Journal of Inclusion Phenomena and Macrocyclic Chemistry (2006) 56: 225–235
DOI 10.1007/s10847-006-9088-z

A Simulation on the Complexation of Cyclodextrins with Phospholipid Headgroups

YAN-MIN YU[1], WENSHENG CAI[1,*] and XUEGUANG SHAO[2]
[1]*Department of Chemistry, University of Science and Technology of China, 230026, Hefei, Anhui, P.R. China;*
[2]*Department of Chemistry, Nankai University, 300071, Tianjin, P.R. China*

Key words: cyclodextrin, flexible docking algorithm, inclusion complex, molecular mechanics, phospholipid headgroup

Abstract

The inclusion complexes of α-, β- and γ-cyclodextrin (CD) with three isolated phospholipid (PI – phosphatidylinositol; PS – phosphatidylserine; and PE – phosphatidylethanolamine) headgroups were studied using a flexible docking algorithm FDOCK based on molecular mechanics (CFF91 force filed). In the three phospholipid headgroups, PI headgroup exhibits the strongest affinity for CD, and the affinity of PS headgroup is greater than that of PE headgroup. By investigating the energy distribution and the complex structure in the inclusion procedure, it can be found that the van der Waals force is the main driving force responsible for the complexation. For the α-CD complex of PI headgroup, more than one inclusion complex should coexist due to the steric hindrance, which is reasonably consistent with the experimental results. Furthermore, analyses of the complex of PS and PE headgroup with α-CD also show that two or three possible complexes may appear in the inclusion process, and the complex structure with full inclusion is of the lowest energy and should be the most stable structure in the mixture. For β- and γ-CD, the energies of the most stable complexes structures for the three phospholipids headgroups were also discussed.

Introduction

Cyclodextrins (CDs) are able to form inclusion complexes with various guest molecules [1, 2]. This property has attracted increasing attention in pharmaceutical field to modify physicochemical properties of drug molecules, such as solubility, stability and bioavailability, to reduce their toxicity and side effects, or to suppress unpleasant taste or smell [3]. The most frequently studied CDs are α-, β-, and γ-CD, consisting of six, seven and eight glucopyranose units, respectively. The number of these units determines the dimension of the torus-shaped hydrophobic cavity, in which guest molecules of suitable size can be accommodated in rapidly established equilibria. Computer simulations are frequently used to rationally explain the experimental findings concerning inclusion and recognition, and strengthen the understanding of inclusion phenomena [4–17]. It is worth noting that hemolytic character is also an important biomedical aspect of CDs, and it can increase the erythrocytic membrane permeability [18]. Therefore, the hemolytic activity of CDs severely limits their parenteral use in medicine. Phospholipids are the major components of biological membranes composed of a hydrophilic (polar) head and a hydrophobic tail. CDs are reported to be able to pull phospholipids out of the cell membrane to form lipid–CD complexes [19–22]. This complex-mediated extraction of lipids from the membrane may result in the rupture of the erythrocytic membrane [18, 23].

A non-specific lipid extraction from the membrane was first proposed as the mechanism responsible for the hemolytic activity of CDs [23]. In 1994, an NMR study demonstrated that α-CD interacts very strongly with phosphatidylinositol (PI), and only weakly with phosphatidylcholine [24]. This result suggests that the nature of the lipid headgroup is one key factor governing the association of CD with phospholipids. Therefore, the interaction of α-CD with multilayers of phospholipids bearing different headgroups was measured by NMR spectroscopy [25, 26], and the interaction of α-CD with PI headgroup was also studied by molecular dynamics [27]. However, the detail about the energy distribution and the most stable complex structures in the process of the phospholipid headgroup passing through the CD cavity is still not clear.

In this paper, the inclusion complexes between isolated headgroups of three phospholipids (PI; PS – phosphatidylserine; and PE – phosphatidylethanolamine) and different CDs (α-, β- and γ-CD) were examined by a flexible docking method, FDOCK [28, 29]. With which a step-by-step inclusion passage of phospholipid

* Author for correspondence. E-mail: wscai@ustc.edu.cn

headgroup through the CD cavity with two orientations was investigated, which can quickly give the information of the energy distribution and the energy barriers in the inclusion procedure. Using the information obtained, the stable binding sites of phospholipid headgroup in the CD cavity can be determined. By investigating the energy distribution and the complex structure of each binding site in the inclusion procedure, it can be found that the van der Waals force is the main driving force responsible for the complexation. For the α-CD complex of PI headgroup, more than one inclusion complex should coexist due to the steric hindrance. While for PS and PE headgroup, the complex of full inclusion is of the lowest energy and should be the most stable structure. For the affinity of the three phospholipid headgroups for each CD, the rank of PI headgroup > PS headgroup > PE headgroup can be obtained. To compare, the complexation of β- and γ-CD with PI, PS, and PE, were also studied.

Theory and method

Molecular modeling and simulation

The initial structures of α-, β- and γ-CD were taken from the crystal structures [30–32]. The original structure of PE headgroup was deduced from dilauroylphosphatidylethanolamine (DLPE) [33] by removing the alkyl chains and glycerol backbone. For PS and PI headgroup, the structures were constructed from PE headgroup, where the ethanolamine headgroup was replaced by serine and inositol, respectively. The inositol group was taken from myo-inositol anhydrate [34, 35]. All the initial structures were energy-minimized using the conjugate gradient method under the CFF91 force field in the Discover module within the insight II software [36]. To help the analysis of the final results, CDs were oriented to have almost all the glycosidic oxygen atoms in the XY plane. The origin of the coordinates was placed at the geometry center of CD, and the direction from the wide side to the narrow side of CD was taken as the positive direction of Z-axis (Figure 1(a)). CDs and all the phospholipid headgroup molecules were oriented with its principal axis along with the Z-axis before calculation.

In order to obtain more detail information about the energy barriers in the inclusion process and the possible binding regions of phospholipid headgroup in the CD cavity, the phospholipid headgroup was moved towards CD along Z-axis step by step with two different orientations and forced to penetrate the CD cavity. Taking the headgroup of PI as an example, Figure 1(b) and (c) are graphical representations of the two orientations, in which the orientation of inositol ring facing the narrow side and the wide side were defined as orientation I and orientation II, respectively. Two orientations were calculated in two independent runs of FDOCK by rotating the molecule to orientation I or orientation II as the initial input structure and keeping the orientation during the moving. The lowest energy structure of every step and the corresponding energies were recorded to a trajectory file for further analysis.

