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Review

Surface engineerings of polyacrylonitrile-based asymmetric membranes towards biomedical applications: An overview

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Abstract

Polyacrylonitrile (PAN) membranes have attracted much attention due to a variety of excellent characteristics, which include thermal stability and tolerance to most solvents, atmosphere, bacteria and photo irradiation. Therefore, their exploitation also covers many fields, like pervaporation, water treatment, enzyme immobilization, and hemodialysis. However, the relative poor hydrophilicity and biocompatibility get the membranes susceptible to protein adsorption and cell adhesion that may cause biofouling and immunoreactions, especially when the membranes are used in blood-related systems. This review presents various surface engineerings for PAN-based membranes, which mainly refers to grafting polymerization (also known as "grafting-from" method), partially hydrolysis, macromolecule immobilization (also known as "grafting-to" method), and enzyme immobilization. To render the membrane surface with biocompatibility for hemodialysis, the concept of mimicking the outer surface of cells is critical. For example, biomacromolecules such as chitosan, heparin or insulin can be tethered on the PAN-based membranes to improve their surface hemocompatibility remarkably. In addition, one of the potential applications of PAN-based membranes is as enzyme immobilization supports. Over 15 relative literatures are listed and briefly discussed with different enzymes, modified supports, and coupling methods. However, the application trial for these enzyme-immobilized membranes is still elementary.

Keywords: Polyacrylonitrile membrane; Surface engineering; Grafting polymerization; Biomedical application; Enzyme immobilization

Contents

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1. Introduction

As a simple, energy-saving and powerful tool for separation and purification processes, membrane technologies have arrested growing interests of industrial managers since the last three decades. It is well known that various membranes are the centers of these technologies. Polyacrylonitrile-based ultrafiltration (UF) membranes constitute a family of such porous products. They obviously turn up to be more advantageous over other conventional membranes in various aspects, such as thermal stability, resistance to most organic solvents, commercial availability, etc. The family has attracted much attention in the biomedical fields like protein filtration and biocatalysts immobilization. Especially, PAN hollow fiber membranes are already used in dialyzers that enable low (e.g. urea, creatinine, etc.) to middle molecule protein (e.g. β_2-microglobulin) removals and high-flux dialysis therapy [1]. Nevertheless, every coin has two sides, and it is also the same with PAN membranes herein. It is well known that in the development of ultrafiltration, membrane fouling has largely and often limited the use in practice. In the progress of filtration, solute molecules deposit on/in the membrane surfaces/pores. The permeate flux is then reduced dramatically, the process has to be stopped, and the fouled membrane must be regenerated or replaced. As pointed out by Nilsson [2], the fouling of UF membranes mainly involves three steps, including solute transfer onto the membrane surface, transfer into the membrane after it either adsorbs or passes through following a set of sorption–desorption events, and surface binding accompanied by structural rearrangement in the adsorbed state. In the case of protein as the solute, factors affecting protein adsorption include the properties of protein molecules such as size, shape, stability, charge and charge distribution; the solution properties such as protein concentration, pH, and ionic strength; and the physical nature of membrane such as its hydrophobicity, charge, and charge density. The interplay between the factors is complex; however, electrostatic, hydrophobic, and entropic effects are often regarded as the main driving forces for surface adsorption. PAN membranes have a relatively hydrophobic surface; therefore, the driving force for protein adsorption is generally attributed to hydrophobicity. This poor hydrophilicity also causes the requirement of anticoagulant injection during hemodialysis. A commonly applied way to adjust the chemical structure for improving the membrane performance is to render the surface hydrophilic, which can obviously decrease the interactions between the proteins and the surfaces. In addition, the hydrophilicity can also protect the membrane surfaces against cell adsorption, which will benefit the dialysis application.

When the membranes are used in the biological environment, hydrophilic/hydrophobic balance is usually emphasized. That means, when applied in vivo, the membrane surfaces must be compatible with the surroundings. Or else, especially in the field of dialysis, serious biological consequences such as thrombosis will happen, which results in the adverse effect of membrane transplantation. Therefore, in this aspect, it is difficult to achieve such request only by creating a hydrophilic surface. Thereafter, the concept of biocompatibility is introduced herein. It refers to any harmful effects induced by the contact of the blood with the dialysis membrane, as well as anaphylactoid reactions. An extended definition of biocompatibility also includes factors related to the patient's clinical conditions and factors associated with the specific supportive technique: i.e. hemodialysis, hemofiltration, etc. [3]. Hemocompatibility is an aspect of dialysis biocompatibility, which has been particularly studied in the context of extracorporeal circulation for cardiopulmonary surgery. How to make the membrane surfaces biologically compatible to the blood component is critical to the development of ultrafiltration membranes used in the hemodialysis. From this point of view, making a biocompatible membrane surface will not only aim to reduce the fouling, but also to prevent the occurrence of pathological changes.

As to the selection of hemodialyser membrane, the history has to be simply introduced. Since the 1970s, a cuprammonium regenerated cellulose (Cuprophan, CU) membrane, made from cotton fibers dissolved in an ammonia solution of cupric oxide, was developed as filtration membrane in hemodialysis. It is relatively porous to fluid and microsolutes but did not allow large molecules (albumin, vitamin B_{12}) to pass freely. It was characteristic of high transports rates, low protein adsorption and high wet strength. However, two shortcomings stood in the way of its commercial application. Firstly, the abundant glycan oligomer in the cellulose could activate the complement system [4,5] and secondly, CU produced a decrease in the immune functions in the blood, leaving the patient more susceptible to infection. Alternatively, membranes from synthetic polymers showed their advantages in these aspects. AN-69 membrane from poly(acrylonitrile-*co*-methallyl sulfonate) was produced by *Rhone Poulenc* in France for hemodialysis. It was more permeable, and coincided with first use of volume controllers to limit plasma water loss during dialysis. Moreover, characterized by complement activation, biocompatibility of AN-69 was improved; in other words, AN-69 were much less activating than cellulose [6]. The negatively charged sulfonate absorbed an intermediate in the complement chain, thus stopping the reaction. However, AN-69 could still adsorb the proteins in the blood and promote blood coagulation despite the presence of repulsion between its negatively charged surface and proteins (at pH above the isoelectric point). Therefore, hemodialysis using AN-69 also required anticoagulant, as other kinds of membranes, regardless of the hydrophilicity and hydrophobicity. Moreover, AN-69 could only be sterilized by radiation, during which small amount of acrylonitrile monomer were found. It was well known acrylonitrile harmed the cells. Today the most widely used membrane is polysulfone membrane for hemodialysis, because after incorporation of a trapped hydrophilic polymer (PVP), the membrane surface was made slightly wettable and showed high biofunctional characteristics and moreover biocompatibility and economical efficiency [7].

