Journal of Chromatography B, 879 (2011) 1605–1609

Contents lists available at ScienceDirect

Journal of Chromatography B

journal homepage: www.elsevier.com/locate/chromb

An efficient molecular docking method for adsorbent screening

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ARTICLE INFO

Article history:
Received 13 December 2010
Accepted 28 March 2011
Available online 6 April 2011

Keywords:
Adsorbent screening
Adsorption separation
Ginkgo biloba extracts
Molecular docking

ABSTRACT

In this study, with flavonol glycosides (FG) and terpene lactones (TL) in ginkgo biloba extract (GBE) as the targets for separation, we investigated the effectiveness of molecular docking in adsorbent screening. Several polyamine-modified methyl acylate-co-divinylbenzene (MA-co-DVB) adsorbent models were built, and their affinity to rutin, quercetin and ginkgolide B (GB) was evaluated via molecular docking. The model of ethylenediamine-modified adsorbent showed the largest difference in affinity between to GB and to quercetin as well as rutin, and thus this adsorbent could have the best separation performance. The results of the subsequently conducted static adsorption and dynamic adsorption experiments correlated well with docking results. Finally, using ethylenediamine-modified MA-co-DVB adsorbent, nearly complete separation of the FG and TL in GBE was simply achieved by one step of adsorption–desorption. Thus, the reported molecular docking method is expected to be helpful for rapid adsorbent screening.

1. Introduction

In the past several decades, solid phase extraction (SPE) method has been widely applied in enrichment/separation of active components of the natural products, pretreatment of biological samples and waste water treatment, etc. [1–6]. To achieve a satisfactory separation, it is important to chose appropriate stationary phase from numerous candidates, and thus a tedious and time-consuming procedure of stationary phase screening is usually needed.

Molecular docking method has been extensively used to analyse and elucidate the intermolecular interactions in host–guest chemistry [7,8]. In principle, adsorption phenomenon is also in the category of host–guest chemistry, therefore, molecular docking also could be employed in analysing the interaction between adsorbent and adsorbate. Additionally, it has been reported that molecular docking is robust in evaluating the affinity between polymers and guest molecules [9]. Hence, to some extent, molecular docking could be utilized for adsorbent screening and alleviating the consumption in screening process.

Ginkgo biloba is one of the oldest medicinal plants. Extracts of its leaves have been extensively exploited for medical application, such as antioxidant and enzyme activity modulator [10–14]. At present, the most widely used product of Ginkgo biloba extracts (GBE) has been standardized with 6.0% terpene lactones (TL) and 24.0% flavonol glycosides (FG), however, pharmacological study has proved the difference in their functions [15].

In this study, with FG and TL in GBE as the targets for separation, we investigated the effectiveness of molecular docking in adsorbent screening. Molecular models of several polyamine-modified MA-co-DVB adsorbents were built, and their affinity to rutin, quercetin and GB was evaluated via molecular docking. The model of ethylenediamine-modified adsorbent showed the largest difference in affinity between to GB and to quercetin as well as rutin, and thus the corresponding adsorbent could have the best separation performance. The results of the subsequently conducted static adsorption and dynamic adsorption experiments correlated well with the docking result.

2. Experimental

2.1. Composition of the adsorbents

The adsorbents were polyamine-modified MA-co-DVB macroporous resins. The polyamines were diethylenetriamine, ethylenediamine, butanediamine, hexanediamine, respectively.

2.2. Computational methodologies

The linear polymer models (LPMs) of the adsorbents were constructed for molecular docking study. Represented molecular structures of the LPMs were listed in Fig. 1. During the computational process, the functional groups on the LPMs were varied

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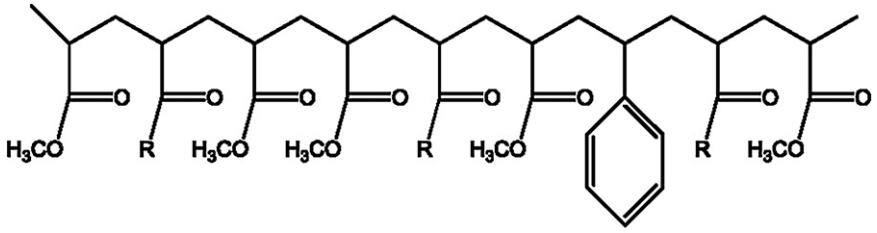
doi:10.1016/j.jchromb.2011.03.053



