Membranes and membrane processes for chiral resolution

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This *critical review* is devoted to an active field of research on chiral separation, membrane-based enantioseparation technique, which has potential for large-scale production of single-enantiomer compounds. Adsorption-type enantioselective membranes and membrane-assisted resolution systems with non-enantioselective solid membranes have attracted much attention recently. The principles and recent developments of both enantioselective liquid and solid membranes and membrane-assisted processes for chiral resolution will be summarized comprehensively in this review, in which the contents are of interest to a wide range of readers in a variety of fields from analytical, organic and medicinal chemistry, to pharmaceutics and materials, to process engineering for fabricating pharmaceuticals, agrochemicals, fragrances and foods, and so on (148 references).

1. Introduction

Chirality of chemical substances is ubiquitous in nature, and the essential substances that form living organisms such as amino acids and sugars are chiral and usually exist as pure enantiomeric forms.[1,2] Early in 1848, Pasteur[3] isolated two enantiomers of tartaric acid that had apparently identical chemical qualities and crystalline forms but acted differently in solution towards polarized light. In the last two decades, enantioseparation technology has developed rapidly[4,5] in response to demand for optically pure compounds in a wide variety of applications. The goal of this review is to introduce the state of the art of membrane-based technology for chiral resolution.

### 1.1 The increasing need for optically pure compounds

The different enantiomers of a chiral drug usually exhibit different pharmacological activities, metabolic effects, metabolic rates, and toxicities due to the high degree of stereo-selectivity of enzymatic reactions and other biological processes.[4–6] In some cases, only one of the enantiomers of a chiral drug contributes to its pharmacodynamic behavior, while the other shows no or a much weaker effect as well as side-effects or even toxicity.[7] A typical example is thalidomide, a racemic drug widely used during the 1960s to treat nausea during pregnancy in many European countries. The D-isomer of thalidomide is a safe sedative, but, unfortunately, the L-form causes severe birth defects and deformities. D-Propoxyphene (trade name "Darvon") is a narcotic analgesic and prescribed for the relief of moderate pain from surgery or major injuries, while L-propoxyphene enantiomer has antitussive properties. Another example is *S*-perindopril (1-[2-[(1-carboxybutyl)amino]-1-oxopropyl]octahydro-1*H*-indole-2-carboxylic acid), which can both suppress tumor growth and angiogenesis and modify clinical features of Parkinson's disease. Its *R*-isoform, however, has less activity.[8] There are many other examples with less toxicity, such as the β-blocker *R*/*S*-propranolol (Prp), D/L-carnitine, and D/L-methotrexate.[9–14]

Therefore, single-enantiomer drugs are urgently needed to improve safety and potency. In recent years, many countries have established strict requirements for patenting new chiral

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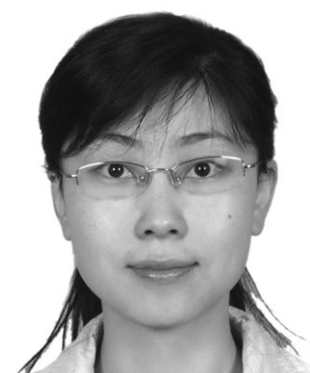
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drugs to ensure their safety and potency.[15–17] For example, the US Food and Drug Administration demands detailed documentation of pharmacological and pharmacokinetic behavior of each enantiomer as well as their combined effects and permits only single-enantiomer drugs to enter the market when the individual enantiomers show differences.[15] Such regulations have greatly encouraged and stimulated the production and sale of single-enantiomer drugs. Although sales of all drugs have continued to increase, the proportion of available drugs that are chiral agents has risen; an estimated 39% (US $151.9 billion) of all drugs sold in 2002 were single enantiomers, which is up from approximately 27% (US $74.4 billion) in 1996.[7] Thus, the sales of chiral drugs are increasing annually at a rate of 7% to 8% and are expected to exceed US $200 billion by 2008.

Besides the use of single enantiomers in the pharmaceutical industry, enantiomerically pure compounds are becoming increasingly significant in the production of other chemical products, such as agrochemicals, fragrances, and foods.[4–6,18–20] Thus, the large-scale production of single enantiomeric compounds is scientifically and economically important.

1.2 Existing methods for obtaining optically pure compounds

Generally, asymmetric synthesis and chiral resolution are the two methods available for preparing enantiomerically pure substances.[20–22] Compared with the currently more popular resolution method, asymmetric synthesis remains expensive and with low overall yields.[4] Although the technologies for chiral resolution have their own difficulties, due to the similar physical and chemical qualities of most pairs of enantiomers, they are effective for the resolution of racemates on both

analytical and industrial scales.[4–6,13,20–24] There are currently four types of chiral resolution methods: crystallization resolution, kinetic resolution, chromatographic separation, and membrane-based separation. The characteristics of these methods are compared in Table 1.

Crystallization resolution[21,22,25] can be divided into direct (or preferential)[26–30] and diastereomeric crystallization methods.[31–45] The direct crystallization method was first employed by Pasteur[3] and is still used to produce some substrates on both small and large scales. Although it can be cheaper and simpler than other methods, it can also be difficult to employ on an industrial scale because the product crystals are mixtures of two different enantiomorphic crystals.[46] This problem can be solved by seeding a supersaturated racemic solution with the desired enantiomer.[46,47] The direct crystallization method is available only when the racemate is a conglomerate. Unfortunately, to the best of our knowledge, only 5% to 10% of all organic racemates form conglomerates. The alternative is to employ the diastereomeric crystallization method, which can resolve a true racemate or racemic compound by using an optically pure resolving agent. Diastereomeric crystallization can be split into four categories based on the molecular recognition in chiral discrimination: diastereomeric salt,[31–34] diastereomeric derivative,[35] inclusion crystallization,[36–41] and metallic complex crystallization.[42–45] Despite these alternatives, identifying an appropriate resolving agent or chiral host compound is a time-consuming task. Furthermore, the appropriate resolving agents or chiral host compounds are often expensive, and they must subsequently be removed from the desired enantiomer.

Table 1 Comparison of currently existing chiral resolution methods

Methods	Advantages	Disadvantages	Possible scale	Ref.
(a) Crystallization resolution				
(a1) Direct or preferential crystallization	Simplicity, low cost	Batch operation, resolving conglomerate	Small- and large-scale	25–30, 46, 47
(a2) Diastereomeric crystallization	Simplicity, wide applicability	Expensive, difficulty in finding appropriate resolving agents	Large-scale, industrial scale	31–45
(b) Kinetic resolution				
(b1) Chemical-mediated	High stability	Low efficiency	Preparative scale, large-scale	50–54, 57
(b2) Enzyme-mediated	High resolving efficiency	Decreasing enzyme activity, narrow application range	Preparative scale, large-scale	48, 49, 52, 55–59
(c) Chromatographic separation				
(c1) Supercritical fluid chromatography	Lower costs,[a] high efficiency, resolving most racemates	Low capacity,	Large-scale	61, 72
(c2) Simulated moving bed chromatography	Continuous operation,[a] high efficiency, resolving most racemates	Low capacity,	Large-scale	64, 65, 72
(c3) Other chromatography	High efficiency, resolving most racemates	Low capacity, expensive, batch operation, slow and labor intensive	Analytical scale, preparative scale	60, 62, 63, 65–69
(d) Membrane-based separation				
	Low cost, energy saving, high capacity, continuous operation and easy scale-up	Low number of transfer units per apparatus	Large-scale, industrial scale	4–6, 9, 10, 13, 14, 70, 71, 75–148

[a] Note: Advantages of (c1) and (c2) were obtained by comparing with high performance liquid chromatography.

Kinetic resolution can resolve two enantiomers on the basis of their different reaction rates with a chiral entity. The chiral entity can be a biocatalyst (*e.g.*, an enzyme or a microorganism)[48,49] or a chemocatalyst (*e.g.*, a chiral acid/base or a chiral metal complex).[50,51] Accordingly, kinetic resolution includes both chemical- and enzyme-mediated kinetic resolution.[48–59] The main disadvantage of enzyme-mediated kinetic resolution, which is frequently used in industrial synthesis,[55] compared with chemical-mediated kinetic resolution is that the catalytic activity decreases over time. Although the theoretical yield of the desired enantiomer from kinetic resolution does not usually exceed 50%, dynamic kinetic resolution can, in principle, convert essentially 100% of the racemate to the desired product.[57–59]

Chromatography techniques for chiral separation include gas chromatography, supercritical fluid chromatography, capillary electrochromatography, and liquid chromatography, the last of which includes thin-layer chromatography and simulated moving bed chromatography.[60–71] Such techniques can separate almost any racemic mixture either on an analytical level or on a preparative level; however, they are generally expensive, inefficient, and must be performed as batch operations. With the development of supercritical fluid chromatography and simulated moving bed chromatography techniques, lower operating costs and continuous operation have been obtained compared with high performance liquid chromatography. Recently, some advances have been made in the fields of supercritical fluid chromatography and simulated moving bed chromatography that have already or sequentially brought chromatographic separation of enantiomers to commercialization.[72]

In summary, crystallization, kinetic, and chromatographic resolution have various advantages but also disadvantages, including high energy consumption, high cost, low efficiency, and discontinuous operation.

1.3 Membrane technologies for chiral resolution

Low-cost, continuous, high-efficiency resolution technology is clearly needed for commercial-scale preparation of enantiomerically pure substances. Membrane technology, fortunately, fulfils this need very well because of its high efficiency, low energy usage, simplicity, convenience for up- and/or downscaling, and continuous operability.[4–6,73,74] Membrane-based chiral resolution can be achieved using either enantioselective or non-enantioselective membranes.[4,5,74] The enantioselective membranes themselves can carry out chiral separation of stereoisomers because they contain chiral recognition sites, and their conformations are usually categorized as liquid or solid. The non-enantioselective membranes have no enantioselectivity themselves and often assist in the separation of enantiomers, for instance, by acting as ultrafiltration membranes. Therefore, non-enantioselective membrane-assisted processes, also called combinatorial methods, are generally combined with other chiral recognition approaches such as enzymatic kinetic resolution, solution systems with micelles, and systems using chiral selectors as complexing agents. There are already some examples of the application of membrane-assisted methods to commercial-scale production,[13] and this approach has attracted a great deal of attention over the last decade.

In this review, we systematically describe the recent progress and achievements in the development of membrane-based chiral-resolution techniques. In section 2, the resolution mechanisms and performance parameters for the membranes are introduced. The techniques and recent developments in chiral separation using enantioselective and non-enantioselective membranes are reviewed in sections 3 and 4, respectively. Finally, section 5 contains a summary and a discussion of the outlook for membrane-based chiral separation.

2. Theories in membrane-based chiral resolution

2.1 Resolution mechanism

Enantioselective membranes are able to resolve optical isomers due to chiral properties such as chiral recognition sites (*e.g.*, chiral side chains, chiral backbones, or chiral selectors). These can be further subdivided into liquid and solid membranes according to the status of the membrane phase. The enantioselective membranes act as selective barriers in the resolution process, and they selectively transport one enantiomer due to the stereospecific interaction between the enantiomer and chiral recognition sites, thereby producing a permeate solution enriched with one enantiomer. The different binding affinities of two enantiomers may be the result of different hydrogen bonding, hydrophobic, Coulombic, van der Waals interactions and steric effects with the chiral sites.[75]

There are two mechanisms of selective transport: facilitated and retarded transport (Fig. 1).[73,76] The separation of enantiomers by liquid membranes is usually based on the facilitated transport mechanism, whereas separation by solid membranes is based on both facilitated and retarded transport mechanisms. The enantioselective solid membranes are therefore categorized according to their mechanisms. Notably, most resolution processes include concurrent sorption- and diffusion-selective transport phenomena. The kind of resolution mechanism depends on the dominant transport process. There are several factors that can influence the classification of the enantioselective membranes. One is the magnitude of the binding affinity force, which is determined by the intrinsic
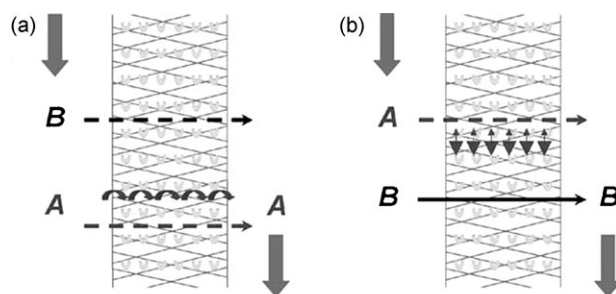
Fig. 1 Schematic illustration of selective transport by enantioselective membranes based on two different resolution mechanisms: (a) facilitated transport mechanism, and (b) retarded transport mechanism. A and B represent two enantiomers of the same compound. The chiral recognition sites in the membranes have a stronger binding affinity for enantiomer A. (Reproduced with permission from ref. 76. © 2004 Elsevier.)

interaction between the analyte and the chiral environment; the others are the kinds of driving force employed and the magnitudes of the employed driving force.