Nevertheless, PAN-based membrane still showed some superiority over polysulfone membrane. The former surface is relatively active and much easier to be modified, and the achieved effect can be maintained much more longer than the latter surface, because the incorporated PVP is more facile to leap out of the membrane. Therefore, considering the past and the perspectives of PAN membranes, an enormous number of researches

have been devoted to the surface engineerings of PAN membranes. Until now, how to improve the surface biocompatibility involves decreasing the roughness, increasing the hydrophilicity, charging, and fabricating microphase separation morphology [8]. The current focus with regard to rendering biocompatibility partially includes the direct reaction occurring on the PAN membrane surfaces followed by immobilizing biomacromolecules or grafting other hydrophilic polymers. Because of the absence of chemical reactive functional groups on the membrane surface, a surface activation process is needed to create reactive sites on it for further grafting or immobilization. Practically, one can generate reactive groups through UV irradiation, plasma treatment, metal ions activation and hydrolysis, etc. In addition, the active groups for immobilization can also be generated by copolymerization of acrylonitrile with other vinyl monomers, in which the amount of introduced groups seems to be more easily controlled. Therefore, considering that the membrane surfaces are mainly modified by grafting or immobilization, the following sections will be developed according to the two modifications.

2. Grafting on the PAN-based membrane surface

The grafting methods can be generally divided into "grafting-to" and "grafting-from" processes [9]. In the case of "grafting-to" method, polymer chains carrying reactive groups at the ends or sides are covalently coupled to the membrane surface. The "grafting-from" method utilizes the active species existing on the membrane surfaces to initiate the polymerization of monomer from the surface towards the outside bulk phase. This technique has a key advantage that the surface properties can be tailored rather flexibly by choosing different grafting monomers. Graft chains with a high density and exact localization can also be introduced easily and controllably. Compared with the physically coated polymer chains, the covalent attachment of polymer chains maintains a long-term stability.

2.1. Surface modifications by plasma treatment and plasma-initiated graft polymerization

Plasma, which is sometimes referred to as the fourth state of matter, generally consists of negatively charged electrons, positively charged ions, and neutral atoms or molecules or both [10]. It contains equal numbers of ions and electrons in a sufficient density so that its Debye shielding length is much smaller than the dimension of medium. Plasma can be typically obtained when gases are excited into energetic states by radio frequency, microwave, or electrons from a hot filament discharge. It is a highly unusual and reactive chemical environment in which many surface reactions can take place. Modification can be rapidly and cleanly achieved due to the formation of various active species on the surface. In practice, plasma-assisted modification has been intensively studied since 1960s [11–14]. Especially in the biomedical field, plasma-assisted surface modification is gaining popularity and with regard to biomaterial engineering. Chu et al. [11] specifically pointed out the advantages offered by plasma-based techniques, which include the improvement of biocompatibility. Siow et al. [13] reviewed the plasma-assisted generation of reactive surfaces for biomacromolecule immobilization and cell colonization with good stability. Therefore, the plasma process and its effect on the polymer surface will not be described in detail and only the treatment of PAN membranes is discussed here.

Ulbricht et al. were the first to apply this technique to modify PAN ultrafiltration membranes. As the first example [15], they treated the membrane surface with low temperature helium or helium/water plasma followed by exposure to air. The results indicated that hydrophilicity for the membrane surface was significantly increased with only minor changes in permeability. Meanwhile, surface analysis showed high concentrations of peroxide species were created, which suggests the surface chemistry underwent fast and intensive changes after excitation and exposure. Usually, plasma treatment is performed on the membrane surface to generate active sites for subsequent grafting. This is why He and water were used here, because they are known to favor the formation of radical sites (i.e. peroxides) and functionalizations on the membrane surface. Under thermal condition, the graft polymerization of hydrophilic monomers such as 2-hydroxy-ethylmethacrylate (HEMA) and acrylic or methacrylic acid was initiated via the decomposition of peroxides [16]. As a result, both for the plasma-treated and HEMA-grafted membrane surfaces, protein fouling was strongly reduced due to less static protein adsorption, and higher permeation fluxes were enabled with the same protein retention. Similarly, Bryjak et al. [17] studied the effect of air plasma treatment on the microporous PAN membranes. They found the surface polarity was increased at low energy treatment.

To prepare hydrophilic PAN-based nanofiltration membranes, Zhao et al. [18,19] studied the grafting modifications of PAN ultrafiltration membranes induced by low temperature plasma. Herein, it is necessary to give a brief introduction of low-temperature plasma, which can be divided into (1) hot plasma, i.e. gas temperature normally of the order of 10^4 K, and (2) cold plasma, i.e. gas temperature normally of the order of 10^2 K. The success of cold plasma in surface treatment relies on its very high electronic temperature and relatively low gas temperature. The former affords a sputtering effect on the surface and the possibility of chemical modification, when the latter, being as low as room temperature, in most cases, enable the matrix to experience such plasma surface modification without loss of their mechanical properties. A hydrophilic monomer, acrylic acid, was introduced on the PAN membrane by argon (Ar) plasma treating and subsequent grafting reaction. It was found Ar plasma radiation did not break the C≡N bonds but caused the scission of C–H bonds, which would favor the next grafting. The surface pores were both affected by grafting and surface etching; the former resulted in smaller pore size due to the coverage of poly(acrylic acid) but the latter increased it. Considering the balance of surface hydrophilization and membrane permeability, a short graft reaction time was recommended. Similarly, *N*-vinylpyrrolidone in aqueous solution was also grafted on the PAN membrane under Ar plasma. At the low concentration range of monomer, the water permeation flux was not significantly affected.