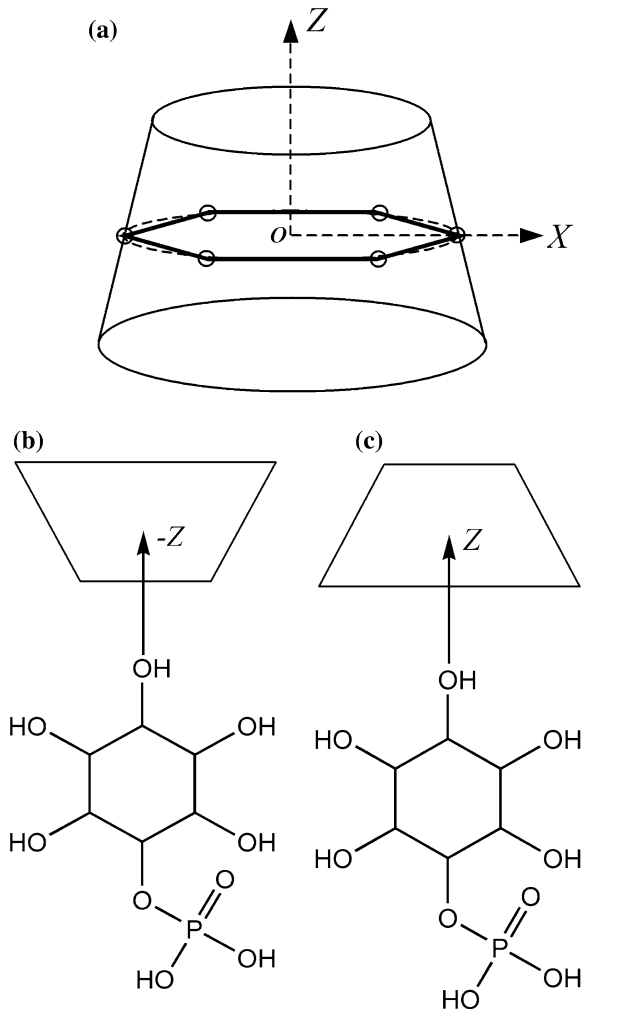


Figure 1. Schematic representation of the CD orientation used in this work (a) and the two orientations (taking the headgroup of PI as an example) of the guest passing through the CD cavity: (b) orientation I, (c) orientation II.

Flexible docking

In this study, the complete conformational flexibility for both host and guest molecules has been taken into account. All the calculations were performed using the flexible docking algorithm FDOCK, which is a combination of fast annealing evolutionary algorithm (FAEA) [37] with the limited memory BFGS (L-BFGS) algorithm. In which, FAEA is used for the global optimization and L-BFGS is used for a local optimization.

In rigid docking, the host and the guest are considered to be rigid, and the parameters to be optimized are the relative position (T_x, T_y, T_z) and the relative orientation (θ, φ, ψ) of the guest molecule in the cavity of CD. It is suitable for less flexible system [38]. While in flexible docking concerning the flexibility of the host and the guest, the internal coordinates, i.e., the bond lengths, bond angles, and dihedral angles, of each molecule also need to be optimized. In this paper, taking orientation I

as an example, the host (CD) was firstly fixed at the original point as in Figure 1b. The guest (phospholipid headgroup) was located at the narrow side of the cavity and 15 Å away from the host, then, it was moved across the cavity along the Z-axis step by step to the wide side. For each step movement, an initial complex structure can be obtained, and the relative position (T_x, T_y, T_z) and the relative orientation (θ, φ, ψ) of the guest molecule in the cavity of CD were firstly optimized by FAEA, and then, the coordinates of the host and guest molecules were optimized with L-BFGS by minimizing the score function to obtain a reasonable conformation. Finally, a random local search procedure was used to optimize both the relative position and orientation of the guest molecule in the cavity, as well as the atomic coordinates of each molecule to obtain the lowest energy structure of this step. The lowest energy structure of each step was saved to a trajectory file for further analysis.

Binding free energy function

In FDOCK, the complete CFF91 force field [39] is adopted to evaluate the energy involved in the complexation process, and an implicit solvent model [40] is used to calculate the solvation energy of the complexation. The empirical binding free energy is given by

$$\Delta G_{\text{total}} = E_{\text{inter}} + \Delta E_{\text{intra}} + \Delta G_{\text{sol}} \tag{1}$$

in which, the interaction energy E_{inter} consists of the van der Waals term E_{vdw} and the electrostatic term E_{elec} between host and guest molecules. ΔE_{intra} is the changed conformational energy of the host and the guest, including the changed energies of bond stretching, angle bending, torsional energy, out of plane bending, and all the cross terms, as in Eq. (2).

$$\begin{aligned}\Delta E_{\text{intra}} = {} & \Delta E_{\text{vdw}} + \Delta E_{\text{elec}} + \Delta E_{\text{bond}} + \Delta E_{\text{angle}} + \Delta E_{\text{tor}} \\ & + \Delta E_{\text{out\_of\_plane}} + \Delta E_{\text{bond\_bond}} + \Delta E_{\text{angle\_angle}} \\ & + \Delta E_{\text{bond\_angle}} + \Delta E_{\text{bond\_dihedral}} \\ & + \Delta E_{\text{angle\_dihedral}} + \Delta E_{\text{angle\_angle\_dihedral}} \\ & + \Delta E_{\text{bond\_bond\_1\_3}}\end{aligned} \tag{2}$$

ΔG_{sol} is the solvation energy calculated by an implicit solvent model based on the solvent accessible surface area (SASA) [41].

In the CFF91 force field, hydrogen bonds are taken as a natural consequence of the standard van der Waals and electrostatic parameters. Therefore, no extra hydrogen bond energy term is used in this study. The CFF91 parameter set is used in the calculations and the distance dependent dielectric constant ($\varepsilon = 4r_{ij}$) is adopted.

Results and discussion

Energy profiles of rigid docking for α-CD complexes

In order to investigate the detail information of energy barriers that exist in the inclusion process, all the molecules were firstly considered to be rigid, and the phospholipid headgroup was forced to penetrate the α-CD cavity along Z-axis from 15 to −15 Å with orientation I and from −15 to 15 Å with orientation II in 300 steps. The two calculations were carried out in independent runs. These results constructed a function of energy with Z-coordinate of the geometry center of the guest. Figure 2(a), (b), and (c) give the energy profiles of PI, PS, and PE headgroup, respectively.

In Figure 2(a), the prominent energy peak shows the existence of a large energy barrier during PI headgroup passing through the cavity of α-CD due to the large volume of PI headgroup. In the case of PS and PE headgroup, it can be found from Figure 2(b) and (c) that the energy profiles have the similar change tendency. For PS headgroup with orientation I, there are two main energy barriers during the moving. The first energy barrier corresponds to the inclusion of the serine group into the narrow side of α-CD, and the second one appears when the phosphate group is moved into the narrow side. The two energy barriers have the contrary order for orientation II. These results reveal the presence of steric hindrance during the complexation.

