**Designing and Preparation of Novel Alkaloid-imprinted Membrane with
Grafting Type and its Molecular Recognition
Characteristic and Permselectivity**

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Abstract A novel polysulfone-based molecularly imprinted membrane (MIM) with graft type (designated as GMIM) was successfully prepared by a combination of film-forming method of immersion-precipitation phase transformation with molecule surface-imprinting technique. The porous asymmetry membrane of chloromethylated polysulfone (CMPSF) was first prepared by a phase inversion method, and then the CMPSF membrane was amination-modified with ethanediamine as reagent, resulting aminated polysulfone membrane AMPSF, on whose surface primary amino groups were contained. Then the graft-polymerization of methacrylic acid (MAA) was realized by initiating of the surface-initiating system of $\text{-NH}_2/\text{S}_2\text{O}_8^{2-}$, obtaining the grafted membrane PSF-g-PMAA. After PSF-g-PMAA membrane adsorbed matrine, the crosslinking reaction of the grafted PMAA was allowed to be carried out with ethylene glycol diglycidyl ether (EGDE) as crosslinker, resulting in the matrine imprinted membrane with graft type, GMIM. The binding characteristics of the imprinted membrane GMIM, the permeability and separation property for matrine were investigated in depth. The experimental results show that the imprinted membrane consists of a thin imprinted layer, a thin skin layer containing channels at nanoscale and a support layer with macroporous structure. The imprinted membrane GMIM has specific recognition selectivity and excellent binding affinity for matrine, and its selectivity coefficient for matrine relative to cytosine is 4.85. More importantly, the imprinted membrane can produce good “gate effect” because of its own structure characteristic, so that it has fine permselectivity for the template, matrine molecule. The separation coefficient of the imprinted membrane GMIM for matrine relative to cytosine as a contrast reaches up to 5.9, displaying the excellent performance of a selectively permeable membrane.

Keywords Film-forming by phase inversion; Molecularly imprinted membrane; Surface imprinting; Alkaloids; Permeation and separation

1. Introduction

Compared with other separation processes, membrane separation technique has also attracted more attentions in many fields of science and technology because it has many advantages, such as energy-saving, flexible to operate continuously, easy to scale up and integrate into other separation processes [1,2]. Two key factors determine the success of a specific membrane separation process, and they are the flux of and the selectivity of the membrane for the target molecule [3,4]. Unfortunately, for the usual commercial membranes, their separation function is only based on a sieving mechanism, and do not allow the selective separation for individual substances [5,6]. Therefore, in the last few years, methods of developing membranes with controlled specificity for individual compounds have attracted much attention, and preparing molecularly imprinted membranes (MIMs) is just a possible solution for this challenge. It is well known that molecularly imprinted polymers (MIPs) are the synthetic antibodies or receptors [7-9], and they have highly specific recognition and binding selectivity for the target (template) molecules. Therefore, it is one of the good strategies to combine molecularly imprinting and membrane technology for developing selectively permeable membranes for the separation of particular target molecules from a mixture of structurally similar compounds. MIM separation technology is of potential prospects in molecular special recognition, the separation of biomacromolecules, the resolution of chiral compounds, the enrichment and separation of pharmacologic active substances, the constitution of sensors, the recovery and reuse of substances, the removal of toxic substances from water body, drug delivery systems, and so on [18-17]. It is obvious that MIM separation technology is of high application value in chemical engineering and biochemical engineering. It can be thought that MIM separation is a novel membrane separation technique or is a state-of-the-art membrane separation technique. As is well known, alkaloids have important physiological and pharmacological activity [18,19], and matrine is one of alkaloids. It is significant to

separate matrine or other alkaloids from the crude extraction solution of plants using MIM separation technology because such separation technology is flexible to operate continuously and easy to scale up compared to other separation processes. The purpose of this study is to develop novel MIMs with high performance with matrine as a model compound.

According to the preparation methods, the present MIMs used in membrane separation can be generally divided into three categories, molecularly imprinted filling membranes (MIFM) [20], molecularly imprinted monolithic membranes (MIMM) [21] and molecularly imprinted composite membranes (MICM) [22]. However, they have their own shortcomings. For example, for MIMM, the obvious shortcomings are poor toughness and mechanical strength as well as extremely low fluxes (not larger than 10^{-4} mol/(m²×h)) as consequences of high degrees of MIP cross-linking [23]; for MICM, although high flux can be obtained, the imprinted films combine with the support membranes (in general, they are micro-filtration membrane [24,25]) via physical interaction, leading to the poorer stability of membranes. Apparently, it is very necessary to design and develop novel MIMs with higher performance.