2.2. Surface modification by photo-induced grafting

It is well known that photo-induced graft polymerization is a desirable method for the surface modification of polymers for a number of reasons. Firstly, photochemically produced triplet states of carbonyl compounds facilitate hydrogen abstraction, so graft polymerization is initiated without prior modification of a surface with conventional or living radical initiators. Secondly, a high concentration of active species is produced locally at the interface between the substrate polymer and the monomer solution. Thirdly, the procedure is relatively simple, energy-efficient, and cost-effective. Fourthly, photo-induced polymerization is well suited for integration with other technologies, such as microcontact printing and photolithography, to produce desired surface chemistry changes in well-defined two-dimensional regions on a surface. In practice, surface modification through the graft polymerization of a variety of monomers initiated by UV energy has been extensively studied. However, the amount of reports with regard to PAN-based membranes is limited and they were mainly carried out in the last century. Because this polymer membrane is intrinsically low-photoreactive, a photoinitiator, such as benzophenone (BP) or benzoin, must be applied (Fig. 1). In the instance of BP, it is either precoated on the membrane surface or present in the solution. When exposed to UV light, photons cause the excitation of BP to a short-lifetime singlet state from where it relaxes to a triplet state. At this point, BP can abstract hydrogen atoms from the polymer by inelastic collision, thereby creating free radicals on the surface which can serve as active sites for a wide range of monomers [20].

As critical examples, Ulbricht and his co-workers systematically explored photo-induced polymerizations on PAN ultrafiltration membranes. In 1995 [21], with the UV-initiated graft polymerization of acrylic acid from gas phase, they realized a heterogeneous surface modification of PAN membranes. The graft polymerization was surface-specific and took place in the upper 5-μm layer of the membrane when the photoinitiator, BP, was pre-coated on the membrane surface. Compared with other membrane materials (e.g. polysulfone), PAN exhibited an advantage of stable pore structure under UV excitation and graft reaction conditions, along with the photoactivation by BP. Due to the coverage of grafted polymer chains, the pore size and porosity were reduced, which decreased membrane permeability and increased solute retention. In the subsequent BP-related study [22], they modified PAN ultrafiltration membranes with either simultaneous or sequential UV irradiation graft polymerizations from aqueous monomer solutions. The former grafting was realized by pre-coating BP on the membrane surface and then polymerizing in monomer solution under UV light, while the latter by the photo-induced formation of radicals on the membrane surface and polymerizing in the solution. In the comparison, parallel homopolymerization induced in the solution could not be suppressed completely for the former case. Hydrophilic monomers used in their work mainly included acrylic acid, 2-hydroxyethyl methacrylate and various poly(ethylene glycol) methacrylates. For both approaches, with sufficient degree of modification, hydrophilicity was increased a lot and very little BSA adsorption was observed. Because of the diminished protein–membrane interactions (adsorption and subsequent aggregation via hydrophobic or charge interactions), protein/protein separations were feasible to apply with poly(ethylene glycol) mechacrylates modified membranes. Furthermore, with BP-assisted photopolymerization, modifiers with varied hydrophilicity and charge were attached on the PAN membrane surface and the modified surface could be tailored to permanently anionic or cationic as well as ionizable or non-ionic hydrophilic, flexible types, which was said to provide guide-lines for the development of new low-fouling UF and nanofiltration membranes [23]. Instead of BP, they also developed a two-step heterogeneous polymer surface modification involving photo-bromination as activation and subsequent UV-induced graft polymerization with acrylic monomers [24]. Even for relatively low degrees of modification, thin and smooth graft polymer layers were created which specifically altered the hydrophilicity of the membrane surface.

In a simple comparison, plasma modification can be applied to various membranes, whereas the substrate may suffer from severe etching. In the case of UV-induced graft polymerization, radicals cannot be generated on all membrane surfaces. It is selective to the matrix and causes much less damage to the membrane surface.

2.3. Surface modification by metal ions-initiated grafting

Polymerization in the presence of inorganic ions and complexes was reviewed by Oster and Yang [25] in as early as 1968. The polymerization of vinyl monomers can be initiated by free

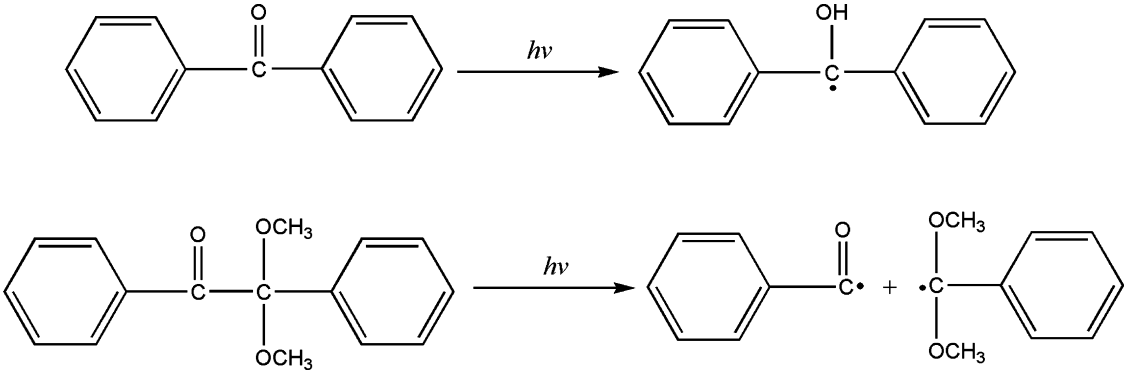

Fig. 1. Generation of radicals from photoinitiators.

radicals or metal ions in their unstable valence states, which are produced by excited electron transfer. This process can take place spontaneously in "redox" initiating systems or can be provoked by externally supplied energy. Based on the theories [26,27], metal ions abstract hydrogen atoms from the monomer, solvent and polymer substrates and then produce abundant free radicals, which initiate graft polymerization and homopolymerization at the same time.

