate to the active site of CA.[27,28]

The complex between topiramate and different isoforms of the CA enzyme is studied experimentally or theoretically,[29–31] but unfortunately, there are
few or no experimental and theoretical studies about rotational isomerism in sulfonamide moiety and the fructopyranose ring of topiramate.\textsuperscript{[32]} In the present study, conformational analysis of the topiramate and topiramate anion (TPM\textsuperscript{−}) in the solution and binding of TPM\textsuperscript{−} to the CA active center was performed using a quantum mechanical approach.

**EXPERIMENTAL**

**Topiramate Synthesis**

Preparation of topiramate involves two steps (Sch. 3). The first step was reaction of 2,3:4,5-bis-\textit{O}-(1-methylethylidene)-\textit{β}-D-fructopyranose 1 with
Scheme 2: Chemical structure of topiramate, 2,3:4,5-Bis-O-(1-methylethylidene)-β-D-fructopyranose sulfamate.

N-protected sulfamoyl chloride 2 in the presence of triethylamine in toluene to produce N-substituted topiramate 3. The second step was hydrolyzing of 3 at pH 3.5 to give topiramate, a white solid with mp 124–126°C.[33,34]

Scheme 3: Schematic presentation of topiramate synthesis.
Preparation of N-[(Diphenylamino) carbonyl]-2,3:4,5-bis-O-(1-ethylethylidene)-β-D-fructopyranose sulamate (3)

Diphenylamine (1.2 g, 7.1 mmol) in toluene (10 mL) was added dropwise to a solution of chlorosulfonyl isocyanate (1.2 g, 8.5 mmol) in toluene (10 mL) at −10°C under argon. The solution was stirred for 30 min, after which the mixture of 2,3:4,5-bis-O-(1-methylethylidene)-β-D-fructopyranose (1.7 g, 6.5 mmol) and triethylamine (1.4 mL, 10 mmol) in toluene was added dropwise. It was stirred at 5°C and the reaction progress was monitored with TLC. The reaction mixture was washed with water (2 × 15 mL). After separation of the organic layer, dried over MgSO4 and evaporated, the crude product was used in the next step without further purification.

Preparation of Topiramate

The crude product obtained above was dissolved in acetone (7 mL) and sodium acetate-acetic acid buffer solution (pH = 3.5). The reaction mixture was heated at 70°C for 2 h. The cooled mixture was diluted with water (7 mL) and 10 M NaOH was added to reach a pH of 13 to 14. Then, it was extracted with t-butyl methyl ether (3 × 5 mL). The aqueous phase was treated with 85% phosphoric acid to adjust the pH to 5.5 to 6, and white crystals precipitated upon cooling to 10°C. The product was collected by filtration and washed with cold water. The purity was improved by recrystallization from a mixture of acetone and water.

NMR Measurements

1H NMR, 13C NMR, COSY, and HMQC spectra of topiramate were obtained at 298 K in CDCl3 (99.99% D) on a Bruker DRX500 operating at 500.133 MHz for 1H and 125.770 MHz for 13C, using a 5-mm broadband inverse probe. All 2D NMR spectra were acquired by the pulsed field gradient method. Two-dimensional correlation spectroscopy (1H-1H COSY) was used to confirm 1H signal assignments (Fig. 1). Heteronuclear multiple quantum correlation (HMQC) and heteronuclear multiple bond correlation (HMBC) were used for 13C signal assignments (Figs. 2 and 3). HMQC and HMBC spectra were recorded using 2048 × 1024 data matrices; the number of scans and dummy scans was 48 and 16, respectively, in all cases. The HMQC and HMBC were recorded with a 2-s interpulse delay. The spectral width was \( sw_1 \times sw_2 = 3255 \times 22,123 \) Hz for all 2D experiments. For Z-only gradients, the G1:G2:G3 = 50:30:40.1 gradient ratios were used for both HMQC and HMBC spectra.
Theoretical Section

Computational Details

Ab initio calculations were carried out with the Gaussian program series 1998.\textsuperscript{34} The geometries of the CA enzyme active site, topiramate and its related anion, and the complex between topiramate and CA were fully optimized employing a hybrid Hartree-Fock density functional scheme and the adiabatic connection method—Becke three-parameter with Lee-Yang-Parr (B3LYP) functional\textsuperscript{35}—of density functional theory (DFT)\textsuperscript{36} with the standard 6-311++G\textsuperscript{**} basis set. Full optimizations were performed without any symmetry constraints. The harmonic vibrational frequencies were computed...
Figure 2: HMQC spectrum of topiramate in CDCl₃ at 298 K.