In this work, a novel molecularly imprinted membrane with grafting type (GMIM) was designed and prepared, and it combines the advantages of the above several MIMs. Porous chloromethylated polysulfone (CMPSF) asymmetry membrane as substrate membrane was first prepared with phase inversion method; amino groups (–NH₂) were introduced onto the surface of the CMPSF membrane via chemical modification; by the initiating of the surface initiating system of –NH₂/S₂O₈²⁻, methacrylic acid (MAA) was graft-polymerized onto the surface of the asymmetry membrane; finally, matrine surface-imprinting was realized with ethylene glycol diglycidyl ether (EGDE) as crosslinker. In fact, the process of preparing matrine surface-imprinted membrane as briefly described above is an imprinting method of “pre-graft polymerizing and post-imprinting”. The obtained molecularly imprinted membranes is novel, and such

imprinted membrane with grafting type has the following characteristics: (1) the thin imprinted layer is grafted on to the substrate membrane (so called as the membrane grafting type), and the imprinted film and the substrate membrane combine together by chemical bond force, leading good stability and mechanical strength of the membrane; (2) there is a great deal of pore channels within the asymmetry membrane, resulting in high flux and fine transport property; (3) the thin imprinted layer afford the membrane excellent permselectivity for the template molecule.

In this study, a novel molecularly imprinted membrane with grafting type, GMIM, was successfully designed and prepared by combination of phase inversion film-forming method with molecule surface-imprinting technology, and it possesses excellent comprehensive performance. The design idea and the preparation route are late-model, and the similar study has not been reported. The research result in this work has obvious science significance and application value in developing selectively permeable membrane technique.

2. Experimental Section

2.1. Material and equipment

Polysulfone (PSF, $\bar{M}_r = 67000$) was of industrial grade and obtained from Shuguang Chemical Plant of Shanghai Plastic Industry Associated Company (China); Chloromethylated polysulfone (CMPSF) was self-synthesized with 1,4-bis (chloromethoxy) butane (BCMB) as chloromethylation reagent [26]; Dimethylacetamide (DMAc) and dimethylformamide (DMF) were purchased from Tianjin Bodi Chemical Co., Ltd. (China); Methacrylic acid (MAA, It was purified by vacuum distillation before use) and ammonium persulfate (APS) were obtained from Sinopharm Chemical Reagent Co., Ltd. (China); Polyethylene glycol 400 (PEG-400) and ethylene glycol diglycidyl ether (EGDE) were supplied by Tianjin Tianli Chemical Co., Ltd. (China); matrine and cytosine with a purity of 98% were obtained from Xian

Bio-Medicine Science and Technology Ltd. (China); The other chemicals used were all of commercial analytical grade, and were purchased from Chinese companies.

The instruments used in this study were as follows: Perkin-Elmer1700 infrared spectrometer (Perkin-Elmer Company, USA); LEO-438VP scanning electronic microscope (SEM, LEO Company, UK); Unic-2602 UV spectrophotometer (Unic Company, Shanghai); Zetasizer Nano-Zeta potential analyzer (Malvern Instrument Company, UK); P1201 high performance liquid chromatograph (HPLC , Dalian Elite Analytical Instruments Ltd., Dalian City, China); THZ-92C constant temperature shaker equipped with gas bath (Shanghai Boxun Medical Treatment Equipment Factory, Shanghai, China); Leica metalloscope (OM, Leica Company, German).

2.2. Preparation and characterization of grafted membrane PSF-g-PMAA

2.2.1. Preparation of substrate membrane

The chloromethylated polysulfone (CMPSF) was first prepared with BCMB as chloromethylation reagent according to the procedure described in Ref. [26], and then the porous asymmetry membrane of CMPSF was prepared by the method of immersion-precipitation phase transformation. The detailed process is described as follows. DMAc (6 mL) was placed into a conical flask followed by adding 1 g of CMPSF, and CMPSF was allowed to be completely dissolved. PEG 400 (0.2 g) as porogen was added, and the solution was stirred for 24 h. After standing 24 h for defoaming, the solution was casted onto a glass plate, and the glass plate was immediately placed in water containing 5% of DMAc at 20 °C so that the solution was coagulated and a membrane formed by the phase inversion technique. The resultant membrane was soaked into distilled water for 48 h, and in the meantime, the water was replaced many times to remove the porogen PEG 400. Finally the porous asymmetry membrane of CMPSF (abbreviated as CMPSF membrane) with thickness of 200 μm was resulted in.

2.2.2. Preparation and characterization of grafted membrane PSF-g-PMAA

By adopting the “surface-initiated graft-polymerization” method similar to that described in Ref. [27], the graft-polymerization of MMA on the surface of CMPSF membrane was realized, and the typical procedure is described as follows. (1) Preparation of AMPSF membrane. CMPSF membranes were soaked in ethanediamine, which was used as reagent and also used as solvent, and the nucleophilic substitution reaction between the chloromethyl groups on the surfaces of CMPSF membranes and the amino group of ethanediamine was made to be carried out at 25 °C for 10 min, obtaining aminated PSF membrane, abbreviated as AMPSF membrane, on which primary amino groups as well as secondary amino groups were contained. (2) Performing graft-polymerization of MMA. Several sheets of AMPSF membranes (0.1 g) were placed in a four-necked flask equipped with a mechanical agitator, a reflux condenser, a thermometer and a N₂ inlet, followed by adding 70mL of the mixture of DMF and water (V:V=1:1), and the AMPSF membranes were allowed to be swelled for 2 h. The monomer MMA (3.3mL) was added, N₂ was bubbled for 30 min to exclude air, and then the temperature of the system was raised to 50 °C. The aqueous solution (5 mL) containing 0.035 g of initiator APS was added, and a surface-initiating system of –NH₂/S₂O₈²⁻ was constituted, for which the group of –NH₂ came from AMPSF membrane, and the ion of S₂O₈²⁻ came from the solution. Thereupon the graft-polymerization of MMA on the membrane surfaces was carried out at 50 °C for 8 h, obtaining the grafted membranes PSF-g-PMAA with a PMAA grafting degree of 9.47mg/cm².