Very few works have been done about the ions initiated grafting on PAN-based ultrafiltration membranes but by Yuan and her co-workers [23,24]. Using ceric ammonium nitrate (Ce^{4+}) as initiator, they grafted acrylamide (AAm) onto the membrane surface of poly(acrylonitrile-*co*-methyl methacrylate) (PANCMMA). A basic consideration is that the functional groups on the main chains of the copolymer could be oxidized to generate radicals by Ce^{4+} [28]. After grafting, the membrane surface got rougher and the pore structure had little change. Therefore, the wettability was improved. In a succeeding work, ferrous ammonium sulfate (Fe^{2+})/H_2O_2 as an initiator system was used to graft AAm on the same copolymer membrane [29]. Similar goal was achieved that the wettability was improved through the attachment of polyacrylamide chains on the membrane surface with the pores slightly blocked. Compared with the Ce^{4+} initiated grafting process, the initiator of Fe^{2+}/H_2O_2 resulted in much higher grafting degree, because the former might use the reactive groups in the early stage but the latter system could generate hydroxyl radicals in the medium that transferred into PAN-based macromolecules on the membrane surface so that the reaction could last for a longer time. Moreover, they brought forward a mechanism to interpret how Fe^{2+}/H_2O_2 initiated this surface grafting (Fig. 2). Simply speaking, some of –CN and –COOR groups in the copolymer were hydrolyzed into –COOH groups by NaOH treatment. Then, Fe^{2+} would be adsorbed on the surface while contacting with the hydrolyzed membrane (reaction 1). H_2O_2 in the medium quickly diffused onto the membrane surface and reacted with Fe^{2+} to give hydroxyl radicals ($OH^{\bullet}$) (reaction 2), which then abstracted hydrogen from the tertiary carbon in the PAN-based backbones to produce a macroradical (reaction 3), which interacted with the monomer to initiate the grafting [30]. The proposed mechanism accorded well with the experimental results.

In addition to these grafting methods, Belfer et al. [31] prepared layered membranes by sequential redox-initiated grafting onto the PAN ultrafiltration membranes. Water-soluble monomers with oppositely charged ionic groups were used. They reported that the proceeding of grafting derived from electrostatic attraction between the grafted polymer and the oppositely charged monomer. Water flux declined in comparison of the layered membranes with the initial UF membrane. By manipulating the conditions affecting grafting, the membranes could exhibit stability over various pH values and storage time. The antifouling property was not discussed in the work; however, it can be expected this property can also be improved due to (1) the reduced adsorption of proteins by hydrophilic graft polymer and (2) the electrostatic repulsion between charged polymer and protein at proper pH. With respect to ionic monomers grafting, Tanioka and his co-workers [32] intensively explored the surface properties of modified PAN membranes by ζ potential measurement. The ζ potential is a potential between the pore surface and the shear plane. It is generated by hydrodynamic flow induced by a pressure difference across a membrane and, therefore, could provide much information on the pore surface properties such as pK, charge density and isoelectric point. The last of which facilitates the interpretation of native surface properties related to protein adsorption and membrane-protein interaction phenomena.

3. Hydrolysis of the PAN-based membrane surface

Partial hydrolysis is one of the most important and most frequently used methods for PAN-based membranes, due to the convenience and favorableness for further modification. It is well known that PAN membrane can maintain its structure and composition when subject to acidic conditions (hydrolyzation is existent, but is rather slow). However, the case is not for the presence of base, because the nitrile group (–C≡N) can be

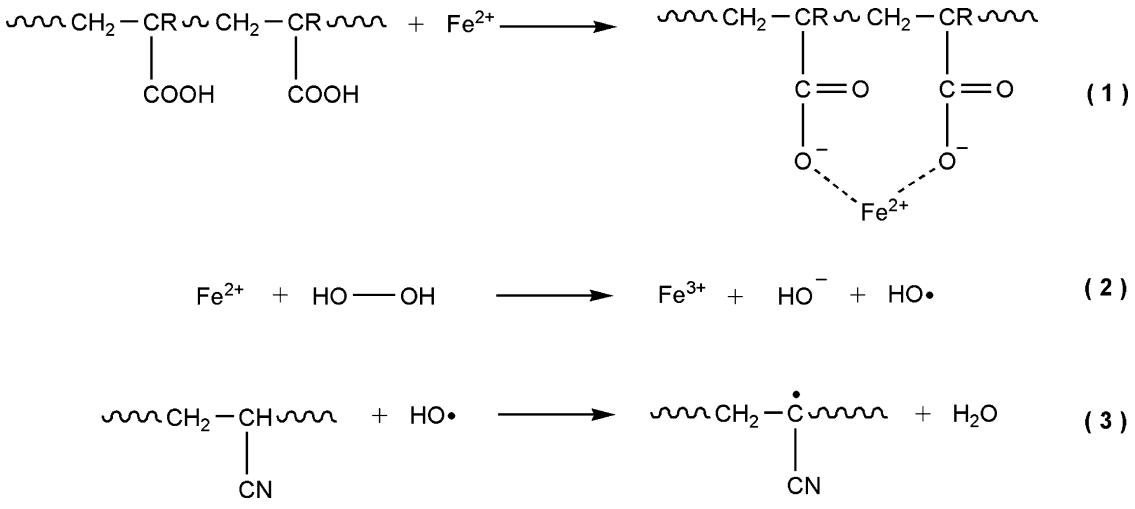

Fig. 2. Mechanism of Fe^{2+}/H_2O_2 initiating grafting reaction on the hydrolyzed PAN-based membrane.