Stronger intermolecular interactions and the use of stronger driving force may help the enantioselective membranes function *via* the retarded transport mechanism. The driving force can be a difference in the concentration, pH, pressure, or electrical potential. The magnitude of the driving force is also important for obtaining an efficient chiral resolution; if the driving force is excessively large or small, it will fail to achieve any enantioseparation.[77–81]

In facilitated transport processes, one enantiomer preferentially adsorbs to the chiral recognition sites in the enantioselective membranes near the feed phase due to a higher binding affinity. From there, it continuously adsorbs and desorbs from one chiral site to the next, and at last is transported toward the stripping phase, usually by concentration-driven permeation or occasionally by an electrical potential difference (Fig. 1a). The other enantiomer, which has no or less specific binding affinity for the chiral environment, passes through the membrane by diffusion.[76] In other words, this transport mechanism is based on the different diffusion rates of two enantiomers and is similar to extraction in liquid. Generally, most chiral liquid and solid membranes composed of a chiral polymer or coated with an enantioselective polymeric layer utilize this class of transport.[73] Typical examples of chiral polymers include polysaccharides, for example chitosan and sodium alginate.[82] This type of transport is also utilized by chiral selector-immobilized membranes with relatively low binding affinities for two enantiomers.

Chiral resolution membranes based on facilitated transport mechanisms are also called diffusion-enantioselective membranes.[73] In these membranes, complete optical resolution is achieved at the initial period of the permeation, and the enantioselectivity decreases with time due to the increasing non-enantioselective diffusion of weaker binding isomers. Moreover, the higher selectivity coefficient should be obtained at the lower feed concentration, and the selectivity usually will decrease as the driving force increases, eventually resulting in a loss of permeation selectivity. Smaller-diameter or denser transmembrane pores are helpful for preventing the non-enantioselective diffusion of one enantiomer and produce higher enantioselectivities at the expense of lower permeability.[76,83] These phenomena show that the facilitated transport process is kinetically driven. The main disadvantage of these diffusion-enantioselective membranes is the inverse proportionality of permeability and permeation selectivity, which are the two most important properties determining the possible scale of the process.[73]

In some cases, when the driving force is a pressure gradient rather than a concentration gradient, the transport mechanism may change into retarded transport. Membranes based on the retarded transport mechanism are also called adsorption-enantioselective membranes,[73] and they usually incorporate chiral selectors. In contrast to the facilitated transport mechanism, retarded transport retains the adsorbed enantiomer in the membrane phase,[76] while permitting the other enantiomer to pass through the membrane more easily due to it having no or lower affinity for the chiral recognition sites

(Fig. 1b). In an adsorption-enantioselective membrane, the binding affinity between chiral recognition sites and enantiomers is stronger than that of a diffusion-enantioselective membrane, and this interaction force always exists between one enantiomer and one chiral site. A pressure difference is the commonly used driving force for such membranes. Separation efficiency of these membranes will be mainly determined by the binding capacity, and the separation factor (α) becomes 1.0 after the chiral recognition sites reach saturation.[76] The adsorption-enantioselective membranes are expected to simultaneously possess relatively high flux and high enantioselectivity, and thus have more potential than diffusion-enantioselective membranes to carry out industrial-scale productions of optically pure compounds.[73]

Some non-enantioselective membranes in chiral resolution systems are only used as supports to capture chiral selectors or to separate particles by size rather than for recognizing one enantiomer in a racemic mixture. In other words, the resolution mechanism for non-enantioselective membranes is quite different from that for enantioselective membranes. Non-enantioselective membranes merely utilize the retention for larger molecules to achieve the enrichment of smaller molecules in the stripping phase. Therefore, a difference in size of the enantiomers is the key to enantioseparation by such non-enantioselective ultrafiltration membranes. For instance, formation of a complex of one enantiomer with a large chiral recognition molecule such as bovine serum albumin (BSA) or the employment of enzymes to selectively hydrolyze one of the enantiomers followed by ultrafiltration of porous non-enantioselective membrane can be used to accomplish enantiomeric separation.

2.2 Performance parameters

To describe the membrane process for chiral resolution, it is essential to introduce or define the performance parameters of the membranes for enantioseparation first. In general, the performance of an enantioselective membrane is described by two performance parameters, namely, permeability and selectivity. As mentioned above, membranes with both high permeability and selectivity are usually desired for large-scale industrial applications.

2.2.1 Permeability. The permeability, also called the permeation coefficient, is usually calculated by

$$P' = \frac{Jx}{C_f - C_s} \tag{1}$$

where P' is the permeation coefficient, J is the normalized flux, x is the membrane thickness, and C_f and C_s refer to the concentrations of the feed phase and stripping phase, respectively. J is defined as the magnitude of the permeation per unit membrane area and per unit time, and can be calculated by the following equation:

$$J = \frac{\Delta C V}{\Delta t A} \tag{2}$$

where ΔC is the change in concentration, Δt is the permeation time, V is the downstream volume, and A is the effective membrane area. Flux is enhanced by employing a

driving force (*e.g.*, a pressure-driven process or electrodialysis), an ultrathin film, or a membrane with a high porosity.[6,83,84]

A solution-diffusion mechanism determines the permeation through the homogeneous dense membranes, which can be described as follows:[73]

$$P = S \cdot D \tag{3}$$

where S and D refer to the sorption coefficient and the diffusion coefficient, respectively. Sorption coefficient is a thermodynamically determined parameter defined as the ratio of the equilibrium membrane concentration (C_m) to the concentration in the bulk liquid (C_l), as shown in eqn (4).[73] It is dependent on several factors such as the materials and structures of the membranes, the location of the active functional groups, and the properties of the analytes.

$$S = \frac{C_m}{C_l} \tag{4}$$

The diffusion coefficient, D, is a kinetically determined parameter[45] that is influenced by the characteristics of both the membranes and the analytes.

As for membranes with tiny pores, the transport mechanism is a pore flow model such as Knudsen diffusion, surface diffusion, and capillary condensation and so on.

2.2.2 Selectivity. The selectivity of enantioselective membranes is another crucial factor representing the purity of the products. In general, the stereoselectivities of membranes can be described by the enantioselectivity (α^P), the separation factor (α), and the enantiomeric purity, which can include the enantiomeric excess (ee), purity (P_u), and recovery (R). Some frequently used factors such as α and *ee* can have multiple expressions due to the transformation of defined equations under specific conditions or calculations based on experimental data obtained from different analytical approaches.

The enantioselectivity of the membranes, α^P, was first defined as the permeability ratio of the two enantiomers.[73] By substituting eqn (3) into the equation for α^P, we obtain

$$\alpha^P = \frac{P_X}{P_Y} = \frac{S_X}{S_Y} \cdot \frac{D_X}{D_Y} = \alpha^S \cdot \alpha^D \tag{5}$$

where the subscripts X and Y denote the enantiomer preferentially transported through the membrane and retained in the feed solution, respectively; and α^S and α^D are the sorption and diffusion selectivities, respectively.

In general, the separation factor, α, is defined as the concentration ratio of the two isomers in the stripping phase divided by that in the feed phase,[14] as shown in eqn (6):

$$\alpha = \frac{[X]_s / [Y]_s}{[X]_f / [Y]_f} \tag{6}$$

where the X and Y indicate the two enantiomers as described in eqn (5); [X] and [Y] refer to the concentrations of X and Y, respectively; and the subscripts *s* and *f* denote the stripping

and feed phases, respectively. If the concentration of the feed solution is set as that at the initial time, the concentrations of the two enantiomers in the feed phase becomes equal ($[X]_f = [Y]_f$), and eqn (6) becomes

$$\alpha = \frac{[X]_s}{[Y]_s} \tag{7}$$

The normalized flux (J) can be calculated according to Fick's law:[85]

$$J = -D \frac{dc}{dx} \tag{8}$$

where dc/dx is the concentration gradient, x is the membrane thickness, c is the concentration fraction, and D is the diffusion coefficient as described in eqn (3).

When the feed solution is well stirred, D_X and D_Y are approximately equal (the subscripts X and Y are the same as defined in eqn (6)). In addition, for the two enantiomers, the membrane thickness is the same. In other words, the flux is only a function of the concentration of the enantiomer in the stripping phase. Therefore, eqn (6) can be rewritten as

$$\alpha = \frac{J_X}{J_Y} \tag{9}$$

where J_X and J_Y are the fluxes of enantiomer X and antipode Y, respectively.

In some previous reports, the separation factor, α, was calculated using equations somewhat different from those shown in eqn (6) and (9). For instance, when a chromatogram was used to analyze the separation efficiency, α could be calculated as follows:[70,71]

$$\alpha_t = (t_X - t_0)/(t_Y - t_0) = k'_X / k'_Y \tag{10}$$

where t_X and t_Y are the retention times of the two enantiomers, respectively; t_0 is the void time; and k'_X and k'_Y are the retention factors of enantiomer X and Y, respectively. Another equation for estimating α is

$$\alpha' = \frac{J_X / J_Y}{C_{l,X} / C_{l,Y}} \tag{11}$$

where $C_{l,X}$ and $C_{l,Y}$ are the concentrations of enantiomer X and antipode Y in the bulk liquid after adsorption equilibrium has been reached, as mentioned in eqn (4).

Another coefficient called operational enantioselectivity for describing the selectivity, α_{op}, is defined as follows:

$$\alpha_{op} = \frac{[X]_{f,o} - [X]_s}{[X]_s} \times \frac{[Y]_s}{[Y]_{f,o} - [Y]_s} \tag{12}$$

The chiral selectivity of membranes is also frequently calculated in terms of the enantiomeric excess (ee) of permeates. The *ee* value is defined as the ratio of the concentration difference and the concentration summation of both enantiomers in the stripping phase:[13]

$$ee = \frac{[X]_s - [Y]_s}{[X]_s + [Y]_s} \times 100 \tag{13}$$

There have been some transformations of the above equation, such as

$$ee' = \frac{E_X - E_Y}{E_X + E_Y} \times 100 \qquad (14)$$

where E_X and E_Y are the extraction efficiencies of enantiomers X and Y, respectively. The extraction efficiency, E, is defined as the ratio of the amount of analyte extracted in the receiving phase to the total amount of analyte in the initial feed phase.[9] When using high performance liquid chromatography with a chiral column and detection with a UV spectrophotometer, the ee value can be determined from the peak areas of the two enantiomers, A_X and A_Y, as shown in eqn (15):[78,82]

$$ee'' = \frac{A_X - A_Y}{A_X + A_Y} \times 100 \qquad (15)$$

Besides these selectivity parameters, some other factors such as the purity, P_u, and the recovery, R, are used to estimate the separation efficiency of the membranes in solution systems.[86] P_u and R are defined as follows,

$$P_u = \frac{[X]_s}{[X]_s + [Y]_s} \qquad (16)$$

and

$$R = \frac{[X]_s}{[X]_{f,o}} \qquad (17)$$

where $[X]_{f,o}$ represents the initial feed concentration of enantiomer X.

3.   Enantioselective membranes for chiral resolution

Enantioselective membranes can achieve enantioseparation by binding the two enantiomers with different affinities. Either the bulk structures of membrane materials or the chiral selectors added to the membranes provide chiral recognition sites. Examples of bulk structures with chiral recognition sites include chiral polymers[78,82,87,88] such as polysaccharides (e.g., chitosan and sodium alginate) and polyamino acid derivatives, and chiral selectors can include proteins (e.g., antibodies or BSA), amino acids and their copper complexes, DNA, polypeptides, enzymes, calix[n]arene, cyclodextrins, and crown ether derivatives.

Enantioselective membranes include liquid membranes and solid membranes. In liquid membrane systems, chiral selectors are directly dissolved in the liquid membrane phase, and the liquid membrane should not be miscible in either the feed or the receiving phases. One of the two isomers is preferentially transported by virtue of mobile chiral carriers in the liquid membrane usually in the presence of a pH or concentration gradient. A typical liquid membrane system consists of an organic solvent with aqueous donating and receptor phases.