Calculation of Binding Energy

To quantify the interaction between the topiramate inhibitor and CA active site in the optimized geometries, the binding energy (BE) and complexation
energy (CE) are evaluated using Eqs. 2 and 3: \[^{[38]}\]

\[
\text{BE} = \frac{E_{\text{CA/Inh}}}{E_{\text{isolated CA}}} - (E_{\text{isolated CA}} + E_{\text{isolated Inh}})
\]

\[
\text{CE} = E_{\text{opt complex}} - (E_{\text{opt CA}} + E_{\text{opt Inh}})
\]

According to Eq. 3, the complexation energy was defined as the difference in energies of the isolated inhibitor and CA active site, at their optimized conformations, from that of the complex.

**RESULTS AND DISCUSSION**

**Geometry Optimization of CA Active Center**

The structure of the CA active center was fully optimized with the B3LYP method using the 6-311++G** basis set without initial symmetry restrictions.
Figure 4: Presentation of the optimized structure of the CA active site in the solvent; the average bond angle of N-Zn-N is 119.74 degrees and the average bond angle of N-Zn-O is 192.32 degrees.

and assuming the $C_1$ point group. The optimized geometry of the CA active center in the gas phase was optimized again by considering the solvent effect ($\epsilon = 78.9$) using the PCM method. Figure 4 shows the optimized structure and some structural details of the CA active center in the solvent. Calculation of vibrational frequencies confirmed the stationary point without the negative eigenvalue observed in the force constant matrix.

**Geometry Optimization of Topiramate**

Topiramate’s structure was also fully optimized with the B3LYP method using the 6-311++G** basis set without initial symmetry restrictions and assuming the $C_1$ point group. The optimized geometry of topiramate in gas phase was optimized again in water at the same level of calculations. Figure 5 shows the optimized structure of topiramate in $H_2O$. A selection of calculated bond
Calculation of vibrational frequencies has confirmed the stationary point without the negative eigenvalue observed in the force constant matrix.

**Calculation of Chemical Shifts and NMR Spin-Spin Coupling Constants of Topiramate**

NMR computations of absolute shieldings were performed using the GIAO method\[39\] at the DFT optimized structure in the presence of chloroform solvent. The \(^1\)H and \(^{13}\)C chemical shifts were calculated by using the corresponding absolute shielding calculated for Me\(_4\)Si at the same level of theory (Table 2). The good agreement between experimental and theoretical chemical shifts shows the reliability of DFT calculations for these series of molecules.

Recent investigations have shown that DFT can be used to calculate reliable \(J_{CH}\), \(J_{HH}\), and \(J_{CC}\) values in carbohydrates without scaling.\[40,41\] In the
Table 1: Presentation of some chemical properties of optimized topiramate in water

| Connected atom       | Topiramate |
|----------------------|------------|
| **Bond distance (Å)**|            |
| S – N1               | 1.68       |
| S = O\(^a\)          | 1.44       |
| S – O4               | 1.62       |
| O4 – C5              | 1.44       |
| C5 – C6              | 1.55       |
| C6 – C7              | 1.55       |
| C6 – O8              | 1.40       |
| **Bond angle (°)**   |            |
| N1 – S = O2          | 105.8      |
| N1 – S = O3          | 112.3      |
| N1 – S – O4          | 100.1      |
| O2 = S = O3          | 122.7      |
| O2 = S – O4          | 110.5      |
| O3 = S – O4          | 104.1      |
| S – O4 – C5          | 117.0      |
| O4 – C5 – C6         | 110.2      |
| O8 – C10 – O9        | 105.8      |
| O8 – C6 – O11        | 111.7      |
| C6 – O11 – C12       | 113.4      |
| O11 – C12 – C13      | 109.3      |
| C13 – C14 – C7       | 102.6      |
| **Dihedral bond (°)**|            |
| N1 – S – O4 – C5     | 75.4       |
| O2 = S – O4 – C5     | –35.8      |
| O3 = S – O4 – C5     | –169.4     |
| S – O4 – C5 – C6     | –170.7     |
| O4 – C5 – C6 – C7    | 48.5       |
| C6 – C7 – O9 – O10   | –43.1      |
| C6 – O8 – C10 – O9   | –2.4       |
| O8 – C6 – C7 – C14   | 173.6      |
| C7 – C6 – O11 – C12  | 59.6       |
| C6 – O11 – C12 – C13 | –50.4      |

\(^a\)Average S = O\(_1\) and S = O\(_2\).