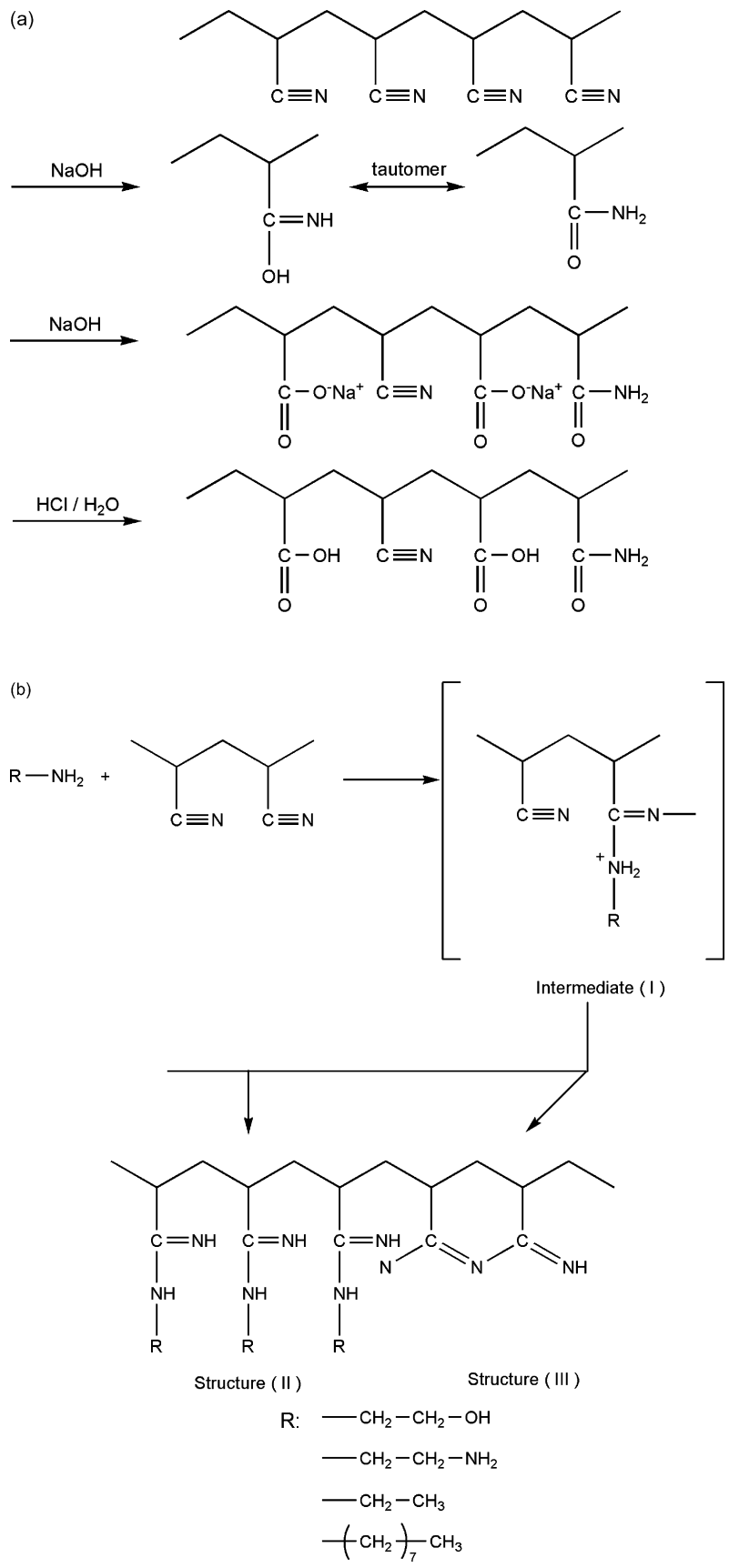

Fig. 3. Reaction mechanism of PAN with (a) NaOH and (b) primary amines.

easily hydrolyzed by NaOH or amine, and converted into carboxyl, acylamide or amide groups. Hydrolyzation can be used to render the PAN membrane surface hydrophilic and charged property, which helps to improve the protein-resistant performance. From the inherence (surface property, structure, etc.) of PAN membrane, hydrolysis has impact on the surface through two aspects, i.e. hydrophilicity and pore size. A possible mechanism for NaOH-induced hydrolysis is indicated in Fig. 3(a) [33]. PAN with a certain content of –COOH is easily swollen when expose to aqueous medium. The swollen macromolecules then become more mobile to move towards the pores. This process decreases the pore size and makes the membrane surface smoother.

The reaction of PAN chain with primary amine is shown in Fig. 3(b) [34]. A possible mechanism for this reaction is:

1. An intermediate (I) could be obtained in the initiation step by the nucleophilic attack of the amine groups in primary amine on the nitrile groups in PAN.
2. The intermediate with its separation of charges could rearrange into imine (structure II).
3. A further reaction might occur simultaneously among the intermediates, and the nitrile groups could form a carbon-nitrogen conjugated structure (structure III).

When the amine used is ethanolamine or ethylenediamine, structure II could enhance the hydrophilicity of PAN because of the increase of hydrophilic –OH or $–NH_2$ groups within the polymer chains. On the contrary, structure III increases the hydrophobicity for the linear sequences of fused rings.