Energy distribution of flexible docking for α-CD complexes

The host and the guest were considered to be flexible, and the changes of bond length, bond angle and dihedral angle were involved in complexation. The energy minimization was performed following the same approach as in the previous section. The added flexibility of the internal coordinates can explore the conformational space in the binding region. Figure 3 gives the energy distribution and the contribution of the component energy term to the total energy in the inclusion procedure for the complexes of the three phospholipid headgroups with α-CD, respectively. From these figures, it can be found that, in the whole process, the change of total energy mainly comes from the van der Waals interaction. While the electrostatic interaction is much weaker than the van der Waals interaction, and the jumps in the electrostatic energy reflect hydrogen bond breaking or reforming. This indicated that the van der Waals interaction makes a main contribution to the total energy, and it is the main driving force responsible for the complexation.

Figure 3(a) gives the energy distribution of the PI headgroup complex. From this figure, it can be seen that the distribution of total energy is not continuous but aggregated into several groups. The position of each group corresponds to a binding region and a locally stable complex structure. By comparing Figure 2(a) with Figure 3(a), it is very clear that the interrupted region in Figure 3(a) is the position where the big energy barrier appears. For PI headgroup, more than one binding region appears in the inclusion process, as it can be seen from Figure 3(a). This indicates that more than one locally stable complex structure exists in the inclusion process.

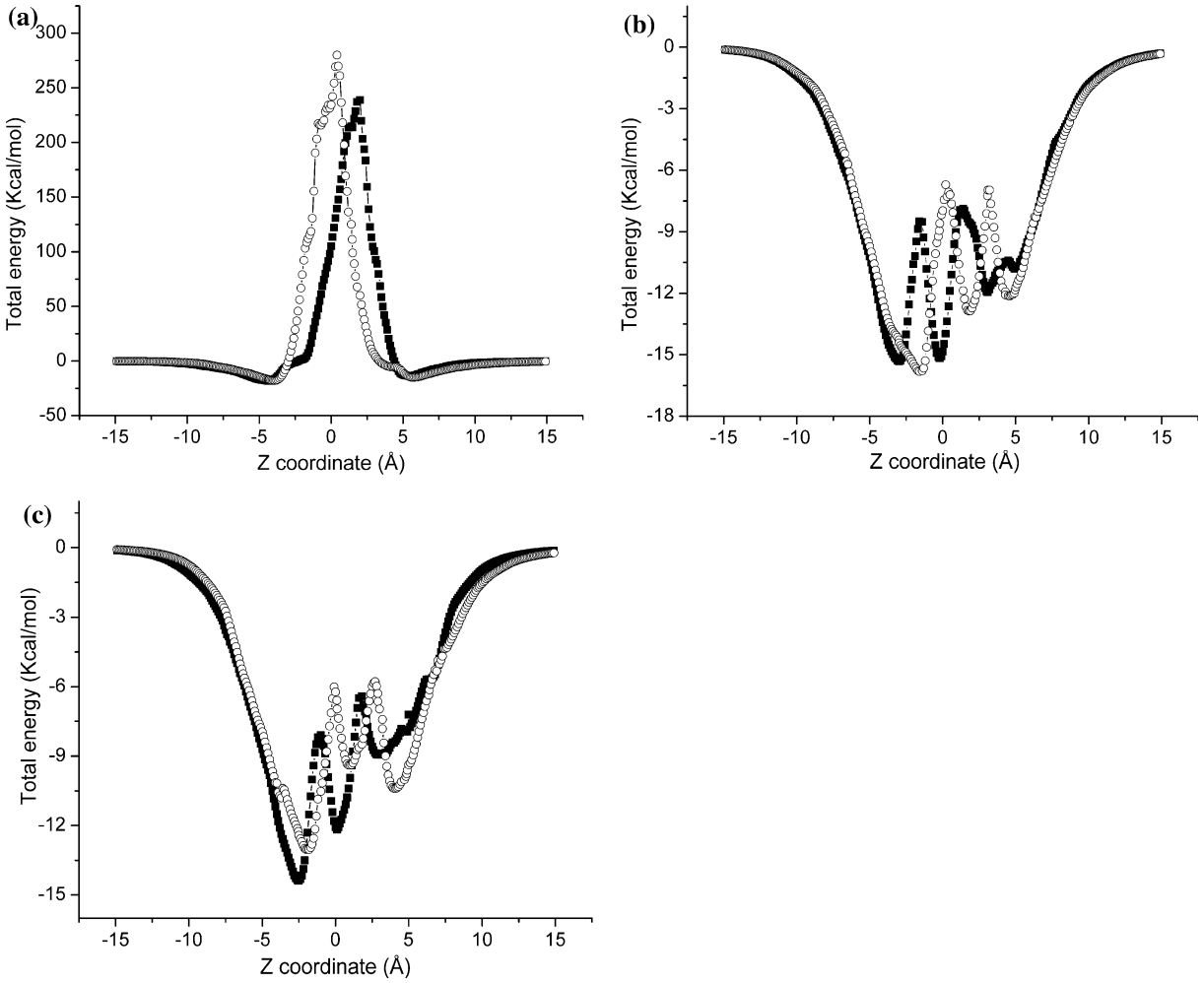


Figure 2. The total energy change profiles for the three phospholipid headgroups passing through α-CD cavity calculated by rigid docking for orientation I (the solid square) and orientation II (the hollow circle). (a) PI headgroup; (b) PS headgroup; and (c) PE headgroup.