LPM code	R
LPM0	$-OCH_3$
LPM1	$-NHCH_2CH_2NHCH_2CH_2NH_2$
LPM2	$-NHCH_2CH_2NH_2$
LPM3	$-NH(CH_2)_4NH_2$
LPM4	$-NH(CH_2)_6NH_2$

Fig. 1. Linear polymer models of the polyamine-modified MA-co-DVB adsorbents.

in sequence, but the composition was kept constant. Rutin and quercetin were employed as the models of FG, while GB was employed as the model of TL (Fig. 2).

All the molecules (rutin, quercetin, ginkgolide B and the LPMs) were generated on Silicon Graphic Indio workstation using Sybyl 6.91 software package. The molecules with Gasteiger-Hückel charges added were energy minimized by Powell's method using Tripos force field with a distance-dependent dielectric constant until a terminating gradient of 0.005 kcal mol^{-1} $Å^{-1}$ was reached. Then each molecule was subjected to simulated annealing for 100 cycles. During the simulated annealing, each molecule was heated to 1000 K for 1000 fs and then cooled to 100 K for 1000 fs. The conformation with the lowest energy for each molecule was selected and minimized again subsequently [16].

The docking studies were performed using Autodock 4.0 program with three different initial binding modes for each model. The binding modes were differed from hydrophobic to hydrogen binding interactions. Typically, the three different initial modes of quercetin interacted with LPM2 were shown in Fig. 3. All the computations were carried out with grid box spacing 0.375 Å centered at the interaction region and a maximum of 500 Lamarckian genetic algorithm runs were performed. The resulted binding structures were clustered with a root mean square deviation of 2.0 Å. The lowest binding energy of each docking mode was analysed.

2.3. Materials

GBE containing 24.2% FG and 6.4% TL was obtained from Tianjin YangCheng High-Tech Natural Product Co., Ltd. Rutin, quercetin and ginkgolide B were purchased from J&K Chemical Ltd. (Beijing, China). Divinylbenzene (55% purity) was obtained from the Chemical Plant of Nankai University (Tianjin, China). All other reagents were of analytical purity and used as received.

2.4. Preparation of adsorbents

The adsorbents were prepared and characterized according to our published method [17], and the schematic synthesis of the adsorbents was shown in Fig. 4. Typically, for ethylenediamine-modified MA-co-DVB adsorbent, 4 g dry MA-co-DVB beads were added to a solution containing 20 mL DMF and 10 mL ethylenediamine and kept stirring overnight. The mixture was then kept refluxing for 9 h. The obtained beads were extracted with ethanol for 24 h and subsequently with water for 24 h and dried in vacuum at 70 °C for 24 h.