Solid membranes can be prepared by casting chiral polymer solutions or mixtures containing added chiral selectors. Alternatively, chiral selectors can be immobilized (e.g., by impregnation, esterification, or grafting) on the surfaces or in the pores of support membranes. Also molecular imprinting can be employed to form molecular recognition sites inside the membranes. The prepared membranes are either self-supporting or attached on base membranes. Membranes as supporting or base structures can be flat or tubular, for example, hollow fibers. The use of hollow fiber membranes usually results in a relatively compact system because of a high membrane area per volume.

Liquid membranes usually offer high mass transfer rates and low chiral selector consumption,[89] whereas solid membranes are usually characterized by long-term stability.

3.1   Enantioselective liquid membranes

There are three types of liquid membranes (Fig. 2), including supported liquid membranes (SLMs),[9,83,90–97] bulk liquid membranes (BLMs),[14,97–100] and emulsion liquid membranes (ELMs).[101,102] Of these, SLMs and ELMs are the two most commonly used. To achieve the chiral resolution by liquid membranes, two key considerations must be taken into account when designing such a system. First, an appropriate chiral selector must be chosen that preferentially binds one of the two isomers, and, second, the solubility of the chiral selector and its complex as well as the free isomers in the feed, membrane, and stripping phases must be considered. In other words, to minimize passive transport, the chiral selector and its complex should be soluble only in the liquid membrane, and the uncomplexed enantiomer molecules should be soluble only in the feeding and receiving phases. In liquid membrane systems, concentration and pH gradients are frequently employed as driving forces, and for SLMs, pressure differences are sometimes used as the driving force.

3.1.1   Supported liquid membranes. In a SLM system, the membrane phase is fixed in place by non-enantioselective materials on both sides, or it fills the pores of a non-enantio-selective porous membrane. The latter type of SLM is also called an immobilized liquid membrane (ILM, Fig. 2a) and is more widely used because it is more easily designed and produces thinner membranes. A variety of base materials are
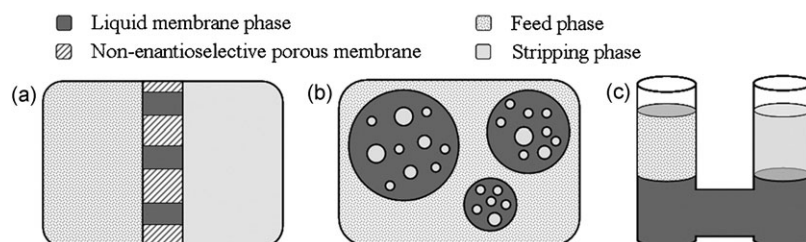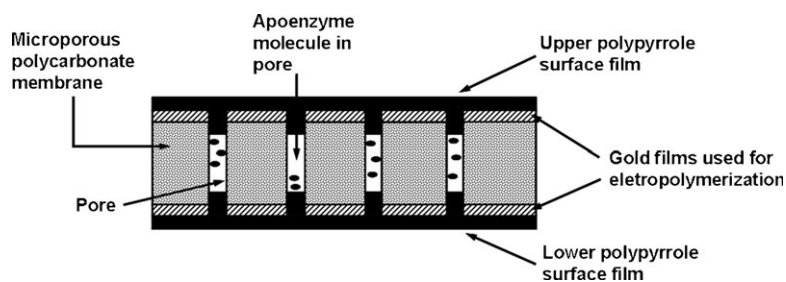

■ Liquid membrane phase   ▨ Feed phase
▨ Non-enantioselective porous membrane   ▢ Stripping phase

Fig. 2   Schematic illustration of different types of liquid membranes: (a) SLM (ILM), (b) ELM, and (c) BLM.

Fig. 3 Schematic cross-section of the sandwich ILM with the apoenzyme entrapped in the pores. (Reproduced with permission from ref. 83. © 1997 Nature Publishing Group.)

available for ILMs, such as polypyrrole, polycarbonate, polypropylene, polysulfone and polyvinylidene difluoride. SLMs offer several advantages compared with ELMs and BLMs. One of the most important is that only a small amount of expensive chiral carrier is required to achieve enantiomeric resolution. In addition, the mechanical strength of the membrane phase of SLMs is higher than those of ELMs and BLMs.

An ILM system using a polypropylene hollow-fiber membrane module was reported by Hadik for the separation of racemic amino acids such as D/L-lactic acid and D/L-alanine (D/L-Ala).[90] In this system, toluene solution containing the chiral selector N-3,5-dinitrobenzoyl-L-phenylalanine-octylester was used as the liquid membrane phase, and it separated hydrophilic lumen- and shell-side aqueous phases. This allowed the preferential transport of the D-enantiomer of the analyte and gave α and ee values of 2.0 and 33.5%, respectively, for D/L-lactic acid and 1.75 and 27.17%, respectively, for D/L-Ala. The maximal flux appeared at the initial stage of operation, which was 1.88 and 1.54×10^{-4} mol m^{-2} h^{-1} for D-lactic acid and L-lactic acid at 20 h, respectively, and 9.19 and 6.13×10^{-5} mol m^{-2} h^{-1} for D-Ala and L-Ala at 11 h, respectively. Similarly, Chmiel et al.[95] enantioseparated a series of N-protected amino acid derivatives by employing ILM based on polysulfone hollow fibres with separation factor between 2 and 4. After five separation steps, 99% D-enantiomer and 99% L-enantiomer could be gained for DNB-D,L-leucine at transmembrane flux of larger than 20 mmol m^{-2} h^{-1}.

For flat ILMs, sandwich membranes are the most commonly used configurations. These systems are composed of a porous membrane impregnated with a chiral selector-containing liquid, which is then surrounded by two relatively dense films. Martin's group[83] and Skolaut et al.[93] proposed a similar strategy for enantioselective ILMs containing physically entrapped enzymes as chiral carriers. The former incorporated D-amino acid oxidase apoenzyme into the sandwich membrane as molecular recognition agent (Fig. 3). Using a 30 nm pore membrane loaded with D-amino acid oxidase apoenzyme, this group was able to achieve the selective transport of D-phenylalanine (D-Phe) from a racemic mixture with a maximal α value of 4.9. Skolaut et al., on the other hand, used two mutant enzymes, Phe ammonia lyase Y109F and histidine ammonia lyase E414A, which maintain their binding affinities for their substrates but have almost no catalytic activity. They studied both the concentration and time dependence of the enantioselectivity in an enzyme-loaded membrane. Surprisingly, when the concentration of D/L-Phe was low in the feed solution (0.1 mM), the L-enantiomer preferentially permeated the Phe ammonia lyase Y109F-immobilized membrane, giving a maximum α value of 2.5 at 30 min. At a much higher concentration (2.5 mM), however, the D-enantiomer was transported faster. This could be due to competition for and longer occupation of the binding sites of the D-enantiomer at high concentrations so that little free enzyme remained available for transporting the L-enantiomer.[93] In contrast, histidine ammonia lyase E414A facilitated the transport of the L-enantiomer at all three concentrations applied (0.1, 1.0, and 2.5 mM). At the lowest concentration, the maximal α value (13.3) was reached at 70 min, whereas at a high concentration, the maximal α value was much lower ($\sim$2 to 3) and was reached within 30 min.

Another kind of flat, circular ILM was prepared by impregnating porous polyvinylidene difluoride membranes with a solution of isopropyl myristate containing the chiral selector N-hexadecyl-L-hydroxyproline. The membranes were then placed between two circular polytetrafluoroethylene blocks with grooves and stabilized on both sides by aluminium blocks.[9] In this system, the enantioseparation of R/S-Prp was greatly influenced by both the pH of the feed phase and the concentration of chiral selector in the membrane phase. The best ee value (5.6%) for R-Prp was obtained after 2 h when the pH was 8 and when the amount of the N-hexadecyl-L-hydroxyproline in the membrane was three times the concentration of the analyte in the initial feed phase.

SLMs often employ the carrier di-(2-ethylhexyl)phosphoric acid for the transport of amino acids. Structurally similar phosphate and phosphonate esters bearing chiral menthol or nopol moieties, however, appear to be poor or moderate carriers, giving α values of only 1.01 to 1.40 for the transport of biogenic amines, amino acids, and amino acids esters. SLMs using di-(2-ethylhexyl)phosphoric acid as a carrier are also effective for the enantiomeric separation of alkyl esters of aromatic amino acids and unusual amino acids.[91,92]

3.1.2 Bulk liquid membranes. In a BLM (Fig. 2c), a relatively thick liquid membrane separates the feed and stripping phases in the absence of a support by virtue of its immiscibility.[14] The principal disadvantage of this technique is the low interfacial surface area and, hence, the slower mass transfer rates compared with SLMs and ELMs.[89] Krieg and coworkers[14] performed the enantiomeric enrichment of a racemic drug chlorthalidone using BLMs containing β-cyclodextrin (β-CD) as a chiral mobile carrier in an aqueous membrane phase. During the initial stages of separation (after 28 h), at a low carrier concentration (1 : 4 β-CD/chlorthalidone) and a pH of 5, the maximal α value was 1.05 for a single BLM and 1.41 with a multiple BLM containing three

membranes and three stripping phases. A BLM has also been reported to achieve enrichment of enantiomers of mandelic acid and phenylglycine.[98] This system employed the chiral carrier cinchonidine dissolved in a mixture of dodecane/decanol (v/v = 1 : 1) as the membrane phase. When the initial concentration ratio of the carrier to analyte in the feed phase was 0.5 and the pH values of the feed and receiving phase were 4–5 and 8, respectively, a maximal α value of 1.5 was reached for L-mandelic acid. Another BLM system was reported by Okada et al.,[103] in which calix[4]arene-derived esters with chiral pendant groups were used to separate L-amino acids with ee values of 5.2% to 26.8%. Calix[n]arene is a macrocyclic compound formed by the condensation of p-alkylphenol and formaldehyde under basic conditions, and it recognizes guest molecules by size and/or specific affinity of functional groups on its framework.[99] Crown ethers are also commonly used as chiral selectors on account of their chirality, side chains, and the rigidity of the micro-environment of the moieties. However, they can only be utilized for the separation of organic compounds containing a primary amine group and should not be utilized together with potassium ion. Aza crown ethers, with a side chain attached to the nitrogen atom in the macrocyclic ring, may enhance and regulate cation-binding properties as well as lipophilicity. Demirel and co-workers[100] studied the enantioselective transport of the sodium and potassium salts of phenylglycine, Phe, and tryptophan (Trp) through chloroform liquid membranes containing diaza crown ethers as chiral carriers. They found that the highest α factor (3.29) was achieved for the sodium salt of L-Trp.

3.1.3 Emulsion liquid membranes. ELMs (Fig. 2b), also called liquid surfactant membranes or double emulsions, contain spherical membrane globules that separate the feed and stripping phases (usually aqueous phases) and are stabilized by surfactants. ELMs have some benefits such as relatively low cost, good stability, and the highest mass transfer rates among the three liquid membrane systems;[89,101,102] however, they suffer from emulsion swelling and leakage, which may lead to lower extraction efficiency and selectivity.[89]

For ELMs, it is vital to choose a proper emulsion stabilizer to ensure membrane stability and to avoid membrane rupture. In addition, the proper choice of chiral carriers can facilitate the preferential transport of one enantiomer through the liquid membrane. Dżygiel and Wieczorek[102] reported that several industrial surfactants are able to stabilize liquid emulsion membranes but that they did not act as carriers for amino acid transport. As a result, the application of an additional carrier, di-(2-ethylhexyl)phosphoric acid, was desirable to facilitate the transport. At the same time, the α value achieved with lecithin, a natural surfactant, was very low.

To separate D-Phe from a racemic mixture, Pickering and Chaudhuri[89,101] developed a chiral ELM using copper(II) N-decyl-L-hydroxyproline as a chiral carrier in a mixture of hexanol:decane (v/v = 1 : 1) as a membrane solvent. A maximum α value of 2.4 was observed in the early stages of extraction when the pH was low in the source phase and high in the membrane phase. This system should provide a low-cost means of producing large quantities of reasonably pure amino acid enantiomers from racemic mixtures.