In the present work, we extend this approach to calculate the \(J\)-coupling constants using the DFT method. \(^1\)H and \(^13\)C NMR spin-spin coupling constants in the DFT optimized structure in the presence of solvent were obtained by finite-field (Fermi-contact) double perturbation theory[^42] calculated at the B3LYP level using the 6-311++G** basis set. Appropriate values for the perturbing fields imposed on the coupled nuclei were chosen to ensure sufficient numerical precision, while still allowing a satisfactory low-order finite-difference representation of the effect of the perturbation. The result of a recent study on heparin disaccharide with O- or N-sulfated (OSO\(_3^-\) or NSO\(_3^-\)) residues showed that the Fermi contact (FC) term was not always dominant and that paramagnetic spin-orbit (PSO), diamagnetic spin-orbit (DSO), and spin-dipolar (SD)
| Chemical shifts  | Exp.       | H_13C | Calc. | Exp. | Coupling constant | Exp. | Calc. |
|------------------|------------|-------|-------|------|-------------------|------|-------|
| 5(H_R,H_S)       | 4.27–4.37  | 4.15–4.25 | 5.7, 14, 13 | 68.5–69.7 | 70–70.9 | 2J_{H_{5R},H_{5S}} | –11.05 | –11.4 |
| 7                | 4.12       | 4.33 | 6 | 100.2 | 101.3 | 2J_{H_{7},H_{14}} | 2.6 | 2.8 |
| 14               | 4.38       | 4.65 | 4.4 | 6 | 109.0 | 110.0 | 2J_{H_{14},H_{13}} | 7.9 | 8.1 |
| 13               | 4.17       | 4.28 | 4.24 | 12 | 59.7 | 61.5 | 2J_{H_{13},H_{12}} | 0.80 | 1.0 |
| 12(CH_2)         | 3.65, 3.85 | 3.82, 3.94 | 10, 17 | 25.9, 26.8 | 24.3, 25.5 | 2J_{H_{13},H_{12}^*} | 2.0 | 2.1 |
| NH_2             | 4.58       | 4.92 | 5.2 | CH_3 | 25.9, 26.8 | 24.3, 25.5 | 1J_{C_{5},H_{5R}} | 146.2 | 146.9 |
| CH_3             | 1.08       | 1.19 | (1.32) | 27.2, 28.1 | 26.1, 26.8 | 1J_{C_{5},H_{5S}} | 151.5 | 152.0 |

\[a\] DFT calculated J values in the optimized topiramate. Data in parentheses were taken from Abbate et al. \[45\]
Figure 6: Presentation of different optimized conformers of topiramate \((R = \text{S(O}_2\text{)}\text{NH}_2)\). A: twist-boat conformer; B: chair conformer; C: alternative chair conformer.

Contributions considerably influenced magnitudes of proton-proton spin-spin coupling constants. Therefore, we consider all contributions to calculate the coupling constants in the topiramate molecule.

**Conformational Properties of Pyranose Ring in the Topiramate Molecule**

According to Figure 6, the pyranose ring in the topiramate molecule could exist in three conformations. The critical parameters are the coupling constants for proton on the pyranose ring: \(2J_{H7,H14} = 2.7\) Hz, \(2J_{H14,H13} = 7.9\) Hz, \(2J_{H13,H12} = 0.8\) Hz, and \(2J_{H13,H12'} = 1.9\) Hz. For chair conformation, B, one would expect \(2J_{H7,H14}\) to be a large “anticoupling” of about 10 Hz and \(2J_{H14,H13}\) to be a small “gauche coupling” of 2 to 3 Hz, which is not the case for topiramate. The alternative chair, C, would exhibit a large coupling for one of the \(2J_{H13,H12}\) values and a small coupling for \(2J_{H14,H13}\). Neither situation is observed in the data for topiramate. To confirm twist-boat conformation for topiramate in solution, the geometry of three conformers, A, B, and C, was optimized, and coupling constants of them were calculated in solution. Table 3 indicates some structural details and some calculated proton-proton spin-spin coupling constants in the pyranose ring for the three conformers. Comparison between experimental and calculated coupling constants for the three conformers shows good
Table 3: Presentation of torsion angles of intraring protons and proton–proton coupling constants in the pyranose ring for conformers A, B, and C