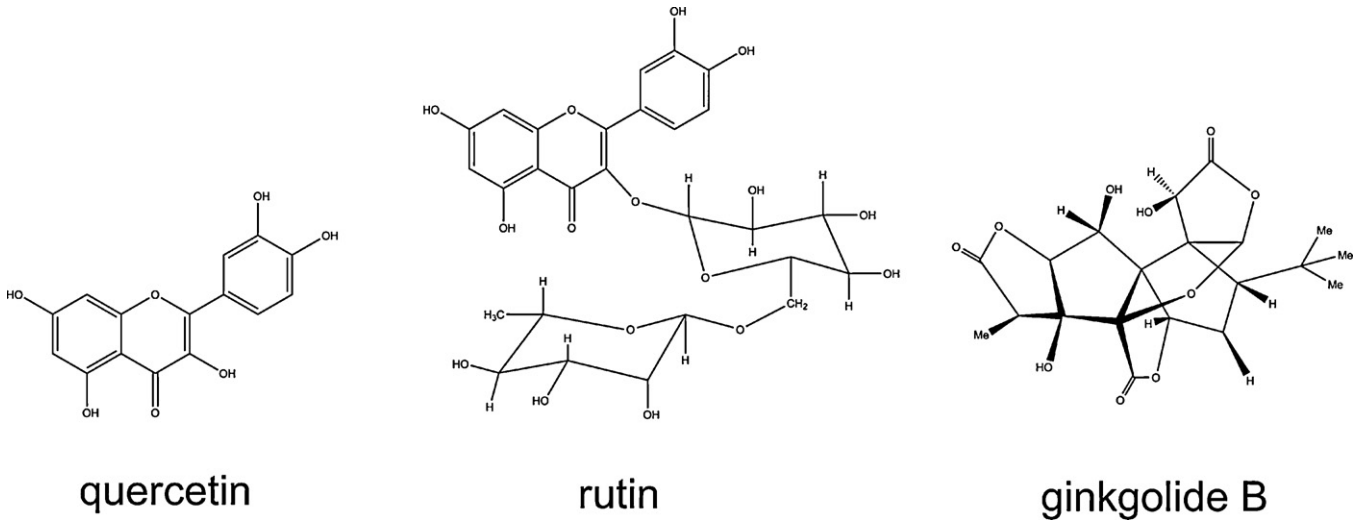


Fig. 2. Molecular structures of rutin, quercetin, ginkgolide B.

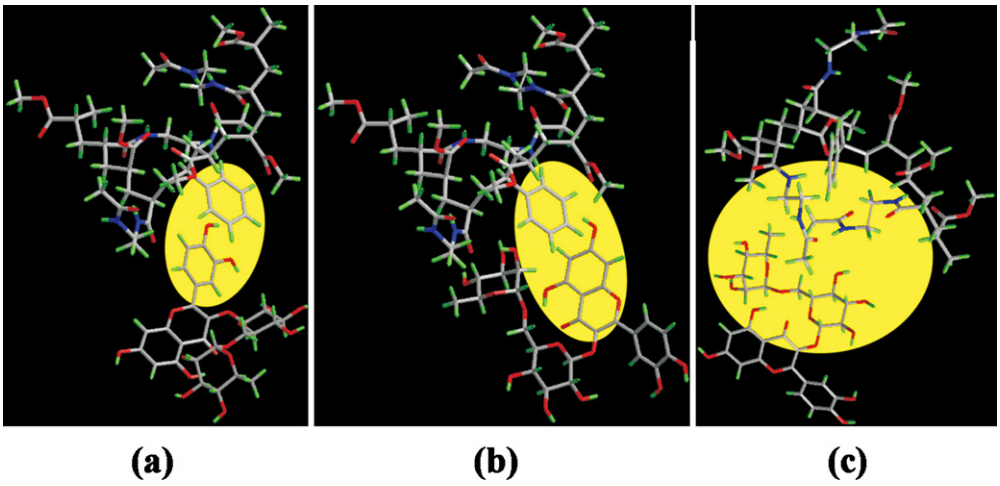


Fig. 3. Selected initial binding modes of rutin to LPM2 (a) and (b): hydrophobic interaction between the aromatic ring of LPM2 and quercetin. (c): Hydrogen binding between the phenol–OH of quercetin and the amide group of LPM2.

2.5. Static adsorption experiments

For each sample, 1 g adsorbent was added to triangle flask containing 50 mL adsorption solution. The flasks were subsequently shaken on an oscillator at 293 K for 10 h. The adsorption solutions were rutin (0.0725 mg/mL), quecertin (0.0072 mg/mL) and ginkgolide B (0.0134 mg/mL) in 10% ethanol aqueous solution, respectively. The equilibrium adsorption capacity Q_e (mg/g) was calculated according to the following equation:

$$Q = \frac{(C_0 - C_1)V}{W}$$

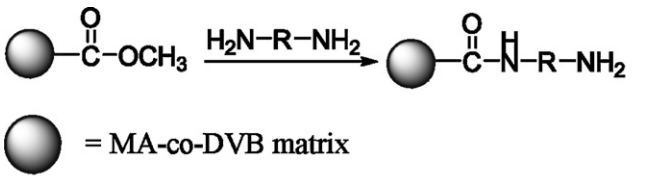
where W is the weight of the dry adsorbent, C_0 is the initial concentration of the adsorbate solution, C_1 is the equilibrium concentration of adsorbate solution, V is the volume of the solution.