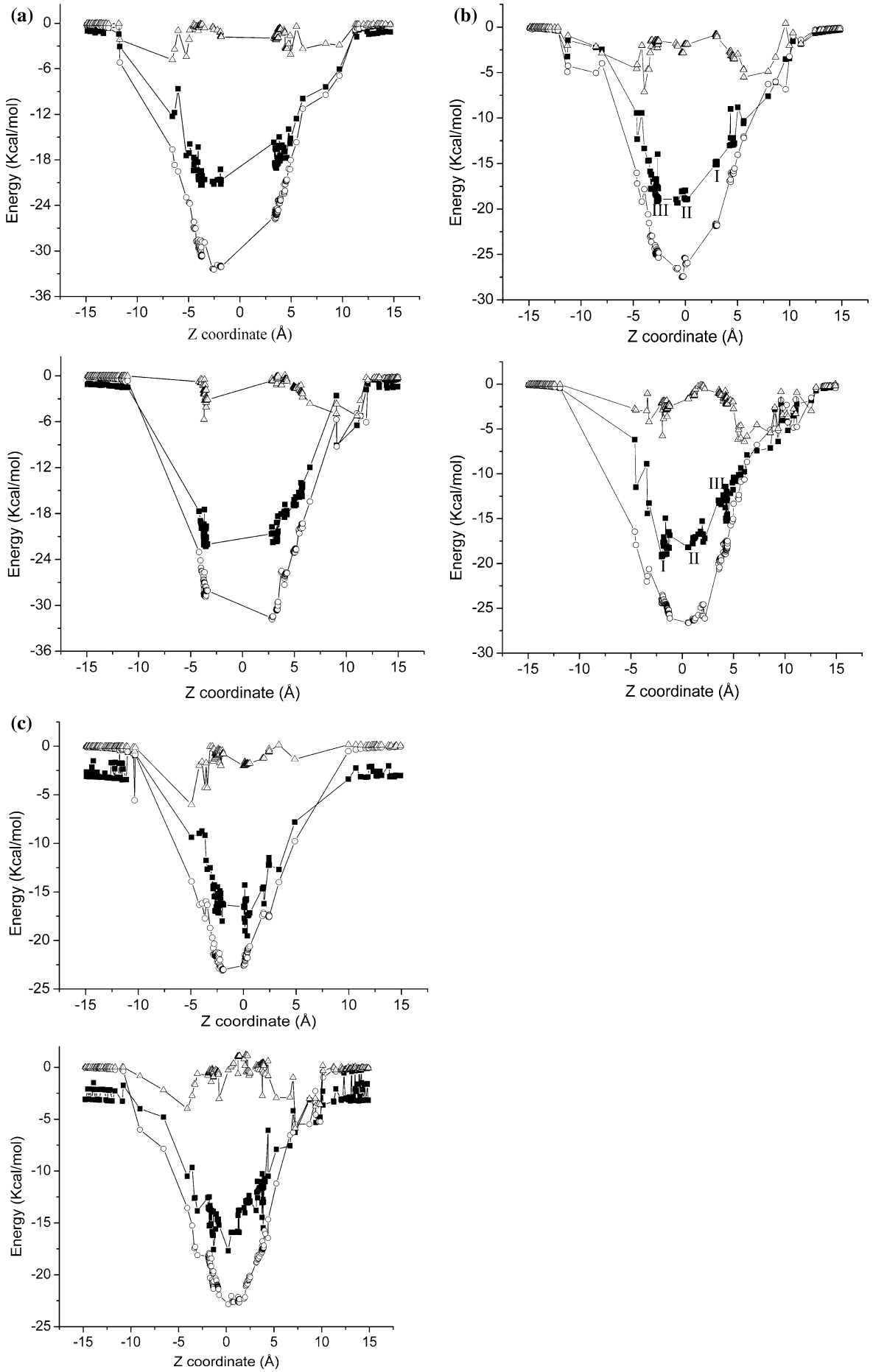
In the case of PS and PE headgroup, the energy distributions are similar, as we can see from Figure 3(b) and (c). For PS headgroup, there are three binding regions in the inclusion process both for orientation I and for orientation II. However, not every binding region is reasonable in the real experiment due to the existence of energy barriers, which will be discussed in the following section.

Structural analysis for the complexes of α-CD with PI headgroup

Figure 4 gives the locally stable complex structures, which are taken from the lowest energy structure of every binding region, in the process of PI headgroup passing through the α-CD cavity with orientation I. The corresponding energies are summarized in the first, second and third rows of Table 1.

Although moving from structure (a) to structure (b), the guest molecule must cross an energy barrier, the movement causes the decrease of total energy. Therefore it is possible that PI headgroup overcomes the energy barrier and forms the structure (b) in the inclusion process. Both structure (a) and structure (b) are in agreement with the NMR experiment [27]. Comparing the three structures, structure (b) is the tightest structure and is of the strongest intermolecular interaction. It is probably that the tight inclusion increases the intermolecular interaction. Although structure (c), in which the phosphate group included into the wide side of the α-CD cavity, is of the lowest total energy, it is a very loose structure and is of the weaker intermolecular interaction comparing with structure (b). This indicates that, structure (c) should be less preferential than structure (b). Comparing structure (a) with (b), it is clear that structure (a), i.e., partially included state, has the higher

Figure 3. The energy distribution of the inclusion complexes for the three phospholipid headgroups passing through α-CD cavity calculated by flexible docking for orientation I (the upper) and orientation II (the lower). The solid squares, hollow circles, and hollow triangles correspond to the distribution of the total energy, van der Waals energy, and electrostatic energy, respectively. (a) PI headgroup; (b) PS headgroup; and (c) PE headgroup.


(a)
(b)
(c)
Energy (Kcal/mol)
Z coordinate (Å)
I
II
III

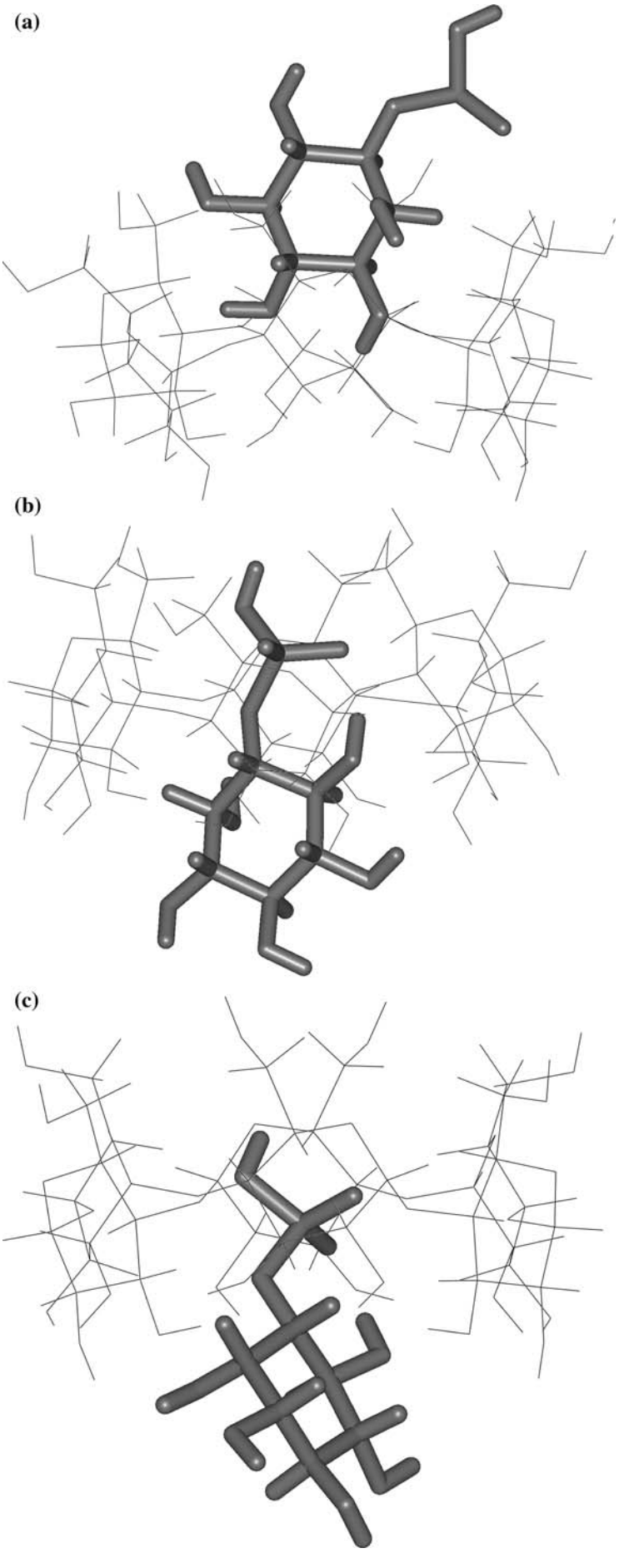

(a)
(b)
(c)