Among these three liquid membrane systems, the most efficient may be the ILM because it has a higher resolution selectivity (ee = 33.5% and α = 13.3). Chiral resolution technology based on liquid membranes is an inexpensive approach with a relatively high selectivity and fast mass-transfer rate; however, the extraction efficiency and selectivity of these membranes are counteracted by their lack of stability and mechanical strength. For instance, a pressure difference is commonly employed as the driving force to enhance the mass-transfer efficiency, but it cannot be effectively employed in liquid membrane systems except for ILMs. Therefore, large-scale production of reasonably optically pure compounds by liquid membrane systems awaits resolution of these shortcomings.

3.2 Enantioselective solid membranes

Enantioselective solid membranes can be categorized into two kinds, namely inherent chiral membranes and membranes functionalized with immobilized chiral selectors. As far as inherent chiral membranes are concerned, they are most often fabricated by casting membrane-forming solutions of chiral polymers (e.g., polyamino acids, polysaccharides, or polypeptides) or of achiral polymers containing chiral selectors. The chiral polymers include those with chiral backbones and/or chiral side chains. In the chiral selector immobilization, chiral selectors or their footprints are usually immobilized on the surface, in the pores, or in bulk configuration on base membranes by impregnation, covalent grafting, transesterification, or molecular imprinting.[104,105]

Enantioselective solid membranes can be separated into two categories: diffusion-enantioselective membranes and adsorption-enantioselective membranes. Usually, membranes made from chiral polymers and/or that bind relatively weakly to enantiomers act as diffusion-enantioselective membranes because they lack one-to-one interactions between the chiral recognition sites and the enantiomers, whereas membranes with added chiral selectors or their footprints usually act as adsorption-enantioselective membranes.

3.2.1 Diffusion-enantioselective solid membranes

3.2.1.1 Enantioselective membranes made from chiral polymers. Similar to cellulose, polysaccharides such as sodium alginate and chitosan contain a large amount of chirally active carbons on the backbone of the ring structure. Kim et al.[82] prepared membranes by crosslinking polysaccharides with glutaraldehyde for enantioseparation of α-amino acids (e.g., Trp and tyrosine (Tyr)) by a pressure gradient. In this system, a lower degree of crosslinking, higher concentration of a feed solution, higher operating pressure, and smaller solute size leads to a lower enantiomeric excess value. As shown in Fig. 4, a maximum ee″ value of over 98% was obtained for D-Trp using a chitosan membrane.

A diffusion-enantioselective polysulfone membrane also separated a mixture of R/S-Prp with an α value of 1.7.[10] This membrane was prepared by sol–gel phase inversion using a casting solution containing the chiral selector N-hexadecyl-L-hydroxyproline (1.2 wt%). The α value decreased after 48 h and became higher when the transport rates were low.
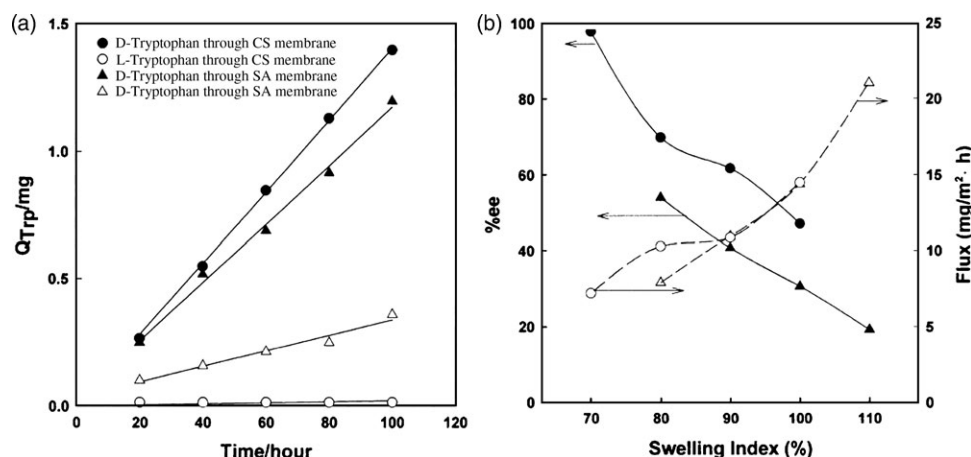
Fig. 4 The effect of time (a) and the swelling indices (b) on enantioseparation of D/L-Trp through chitosan (CS: ●, ○) and sodium alginate (SA: ▲, △) membranes. Swelling index is the increase degree of mass of the membrane swollen in water. The concentration of the feed solution and the operating pressure were 0.49 mM and 0.1 MPa, respectively. The swelling indices values were 70% and 80% for chitosan and sodium alginate membranes, respectively. (Reproduced with permission from ref. 82. © 2003 Elsevier.)

To separate enantiomers from racemic mixtures of amino acids or chiral drugs, the Aoki group[106–108] synthesized optical resolution membranes from norbornadiene and disubstituted acetylene polymers that both have optically active pinanyl groups. For example, (+)-poly{l-[dimethyl(10-pinanyl)silyl]-l-propyne} ((+)-PDPSP) membranes allowed selective permeation of a racemic mixture of Trp, 1,3-butanediol, and other compounds. This process could be maintained for more than 600 h by concentration-driven permeation with ee'' values between 12% and 48%. Also, (+)-poly{2-[dimethyl-(10-pinanyl)silyl]norbornadiene} ((+)-PDPSN) membranes were successfully used to optically separate racemic Prp, a hydrophobic compound that could not be separated by (+)-PDPSP membranes. These (+)-PDPSN membranes separated R-Prp from a racemic mixture with an ee'' value of 45%, which was sustained for as long as 2000 h. Furthermore, the (+)-PDPSN membranes were more flexible than the (+)-PDPSP membranes because they have more flexible main chains. This led to a higher P' value for Trp than that attained with the (+)-PDPSP membranes. Moreover, these tough (+)-PDPSN membranes are expected to allow the use of a pressure difference as the driving force for separation.[107]

Diphenylacetylenes with chiral pinanyl groups are easily employed for forming chiral polymeric membranes because of their excellent solubilities and film-forming properties as well as their helical conformations. Poly{(−)-1-4-[dimethyl-(10-pinanyl)silyl]phenyl-1-propyne} membranes showed a higher enantioselectivity (α^P = 3.16) toward R-Trp than membranes made from two enantioselective disubstituted acetylene polymeric membranes, namely, poly{(−)-1-4-[dimethyl(10-pinanyl)silyl]-phenyl-2-phenylacetylene} and poly-{(−)-1-3-[dimethyl(10-pinanyl)silyl]-phenyl-2-phenylacetylene}, which have thermally stable helical conformations. This was probably due to the greater stiffness of poly{(−)-1-4-[di-methyl(10-pinanyl)silyl]-phenyl-2-phenylacetylene} and poly-{(−)-1-3-[dimethyl(10-pinanyl)silyl]-phenyl-2-phenylacetylene} so that they contained more and larger molecular-scale voids than poly{(−)-1-4-[dimethyl(10-pinanyl)silyl]phenyl-1-propyne}.[108]

Teraguchi and Masuda[109] proposed a strategy for preparing poly(diphenylacetylene) by desilylation of poly{(−)-1-4-[di-methyl(10-pinanyl)silyl]-phenyl-2-phenylacetylene}. The resulting poly(diphenylacetylene) maintained the helical structure of poly{(−)-1-4-[dimethyl(10-pinanyl)silyl]-phenyl-2-phenylacetylene} and became insoluble only in organic solvents. Therefore, it is expected that poly(diphenylacetylene) can be used to fabricate chiral membranes by the solution casting method. "Depinanylsilylation" of poly(diphenylacety-lenes) membranes was effective for permeability enhancement (e.g. 1.45×10^{-9} m^2 h^{-1} vs. 1.76×10^{-11} m^2 h^{-1} of original membranes toward R-2-butanol), but led to decreased α^P value (e.g. 3.83 vs. 9.24 of original membranes toward R-2-butanol).[110,111]

Recently, polyelectrolyte multilayer membranes, which are composed of charged and optically active polyelectrolyte layers, have been used for chiral separations. These membranes have high permeation rates due to their thinness and moderate selectivity. For example, Rmaile and Schlenoff[85] prepared a series of multilayer membranes made from polypeptide polyelectrolytes, such as L- and D-poly(lysine), poly-(glutamic acid) (poly(Glu)), poly(N-(S)-2-methylbutyl-4-vinyl pyridininum iodide), and poly(styrene sulfonate). L- or D-ascorbic acid, 3-3(3,4-dihydroxyphenyl)-D/L-Ala, and a chiral viologen (a geometric isomer, not an enantiomer) were employed as the chiral probes. Combination of the L-form of one polyelectrolyte with the D-form of its oppositely charged partner may eliminate the enantioselectivity.

3.2.1.2 Enantioselective membranes made by addition of chiral selectors. Lee and coworkers[84] developed the first antibody-immobilized nanotube membranes for enantioseparation of a racemic drug, 4-[3-(4-fluorophenyl)-2-hydroxy-1-[1,2,4]triazol-1-yl-propyl]-benzonitrile. The immobilized antibody facilitated the transport of the selectively bound RS-enantiomer relative to the SR-antipode. For an alumina membrane with a pore size of 35 nm, a maximal α value of 2.6 was obtained at the lowest concentration of the feed solution. When the pore size was decreased to 20 nm, the α value
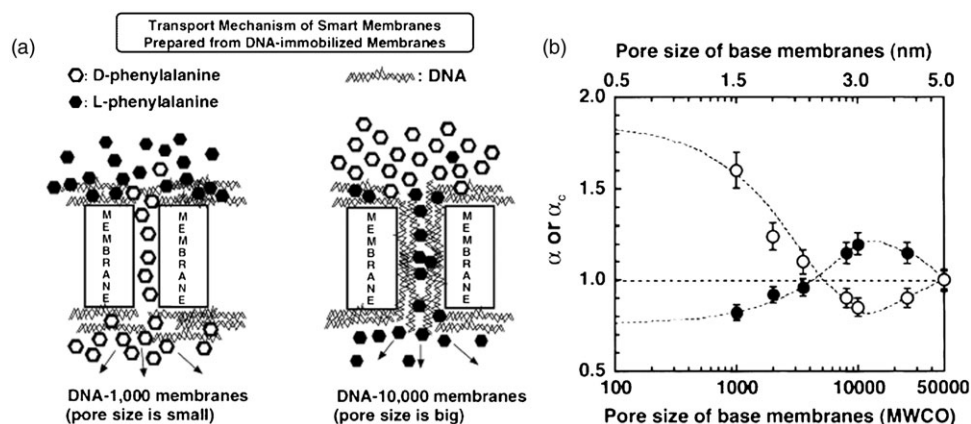
Fig. 5 The mechanism (a) and the separation factor (b) in the permeate ($\bigcirc$; α) and concentrate ($\bullet$; α_c) solutions regulated by the MWCO of the base membranes in optical resolution of D/L-Phe by immobilized DNA membranes at pH 7.0 and 25°C. α_c is defined as the concentration ratio of D-Phe to L-Phe in the feed solution after starting the experiment. (Reproduced with permission from ref. 114 and 115. © 2003 and 2005 Elsevier.)

increased to 4.5 and the fluxes of both enantiomers declined. This confirmed that the mechanism was based on facilitated transport. The content of solvent dimethyl sulfoxide altered the binding affinity and thus influenced the flux of the two enantiomers as well as the α value.

Although DNA can bind both L- and D-amino acids, it has a higher affinity toward the former (*e.g.*, L- *vs.* D-Phe).[112–116] In DNA-immobilized cellulose and chitosan membranes, the pore size influences the preferential permeation of the isomer under a pressure gradient, as shown in Fig. 5, where α_c was defined as the concentration ratio of D-Phe to L-Phe in the feed solution after starting the experiment and shows the opposite tendency as α. For example, due to an interaction between L-Phe and DNA immobilized on the surface of the cellulose membranes, D-Phe preferentially entered the pores of the membranes when the pore size was less than 2.0 nm (molecular weight cut-off [MWCO] < 5000) and then permeated through the membranes. In contrast, when the pore size was larger than 2.0 nm (MWCO > 5000), L-Phe preferentially permeated through the membranes due to immobilization of DNA both on the surfaces and inside the pores. Interestingly, the opposite effect was observed when the membranes were used to separate D- and L-Trp due to the different affinities of the two amino acids for DNA. Higuchi *et al.*[115] proposed using multiple steps of ultrafiltration through DNA-immobilized membranes to achieve a high degree of enantioseparation. For example, they showed that only four stages are needed to obtain 99% purity and a relatively high α value of 4. Therefore, it is expected that multistage separation will be useful for large-scale applications.