| Conformer | A      | B      | C      |
|-----------|--------|--------|--------|
| Torsion angle (°) |        |        |        |
| H7C7C14H14 | −54.08 | 176.17 | 61.07  |
| H14C14C15H15 | 176.02 | −58.33 | 55.42  |
| H13C13C12H12 | 50.22  | 54.50  | −174.10|
| H13C13C12H12' | −86.74 | −65.05 | −56.50 |
| Coupling constant (Hz) |        |        |        |
| $2J_{H7,H14}$ | 2.8    | 9.7    | 2.2    |
| $2J_{H14,H13}$ | 8.1    | 2.6    | 1.7    |
| $2J_{H13,H12}$ | 1.0    | 1.4    | 9.6    |
| $2J_{H13,H12}'$ | 2.0    | 2.1    | 2.0    |

agreement between conformer A, twist-boat, and experimental data. Thus, topiramate appears to adopt a twist-boat structure in solution. A single-crystal X-ray analysis of topiramate shows a similar twist structure in the solid state.\(^{[44]}\)

The results of previous $^1$H NMR studies in cyclic diacetals of pyranose with a “cis-anti-cis” arrangement\(^{[45]}\) and topiramate\(^{[46]}\) showed that it tends to adopt a twist-boat conformation, which is in agreement with our results. It appears that the molecule is a combination of a large, globular hydrophobic region and a small hydrophilic $\text{SO}_2\text{NH}_2$ unit. We suggest that the nature and disposition of these two segments are important for the biological activity of topiramate.

**Calculation of Deprotonation Enthalpy of Topiramate**

According to reaction 1, the anionic form of topiramate binds to the Zn$^{2+}$ ion to form the complex between the active center of the CA enzyme and inhibitor, so in the next step, the anionic form of topiramate is constructed by detaching the proton from the NH$_2$ group. Total enthalpy of the studied species M and H(M) at the temperature T is usually estimated from Eq. 4:\(^{[47–49]}\)

\[
H(M) = E_0 + ZPE + E_{\text{trans}} + E_{\text{rot}} + E_{\text{vib}} + RT,
\]  

(4)

where $E_0$ is the calculated total electronic energy; ZEP stands for zero-point energy; and $E_{\text{trans}}, E_{\text{rot}},$ and $E_{\text{vib}}$ are the translational, rotational, and vibrational contributions to the enthalpy, respectively. Finally, RT represents the PV-work term and is added to convert the energy to enthalpy.

The DE is equal to $H_a + H_p - H_{ih}$, where $H_a$ is the enthalpy of the anion generated by proton abstraction, $H_p$ is the enthalpy of proton, and $H_{ih}$ is the enthalpy of the inhibitor, which is calculated by considering the solvent effect.
The DE value for the anion formed by proton abstraction from topiramate by considering the zero-point energy equals $-307.45$ kcal/mol, so deprotonation of topiramate is exothermic and topiramate has an acidic property.

**Conformational Analysis Around the S-N Bond in Deprotonated Topiramate**

Since very little is known experimentally or theoretically about the sulfonamide anion in the topiramate molecule, we focused on the conformational preferences of the anion. By employing a scan procedure, the potential energy variations were obtained while the H-N-S-O dihedral angle, as a proper reaction coordinate, was changed from 0 to 360 degrees in steps of 10 degrees with the results pictured in Figure 7. Figure 7 indicates that the minimum energy for the topiramate anion would have a H-N-S-O torsion angle of approximately 270 degrees. To ensure that the stationary points have been identified correctly, the geometry of structures with a torsion angle H-N-S-O of 0, 180, and 270 degrees was fully optimized in the gas phase and then in water. The results of frequency calculations confirm that the two conformers with a torsion angle H-N-S-O of 270 and 180 degrees without imaginary frequency refer to local minimum and intermediate, respectively, whereas the 0-degree conformer with one imaginary frequency refers to the transition state. Comparison of energy levels between different conformers after full optimization in water is shown in Figure 8. Some structural details of these three conformers are presented in Table 4.

It is noticeable that the valence geometry of the topiramate anion is rather different from the neutral molecule.\[50,51\] For example, the S-N1 bond length
decreases from 1.68 Å in neutral molecules (Table 1) to 1.56 Å in the anionic form of topiramate (Table 4), while the S=O bonds decrease from 1.47 Å in the anion to 1.44 Å in the neutral topiramate molecule. In addition, the O2=S=O3 bond angle in the anionic form of topiramate is 118 degrees, as opposed to 122 degrees in the neutral molecule. All of these differences are consistent with an increased S-N bond character and delocalization of the negative charge in the anion. Therefore, the lone pairs on the nitrogen anion can be delocalized into the \( \sigma^* \) orbital of the S-O bond.