The following characterizations for the grafted membrane PSF-g-PMAA were done. (1) The Fourier Transform Infrared Spectroscopy (FTIR) was determined with KBr pellet method to confirm the chemical structure of the grafted membrane PSF-g-PMAA. (2) The cross-section structures were examined by SEM, and the surface morphologies were examined by OM. (3) The grafting degree of PMAA on the surface of the grafted membranes PSF-g-PMAA mentioned above was determined by weighting method, and it results

from subtracting the weight of the dried AMPSF membrane from the weight of the dried PSF-g-PMAA membrane.

2.3. Preparation and characterization of matrine-imprinted membrane with grafting type

2.3.1 Preparation

The grafted membrane PSF-g-PMAA with a surface area of 8 cm² was placed into an aqueous solution of matrine with a concentration of 1.4 g/L, and the pH value was adjusted to 8 with diluted NaOH solution. This mixed system was placed in a constant temperature oscillator for 8 h to let the adsorption of the grafted membrane towards matrine reached saturation. This membrane adsorbed matrine was placed in a mixture of water and ethanol (V:V=1:1), in which matrine with a concentration of 1.4 g/L was contained to prevent the disadsorption of matrine from the membrane, and the crosslinker EGDE (0.05 mL) was dissolved. The crosslinking reaction of PMAA on the grafted membrane PSF-g-PMAA was allowed to be conducted at 45 °C for 10 h with stirring. After finished the crosslinking reaction, the membrane was washed repeatedly with a diluted HCl solution containing NaCl to remove the template matrine molecules, resulting in the matrine molecule-imprinted membrane with grafting type (denoted as GMIM). Under the same condition, the crosslinking reaction of PMAA on the surface of the grafted membrane PSF-g-PMAA was also conducted in absence of matrine, obtaining the non-imprinted membrane (NMIM).

2.3.2. Characterization

For the imprinted membranes GMIM, the FTIR was determined with KBr pellet method to confirm their chemical structure, the cross-section structures were examined by SEM, and the surface morphologies were observed by OM.

2.4. Examining binding characteristics of imprinted membrane GMIM for matrine

2.4.1. Isothermal binding experiments

The aqueous solutions of matrine were prepared in a range of 0.2~1.4 g/L. The binding kinetics experiment of GMIM for matrine was first performed, and the time of binding equilibrium was determined to be 8 h. On this basis, the isothermal binding experiments were conducted. Matrine solutions of 25 mL with different concentrations were placed into a number of conical flasks with cover, and the pH values of these solutions were adjusted to 8. Quadrate sheets of GMIM membrane with a total area of 4 cm² were placed into these solutions, respectively, and these solutions were shaken for 8 h in a constant temperature oscillator with a rotation speed of 60 rpm. After standing, the supernatants were taken, the matrine concentrations in the supernatants were determined by ultraviolet spectrophotometry at 204 nm, and the equilibrium binding amounts Q_e (mg/cm²) of matrine were calculated according to Eq. (1).

$$Q_e = \frac{V(C_0 - C_e)}{A \times 2} \quad (1)$$

where C_0 (g/L) and C_e (g/L) are the initial and equilibrium concentration of matrine, respectively; V (mL) is the volume of matrine solution, and A (cm²) is the area of the imprinted membrane GMIM.

Cytisine is also an important alkaloid like as matrine. Furthermore, they coexist in the plants of sophora alopecuroides, and their structures have similarity in a certain degree. Therefore, in this work, cytisine was selected as the contrast substance to examine the binding and permeation selectivity of the imprinted membrane GMIM for matrine. The molecular structures of the two substances are schematically expressed in Scheme 1. With the same method like as that for matrine, the binding isotherms of the imprinted membrane GMIM for cytisine was determined. The concentrations of cytisine in the supernatants were also determined with spectrophotometry at 304 nm.

Scheme 1

2.4.2. Binding selectivity experiments

In order to further examine the binding selectivity of the imprinted membrane GMIM for matrine, the competitive adsorption experiments were carried out. A binary mixed aqueous solution, matrine/cytisine, was prepared and pH value was adjusted to 8, and in the solution, the concentrations of each component were the same, 0.6 g/L. Such mixed solutions of 20 mL were taken and placed in a conical flask with cover, followed by adding the imprinted membrane GMIM with a certain area. The adsorption system was shaken in a constant temperature oscillator for 8 h. After standing, the concentrations of the two alkaloids in the supernatant were determined by dual-band UV spectrophotometry. The distribution coefficient for each substance in the binary mixed solution was calculated according to Eq. (2), and this equation was originated from Ref. [28, 29].

$$K_d = \frac{Q_e}{C_e} \quad (2)$$

where K_d represents the distribution coefficient (mL/cm²) of a substance; Q_e (mg/cm²) is the equilibrium binding quantity of this substance; C_e (g/L) is the equilibrium concentration of this substance.