Carboxyl groups derived from the partial hydrolysis of PAN with NaOH are facile to be ionized by surrounding water, so their generation does not only improve the hydrophilicity, but also results in negative surface charge for the PAN-based membranes. Godjevargova and Dimov [35] investigated the permeability (vitamin B_{12}) and protein adsorption (albumin solution) behavior of PAN-based membranes modified by hydroxylamine (NH_2OH), diethylaminoethyl methacrylate (DEAEM) and NaOH (DEAEM + NaOH). The grafting of DEAEM was initiated by an oxidation reduction system of H_2O_2–Fe(II). The nitrile groups were converted by NH_2OH into primary amine, oximes (simply speaking, $–NH_2$ and –OH, respectively, added to C and N atoms of –C≡N to form primary amine and oxime) and by DEAEM into tertiary amino group. They got that the modified membranes gave better permeability coefficient of vitamin B_{12} than the original membrane with the exception of DEAEM-grafted membranes, while the increase in hydrophilicity did not lead to the expected increase in permeability coefficient, an effect attributed to the decrease in the pore volume in the selective layer; partially hydrolyzed membrane showed the smallest adsorption of proteins. The amount of protein adsorbed on the membrane surface was determined by the membrane charge instead of the membrane hydrophilicity. Above the isoelectric point (IEP), albumin with negative charges repulsively interacted with the negatively charged membrane surface. Therefore, the hydrolyzed membrane surface showed small amount of protein adsorption but the DEAEM-grafted membrane surface resulted in higher adsorption, because the latter consumed some carboxyl groups on the membrane surface.

Yang and Tong [36] studied the ultrafiltration of proteins through the ionized PAN hollow fiber membrane with hydrolyzation and investigated its capability of separating two proteins having two similar molecular weight but different IEP. After treating in definite conditions including hydrolyzing time, temperature and NaOH concentration, charge density on the membrane surface could reach highest, while too high concentration of NaOH could cause severe degradation of the hollow fiber. Due to the pH-sensitive poly(acrylic acid) (PAA) layer, the hydraulic permeability and the ultrafiltration performance of proteins were greatly affected by pH. For both buffer solution and protein solution, the permeability at pH above 5 was about 1/3 of that at pH below 5. It was possibly due to the swollen and thicker PAA layer at higher pH. The retention of both proteins (myoglobin and cytochrome *c*) increased with the ionic density of PAN hollow fiber and reached the minimum at the IEP for the hydrolyzed membranes. It is known that, when the pH is equal to the IEP, the protein is neutral and its size is the smallest. Most of the pores on the ionic PAA layer were large enough for single particles of myoglobin and cytochrome c to pass through, despite the electrostatic interaction. When the surface charge density was higher, the electrostatic interaction was stronger and the retention was hence increased. Their results indicated that using the hollow fiber at varied pH, the selectivity towards a mixture of proteins could therefore be controlled.

Bryjak et al. [37] also modified a porous PAN membrane with NaOH. They found that the average pore diameter changed from 2.6 to 0.6 nm and the modified membranes were less prone for protein deposition, in which fouling caused a pore diameter reduction of 80% for the untreated and 20% for the modified membranes. As is known, transformation of nitrile to carboxylic group on the membrane surface may result in a decrease of pore lumen up to filling them with a PAA-like gel. However, when the pores in the membrane are small enough and the membrane material is charged, such a membrane may work in the nanofiltration mode. Results indicated that a membrane immersed in the modification bath was capable of rejecting about 50% of calcium while the unmodified membrane rejected no salt. Lohokare et al. [38] compared the effects of different treatment modes on the hydrolyzation with organic (ethanolamine/triethylamine) and inorganic bases (NaOH/KOH). The modes included dead-end and cross-flow one. It was found the extent of change in permeation was highly dependent on the treatment mode and treatment temperature. A maximum increase of 152% in water flux was achieved by dead-end mode within 20 h, while cross-flow offered maximum increase of 230% in just 2.5 h duration at 45 °C, because cross-flow mode did not cause a noticeable pore swelling. As a typical example in application, Oh et al. [33,39] functionalized PAN UF membranes by NaOH aiming at the preparation of nanofiltration composite membranes, in which polyacrylamide active layers were interconnected with support layers via the formation of ionic bonds. Kim et al. [40] also studied the effect of hydrolysis of annealed membrane using different concentrations of NaOH and CH_3ONa.

4. (Bio-)macromolecule immobilization on the PAN-based membrane surface

Surface immobilization, also referred to as "graft-to" method mentioned previously, involves the covalent attachment of designated macromolecular chains on the membrane surface. This usually requires the presence of reactive groups on the membrane, which can be generated through hydrolysis or copolymerization. The former method has been described in detail, and the latter one can be realized using vinyl monomers with reactive groups, such as acrylic acid, maleic anhydride and 2-hydroxyethyl methacrylate. The obvious advantage of surface immobilization over surface polymerization is that more kinds of macromolecules are available for selection, including natural polymers and synthesized polymers that are hardly obtained by radicals-initiated polymerization (e.g. PEG). The principle is only based on that (1) the immobilized polymers possess active groups and (2) the selected polymers are hemocompatible when used in hemodialysis, such as chitosan, heparin, some kinds of proteins, PEG, etc.

Hemodialysis is an important clinical procedure for the removal of toxic biological metabolites in patients with end-stage renal disease. The key element of a hemodialyzer is the semipermeable membrane, which allows for selective transport of low molecular weight biological metabolites from blood. Therefore, the discussion below is mainly focused on functionalizing PAN-based membranes for the purpose of hemodialysis.

4.1. Homo-surface immobilization

It was mentioned that PAN membranes are susceptible to alkaline condition and the nitrile groups can be hydrolyzed into carboxyl groups. Therefore, macromolecule immobilization can be carried out via the reaction of reactive carboxyl groups on the membrane and suitable polymer chains. With respect to this immobilization process on the hydrolyzed-PAN membrane, Yang et al. [39,40] investigated the effect of chitosan/heparin polyelectrolyte complex (PEC) and some proteins on the surface properties.