Figure 4. Locally stable complex structures between α-CD and PI headgroup with orientation I, in which plot (a), (b), and (c) represent the corresponding structure (a), (b), and (c) for PI headgroup with orientation I listed in Table 1, respectively.

total energy and the weaker intermolecular interaction, and should be less stable. Therefore, for the α-CD complex of PI headgroup with orientation I, more than one complex structure should be coexistent. Structure (b), i.e., full inclusion, should be the most stable structure. It is consistent with the simulation result of the interaction between complete PI and α-CD by molecular dynamics [42].

For orientation II, it can be obtained from Figure 2(a) that, the steric hindrance exists when the inositol ring was located at the narrow side of α-CD cavity. Therefore, it is very hard to form the full inclusion structure in the inclusion process. From the lower part of Figure 3(a), it can be seen that two binding regions exist in the process of PI headgroup penetrating the α-CD cavity with orientation II. The lowest energy structures taken from every binding region are shown in Figure 5, and the corresponding energies are also listed in Table 1. From the total energy and the intermolecular interaction of the inclusion complexes, it can be seen that structure (a) is the most energetically favorable. Moreover, the further movement from structure (a) to structure (b) must cross a big energy barrier and cause energy increasing. Therefore, the probability for the occurrence of structure (b) should be very little, and structure (a) should be the most stable structure for orientation II.

On the other hand, structure (a) existing in orientation II is a loose structure and the corresponding intermolecular interaction is weaker than that of the most stable structure existing in orientation I. Therefore, structure (a) that exists in orientation II, in which the inositol ring partially inserts into the wide side of α-CD, is less preferential than the most stable structure that exists in orientation I.

Complex structures of α-CD with PS headgroup and PE headgroup

For PS headgroup with orientation I, the most stable complex structure is given in Figure 6(a), and the corresponding energies can be found in Table 1. From the upper part of Figure 3(b), it can be seen that there are three binding regions in the inclusion process of α-CD with PS headgroup with orientation I. Every binding region corresponds a locally stable complex structure. The lowest energy structure of binding region II, in which PS headgroup is fully included into the α-CD cavity as shown in Figure 6(a), is a tight full inclusion structure. Moreover, it has the lowest total energy and the strongest intermolecular interaction comparing with that of binding region I and III. Though the movement from binding region I to binding region II must cross an energy barrier, the barrier is not so big as it can be seen from Figure 2(b), and the movement can decrease the energy. Therefore, the lowest energy structure in binding region II should be the most stable structure in the inclusion process.

Table 1. The energies obtained by FDOCK for the complexes of the three phospholipid headgroups with α-, β- and γ-CD[a]

Guest	Host	Orient	ΔG_{total}	E_{inter}	E_{vdw}	E_{elec}	ΔG_{sol}	ΔE_{intra}	Z[b]
PI	α-CD	I (a)	−19.065	−27.797	−25.591	−2.206	5.648	3.085	3.536
		I (b)	−21.181	−33.269	−32.440	−0.829	8.773	3.314	−2.528
		I (c)	−21.320	−31.072	−30.728	−0.344	8.174	1.578	−3.763
		II (a)	−22.282	−31.238	−28.130	−3.108	7.017	1.939	−3.550
		II (b)	−21.483	−30.885	−30.579	−0.306	7.527	1.875	3.354
PS	α-CD	I	−19.317	−28.578	−26.605	−1.973	8.725	0.536	−0.786
		II	−19.242	−26.434	−24.332	−2.102	7.576	−0.383	−2.001
PE	α-CD	I	−19.529	−23.532	−21.791	−1.741	6.091	−2.088	0.370
		II	−17.670	−23.082	−22.815	−0.267	6.213	−0.801	0.204
PI	β-CD	I	−28.119	−36.787	−35.022	−1.765	7.306	1.362	−0.903
		II	−27.646	−39.135	−32.278	−6.857	7.484	4.006	1.015
PS	β-CD	I	−19.741	−27.277	−25.594	−1.683	7.220	0.315	−0.721
		II	−19.650	−28.060	−24.634	−3.426	7.694	0.716	−0.077
PE	β-CD	I	−18.557	−21.946	−20.920	−1.026	5.576	−2.188	−0.692
		II	−18.066	−21.992	−20.946	−1.046	5.343	−1.416	−1.089
PI	γ-CD	I	−25.946	−35.092	−33.140	−1.952	7.864	1.283	−0.207
		II	−27.021	−37.909	−32.853	−5.056	8.007	2.881	0.128
PS	γ-CD	I	−19.339	−26.812	−25.192	−1.620	7.631	−0.157	0.030
		II	−18.508	−25.972	−24.697	−1.275	7.691	−0.226	−0.191
PE	γ-CD	I	−15.585	−20.125	−19.187	−0.938	5.389	−0.849	−0.273
		II	−15.847	−18.924	−18.349	−0.575	4.724	−1.646	−0.466

[a]The energy unit is kcal/mol, and $\Delta G_{total} = E_{inter} + \Delta G_{sol} + \Delta E_{intra}$, $E_{inter} = E_{vdw} + E_{elec.}$
[b]The unit of Z values is Å.