The selectivity coefficient k of the imprinted membrane GMIM for matrine relative to cytisine was obtained from the distribution coefficient data according to Eq. (3) [28, 29]. The value of k marks the recognition selectivity of GMIM for the template matrine, and it is one of the most important parameters of imprinted materials.

$$k = \frac{K_d(\text{matrine})}{K_d(\text{cytisine})} \quad (3)$$

2.5. Examining permselectivity of imprinted membrane GMIM for matrine

2.5.1 Permeation experiment for single component

The permeation equipment was self-designed, and it was constituted by two glass compartment

cells with H-shaped. The imprinted membrane GMIM was fixed at the position of ground joint. The matrine solution of 1.4 g/L (100 mL and pH=8) was placed in the left compartment and the pure solvent (100 mL) was placed in the right compartment. At room temperature, both compartments were stirred by a magnetic stirrer at a constant rate. The concentrations of matrine transported through the membrane at different times were determined using the UV spectrophotometry. The permeation amount of matrine, Q (mg/cm^2), was calculated by Eq. (4) [30], and the permeation curve of matrine (permeation amount of matrine as a function of time) was plotted. At the same time, the permeation curve of the contrast substance cytosine was also figured.

$$Q = \frac{C \times V}{A} \quad (4)$$

where the C (mg/mL) represents the concentration of the permeated matrine or cytosine, V (mL) is the volume of the solvent in the compartment, and the A (cm^2) is the valid surface area of the membrane.

2.5.2 Permeation experiment for mixed solution

A binary mixed solution (pH=8), matrine/cytosine, was prepared, and in this mixed solution, the concentrations of the two components were the same, 1.4 g/L. By using the self-designed permeation equipment, the competitive permeation experiment was conducted, and the permeation curves of the two components were plotted. The separation coefficient of the imprinted membrane GMIM for matrine was calculated according to Eq. (5) [31].

$$\alpha = \frac{C(\text{matrine})}{C(\text{cytosine})} \quad (5)$$

where $C(\text{matrine})$ and $C(\text{cytosine})$ represent the concentrations of matrine and cytosine in the permeate solution at certain time, respectively.

3. Results and discussion

3.1. Physical-chemical process to prepare imprinted membrane GMIM

(1) Based on preparing chloromethylated polysulfone CMPSF, the casting solution of CMPSF was prepared with DMAc as solvent, at the same time, the porogen PEG 400 was added. The porous asymmetry substrate membrane of CMPSF was prepared with immersion-precipitation phase transformation method, and the resulted membrane was constituted by a dense skin layer and a macroporous support layer. (2) Through the substitution reaction between the chloromethyl group of CMPSF membrane and the amino group of ethanediamine, ethanediamine was bonded onto the surface of asymmetry substrate membrane of CMPSF so that amino group $-NH_2$ was introduced on the membrane, resulting the modified membrane AMPSF. (3) A surface-initiating system of $-NH_2/S_2O_8^{2-}$ was constituted [27], and free radicals were produced on the surface of AMPSF membrane. As a result, the graft-polymerization of MAA was initiated, resulting in the grafted membrane PSF-g-PMAA. (4) At pH 8, by right of the electrostatic interaction between the dissociated carboxyl group of the grafted PMAA and the protonated N atom in matrine molecule as well as the hydrogen bonding, the adsorption of matrine on the grafted membrane PSF-g-PMAA reach saturation. (5) Finally the grafted macromolecules PMAA are crosslinked with EGDE as crosslinker, and the adsorbed matrine molecules are wrapped into the crosslinking networks, and so the imprinting of matrine is realized by using the “pre-graft polymerizing and post-imprinting” method. After washing out the template matrine molecules, a great deal of the matrine imprinted caves left in the thin polymer layer on the surface of the membrane, forming matrine-imprinted membrane with grafting type, GMIM. The entire physical-chemical process of preparing GMIM can be schematically illustrated in Scheme 2.

Scheme 2

3.2. Characterization of imprinted membrane GMIM

3.2.1. Infrared spectra

Fig.1(A) presents the infrared spectra of the substrate membrane of CMPSF and the modified membrane AMPSF, and Fig.1(B) presents the infrared spectra of the grafted membrane PSF-*g*-PMAA and the imprinted membrane GMIM.

Fig. 1 (A)

Fig. 1 (B)

In the spectrum of the substrate membrane CMPSF, except for all of the characteristic absorption bands of polysulfone, the characteristic absorption bands of chloromethyl group appear at 1440 and 667 cm^{-1} , and the former is attributed to the in-plane bending vibration absorption of C-H bond of the chloromethyl group, and the latter is assigned to the stretching vibration absorption of C-Cl bond of the chloromethyl group. In the spectrum of the ammoniated membrane AMPSF, the two bands described above have been weakened dramatically, and at the same time, the symmetric and antisymmetric stretching vibration absorptions of primary amine group appear at 3588 and 3367 cm^{-1} , respectively, indicating that the amination of the membrane has been realized.