**DFT Calculations for Complexes Between CA Active Center and Topiramate**

To follow the complexation process between the inhibitor and CA active site, the potential energy variations were obtained using the B3LYP/6-311++G* method while the distance between the N atom of the most stable conformer of the topiramate anion and Zn\(^{2+}\), as a proper reaction coordinate, was decreased from 5 Å in steps of 0.3 Å. The binding energy with respect to change in distance is presented in Figure 9. The negative BE change upon
complexation clearly demonstrates that the CA active site can form a stable complex with inhibitor. Therefore, in the presence of a deprotonated inhibitor, an enzyme/inhibitor complex forms (Eq. 1). According to Figure 9, the optimum distance between Zn\(^{2+}\) and the N atom is 1.93 Å.

| Connected atom | Topiramate anion | Intermediate | Transition state |
|----------------|------------------|--------------|-----------------|
| Bond distance (Å) | | | |
| S – N1          | 1.56             | 1.54         | 1.55            |
| S = O          | 1.47             | 1.46         | 1.46            |
| S – O4          | 1.76             | 1.75         | 1.75            |
| O4 – C5         | 1.42             | 1.41         | 1.41            |
| C5 – C6         | 1.55             | 1.55         | 1.55            |
| Bond angle (°)  | | | |
| N1 – S = O2     | 114.6            | 117.3        | 118.5           |
| N1 – S = O3     | 113.7            | 114.2        | 115.2           |
| N1 – S – O4     | 106.0            | 107.2        | 106.3           |
| O2 = S = O3     | 118.4            | 120.2        | 119.3           |
| O2 = S – O4     | 103.0            | 104.2        | 102.6           |
| O3 = S – O4     | 98.2             | 99.3         | 98.9            |
| S – O4 – C5     | 114.0            | 115.2        | 115.0           |
| O4 – C5 – C6    | 112.5            | 113.5        | 113.0           |
| Dihedral bond (°) | | | |
| H–N1–S–O4       | 270.1            | 179.5        | 1.1             |
| N1–S – O4 – C5  | 57.94            | 55.21        | 54.4            |
| O2 = S – O4 – C5| –62.80           | –59.3        | –60.5           |
| O3 = S – O4 – C5| 175.41           | 177.5        | 176.3           |
| S – O4 – C5 – C6| –145.78          | –148.3       | –147.3          |

\(^{a}\) Average S = O\(_1\) and S = O\(_2\).

Figure 9: Binding energy for complexation of topiramate anion with the CA active site.
To calculate the complexation energy, the geometry of the complex was fully optimized at the Zn-N optimum distance (Fig. 10B). As the calculated results indicate, substitution of the anionic form of inhibitor in place of the water molecule at the CA enzyme active center is energetically exothermic, $\Delta E = -192.85$ kcal/mol. Optimized geometry of the complex indicates that the CA/TPM complex has a tetrahedral geometry. Table 5 shows some structural details of this complex according to numbering in Figure 10A.

A comparison of some structural details of the CA/TPM complex with a native active center of CA indicates that a significant conformational rearrangement is necessary in the active site to allow binding of the inhibitor. This has never been evidenced in CA-inhibitor adducts before, as the active site of this protein is highly rigid. For example, according to Table 5, the average bond angle between nitrogen atoms of histidine and zinc, N-Zn-N, is about 107 degrees rather than 119.74 degrees in CA-OH$_2$; the average bond angle of N(TPM)-Zn-N(His) is about 114 degrees as compared to 92.3 degrees, which is the bond angle of O(OH2)-Zn-N(His) in CA-OH$_2$. Therefore, inhibitor binding to the zinc ion generates a structure that closely resembles a tetrahedral geometry.

**Analysis of Thermodynamic Properties of CA/TPM Complexes**

No experimental data of thermodynamic functions such as standard enthalpies of complexation ($\Delta H^{\circ}_{\text{com}}$) and the standard Gibbs free energies of
Table 5: Presentation of some structural details for complexes in water