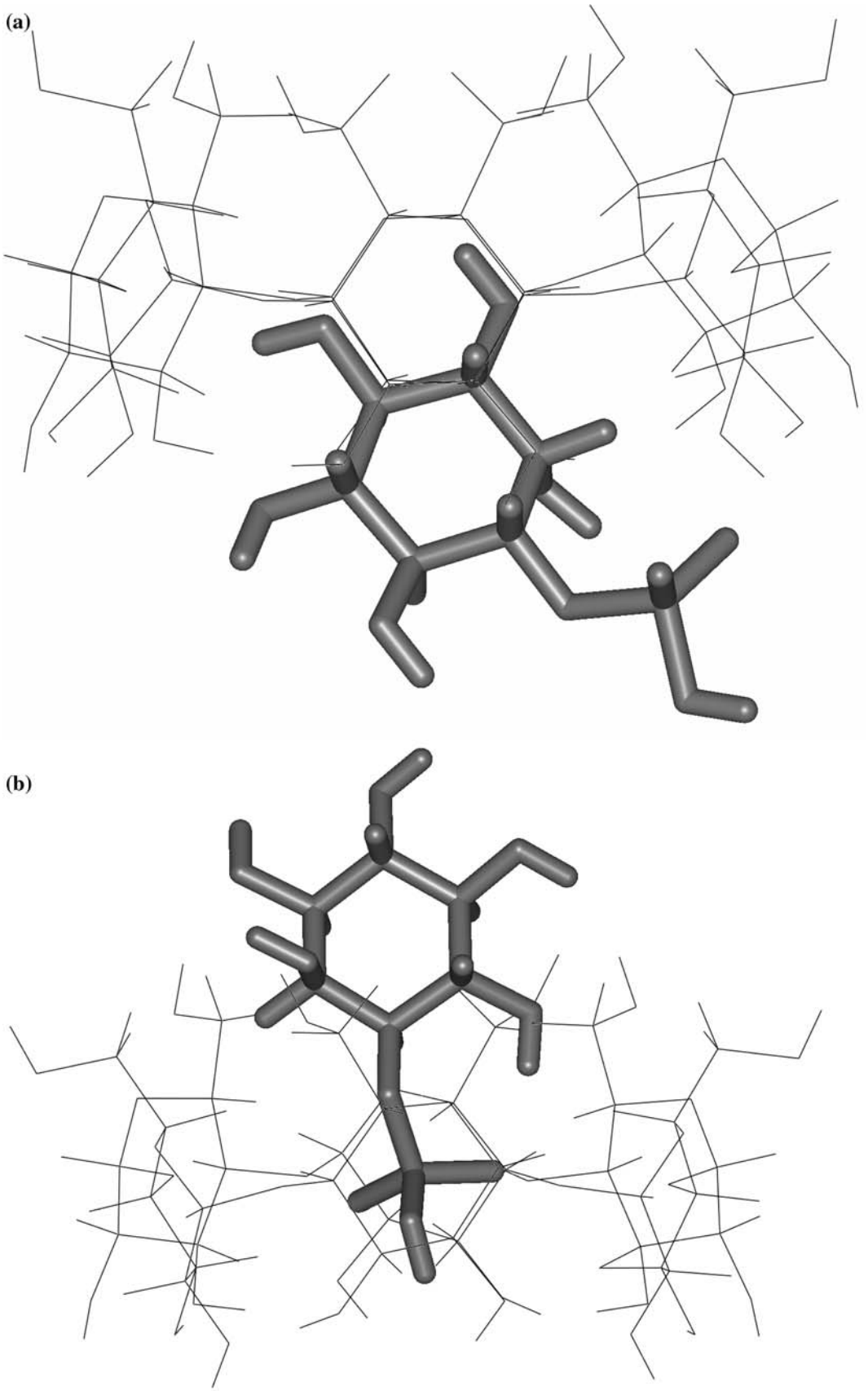


Figure 5. Locally stable complex structures between α-CD and PI headgroup with orientation II, in which plot (a) and (b) represent the corresponding structure (a) and (b) for PI headgroup with orientation II listed in Table 1, respectively.

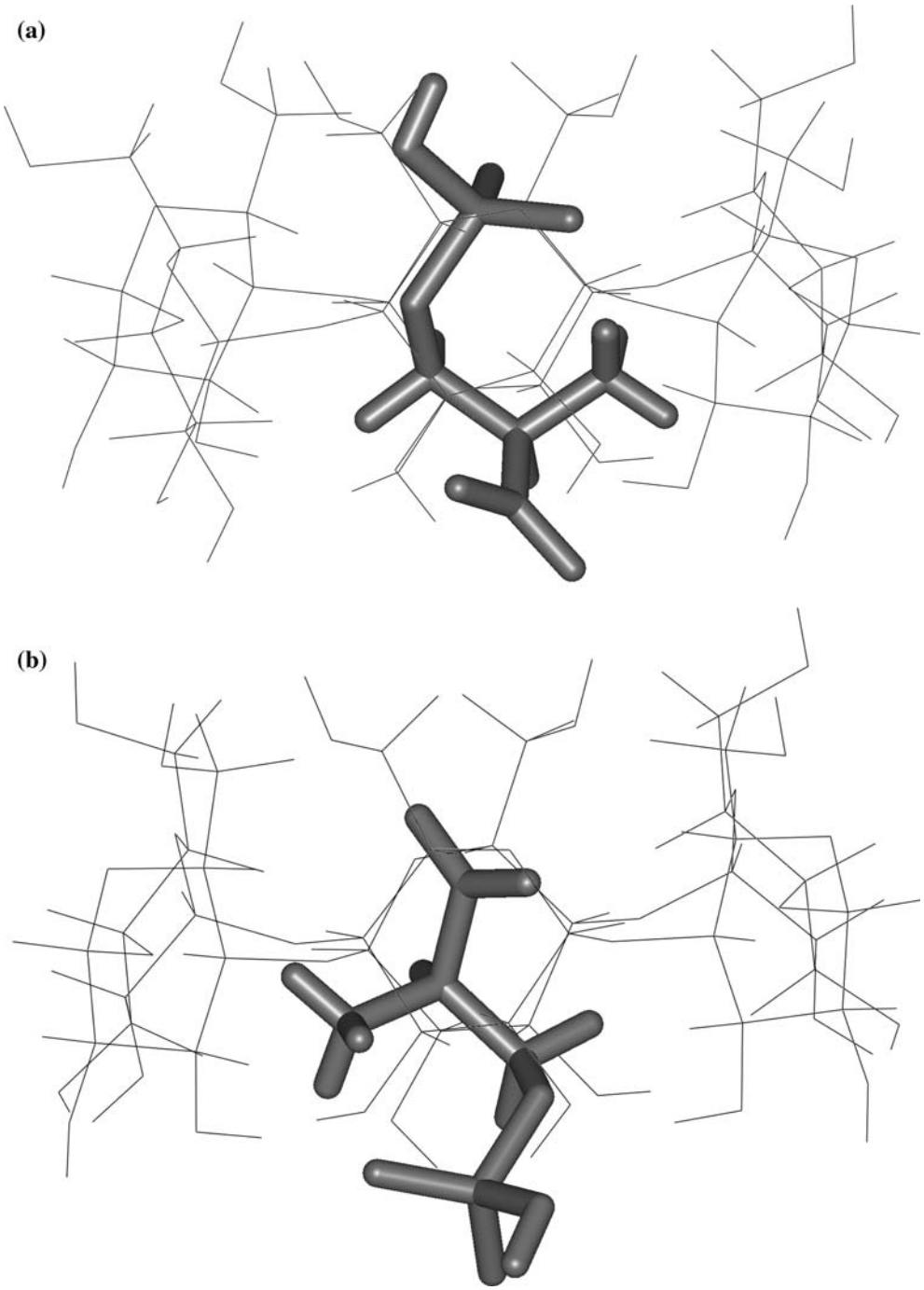


Figure 6. The most stable complex structure between α-CD and PS headgroup with orientation I (a) and orientation II (b).