In the spectrum of the grafted membrane PSF-*g*-PMAA, the three characteristic absorption bands of carboxyl group appear at 1715, 3430 and 964 cm^{-1} , respectively, demonstrating that the surface-graft-polymerization of MAA has been realized. The first is attributed to the stretching vibration absorption of carbonyl group of carboxyl group, the second is assigned to the stretching vibration absorption of the

hydroxyl group of carboxyl group, and the third is ascribed to the out-of-plane bending vibration absorption of the hydroxyl group of carboxyl group. In the spectrum of the imprinted membrane GMIM, there appears an absorption band at 1548cm^{-1} , and it is corresponding to the characteristic absorption of the methylene group $-\text{CH}_2-$, which comes from the crosslinking bridge EGDE, showing the crosslinking reaction between the grafted PMAA has been produced. The above spectrum data fully show that matrine-imprinted membrane with grafting type (GMIM) has been obtained with the porous asymmetry CMPSF membrane as substrate membrane and via several polymer reactions including amination, graft-polymerization and crosslinking reactions.

3.2.2. Morphology and pore structure

Fig. 2 gives the SEM images of the cross-sections of the substrate membrane CMPSF (A) and the imprinted membrane GMIM (B), and Fig. 3 gives the OM images of the surfaces of two membranes (A for CMPSF membrane and B for GMIM membrane).

Fig. 2

Fig. 3

It can be seen that the substrate membrane CMPSF had a typical asymmetric structure, which consisted of a thin and dense skin layer and a large porous support structure containing finger-like pores. In this work, the porogen PEG 400 was added in the film-forming system, and it plays two roles: (1) it induces the formation of micropores at the nanoscale in the skin layer, and inhibits the formations of macropores [32]; (2) it accelerates the linking of nano-channels [33], resulting in the formation of the interconnected

channels at the nanoscale.

It can be found by comparing Fig. 2 (B) and Fig. 2 (A), for the imprinted membrane GMIM, the dense skin layer has been obviously thickened, and it is caused by the graft-polymerization of MAA and the crosslinking of the grafted PMAA during the imprinting processes. It is the imprinted layer that leads to the thickening of the skin layer. Besides, it can be seen by comparing Fig. 3 (B) and Fig. 3 (A) that the surface of the substrate membrane is smoother, whereas the surface of the imprinted membrane GMIM becomes to be rougher, which is caused by the graft-polymerization and crosslinking/imprinting process.

3.3. Interaction between grafted membrane PSF-g-PMAA and matrine

It is a key issue for the molecule imprinting system that there exists strong interactions between host-guest, i.e., between functional monomer or functional polymer and template molecule. For this work, the “pre-graft polymerizing and post-imprinting” method was used to prepare matrine-imprinted membrane, and so the interaction between the grafted membrane PSF-g-PMAA and matrine was first studied. The isothermal adsorption experiment of the grafted membrane PSF-g-PMAA for matrine was conducted, and the adsorption isotherm at pH 8 is presented in Fig. 4.

Fig. 4

Fig. 4 displays that PSF membrane has no adsorption ability for matrine at all. However, at pH 8, the grafted membrane PSF-g-PMAA exhibits very strong adsorption ability for matrine, and the adsorption capacity reaches 1.34 mg/cm², displaying that there is a strong interaction between the grafted membrane PSF-g-PMAA and matrine molecule. This is the fundamental basis of imprinting matrine on the surface of the grafted membrane PSF-g-PMAA. The strong interaction is mainly the electrostatic interaction between

the disassociated carboxyl groups of the grafted PMAA and the protonated N atoms in, and there is also the synergistic effect of hydrogen bonding. The binding mode of the grafted PMAA with matrine molecule can be clearly seen in Scheme 2.

In order to determine the optimum pH value for surface-imprinting, the isothermal adsorption experiments of the grafted membrane PSF-g-PMAA for matrine at different pHs were conducted, and the saturated adsorption amount (Q_m) as function of pH value is inserted into Fig. 4 as a small figure. It can be observed that the adsorption capacity of the grafted membrane PSF-g-PMAA for matrine first increases and then decreases with the increase of pH value, and at pH 8, there is a maximum adsorption capacity. This change rule reflects the variation of the interactions between host-guest with pH value of the medium. At lower pH, although the protonation degree of the N atoms in matrine molecule is high, ionization degree of the carboxyl groups of the grafted PMAA is very small, and the electrostatic interaction between host-guest is weaker, leading to low adsorption capacity. Along with the increase of pH, the ionization degree of the carboxyl groups increases, and the electrostatic interaction between host-guest strengthens, leading to the increase of the adsorption capacity. However, the protonation degree of the N atoms in matrine molecule decreases along with the increase of pH, and it will negatively affect the electrostatic interaction between host-guest. This negative effect does not emerge as $\text{pH} < 8$, but it begins to show as $\text{pH} > 8$, leading to the turning of the adsorption capacity at pH 8. Here, there exists the maximum electrostatic interaction between host-guest as well as hydrogen binding as shown in Scheme 2, and so $\text{pH}=8$ is selected as the suitable pH value for the surface-imprinting process.