2.6. Dynamic adsorption of GBE

2 g GBE was dissolved in 200 mL water at 70 °C. After cooled to room temperature, the adsorption solution was obtained by filtration.

15 mL wet adsorbents were packed in glass column (Φ = 15 mm), respectively. Subsequently, the columns were subjected to elution using the adsorption solution at a flow rate of 40 mL/h. When the elution finished, the columns were washed with de-ionized water and then desorbed with 80% (v%) ethanol aqueous solution.

The effluent and desorption solutions were vacuum dried, then the content of FG and TL in the effluent and desorption solution was determined by HPLC, respectively.



Adsorbent code	R
A0	-
A1	$-CH_2CH_2NHCH_2CH_2-$
A2	$-CH_2\ CH_2-$
A3	$-(CH_2)_4-$
A4	$-(CH_2)_6-$

Fig. 4. Synthesis of the polyamine-modified MA-co-DVB adsorbents.

2.7. HPLC analysis of FG and TL

All analyses were carried out using Waters 510 High Performance Liquid Chromatography pump on a reversed column packed with Nova-park C18 (5 μ, 4.6 mm × 200 mm).

Detection of FG was carried out using a UV-Detector. The detection wavelength was 368 nm. The mobile phase contained methanol, water and phosphoric acid (55:45:0.3, v:v:v). The flow rate was 0.8 mL/min.

Detection of TL was carried out using an Evaporative Light-scattering Detector. The mobile phase contained methanol, water and tetrahydrofuran (60:30:10, v:v:v). The flow rate was 1.0 mL/min.

3. Results and discussions

Previously, we have found that the diethylenetriamine-modified MA-co-DVB porous resin (8% DVB) worked efficiently in the separation of FG and TL in GBE [17]. Whether there were other type of polyamine-modified adsorbents that have comparable or better performance caught our interest. Hence, in the present work we designed several polyamine-modified MA-co-DVB adsorbents, and evaluated their performance using molecular docking method. The polyamines were diethylenetriamine, ethylenediamine, butanediamine, hexanediamine, respectively. Quercetin, rutin and GB are elementary components of flavonoids, flovonol glycosides and terpene lactones, respectively. Thus, they were selected as models of the adsorbates for the sake of simplicity and convenience of docking study, while linear polymers with the structural characteristics of the adsorbents were employed as the models of the adsorbents.

After the conformation and energy of all the model molecules were optimized, each adsorbate molecule was fitted to the LPMs using three different modes as shown in Fig. 3. Subsequently, the fitted molecular pairs were subjected to 500 cycle blind docking on Autodock. The average of the three separately obtained lowest energies was used to evaluate the affinity between the adsorbate molecules and the adsorbents. The docking results are listed in Table 1.

Table 1
Average of the binding energy between LPMs and the adsorbate molecules.

LPM code	Average binding energy E (kcal/mol)		
	Rutin	Quercetin	Ginkgolide B
LPM0	−25.199	−20.019	−18.434
LPM1	−28.796	−20.594	−15.269
LPM2	−34.588	−23.993	−11.386
LPM3	−37.032	−26.368	−15.617
LPM4	−42.568	−24.828	−10.875

Table 2
Results of the static adsorption experiment.

Adsorbent code	Adsorption capability (mg/g)		
	Rutin	Quercetin	GB
A0	0.47	0.047	0.381
A1	1.50	0.096	0.107
A2	2.57	0.127	0.032
A3	2.48	0.146	0.127
A4	2.01	0.136	0.119

From Table 1, it can be seen that LPM1-4 show decreased binding energy for rutin and quercetin in comparison with the LPM0, indicating the higher affinity between polyamine-modified adsorbents and rutin as well as quercetin, this is probably because that rutin and quercetin possess several active phenol–OH groups, which can effectively form hydrogen bonds with the amide groups on the adsorbents. In contrast, the binding energy between LPM1-4 and GB increased after modification. This is probably because the adsorption of the GB mainly depends on the hydrophobic effect [17], but the modification make the adsorbents more hydrophilic, and thus reduced the affinity between the adsorbents and adsorbates.