For orientation II, the most stable structure is given in Figure 6(b), and the corresponding energy values can also be found in Table 1. As shown in the lower part of Figure 3(b), there are also three binding regions in the inclusion process for PS headgroup with orientation II. The lowest energy structure of binding region I is the most energetically favorable in the whole inclusion process, and no energy barrier exists in the process of reaching the biding region I. The further movement from binding region I to binding region II and III must overcome the energy barriers and result in the energy increasing. Therefore, the probability for the occurrence of binding region II and III should be very little. Therefore, the lowest energy complex structure of binding region I should be the most stable structure for the α-CD complex of PS headgroup with orientation II.

The inclusion process of PE headgroup is very similar with PS headgroup. The most stable structures, in which PE headgroup is fully included into the α-CD cavity, are shown in Figure 7, and the corresponding energy values are also listed in Table 1. From the energy values, it can be seen that the affinity of PE headgroup for α-CD is slightly less than that of PS headgroup.

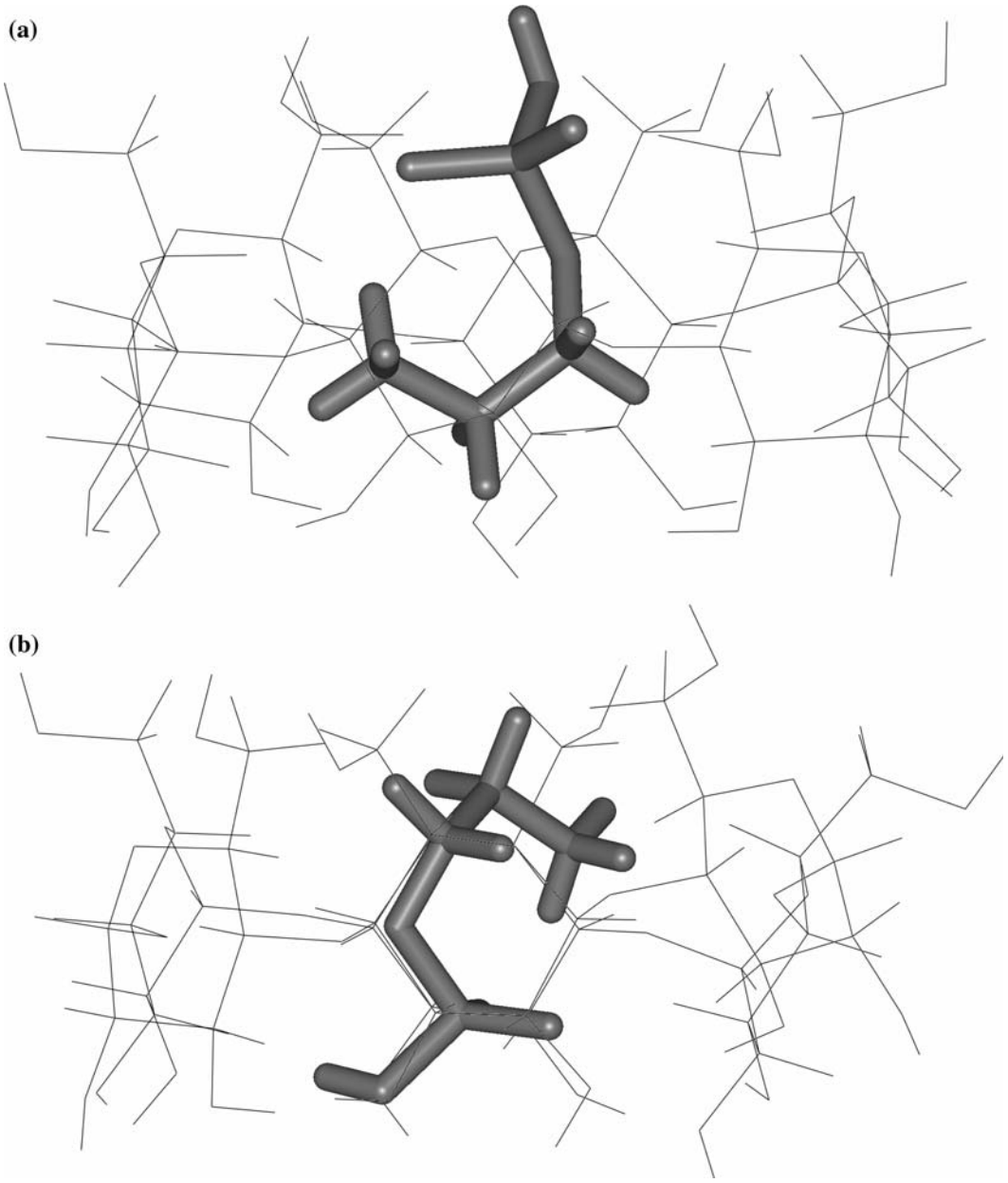


Figure 7. The most stable complex structure between α-CD and PE headgroup with orientation I (a) and orientation II (b).

The interactions of β- and γ-CD with the phospholipid headgroups

The interactions of the three phospholipid headgroups with β- and γ-CD were also investigated with the same method. The calculated results show that no prominent energy barrier appears, and only one binding region exists in the process of the phospholipid headgroup passing through β- or γ-CD cavity. It is mainly due to the cavity volume of β-CD and γ-CD is larger than that of α-CD, and allows phospholipid headgroup molecule to penetrate the CD cavity without barrier. Therefore, the structure with the lowest energy in the whole inclusion process should be the most stable complex structure. The energy values of the most stable complexes structures for the three phospholipids headgroups with β- and γ-CD are also listed in Table 1, from which, it is clear that the affinity of the three phospholipids headgroups for CD displays the rank of PI headgroup > PS headgroup > PE headgroup both for β- and γ-CD. It is of the same rank with α-CD, which is consistent with the experimental result [26].

For the complexes of PI headgroup with different CDs, if only the energy values were concerned, the complexes stability rank of the three CDs is β- > γ- > α-CD. The complexes energy values of the three CDs with PS headgroup are very close. While for PE headgroup with smaller size, the complexes stability rank of the three CDs is α- > β- > γ-CD. Furthermore, for the complex of γ-CD with phospholipid headgroup, the guest molecule prefers to position tilted in the cavity, which can increases the van der Waals interaction. However, in the real membrane system, the structure is probably hard to be formed due to the constraints of the membrane system.