3.4. Binding and recognition property of imprinted membrane GMIM for matrine

3.4.1. Isothermal binding curve

The adsorption experiments with batch method were first performed using the non-imprinted

membrane NMIM and the imprinted membrane GMIM, respectively, and Fig. 5 and Fig. 6 give the adsorption isotherms of NMIM and the binding isotherms of GMIM for matrine and cytosine, respectively.

Fig. 5

Fig. 6

Fig. 5 displays that the non-imprinted membrane NMIM have high adsorption capacities for both matrine and cytosine (0.76 mg/cm^2 for matrine and 1.01 mg/cm^2 for cytosine, respectively) because there also exists strong electrostatic interaction as well as hydrogen binding between the non-imprinted membrane NMIM and cytosine molecule. Apparently, the non-imprinted membrane NMIM has no adsorption selectivity for the two alkaloids.

However, Fig. 6 clearly presents the following facts. The binding capacity of the imprinted membrane GMIM for cytosine has significantly reduced, and it decreases from 1.01 mg/cm^2 to 0.33 mg/cm^2 . It is obvious that the imprinted membrane GMIM hardly recognizes and binds the contrast substance cytosine. However, for the template molecule matrine, the imprinted membrane GMIM not only still maintains high binding capacity, but also the binding capacity is even further increased (from 0.76 mg/cm^2 to 1.09 mg/cm^2), displaying the specific recognition selectivity and excellent binding affinity of the imprinted membrane GMIM for matrine.

The reason for the above facts can be explained as follows. There is a great quantity of the imprinted caves of matrine molecule within the thin grafted polymer layer on the surfaces of the imprinted membrane GMIM. These caves are highly matched with matrine molecule in size, shape and spatial

arrangement of action sites, and it leads to the specific recognition ability and strong binding action of the imprinted membrane GMIM towards matrine molecule. However, these imprinted caves are unmatched with cytosine molecule in size, shape and spatial arrangement of action sites, so these imprinted caves are difficult to accept cytosine molecules. Therefore, the imprinted membrane GMIM does not recognize basically the contrast substance, cytosine molecules, leading to very low binding capacity.

3.4.2. Recognition selectivity

A binary mixed solution, matrine/cytisineand, was prepared, and the competitive adsorption experiment of the imprinted membrane GMIM was conducted. Table 1 summarizes the data of the distribution coefficients K_d and selectivity coefficients k .

Table 1

The data in Table 1 display that the selectivity coefficients of the non-imprinted membrane NMIM for matrine relative to cytosine (0.915) is closed to 1. It implies that the adsorption abilities of NMIM for the two components in the binary mixed solution are the basically same, namely NMIM has no any adsorption selectivity. However, the selectivity coefficients of the imprinted membrane GMIM for matrine relative to cytosine is 4.854, and it further confirm that the imprinted membrane GMIM competitively adsorbs matrine, namely it has a special recognition and binding selectivity for matrine. As compared with some molecularly imprinted composite membranes, which are the most extensively studied imprinted membranes, the imprinted membrane GMIM possesses better binding selectivity for the template. For example, for curcumin-imprinted composite membrane, the selectivity coefficient of the composite membrane for curcumin relative demethoxycurcumin, which is the analog of curcumin, is only 1.5 [34].

3.5. Permeation and separating property of imprinted membrane GMIM

3.5.1. Permeation curve of single component

The permeation experiment of the single component for matrine or cytosine was carried out by using the self-designed permeation equipment. Fig.7 and Fig. 8 give the permeation curves as the solutions of the two alkaloids permeate across the non-imprinted membrane NMIM and the imprinted membrane GMIM, respectively.

Fig. 7

Fig. 8

It can be observed that as the solutions of the two alkaloid solutions permeate across the non-imprinted membrane NMIM, their permeation rate are basically similar, and in 24 h, the permeation amounts are about 50 mg/cm², indicating that the non-imprinted membrane NMIM for the permeation of the two alkaloids has no selectivity. However, the permeation curves shown in Fig. 8 have changed dramatically. The permeation rate of matrine is much higher than that of the contrast substance cytosine. The permeation amount of matrine in 24 h reaches 49 mg/cm², whereas that for cytosine is only 18 mg/cm², fully displaying the permselectivity of the imprinted membrane GMIM for matrine. The reason for this is still that the imprinted caves within the imprinted layer are highly matched with matrine molecule in size, shape and spatial arrangement of action sites. Under the dual effects of concentration gradient driving and “gate effect”[31], matrine molecules are first adsorbed onto the surface of GMIM, and then these template

molecules smoothly and continually pass through the channels and diffuse to the blank compartment cell (the detailed permeation mechanism will be further described below). Conversely, the diffusion of the contrast substance molecules, cytosine molecules, is seriously impeded by the “gate effect” because of the mismatching of the imprinted caves, leading to low permeation amount of cytosine.

3.5.2. Permeation curve for mixed solution

To fully estimate the mechanism of selective permeation of the imprinted membrane GMIM, competitive permeation experiment was also conducted. A binary mixed solution (pH=8) of matrine and cytosine, in which the concentration of each component was the same, 1.4 g/L, was allowed to pass through the imprinted membrane GMIM. Fig. 9 presents the permeation curves.