Chitosan, the principal derivative of chitin, is obtained by *N*-deacetylation to a varying extent that is characterized by the degree of deacetylation, and is consequently a copolymer of *N*-acetyl-D-glucosamine and D-glucosamine. Chitosan has been identified as biocompatible, biodegradable, non-toxic, physiologically inert, antibacterial properties, etc. Moreover, chitosan is one of the most plentiful, renewable organic resources and thus can be commercially obtained at a relatively low cost from the shells of shellfish, wastes of the seafood processing industry [41]. Despite so many advantages, chitosan enhances plasma protein adsorption, platelet adhesion and activation, and thrombus development; in other words, chitosan is an effective hemostatic agent [42,43], possibly due to electrostatic attraction. Therefore, chitosan cannot be applied for contacting blood unless it is modified. Contrary to chitosan, heparin is the most widely used blood anticoagulant. Heparin, a mixture of variably sulfated polysaccharide chains composed of repeating units of D-glucosamine and either L-iduronic or D-glucuronic acids, is commonly used in hemodialysis. The presence of sulfate groups in the carbohydrate moiety gives heparin its highly negative charge. Heparin binds to antithrombin III, accelerating the enzyme-neutralizing effect of this serine protease inhibator, and prevents thrombin formation. In the presence of heparin, the interaction of antithrombin with thrombin is virtually instantaneous. The heparin–antithrombin complex inhibits the conversion of other coagulation proteins to active serine proteases (Factors XII, XI, IX and X). Considering these fundamentals, the chitosan/heparin complex (PEC) was covalently immobilized onto the surface of hydrolyzed PAN membrane [44,45]. Briefly, the immobilization process involved that the reaction of chitosan with 1-ethyl-3-(3-di-methylaminopropyl) carbodiimide (EDC)-activated carboxyl groups on the membrane and the attachment of heparin onto immobilized chitosan spacer by glutaraldehyde. A series of characterizations and evaluations were performed, revealing that the surface hydrophilicity was increased, thrombus formation was reduced, the proliferation of bacterial was suppressed and the permeability was retained. Therefore, the conjugation of chitosan with heparin simultaneously endowed the membrane surface with anticoagulation activity and antibacterial activity, the latter of which was emphasized because anticoagulant must be injected continuous during hemodialysis. And the conjugate covalent immobilization possessed additional advantage of reducing release of heparin from the surface, comparing with physical or ionic bonding. The undesirable release into the blood can eventually leads to the disadvantages such as systemic heparinization and increases the risk of abnormal hemorrhage or the symptom of thrombocytopenia [46]. The less loss of heparin also reduced the injection of heparin in clinical applications, and even resulted in the heparin-free therapy. This work was meaningful in potentializing the application of PAN membrane in hemodialysis.

Yang and co-workers [47] also covalently immobilized plasma proteins onto hydrolyzed PAN membrane to evaluate the hemocompatibility and anaphylatoxin formation. Platelet-adhesion-inhibiting human serum albumin (HSA) and platelet-adhesion-promoting collagen (COL) were used as typical model proteins. Both immobilizations increased the hydrophilicity of the membrane surface, but just a little. Immobilized HSA reduced the platelet adhesion, fibrinogen adsorption, prolonged the blood coagulation times, as well as reduced the leukopenia and anaphylatoxin formation. On the other hand, COL immobilizing on PAN membrane exhibited the opposite effect when contacting with blood, though induced less increase of C3, C4 antigens of serum. During the process of hemodialysis, the plasma proteins will always more or less adsorb onto the surface of hemodialyzer, even after intensive washing. Therefore, the results helped to explain why reused hemodialyzers were beneficial to hemodialysis therapy. That was because the reused hemodialyzers usually suppressed the leukopenia, anaphylatoxin formation and avoided inducing the first use syndrome. It was also obtained that HSA immobilization on the surface of PAN membrane was useful and practicable in improving the hemocompatibility and diminishing the anaphylatoxin formation during hemodialysis treatment.

4.2. Co-surface immobilization

Acrylonitrile has an excellent advantage of easy copolymerization with other vinyl monomers, which is concluded according to that it possesses relative high values of Q (reactive activity) and e (polarity). So far, to our knowledge, the copolymerization may benefit to the improvement of surface hemocompatibility in two aspects: on one hand, the incorporation of hydrophilic and/or biocompatible comonomers may endow the acrylonitrile-based copolymer with less affinity to blood components; on the other hand, the reactive groups can be facile to introduce via the comonomers for the further immobilization of other (bio-) macromolecules. Concerning the tethering of biomacromolecules on the reactive groups-contained PAN-based membranes, our group was devoted to creating a biomimetic membrane surface and systematically investigating its effect on the hemocompatibility [48–56].

The copolymers were synthesized with a water-phase precipitation copolymerization process (WPPCP). Briefly speaking, the copolymerization occurred in water medium and the water-insoluble products were obtained by precipitation and filtration. WPPCP has main advantages over solution copolymerization such as high molecular weight, relatively high monomer conversion and environmental friendship. Chemical incorporation of α-allyl glucoside [57,58] as well as *N*-vinyl-2-pyrrolidone [59,60] into PAN chains had proven to resist protein adsorption and platelet or macrophage adhesion very well. Copolymerization of acrylonitrile with maleic anhydride (MA) and 2-hydroxyethyl methacrylate increased the hemocompatibility limitedly; however, biomacromolecules could be further bound onto the membrane surface via a reaction between the introduced reactive groups and the functional groups of biomacromolecules. The synthesis of poly(acrylonitrile-*co*-maleic anhydride) (PANCMA) and PANCHEMA are indicated in Fig. 4.

4.2.1. Tethering PEG onto PANCMA membrane [48–51]

For tethering PEG on the PANCMA membrane, the anhydride groups had to be regenerated from –COOH as shown in Fig. 5, because the monomer of maleic anhydride was easy to hydrolyze into maleic acid when exposed to water. Poly(ethylene glycol) (PEG) is an uncharged polyether with the chemical formula, H–$(OCH_2CH_2)_n$–OH, which is the simplest structure of water-soluble polymers and is well known for its extraordinary ability to resist cell adhesion and protein adsorption because of its hydrophilicity, large exclusion volume, and unique coordination with surrounding water molecules in an aqueous medium [61]. In addition, PEG has the unique properties of nontoxicity and nonimmuno-genecity. PEG is useful for rendering surfaces inert to the adsorption of proteins and to the adhesion of platelets and other cells, and the resistance of PEG-coated surfaces increases with increasing density and length of the chains in the surface-grafted film [62,63].