| Connected atoms | Bond distance (Å) | Bond angle (°) | Dihedral angle (°) |
|-----------------|------------------|---------------|-------------------|
|                 | N1 – Zn          | 1.95          |                   |
|                 | N1 – S           | 1.62          |                   |
|                 | S = O<sup>a</sup> | 1.46          |                   |
|                 | S – O4           | 1.66          |                   |
|                 | Zn – N<sup>b</sup>| 2.03          |                   |
|                 | S – N1 – Zn      | 124.42        |                   |
|                 | O4 – S – N1      | 105.87        |                   |
|                 | O3 = S – N1      | 112.29        |                   |
|                 | O2 = S – N1      | 106.96        |                   |
|                 | O2 = S = O3      | 121.04        |                   |
|                 | N1 – Zn – N<sup>c</sup> | 114.87 |                   |
|                 | N – Zn – N<sup>d</sup> | 107.15 |                   |
|                 | O4–S–N1–Zn<sup>−</sup> | −123.83 |                   |
|                 | O2 = S – N1–Zn<sup>−</sup> | −10.65 |                   |
|                 | O3 = S – N1 – Zn<sup>−</sup> | 126.38 |                   |
|                 | S – N1 – Zn – N5<sup>−</sup> | −20.75 |                   |
|                 | S – N1 – Zn – N6 | −143.30       |                   |
|                 | S – N1 – Zn – N7 | 104.43        |                   |

<sup>a</sup>Average S = O<sub>2</sub> and S = O<sub>3</sub>.
<sup>b</sup>Average Zn – N<sub>5</sub>, Zn – N<sub>6</sub> and Zn – N<sub>7</sub>.
<sup>c</sup>Average N<sub>1</sub> – Zn – N<sub>5</sub>, N<sub>1</sub> – Zn – N<sub>6</sub> and N<sub>1</sub> – Zn – N<sub>7</sub>.
<sup>d</sup>Average N<sub>5</sub> – Zn – N<sub>6</sub>, N<sub>5</sub> – Zn – N<sub>7</sub> and N<sub>6</sub> – Zn – N<sub>7</sub>.

formation (ΔG<sup>°</sup><sub>com</sub>) for the CA/topiramate complex are available, so ΔU<sup>°</sup><sub>com</sub>,
ΔH<sup>°</sup><sub>com</sub>, ΔS<sup>°</sup><sub>com</sub>, and ΔG<sup>°</sup><sub>com</sub> were calculated for the CA/TPM complex according to Eq. 1. The standard enthalpy change of the reaction (ΔH<sup>°</sup><sub>com</sub>) is given as:

$$\Delta H_{\text{com}} = [H_{\text{complex}} + H_{\text{H2O}}] - [H_{\text{E_zn(II)OH2}} + H_{\text{inhibitor}}]$$

(5)

Which total standard enthalpies of the studied species, at the temperature T estimated from the expression 4.

Similarly, ΔS<sup>°</sup><sub>com</sub> was obtained with:

$$\Delta S_{\text{com}} = [S_{\text{complex}} + S_{\text{H2O}}] - [S_{\text{E_zn(II)OH2}} + S_{\text{inhibitor}}]$$

(6)

Table 6: Calculated thermodynamic properties of CA/TPM complex in water

| ΔE (kcal/mol) | ΔH (kcal/mol) | ΔS (cal/mol.K) | ΔG (kcal/mol) |
|--------------|---------------|----------------|---------------|
| −192.85      | −192.85       | −5.73          | −194.56       |
According to the thermodynamic equation, $\Delta G = \Delta H - T \Delta S$, the $\Delta G_{\text{com}}$ was calculated. The computed thermodynamic properties for the CA/TPM complex are reported in Table 6. The results indicate that substitution of the anionic form of inhibitor in place of the water molecule at the active center of the CA enzyme is energetically exothermic.

**CONCLUSION**

Ab initio calculations have been carried out to study the complexation between the most stable conformer of the topiramate anion in solution, a potent inhibitor, with the active site of the CA enzyme. The results show that the sulfonamide anion moiety of topiramate binds to the $\text{Zn}^{2+}$ ion in CA through electrostatic interaction. The results of conformational analysis of the pyranose ring in topiramate and sulfonamide moiety in the topiramate anion indicate that the twist-boat conformer is the most stable conformer of the pyranose ring as compared to the chair and alternative chair conformers in the solvent. In addition, the preferred O-S-N-H torsion angle is about 270 degrees in the most stable anionic form of topiramate in solution. According to the frequency calculation results, the conformer with O-S-N-H torsional angles of 180 and 0 degrees are intermediate and transition states that are 6.3 kcal/mol and 15.2 kcal/mol higher than the global minimum. The results of thermodynamic calculations show that the substitution of the anionic form of inhibitor in place of the water molecule at the active center of the CA enzyme is energetically exothermic. We hope that these data may be useful for the design of new CAIs.

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