Fig. 9

Fig. 9 clearly displays that in 24 h, in the receiving cell (original blank cell), the permeation amounts of matrine and cytosine are 47 mg/cm² and 8 mg/cm², respectively, and the separation coefficient α is equal to 5.9. Apparently, matrine molecule preferentially and competitively penetrates through the imprinted membrane GMIM. As compared with some molecularly imprinted composite membranes, the imprinted membrane GMIM possesses better selective permeability. For example, for artemisinin-imprinted composite membrane, the separation coefficient α relative to artemether, which is the analog of artemisinin, is only about 1.6 [35]. The selective permeation mechanism of the template molecule can schematically expressed in Scheme 3.

Scheme 3

By the driving of the concentration difference, both the template molecule and the contrast substance molecule have the trend to diffuse towards the blank cell. However, as they diffuse to the surface of the imprinted membrane, matrine molecules are preferentially absorbed because of the selective recognition and binding of the imprinted caves, and this is a key step [36]. Then matrine molecule moves from one cave to the next one within the imprinted layer, and then passes across the skin layer because the channels in the imprinted layer and the channels at nanoscale in the skin layer are interconnected as shown in Scheme 3. Finally, matrine molecules enter into the support structure with macropore, and pass into the receiving cell.

Unlike the template molecules, the contrast molecules cannot be adsorbed and cannot pass through the imprinted membrane GMIM smoothly because they have neither a space configuration matching with the imprinted caves of the GMIM nor groups matching with functional groups inside the caves of the GMIM, leading to very low permeation amount. In a word, the “gate effect” plays a key role in the holding back cytosine to pass across the imprinted membrane GMIM.

4. Conclusions

In this work, by elaborate molecular design, film-forming method of immersion-precipitation phase transformation was well combined with molecule surface-imprinting technique, and polysulfone-based molecularly imprinted membrane with graft type, GMIM, was successfully prepared. The imprinted membrane GMIM consists of a thin matrine imprinted layer, a thin skin layer containing channels at nanoscale and a support layer with macroporous structure. GMIM membrane combines the advantages of the imprinted monolithic membrane and the imprinted composite membrane. GMIM membrane not only is

stable, which comes from the chemically linking between the support membrane and the grafted film, but also it has high flux because of the channels within its interior. The great deal of the matrine imprinted caves in the imprinted layer lets the imprinted GMIM to have specific recognition selectivity for matrine molecule, and its selectivity coefficient for matrine relative to cytosine reaches 4.85. Under the dual effects of concentration gradient driving and “gate effect”, the imprinted GMIM exhibits excellent selective permeation performance, and the separation coefficient relative to cytosine reaches up to 5.9. In a word, the investigation result in this work supplies a new and effective way for preparing imprinted membrane with high performance, and it is significant for the development of selectively permeable membrane technique.

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References

- [1] Sueyoshi Y, Fukushima C, Yoshikawa M. *J Membr Sci* 357 (2010) 90–97.
- [2] Meng M-J, Feng Y-H, Zhang M, Ji Y-J, Dai J-D, Liu Y, Yu P, Yan Y-S. *Chem Eng J* 231(2013) 132–145.
- [3] Zhai Y-H, Liu Y-W, Chang X-J, Ruan X-F, Liu J-L. *React Funct Polym* 68 (2008) 284–291.
- [4] Sueyoshi Y, Fukushima C, Yoshikawa M. *J Membr Sci* 357(2010) 90–97.
- [5] Wang X-J, Xu Z-L, Feng J-L, Bing N-C, Yang Z-G. *J Membr Sci* 313(2008) 97–105.
- [6] Ulbricht M. *J Chromatogr B* 804 (2004) 113–125.
- [7] Kryscio D R, Peppas N A. *Acta Biomaterialia*, 8(2012) 461-473.
- [8] Prasad B B, Sharma P S, Lakshmi D. *Journal of Chromatography A*, 1173(2007) 18-26.
- [9] Ng S M, Narayanaswamy R. *Sensors and Actuators B: Chemical*, 139(2009)156-165.
- [10] Pegoraro C, Silvestri D, Ciardelli G, Cristallini C, Barbani N. *Biosens Bioelectron* 24 (2008) 748–755.
- [11] Chen R-R, Qin L, Jia M, He X-W, Li W-Y. *J Membr Sci* 363(2010) 212–220.
- [12] Itou Y, Nakano M, Yoshikawa M. *J Membr Sci* 325 (2008) 371–375.
- [13] Kochkodan V, Hilal N, Melnik V, Kochkodan O, Vasilenko O. *Adv Colloid Interface Sci* 159 (2010) 180–188.
- [14] González G P, Hernando P F, Alegría J S D. *Anal Chim Acta* 638(2009) 209–212(2009).
- [15] Wang X-W, Zhang L, Ma C-L, Song R-Y, Hou H-B, Li D-L. *Hydrometallurgy* 100 (2009) 82–86.
- [16] Takeda K, Kobayashi T. *J Membr Sci* 275(2006) 61–69.
- [17] Suede R, Bodhibukkana C, Tangthong N, Amnuaikit C, Kaewnopparat S, Srichana T. *J Controlled Release* 129 (2008) 170–178.
- [18] Sun C, Liu H-Z. *Anal Chim Acta* 612(2008) 160–164.
- [19] Sun S-W, Lee S-S, Chen L-Y, Chen C-K. *J Chromatogr A* 767 (1997) 277-284.
- [20] Zhang Y-Q, Shan X, Gao X-Q. *Sep Purif Technol* 76 (2011) 337–344.
- [21] Hilal N, Kochkodan V, Busca G, Kochkodan O, Atkin B P. *Sep Purif Technol* 31 (2003) 281–289.
- [22] Wang P, Hu W-M, Su W-K. *Anal Chim Acta* 615(2008) 54–62.
- [23] Sergeyeva T A, Brovko O O, Piletska E V, Piletsky S A, Goncharova L A, Karabanova L V, Sergeyeva L M, El'skaya A V. *Anal Chim Acta* 582 (2007) 311–319.
- [24] Kochkodan V, Hilal N, Melnik V, Kochkodan O, Remizovska A. *J Membr Sci* 377(2011) 151– 158.