The enantioseparation of racemic kynurenine has been accomplished using BSA-grafted nylon hollow fiber membranes with pore sizes between 0.1 and 1.2 μm and a diameter of 2.5 cm.[86] During concentration-driven experiments, the maximal *ee* value for L-kynurenine was close to 30% but decreased to 0 after 2 h due to the nonselective diffusion of both enantiomers. For the diffusion-enantioselective membranes, however, it may be possible to prevent such diffusion by reducing the pore size of the membranes and creating multilayered BSA-grafted membranes.

Wang *et al.*[117] prepared cation-exchange and anion-exchange enantioselective membranes by blending poly(vinyl alcohol), β-CD and ion-exchange materials according to the ratio of 8:3:3. During electrodialysis experiments, cation-exchange membranes preferentially transport D-4-hydroxy-phenylglycine cation with maximal α of 1.34 in acid medium; while anion-exchange membranes preferentially transport L-4-hydroxyphenylglycine anion with maximal α of 0.84 in base medium.

Those chiral selectors usually used in liquid membranes, such as *N*-dodecyl-4(*R*)-hydroxy-L-proline, were also covalently bound on polysulfone matrix to fabricate enantioselective membranes, with α value of 1.1 toward *S*-Prp at 96 h.[118]

Because diffusion-enantioselective solid membranes selectively transport one of the enantiomers based on facilitated transport, the maximal selectivity is always obtained at the initial stage of the permeation, at a lower concentration of the feed solution, and using a membrane with a smaller pore size. The enantioselective permeation of diffusion-enantioselective membranes is always low because a concentration gradient is frequently used as the driving force. The main disadvantage of this kind of membrane is the reverse relationship between the selectivity and the flux, which substantially limits the industrial-scale application of diffusion-enantioselective membranes.

3.2.2 Adsorption-enantioselective solid membranes. Unlike diffusion-enantioselective membranes, adsorption-enantioselective membranes allow simultaneous enhancement of the flux and selectivity. These adsorption-enantioselective membranes, however, have some shortcomings too. For example, their selectivity becomes 1.0 after the chiral sites become saturated. This can be resolved to some extent by adjusting the driving force, increasing the chiral selector concentration in the membrane, or by using a so-called swing adsorption approach.

3.2.2.1 Enantioselective membranes made by addition of chiral selectors. BSA has a high binding affinity for L-form enantiomers. To separate enantiomers from a mixture of D- and L-Trp, Saito *et al.*[70,71,119–121] prepared porous hollow-fiber membranes containing multiple layers of adsorbed BSA
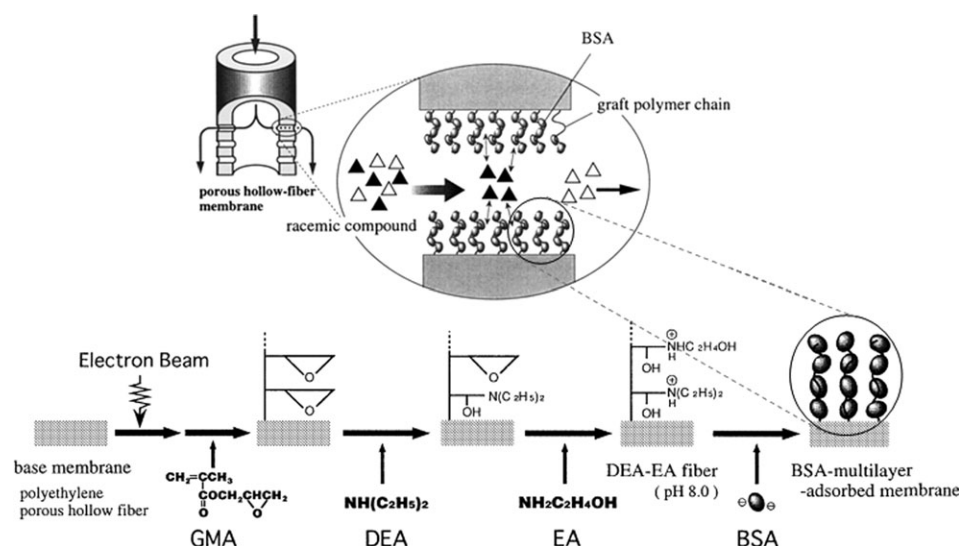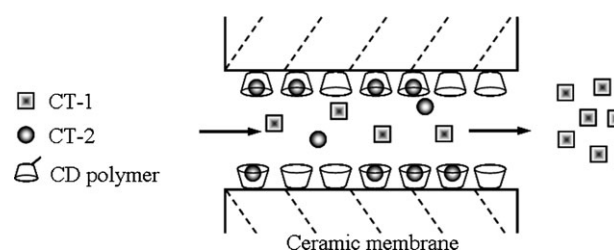
Fig. 6 Schematic illustration of the preparation steps and principle of chiral separation by a porous BSA-multilayered hollow fiber membrane. The ▲ and △ symbols represent the L- and D-form enantiomers, respectively. (Reproduced with permission from ref. 119. © 1999 Elsevier.)

(Fig. 6). The membranes were fabricated by first reacting the epoxy groups of grafted poly(glycidyl methacrylate) chains with diethylamino groups, after which the unreacted epoxy groups were neutralized with 2-hydroxyethylamine to reduce nonselective adsorption of BSA. Because the resulting grafted diethylamino-2-hydroxyethylamino chains are positively charged, they fully extended to the interior of the pores of the hollow-fiber membranes due to electrostatic repulsion. Thus, negatively charged BSA was adsorbed in the pores when the membrane was permeated with a BSA-containing buffer. The number of layers of BSA ranged from 3 to 11 and increased with the concentration of diethylamino groups. Use of a multilayered structure increased the number of binding sites for BSA and thus led to a higher separation factor compared with monolayer-adsorbed membranes (e.g. α_t = 6.6 and 2.9 for four-layer- and monolayer-adsorbed membranes, respectively). The BSA-multilayered membranes were also capable of rapid enantiomeric separation. To avoid loss of BSA from the grafted chains, BSA was crosslinked with glutaraldehyde. Porous hollow fiber membranes with three layers of crosslinked immobilized BSA gave a high separation factor (e.g. α_t = 12 for L-Trp).

Because proteins contain a wide variety of functional groups and binding sites, nonspecific binding is inevitable when they are used as chiral selectors. Therefore, other macromolecules such as cyclodextrins may be more suitable for the resolution of chiral compounds. For example, a β-CD polymer-impregnated ceramic membrane was reported for the stereoselective separation of racemic chlorthalidone (Fig. 7).[122] Although some of the β-CD was lost during concentration-driven permeation, an average α value of 1.24 was obtained for racemic chlorthalidone. As mentioned above, the saturation of the chiral recognition sites in adsorption-enantioselective membranes can eliminate the enantioselectivity. To solve this problem, a concentration swing adsorption approach was developed by Krieg et al.,[122] which allows continuous processing and simultaneously producing both optically pure enantiomers. In this method, two membranes are required for

alternate use. One is replaced by the other when adsorption sites of the former membrane are saturated with excess chlorthalidone-2 and the stripping phase contains excess chlorthalidone-1. The former membrane is then washed to release the adsorbed enantiomers (most of which are chlorthalidone-2) from β-CD polymer. When the latter membrane reaches the adsorption saturation, it is substituted by the regenerated membrane and then washed. Thus, the enantioseparation process can be operated continuously by repetitively exchanging the two membranes. β-CDs were usually covalently bound on the membrane substrates to separate D/L-Trp. A β-CD immobilized cellulose membrane with MWCO of 1000 exhibited an α value of 1.10 toward D-Trp.[123]

Recently, a novel thermo-responsive enantioseparation membrane was developed by grafting both poly(N-isopropylacrylamide) (PNIPAM) and β-CD onto Nylon-6 porous membrane substrates in the authors' group.[124] The ee'' value of the thermo-responsive membrane for enantioseparation of D/L-Trp at a temperature lower than the lower critical solution temperature (LCST, around 32 °C) of PNIPAM was larger than that of membranes without thermo-sensitivity. On the other hand, the decomplexation ratio of the thermo-responsive membrane after D/L-Trp resolution at a temperature above the LCST was much higher than that of membranes without thermo-sensitivity. Therefore, for this membrane,

Fig. 7 Retarded transport of racemic chlorthalidone (CT) across a β-CD polymer-impregnated ceramic membrane. (Reproduced with permission from ref. 122. © 2000 Elsevier.)
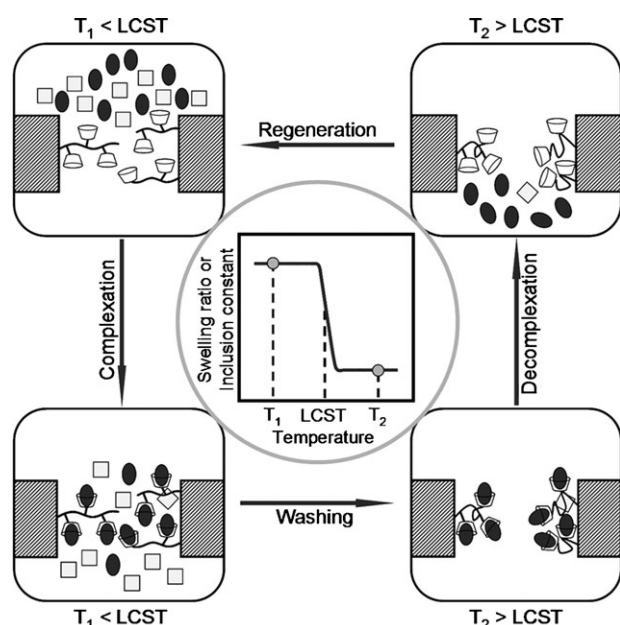
Fig. 8 Schematic illustration of the concept of thermo-responsive enantioseparation membrane and membrane process. One of two enantiomers can form a complex with β-CD molecules and be separated at temperature T_1, which is lower than the lower critical solution temperature (LCST) of poly(N-isopropylacrylamide); after the resolution, the enantiomer in the complex is separated and the membrane is regenerated at temperature T_2, which is higher than the LCST. (Reproduced with permission from ref. 124. © 2008 Wiley-VCH Verlag GmbH & Co. KGaA.)

enantioseparation with a high selectivity and membrane regeneration with a high decomplexation ratio were achieved by simply changing the operating temperature, as shown in Fig. 8. The maximal ee'' values of enantioselective membrane with grafting yield of PNIPAM being 107 mg cm^{-2} and that of β-CD being 23 mg cm^{-2} were 21% at 25 °C and 8% at 40 °C, respectively. For the membrane only immobilized β-CD with grafting yield of 23 mg cm^{-2} but without any grafted PNIPAM, the maximal ee'' value was about 16% at both 25 °C and 40 °C. During membrane regeneration, the decomplexation ratio of the former membrane in water was as high as about 75% at operation temperature of 40 °C in one wash, compared with 20% for the latter membrane with only immobilized β-CD but no grafted PNIPAM. Such an enantioseparation membrane could provide easy operation for the chiral resolution and decomplexation.

Chu *et al.*,[125] investigated the effect of β-CD content in chitosan/β-CD composite membranes with semi-interpenetrating networks on α value and permeate flux in enantioseparation of D/L-Trp. With the weight ratio of β-CD polymer to chitosan increased from 0 to 0.6, the α value decreased from 1.47 to 1.15; however, D-Trp flux increased from 0.0289 to 0.0388 mg cm^{-2} and meanwhile, L-Trp flux increased from 0.0197 to 0.0338 mg cm^{-2}.

Another type of membrane was fabricated by Tone *et al.*,[126,127] who grafted four kinds of terpenes on 0.2 μm pore cellulose acetate membranes by plasma polymerization. The resulting membranes were used to resolve optical isomers from racemic mixtures of Trp, Phe, and Tyr. The α values

decreased as the volume flux increased because, at higher operating pressures, both enantiomers permeated through the membrane without interacting with L-menthol. The maximal α value of L-menthol- and citronellol-grafted membranes was 8 and 9.5, respectively, for D-Trp in pressure-driven ultrafiltration experiments (ΔP = 0.02 to 0.3 MPa), and the volume flux was 1.5×10^{-8} and 4.7×10^{-8} m s^{-1}, respectively.