To examine whether the binding energy in molecular docking could reflect the affinity between the adsorbents and the adsorbates, we prepared these polyamine-modified adsorbents, and the adsorption of rutin, quercetin and GB using these adsorbents was investigated via static adsorption experiment, and the results are summarized in Table 2. It can be seen that the polyamine-modified adsorbents (A1–A4, corresponding to LPM1-4) indeed showed increased adsorption capacity for rutin and quercetin in comparison with the unmodified adsorbent (A0, corresponding to LPM0), while they showed remarkable decrease in adsorption capacity for GB. It should be noted that A2 showed more significant decrease in adsorption capacity for GB in comparison with the others.

Considering that the separation of FG and TL in GBE mainly depends on the difference in affinity between the adsorbents to FG and to TL rather than the absolute adsorption capability of the adsorbents, we investigated the difference of the binding energy (ΔE) between the adsorbent models to GB and to rutin as well as quercetin (Table 3). It can be seen that LPM1–LPM4 showed increased ΔE in comparison with LPM0, and LPM2 showed the largest ΔE. Therefore, the ethylenediamine-modified adsorbent is expected to exhibit the best performance on the separation of FG and TL in GBE.

Table 3
Difference between the binding energy of LPMs to GB and to rutin as well as quercetin.

LPM code	ΔE_1 (kcal/mol)[a]	ΔE_2 (kcal/mol)[b]
LPM0	6.765	1.585
LPM1	13.527	5.325
LPM2	23.202	12.607
LPM3	22.415	10.751
LPM4	21.693	3.953

[a] ΔE_1 is the difference between the binding energy of the LPMs to GB and to rutin.
[b] ΔE_2 is the difference between the binding energy of the LPMs to GB and to quercetin.

Table 4
Results of the dynamic adsorption experiments[a].

Adsorbent code	Initial sample	Desorption solution	Effluent solution
	FG:TL (m:m)	FG:TL (m:m)	FG:TL (m:m)
A0	4.09:1	4.87:1	4.26:1
A1		47.2:1	0.688:1
A2		169.6:1	0.0049:1
A3		16.5:1	0.0074:1
A4		15.1:1	0.524:1

[a] Flow rate was 40 mL/h, total volume of the adsorption solution was 40 mL for each sample.

To further check the validity of the computer-assisted adsorbent screening, the corresponding adsorbents were employed for the dynamic adsorption of GBE solution. Table 4 lists the results of the dynamic adsorption experiment. It can be seen that the unmodified MA-co-DVB adsorbent (A0) did not show obvious selectivity for FG or TL, as indicated by the small variation of the value FG/TL in either desorption solution or effluent solution in comparison with the initial sample. In the case of the polyamine-modified adsorbents (A1–A4), the value FG/TL in desorption solution increased significantly in comparison with initial adsorption solution, while decreased significantly in effluent solution. This indicate that the polyamine-modified adsorbents could effectively adsorb FG molecules, and FG were mainly accumulated in desorption solution, while TL molecules could not form effective association with the adsorbents, and hence mainly accumulated in effluent solution. It should be noted that FG can hardly be detected in the effluent solution of A2 and A3, therefore, completely separation of FG and TL could be achieved using these adsorbents. Considering the content of FG in the desorption solution of A3 is lower than that of A2, hence, the ethylenediamine-modified MA-co-DVB adsorbent gave the best performance for the separation of FG and TL in GBE, which correlated well with the molecular docking results.

4. Conclusions

Molecular docking method was demonstrated to be effective in adsorbent screening. The binding energy obtained from molecular docking could reflect the affinity between the adsorbents and the adsorbates as well as the adsorption capacity of the adsorbents. In the separation of FG and TL in GBE, the ethylenediamine-modified MA-co-DVB adsorbent, which showed the largest difference in the affinity to FG and TL in molecular docking, indeed exhibited the best performance in the dynamic adsorption experiment. And by using this adsorbent, nearly complete separation of FG and TL in GBE was simply achieved by one step of adsorption–desorption. Therefore, molecular docking method is expected to alleviate the consumption in the tedious and time consuming process of adsorbent screening.

Acknowledgement

This project is supported by National Natural Science Foundation of China (Grant No. 20634030).

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