Conclusion

The interactions of three different phospholipid headgroups with α-, β-, and γ-CD were studied using a molecular docking method FDOCK. The results show that, the van der Waals force is the main driving force responsible for complexation. For PI headgroup with α-CD, more than one inclusion complex should be coexistent due to the steric hindrance. By comparing the results of two orientations, it was found that the full inclusion occurs in orientation I rather than orientation II. While for PS and PE headgroup with α-CD, the complex structure with full inclusion is of the lowest energy and should be the most stable complex structure. Comparing the affinity of three phospholipid headgroups for CD, the rank of PI headgroup > PS headgroup > PE headgroup can be obtained. This study may be helpful to better understand the interactions of CDs with phospholipids and synthesize non-hemolytic derivatives.

Acknowledgements

This study is supported by the National Natural Science Foundation of China (NNSFC, No. 20172048), the outstanding youth fund of NNFSC (No. 20325517), and the Teaching and Research Award Program for Outstanding Young Teachers (TRAPOYT) in Higher Education Institute, MOE, P.R.C.

References

1. J. Szejtli: *Chem. Rev.* **98**, 1743 (1998).
2. K.B. Lipkowitz: *Chem. Rev.* **98**, 1829 (1998).
3. K. Uekama, F. Hirayama, and T. Irie: *Chem. Rev.* **98**, 2045 (1998).
4. T.W. Mu, L. Liu, X.S. Li, and Q.X. Guo: *J. Phys. Org. Chem.* **14**, 559 (2001).
5. P. Franchi, M. Lucarini, E. Mezzina, and G.F. Pedulli: *J. Am. Chem. Soc.* **126**, 4343 (2004).
6. I. Bea, C. Jaime, and P. Kollman: *Theor. Chem. Acc.* **108**, 286 (2002).
7. J. Varady, X.W. Wu, and S.M. Wang: *J. Phys. Chem. B* **106**, 4863 (2002).
8. Y. Choi and S. Jung: *Carbohydr. Res.* **339**, 1961 (2004).
9. J.M. Madrid, J. Pozuelo, F. Mendicuti, and W.L. Mattice: *J. Colloid Interf. Sci.* **193**, 112 (1997).
10. M. Cervero and F. Mendicuti: *J. Phys. Chem. B* **104**, 1572 (2000).
11. I. Pastor, A. Di Marino, and F. Mendicuti: *J. Phys. Chem. B* **106**, 1995 (2002).
12. J.M. Madrid, M. Villafruela, R. Serrano, and F. Mendicuti: *J. Phys. Chem. B* **103**, 4847 (1999).
13. J.M. Madrid, F. Mendicuti, and W.L. Mattice: *J. Phys. Chem. B* **102**, 2037 (1998).
14. H. Dodziuk, C.M. Demchuk, A. Bielejewska, W. Kozminski, and G. Dolgonos: *Supramol. Chem.* **16**, 287 (2004).
15. H. Dodziuk, O. Lukin, and K.S. Nowinski: *J. Mol. Struct. (Theochem)* **503**, 221 (2000).
16. H. Kim, K. Jeong, S. Lee, and S. Jung: *Bull. Korean Chem. Soc.* **24**, 95 (2003).
17. W.S. Cai, B.Y. Xia, X.G. Shao, Q.X. Guo, B. Maigret, and Z.X. Pan: *Chem. Phys. Lett.* **342**, 387 (2001).
18. J. Szejtli, T. Cserhati, and M. Szogyi: *Carbohydr. Polym.* **6**, 35 (1986).
19. S.L. Niu and B.J. Litman: *Biophys. J.* **83**, 3408 (2002).
20. K. Tanhuanpaa and P. Somerharju: *J. Boil. Chem.* **274**, 35359 (1999).
21. K. Tanhuanpaa, K.H. Cheng, K. Anttonen, J.A. Virtanen, and P. Somerharju: *Biophys. J.* **81**, 151 (2001).
22. T.G. Anderson, A. Tan, P. Ganz, and J. Seelig: *Biochemistry* **43**, 2251 (2004).
23. Y. Ohtani, T. Irie, K. Uekama, K. Fukunaga, and J. Pitha: *Eur. J. Biochem.* **186**, 17 (1989).
24. F. Fauvelle, J.C. Debouzy, R. Nardin, and A. Gadelle: *Bioelectrochem. Bioenerg.* **33**, 95 (1994).
25. F. Fauvelle, J.C. Debouzy, S. Crouzy, M. Goschl, and Y. Chapron: *J. Pharm. Sci.* **86**, 935 (1997).
26. J.C. Debouzy, F. Fauvelle, S. Crouzy, L. Girault, Y. Chapron, M. Goschl, and A. Gadelle: *J. Pharm. Sci.* **87**, 59 (1998).
27. S. Crouzy, F. Fauvelle, J.C. Debouzy, M. Goschl, and Y. Chapron: *Carbohydr. Res.* **287**, 21 (1996).
28. W.S. Cai, X.X. Yao, X.G. Shao, and Z.X. Pan: *J. Incl. Phenom. Macro. Chem.* **51**, 41 (2005).
29. W.S. Cai, Y.Y. Yu, and X.G. Shao: *J. Mol. Mod.* **11**, 186 (2005).
30. P.C. Manor and W. Saenger: *J. Am. Chem. Soc.* **96**, 3630 (1974).
31. C. Betzel, W. Saenger, B.E. Hingerty, and G.M. Brown: *J. Am. Chem. Soc.* **106**, 7545 (1984).
32. K. Harata: *Bull. Chem. Soc. Jpn.* **60**, 2763 (1987).
33. M. Elder, P. Hitchcock, R. Mason, and G.G. Shipley: *Proc. R. Soc. Lond. A* **354**, 157 (1977).
34. I.N. Rabinowitz and J. Kraut: *Acta Cryst.* **17**, 159 (1964).
35. T.R. Lomer, A. Miller, and C.A. Beevers: *Acta Cryst.* **16**, 264 (1963).
36. Accelrys Inc.: *Insight II 2005*, Accelrys Software Inc, San Diego (2005).
37. W.S. Cai and X.G. Shao: *J. Comput. Chem.* **23**, 427 (2002).
38. B.Y. Xia, W.S. Cai, X.G. Shao, Q.X. Guo, B. Maigret, and Z.X. Pan: *J. Mol. Struct. (TheoChem)* **546**, 33 (2001).
39. J.R. Maple, U. Dinur, and A.T. Hagler: *Proc. Natl. Acad. Sci. USA* **85**, 5350 (1988).
40. F. Fraternali and W.F. van Gunsteren: *J. Mol. Biol.* **256**, 939 (1996).
41. W. Hasel, T.F. Hendrickson, and W.C. Still: *Tetrahedron Comp. Method.* **1**, 103 (1988).
42. M. Goschl, S. Crouzy, and Y. Chapron: *Eur. Biophys. J.* **24**, 300 (1996).