3.2.2.2 Molecular imprinting membranes. Molecularly imprinted membranes (MIMs) are fabricated by incorporating optically pure print or template molecules into the membranes and then extracting the template molecules to form voids that recognize both the template molecules and the family or analogue of the print molecules. When the racemic solution permeates through the membranes, the print molecules and their analogs are selectively adsorbed to the print sites and the other enantiomers are excluded. MIMs are considered as adsorption-enantioselective membranes due to the one-to-one intermolecular interactions between the chiral recognition sites and the enantiomers. There are some disadvantages of MIMs. For example, optically pure enantiomers for use as template molecules can be difficult to find, the enantiomers can only recognize the print molecules and their families, and the membranes typically have a low loading capacity and efficiency.[4] Therefore, intensive study is still needed before MIMs can be applied commercially.

Using a solvent evaporation method, Yoshikawa and co-workers[77,79–81,128] fabricated cellulose acetate MIMs and myrtenal-containing polysulfone MIMs with N-α-Z-D-Glu or N-α-Z-L-Glu as print molecules, and MIMs made from copolymers of acrylonitrile and styrene bearing tetrapeptide derivatives (*e.g.*, HGlu(OBzl)-Gln-Lys(4-Cl-Z)-Leu-CH$_2$–), tripeptide resins (*e.g.*, HGlu(OBzl)-Glu(OBzl)-Glu(OBzl)-CH$_2$–), or oligopeptide tweezers in presence of print molecules such as Boc-L-Trp, Acetyl-L-Trp or Boc-D-Trp. Membranes prepared from the L-amino acid-containing derivatives produced few chiral recognition sites when a D-isomer was used as the print molecule, but the MIMs containing oligopeptide tweezers (L-amino acid residues) successfully formed chiral recognition sites for the D-isomer.[80,81] The resulting membranes showed adsorption selectivity toward the print molecule and its analog (Fig. 9). The α' value of these membranes toward print molecules or their families ranged from 1.5 to 5.0 in enantioselective electrodialysis experiments, and the total flux increased as the applied potential difference increased.

Enantioselective chitosan MIMs crosslinked with γ-glycidoxypropyltrimethoxysilane and with L-Phe as the print molecule were prepared by a modified sol–gel process.[129] An α^P value of 4.5 was achieved during resolution of D/L-Phe. To obtain higher flux than homogenous membranes, Son *et al.*[130] synthesized composite MIMs by interfacial polymerization of piperazine and trimesoyl chloride on the surface of polysulfone ultrafiltration membranes, and with D-serine as the print molecule. D-Serine appeared to permeate faster than L-serine through the prepared membrane, and the maximal ee value of serine racemates was close to 80% when operating time reached 60 h.

To replace the costly practice of bulk polymerization followed by grinding, sieving and sedimenting during the
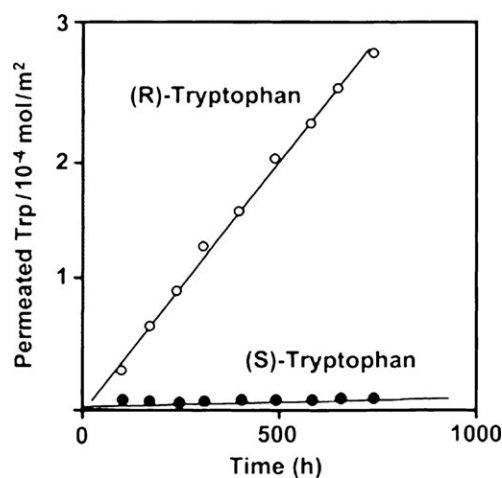
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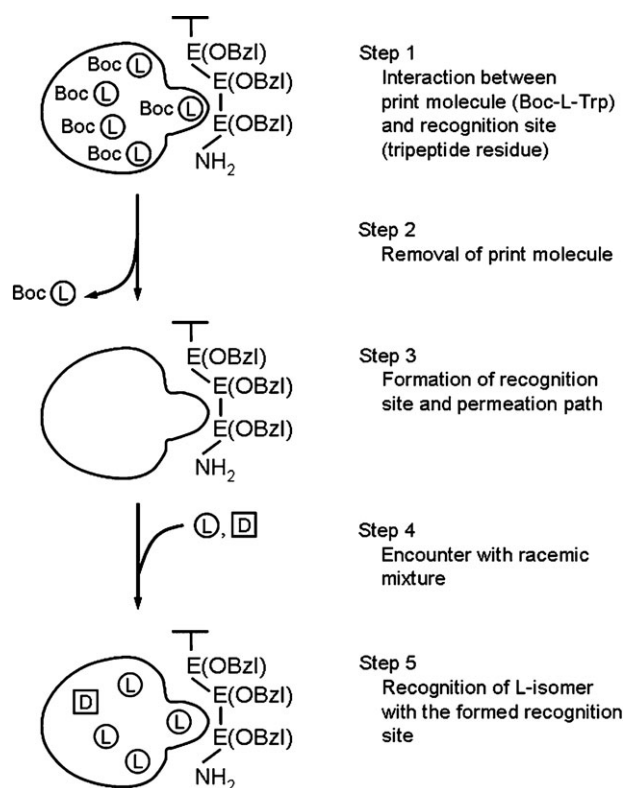
Fig. 9 The concept of alternative molecular imprinting. (Reproduced with permission from ref. 77. © 2001 Elsevier.)


Fig. 10 The time dependence of D/L-Trp permeation from a racemic mixture at 34 °C. (Reprinted with permission from ref. 132. © 1990 American Chemical Society.)

generation of molecularly imprinted polymers, a faster and more efficient technique for screening combinatorial libraries of molecularly imprinted polymers was recently developed.[131] MIM solutions were cast on polytetrafluoroethylene membranes in microfiltration modules, and the membranes were then evaluated in terms of affinity towards the target molecule (template), R-(−)-phenylbutyric acid. This technique accelerates the process of identifying optimal concentrations of crosslinkers and porogens when screening different molecularly imprinted polymers. Further development of this technique is necessary to obtain quantitative screening and higher reproducibility.

Although the enantioselectivity of MIMs is currently relatively low and the α value does not yet exceed 5, higher selectivity with this method will be possible if its limitations such as low loading capacity and low efficiency are improved.

3.2.2.3 Enantioselective membranes made from chiral polymers. In the 1990s, Maruyama et al.[132] reported the complete enantioseparation of a mixture of D- and L-Trp using membranes made of a poly-(L-glutamate) derivative with (n-nonylphenoxy)-oligo(oxyethylene) side-chains (Fig. 10). Similarly, enantioselective membranes have been fabricated by the solution casting method with polyamino acid derivatives prepared by transesterifying poly(γ-methyl-L-glutamate).[78,87] During pressure-driven permeation (ΔP = 0.1 or 0.2 MPa), these membranes showed a maximal ee″ value of 20% toward D-Trp at the beginning stage, which decreased as the enantioselective recognition sites saturated. When the driving force was changed to an electrical potential difference (e.g., ΔE = 4 V), however, a constant enantioselectivity value was obtained (α^P = 3.0) for the separation of N-acetyl-D-Trp from a racemic mixture of N-acetyl-D/L-Trp using the poly(γ-methyl-L-glutamate) membrane. The permeation rate of acetyl-Trp in electrodialysis was higher than that of Trp in pressure-driven permeation. However, electrodialysis is ineffective when the net charge of the analyte is zero, such as in Trp. For freestanding poly{γ-[3-(pentamethyldisiloxanyl)propyl]-L-glutamate} membranes,[87] the ee″ value was 16% toward L-Trp, and the permeation rate was high (10^{-6} g m^{-1} h^{-1}) at a pressure difference of 1.0 MPa. The enantioselective permeation was stable for more than 160 h. These results showed that short siloxane side chains are required for the asymmetric centers in the backbone to recognize enantioselectively the permeating solutes.

Lee and Frank[104] modified polyvinylidene difluoride membranes by vapor deposition and then used them to fabricate poly(L-Glu) (PLGA)-modified membranes and polyglutamates membranes with triethylene glycol monomethyl ether side chains (PLTEG) by debenzylation or ester exchange reactions, respectively (Fig. 11). The α^P values of the PLGA-modified membranes for chiral α-amino acids (e.g., Trp, Phe, and Tyr) and chiral drugs (e.g., Prp, atenolol, and ibuprofen) ranged from 1.04 to 1.47 for the L-isomers. When the solvent contained higher ethanol concentrations and higher acidity, the PLGA chains changed into α-helical conformations, resulting in a higher selectivity of permeation. Experimental results indicated that chemisorbed PLGA-modified membranes showed 5% to 23% higher enantioselectivity than physisorbed membranes, which is due to the enhanced interaction between the chiral compounds and the surface-bound polypeptides. On the other hand, PLTEG-modified membranes allowed preferential permeation of D-Trp with α^P value of 1.29.

Adsorption-enantioselective membranes usually have stronger one-to-one interactions between the chiral recognition sites and enantiomers and often employ a pressure or electrical potential difference as the driving force. Adsorption-enantioselective membrane technology may be the most promising approach for

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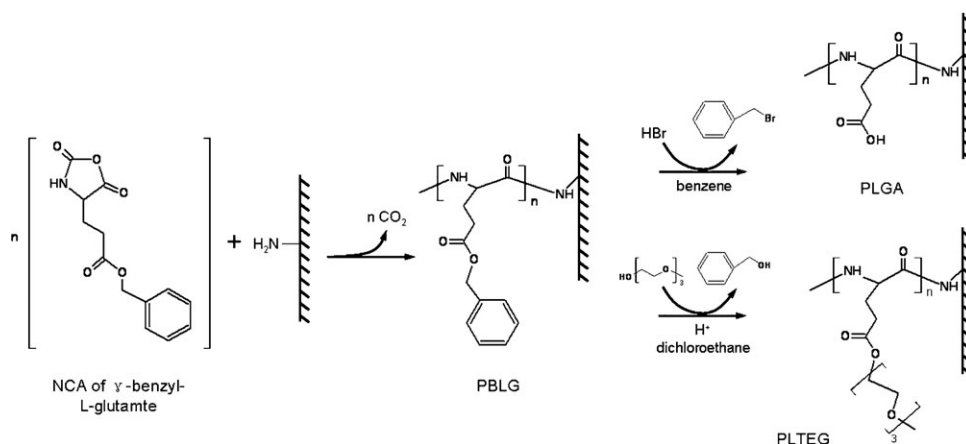
Fig. 11 Steps for preparation of PLGA and PLTEG membranes by debenzylation or ester exchange reaction from poly(γ-benzyl-L-glutamate). (Reproduced with permission from ref. 104. © 2002 Elsevier.)

achieving high flux, selectivity, efficiency, and stability, as well as for application in industrial-scale chiral resolution.

4. Non-enantioselective membranes in chiral resolution

Non-enantioselective membranes combined with other chiral recognition techniques, referred to as membrane-assisted resolution techniques, are an alternative method of carrying out enantioseparations. In this technique, non-enantioselective membranes served as barriers or filtration mediums to selectively separate optically pure enantiomers from a solution containing a racemic mixture and stereoselective binding agents. The non-enantioselective membranes can also be combined with enzymatic reactions or act as supports for functional layers. Importantly, this technique is economical and convenient. Due to the intrinsic advantages of membranes, such as energy-saving and easy scale-up, this technique is suitable for industrial-scale production of optically pure compounds; therefore, it has attracted a great deal of attention. As with enantioselective membranes, there are two main classes of non-enantioselective membranes applied to enantioseparation including liquid and solid formats. The combinatorial approach used with these membranes includes systems with and without chemical reaction.

### 4.1 Membrane systems without chemical reaction

Investigations on combinatorial chiral resolution with membrane systems in the absence of chemical reactions are conducted in solution systems. In a solution system, chiral selectors selectively bind one of the enantiomers to form the diastereoisomer complex, which usually has a higher molecular weight and therefore can be retained by a membrane with a suitable MWCO. The free enantiomers, which have a lower molecular weight, pass through the membrane. The complexed molecules are then released by changing the operating parameters.[133,134] Thus, two reasonably pure enantiomers can be obtained separately. The key to performing chiral resolution in such a system is choosing a chiral selector and membrane with binding abilities and appropriate sizes. In other words, the chiral selector should easily and selectively form a complex with one enantiomer and

the chiral selector and chiral selector–enantiomer complex should be big enough to be retained by the porous membrane.