- [25] Meng M-J, Feng Y-H, Zhang M, Ji Y-J, Dai J-D, Liu Y, Yu P, Yan Y-S. Chem Eng J 231(2013) 132–145(2013).
- [26] Gao B-J, Qiao Z-W, Chen T. Mater Chem Phys 143 (2014) 1119-1130.
- [27] Gao B-J, Zhang Y-Q, Xu Y. Hydrometallurgy, 150(2014) 83-91.
- [28] Ersöz A, Say R, Denizli A. Anal Chim Acta 502 (2004) 91–97.
- [29] Say R, Ersöz A, Türk H, Denizli A. Sep Purif Technol 40(2004) 9–14.
- [30] Chen R-R, Qin L, Jia M, He X-W, Li W-Y. J Membr Sci 363(2010) 212–220.
- [31] Zhang Y-Q, Shan X, Gao X-Q. Sep Purif Technol 76 (2011) 337–344.
- [32] Ma Y-X, Shi F-M, Ma J, Wu M-N, Zhang J, Gao C-J. Desalination 272(2011) 51–58.
- [33] Chakrabarty B, Ghoshal A K, Purkait M K. J Membr Sci 309 (2008) 209–221.
- [34] Wang P, Hu W-M, Su W-K. Analytica Chimica Acta, 615(2008)54–62.
- [35] Wu Y-L, Yan Y-S, Pan J-M, Dai X-H, Shi W-D, Meng M-J. Chinese Chemical Letters, 25 (2014) 273–278.
- [36] Vatanpour V, Madaeni S S, Zinadini S, Rajabi H R. J Membr Sci 373(2011) 36–42.

Figure legend

Scheme 1 Schematic expression of chemical structures of matrine and cytosine

Scheme 2 Chemical process to prepare imprinted membrane GMIM

Scheme 3 Mechanism of selective permeation of matrine across imprinted membrane GMIM

Table 1 Distribution coefficient and selectivity coefficient data for matrine/cytosine system

Fig. 1 (A) FTIR spectra of substrate membrane CMPSF and amination membrane AMPSF

Fig. 1 (B) FTIR spectra of grafted membrane PSF-g-PMAA and imprinted membrane GMIM

Fig. 2 (A) SEM photographs of cross section of substrate membrane CMPSF

Fig. 2 (B) SEM image of cross section of imprinted membrane GMIM

Fig. 3 (A) OM photographs of surfaces of substrate membrane CMPSF

Fig. 3 (B) OM photographs of surfaces of imprinted membrane MIGM

Fig. 4 Adsorption isotherms of grafted membrane PSF-g-PMAA and PSF membrane for matrine

Temperature: 25°C; pH=8

Fig. 5 Adsorption isotherms of NIM for matrine and cytosine

Temperature: 25°C; pH=8

Fig. 6 Binding isotherms of GMIM for matrine and cytosine

Temperature: 25°C; pH=8

Fig. 7 Permeation curves of matrine and cytosine across NIM

Temperature: 25°C; pH=8

Fig. 8 Permeation curves of matrine and cytosine in single component solution across GMIM

Temperature: 25°C; pH=8

Fig. 9 Permeation curves of matrine and cytosine in mixed solution across GMIM

Temperature: 25°C; pH=8

Matrine

Cytisine

Scheme 1

(1) Forming asymmetric porous CMSUF membrane by phase inversion

(2) Amination of chloromethylated polysulfone membrane

(3) Production of free radical on surface of asymmetric membrane

(4) Graft-polymerization of MAA on surface of membrane

(5) Forming Grafted MIM

Scheme 2

Scheme 3

Fig. 1 (A)

Fig. 1 (B)

Fig. 2 (A)

Fig. 2 (B)

Fig. 3 (A)

Fig. 3 (B)

Fig. 4

Fig. 5

Fig. 6

Table 1

Adsorb material	NIM		GMIM	
Adsorbate	Matrine	Cytisine	Matrine	Cytisine
$K_d(\text{mL}/\text{cm}^2)$	0.514	0.562	1.136	0.234
k	0.915		4.854

Fig. 7

Fig. 8

Fig. 9