Racemic mixtures of ibuprofen, Trp, and a kynurenine (a Trp analog) have been separated using BSA as the free complex agent in a solution system.[86,133–135] A simulation indicated that the zwitterionic D- and L-isomers compete for a single unprotonated BSA when the pH is between 7 and 11, which agrees well with theoretical calculations.[86,133,135] Purification of the individual enantiomers depended on their initial concentrations ($[D]_o$ and $[L]_o$), the initial concentration of BSA ($[BSA]_o$) and pH of the solution. The purity and recovery of the D- and L-forms, respectively, reached 80% and 75% for Trp and 85% and 80% for kynurenine at $[BSA]_o = 1.5 \times 10^{-4}$ M, $[D]_o = [L]_o = 1 \times 10^{-4}$ M, and pH 9.5 using polysulfone membranes (MWCO = 10 000 Da) in the filtration experiments.[86,133] A two-stage diafiltration system using tangential flow through microfiltration membranes (Fig. 12) achieved a yield greater than 90%.[135] In the enantioseparation experiments of racemic ibuprofen, both selectivity and solute binding increased as the BSA content in the feed solution increased, and the maximal enantiomeric excess was obtained between pH 9.0 and 9.2.[134] Inclusion of organic solvents such as acetonitrile and methanol at a concentration of less than 15 vol% in the feed solution suppressed nonspecific interactions, and after six stages of separations, the permeate solution contained more than 95% of S-ibuprofen.

The optical resolution of a racemic mixture of Phe was investigated in a DNA solution system using ultrafiltration through polyacrylonitrile hollow fiber membranes with a MWCO of 13 000.[112,113] The DNA preferentially bound to L-Phe so that only the D-form could pass through the membrane. When the concentration of DNA was low (0.01–0.5 ppm), the α value was near 1.0 but fluctuated due to conformational changes depending on the time of permeation.

A multi-stage countercurrent membrane process was developed for enantioseparation of racemic Trp in a solution system (Fig. 13)[136]. In this system, one enantiomer is bound by α-CD, and it is transported with the liquid flow. The free enantiomers pass through the membranes in the opposite direction (to the left side) due to electrodialysis. Fig. 14 shows that, when the selectivity of the chiral selector is fixed, the ee value increases
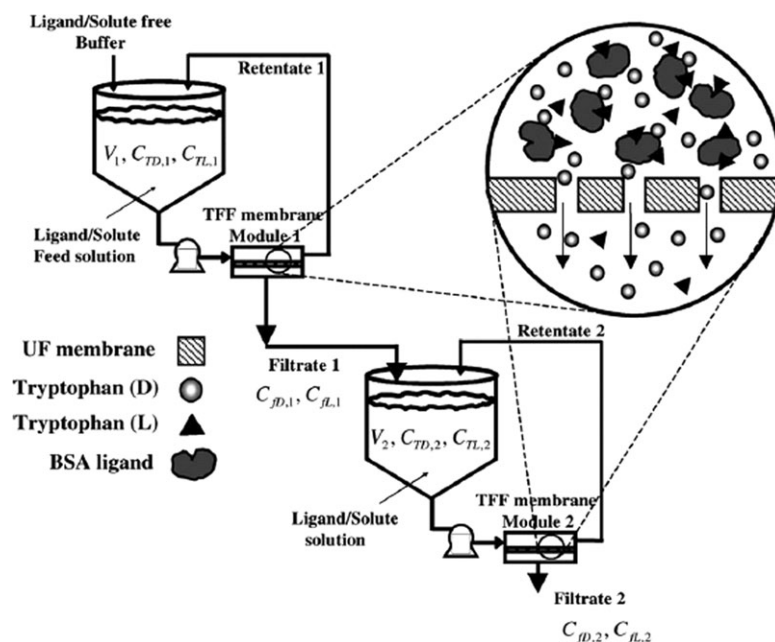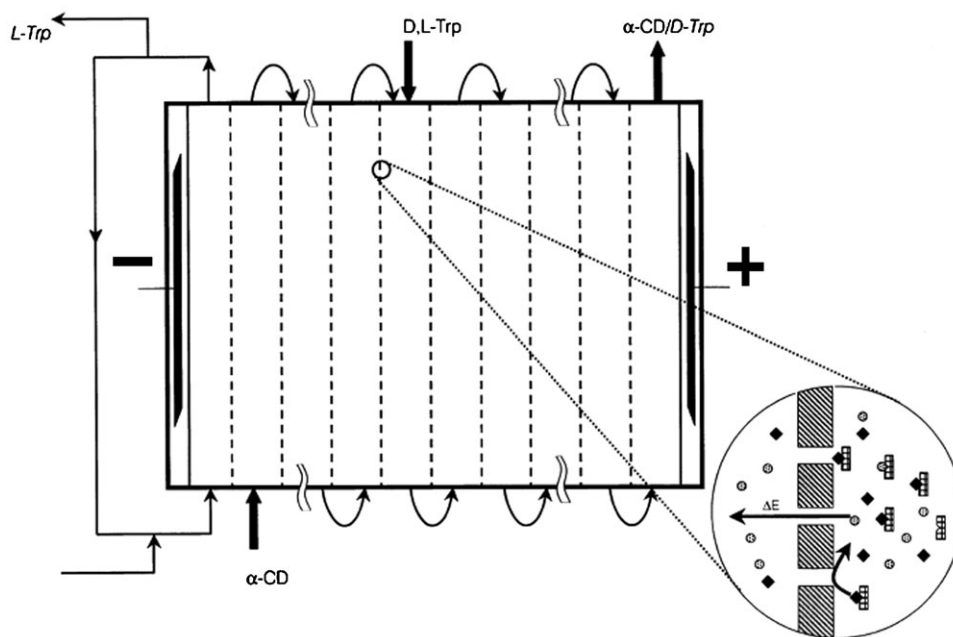
Fig. 12 Schematic illustration of the two-stage solution system containing BSA as a complexing agent with tangential flow filtration. (Reproduced with permission from ref. 135. © 2002 Elsevier.)

as the number of membrane compartments increases. Although the α value in this system for D-Trp was low (1.12), the *ee* value exceeded 99% when it included 250 membrane compartments.

Like BSA and DNA, micelles containing chiral selectors can be used as chiral complexing agents. De Bruin *et al.*[137] separated racemic α-amino acids such as Phe, phenylglycine, *O*-methyltyrosine, isoleucine, and leucine from dilute solutions using cholesteryl L-glutamate Cu(II) as a chiral selector in a novel micelle-enhanced ultrafiltration process. The maximal

α_{op} value was 14.5 for D-phenylglycine. The performance of this system was determined by the hydrophobicity of the racemic amino acid and the stability of the complex formed by the amino acid enantiomer and the chiral selector.

This combinatorial system can include not only non-enantioselective solid membranes but also non-enantioselective liquid membranes. Keurentjes *et al.*[75] used countercurrent fractionation and ILM technology to develop a combinatorial process for separating racemic mixtures that can be easily scaled up or down

Fig. 13 Schematic illustration of a multi-stage counter-current process in a solution system containing α-CD for enantioseparation of a racemic Trp mixture by electrodialysis. (Reproduced with permission from ref. 136. © 2002 Elsevier.)
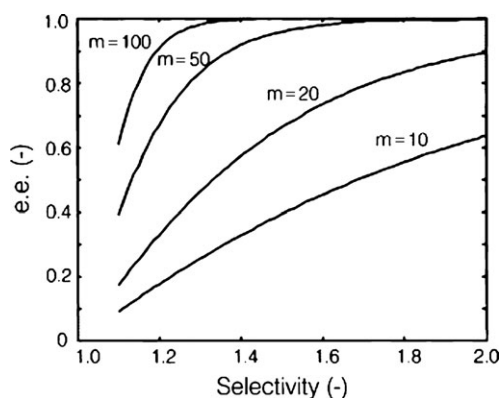
Fig. 14 Estimated effect of the selectivity of the chiral selector and the number of membrane compartments (*m*) on the *ee* value. The feed flow concentration is 20 mM. (Reproduced with permission from ref. 136. © 2002 Elsevier.)

(Fig. 15). Unlike other enantioselective liquid-membrane processes, the chiral selectors were dissolved in the feed phase (organic phase) rather than contained in the membrane phase (aqueous phase), and they could not pass through the aqueous liquid membrane. The liquid membrane was immobilized in the pores of a hollow-fiber membrane separating two miscible organic phases containing chiral selectors for either the *R*- or *S*-enantiomer, respectively, and it was permeable only to the enantiomers. A bench-scale model of this system achieved recoveries of 99% or higher for both enantiomers.

Another kind of non-enantioselective membrane has been used as a support in chiral-resolution systems without chemical reactions. Brunner and coworkers[138,139] developed molecularly imprinted nanospheres of poly[(methylacrylic acid)-*co*-(ethylene glycol dimethacrylate)] synthesized by mini-emulsion polymerization with D- or L-boc-Phe anilide (BFA) as the molecular

template. These imprinted nanospheres provided a large surface area because of their small size. Binding experiments showed that at 20 μmol total BFA, 10-fold more L-BFA than D-BFA was adsorbed by L-isomer-imprinted particles. The difference of binding L-enantiomer was 4-fold in the L-imprinted polymer than in the nonimprinted polymer. A composite membrane was generated by depositing the nanoparticles between two membranes.[138] In this case, although the dense particle layer led to a low flow rate, it helped to establish a chemical equilibrium due to the relatively slow selective binding.

4.2 Membrane systems with chemical reaction

In general, combinatorial systems involving chemical reactions can be created by combining non-enantioselective membranes with catalyzed kinetic resolution. When the catalyst is an enzyme, the system is also referred to as an enzyme membrane reactor (EMR). In a continuous process, a biocatalyst is always required for both high efficiency and low cost. The enzyme can be either immobilized on the membranes or dissolved in the feed solution. Lipases, such as those from *Pseudomonas* sp., *Pseudomonas cepacia*, and *Candida rugosa*, are commonly used. Immobilization of these enzymes can enhance their stability in these systems.[140]

Koter and Ceynowa[141] reported bifunctional membranes prepared by filling the pores of polyamide membranes with imprinted polymers and covalently immobilizing an enzyme layer on its surface. The mechanism of the overall separation of the enantiomers within the bifunctional membrane is shown schematically in Fig. 16. The layer of the imprinted polymer facilitated the transport of the template (1*R*,2*S*)-(−)-*trans*-2-phenyl-1-cyclohexanol) and related analogs. The enantiomeric enrichment was catalyzed by a lipase, yielding a high *ee* value (98.5%) for the 1*R*,2*S*-ester.
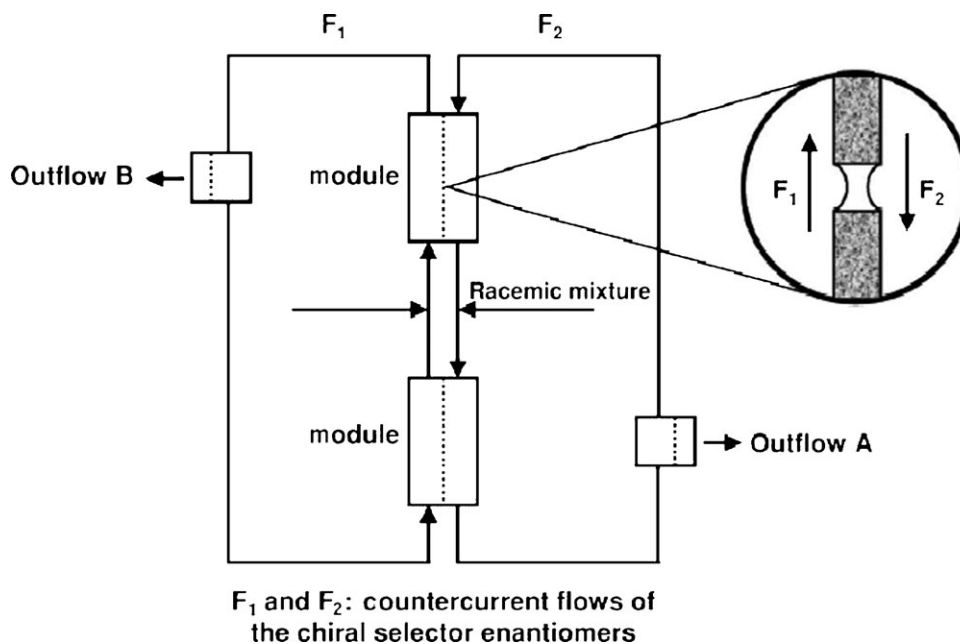
Fig. 15 Schematic illustration of the combinatorial enantioseparation process based on a countercurrent fractionation and ILM technology. (Reproduced with permission from ref. 4 and 75. © 1996 and 2001 Elsevier.)
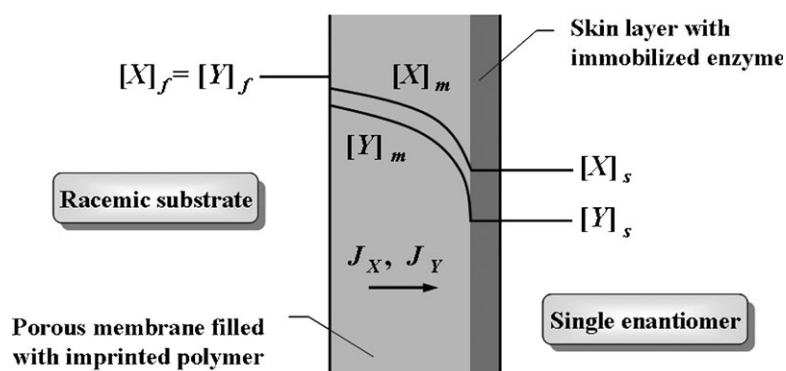
Fig. 16 Schematic illustration of concentration profiles of racemates occurring during the transport through an enantioselective bifunctional membrane. [X] and [Y] are the concentrations of enantiomers X and Y, respectively; the subscripts s and f denote the stripping and feed phases, respectively. J is normalized flux. (Reproduced with permission from ref. 141. © 2003 Elsevier.)

Sakaki and coworkers[142] applied a lipase-immobilized capillary EMR for the optical resolution of a racemic mixture of 2-hydroxyoctanoic acid. The S-isomers of the methyl and butyl esters of 2-hydroxyoctanoic acid were preferentially hydrolyzed by *Pseudomonas cepacia* and *Candida rugosa* lipases, respectively. For example, a biphasic membrane reactor with immobilized *Pseudomonas cepacia* lipase, gave *ee* values of 95% and 99%, respectively, for the R- and S-isomers of 2-hydroxyoctanoic acid.

Another capillary EMR was used to kinetically resolve a racemic mixture of 2-(2-fluoro-4-biphenyl)propanoic and (4-isobutylphenyl)propanoic acids as well as their esters.[140] The EMR was prepared by immobilizing lipase from *Pseudomonas* sp. on the surface layer of an asymmetric polyamide capillary membrane. The catalytic activity of the immobilized enzyme was 27% of the native lipase and was stable for at least 1 month. The best resolution of the acid racemates and an *ee* value exceeding 99% was achieved by esterification followed by hydrolysis of the formed esters. Despite these promising results, a convenient method for separating the esterified product is still needed for this system. Wang *et al.*[143] compared the enantioselectivity, activity and half-life of immobilized enzyme in a biphasic EMR system with those of a free lipase system during enantioseparation of the racemic ibuprofen. Better *ee* values (up to 83.5%), 373% longer half-life, and 60% more activity than in the free lipase system were obtained in the immobilized lipase biphasic EMR system.

On the other hand, compared with enzyme-immobilized EMRs, ultrafiltration systems containing free lipase do have several advantages, including easy addition of fresh lipase, homogenous distribution of catalysts, essentially native enzyme activity, easy setup, and low cost.[144] The mechanism of ultrafiltration separation systems containing free lipase is similar to that in the solution systems without chemical reactions. The enzyme selectively catalyzes one of the two enantiomers, and the reaction product permeates through the non-enantioselective membrane, while both the unreacted enantiomer and enzyme are retained by the membrane because they are too large to pass through the membrane pore.

An ultrafiltration EMR system employing stereospecific hydrolysis by L-aminoacylase was reported for the separation of D- and L-butyrine from *N*-acetyl-D/L-butyrine.[144] Also,

Liese and coworkers[145] described the kinetic resolution of ethyl R/S-2-hydroxy-4-phenylbutyrate using *Pseudomonas cepacia* lipase in conjunction with membrane separation, which is commercially useful because the R-isomer is an important intermediate in the synthesis of several angiotensin converting enzyme inhibitors. The kinetic resolution of this system could be enhanced by continuously removing the product, and enzyme consumption could be reduced by performing the separation in repetitive batch mode. The *ee* value for this system was more than 99.5% with a yield of 275 g L^{-1} d^{-1}. Another system was reported by Long and coworkers,[146] who simulated the enzymatic hydrolysis of racemic ibuprofen ester by a lipase from *Candida rugosa* in a hollow-fiber EMR system. The EMR was reported that has been successfully applied to the commercial preparation of over 75 metric tons of optically pure diltiazem intermediates per year.[13]

Similar to the combinatorial resolution technique in solution systems without chemical reactions, liquid membranes have occasionally been used in systems involving chemical reactions. Goto and coworkers[147,148] developed a novel SLM in which a surfactant–enzyme complex effectively catalyzing esterification is encapsulated in the organic liquid membrane phase. This system, illustrated in Fig. 17, allowed efficient and highly enantioselective separation of S-ibuprofen and L-Phe from their racemic mixtures. After 48 h, the maximum *ee* value for L-Phe exceeded 99% in α-chymotrypsin-facilitated SLM, whereas the *ee* value was 91% for S-ibuprofen when a surfactant–lipase complex was used in the SLM. The maximum permeate fluxes for these two systems were 0.18 and 0.58 mol m^{-2} h^{-1}, respectively. Thus, use of the appropriate enzymes in this SLM system can allow the enantioseparation of various organic compounds.

Recently, Oshima *et al.*[99] developed a BLM system that successfully carried out the enantioselective hydrolysis of hydrophobic amino acid esters. This system included an enzymatic reaction combined with transport of the unhydrolyzed D-form ester in a BLM system containing calix[6]arene hexacarboxylic acid as the mobile chiral carrier. In the enzyme reaction stage, the L-form ester was selectively hydrolyzed to the free amino acid by a surfactant-α-chymotrypsin complex. As a result, the L-amino acid remained in the feed phase, while the unhydrolyzed D-form ester was preferentially transferred by calix[6]arene
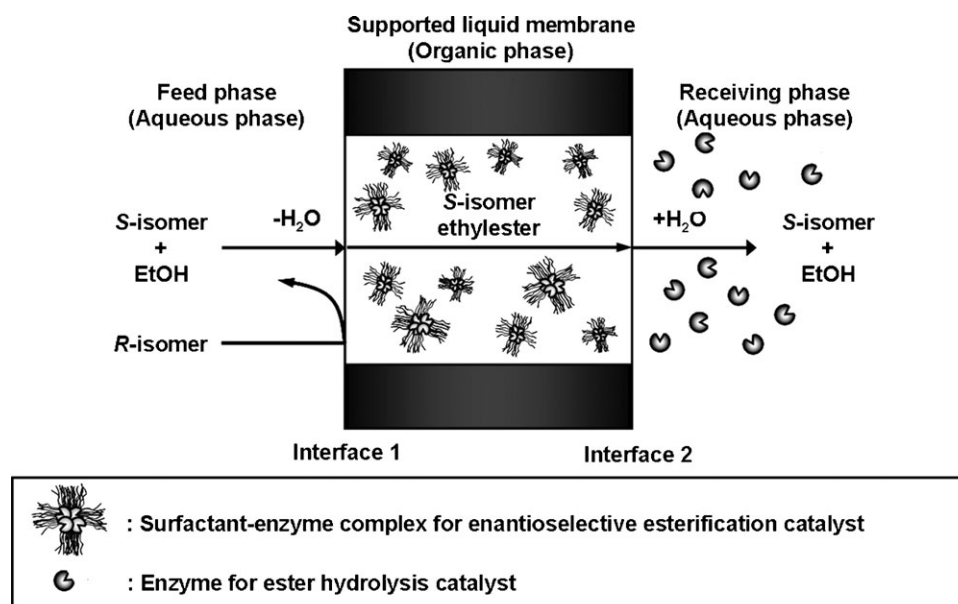
Fig. 17 Schematic illustration of the resolution process through an EMR based on a SLM. (Reprinted with permission from ref. 147. © 2004 American Chemical Society.)

hexacarboxylic acid. The *ee* value after 24 h of operation reached 86.6% for the D-form in the receiving phase.

A membrane-assisted technique can combine the advantages of each combinatorial technique given the appropriate design and operating conditions. These systems can achieve reasonable selectivity by using multi-stage processes and are expected to be feasible for large-scale chiral separations.

5. Summary and outlook

The large-scale production of optically pure substances is urgently needed for scientific studies and industrial preparations of pharmaceuticals, agrochemicals, fragrances, and foods. Current resolution techniques, including crystallization, kinetic resolution, and chromatographic separation can be expensive or inefficient, which can hinder their industrial application, excepting that supercritical fluid chromatography and simulated moving bed chromatography recently have presented promising potentials. Membrane-based enantioseparation techniques have attracted a great deal of attention for industrial application because they are easy to scale-up, save energy, can be continuously operated, and are efficient. For example, EMR has been successfully applied to the large-scale production (over 75 metric tons per year) of optically pure diltiazem intermediates.[13] Thus, membrane-based resolution has a great potential for fulfilling industrial needs.

Enantioselective liquid membranes are usually inexpensive, highly selective, and capable of rapid mass transfer. Enantioselective solid membranes, however, are more stable and stronger than enantioselective liquid membranes. Among the enantioselective liquid membranes, ILMs are the most intriguing because they combine both the high selectivity and rapid mass transfer of liquid membranes with the stability and mechanical strength of solid membranes, and they are expected to be useful for the resolution of racemates with relatively high selectivity. Among the enantioselective solid membranes, adsorption-type enantio-

selective membranes may provide both high flux and high selectivity. Although the permeability and selectivity in previous reports are probably not high enough for large-scale application because of relatively low binding capacities and relatively small difference of binding affinity between chiral selectors and two enantiomers, adsorption-type enantioselective membranes may be one of the most promising approaches for industrial-scale production of optically pure compounds.

Membrane-assisted resolution systems with non-enantioselective membranes can combine the advantages of each of these techniques in a combinatorial system. This technique has recently attracted a great deal of attention, especially for EMR and chiral selector-containing solution resolution systems. Together, experimental results, theoretical calculations, and model simulations indicate that multi-stage membrane-assisted resolution processes can achieve both high optical purity and high yield.

In summary, because of the intrinsic features of membrane technology, membrane-based chiral resolution is a promising method for the preparation of single-enantiomer materials. The technique can achieve relatively high selectivity and high yield by employing multi-stage processes, even when the individual stages do not.

Abbreviations

A	effective membrane area
A_X, A_Y	the peak areas of the X and Y enantiomers
BFA	boc-Phe anilide
BLM	bulk liquid membrane
BSA	bovine serum albumin
c	concentration fraction
C_f	concentration of the feed phase
C_l	concentration in the bulk liquid
C_m	equilibrium membrane concentration
C_s	concentration of the stripping phase

(continued)

ΔC	concentration difference
D	diffusion coefficient
E	extraction efficiency
ΔE	electrical potential difference
ee, ee', ee''	enantiomeric excess
ELM	emulsion liquid membrane
EMR	enzyme membrane reactor
f	the feed phase
Glu	glutamic acid
ILM	immobilized liquid membrane
J	normalized flux
k'	retention factor of enantiomers
MIMs	molecularly imprinted membranes
MWCO	molecular weight cut-off
P	permeation
P'	permeability or permeation coefficient
P_u	purity
ΔP	transmembrane pressure difference
PDPSN	poly{2-[dimethyl-(10-pinanyl)silyl]-norbornadiene}
PDPSP	poly{l-[dimethyl(l0-pinanyl)silyl]-l-propyne}
Phe	phenylalanine
PLGA	poly(L-glutamate)
PLTEG	poly(glutamate) with triethylene glycol monomethyl ether side chains
Prp	propranolol
R	recovery
s	the stripping phase
S	sorption coefficient
SLM	supported liquid membranes
t	retention time of the enantiomers
t_0	void time
Δt	permeation time
Trp	tryptophan
Tyr	tyrosine
V	downstream volume
X, $_X$	enantiomer preferentially transported through the membrane
x	membrane thickness
Y, $_Y$	enantiomer retained in the feed solution
$\alpha, \alpha_c, \alpha_t, \alpha'$	separation factor
α^D	diffusion selectivity
α^S	sorption selectivity
α^P	enantioselectivity
α_{op}	operational enantioselectivity
α-CD	α-cyclodextrin
β-CD	β-cyclodextrin

Acknowledgements

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