To achieve the immobilization of PEG onto the membrane, the converted anhydride groups underwent an esterification reaction with PEG containing hydroxyl end groups. Hydrophilicity and resistance to protein adsorption and platelet adhesion were all enhanced. The effect of molecular weight of PEG was especially and extensively studied on the PANCMA membrane surface properties. It was found that, the membrane modified with PEG 400 (M_w = 400 g/mol) showed the best hydrophilicity and lowest protein adsorption, platelet adhesion as well as macrophage attachment among those with PEG with molecular weight of 200–1000 g/mol (Fig. 6). Exposure of blood to a foreign material almost immediately leads to the adsorption of plasma proteins, which is proposed to involve two steps. The first occurs on the surface as a result of preferential competitive adsorption of high molecular weight proteins, such as albumin, fibrinogen/fibrin, fibronectin and globulins. The second step is absorption in the body of the materials of proteins with low and medium molecular weights, including cytokines and anaphylatoxins. Fibrinogen translates into insoluble fibrin, which is deposited on the surface and forms a mesh of fibrils. In contact with fibrin, platelets are activated and aggregate, giving rise to a cooperative interaction leading to blood clotting. Simultaneously, blood cells are attracted by the thrombus, invade it and contribute to further fibrin formation and platelet recruitment through enzymatic release. These series processes contribute to the formation of a biological membrane that spread over the material and called “protein cake”, which includes plasma

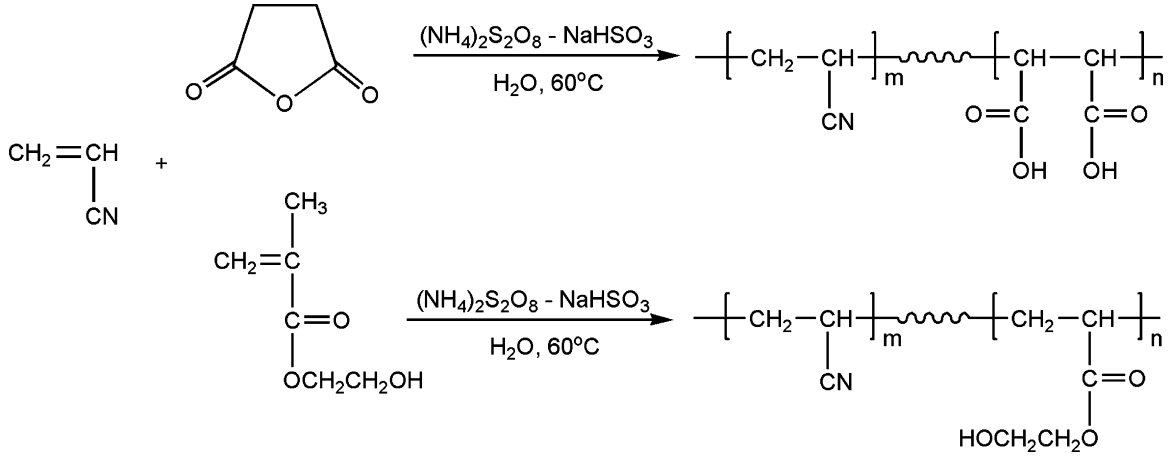

Fig. 4. Schematic representation for the synthesis protocols of PANCMA and PANCHEMA.

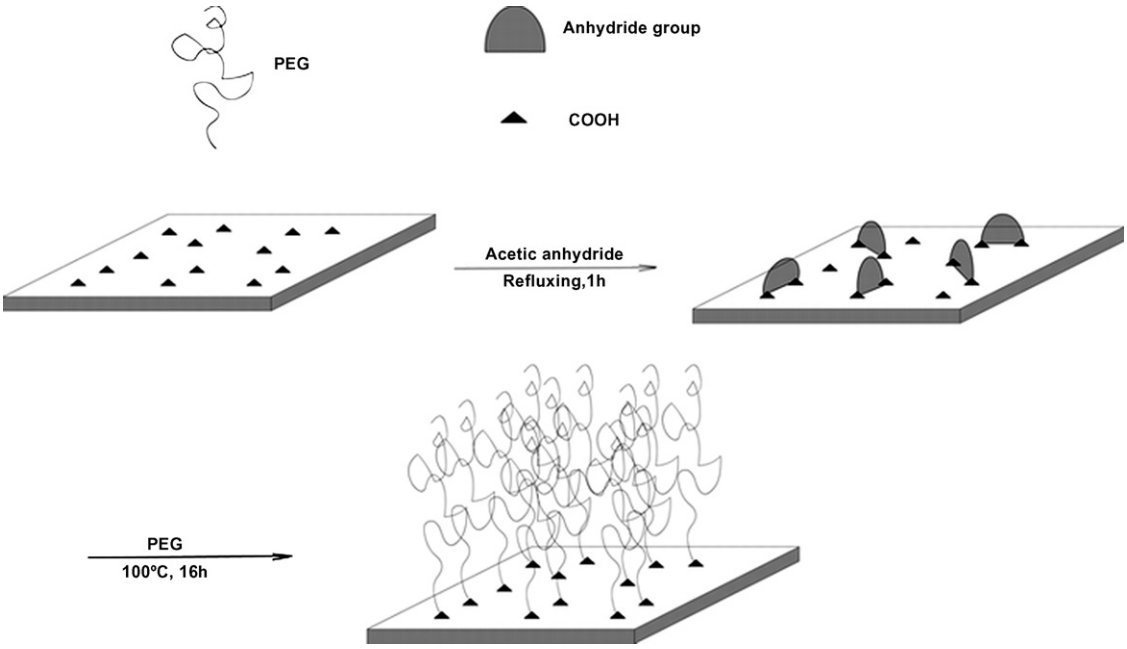

Fig. 5. Schematic representation for the surface modification of PANCMA membrane by PEG tethering.

proteins resulting in further thrombogenesis, strong platelet aggregation and release of procoagulants like platelet Factor IV [3]. Therefore, platelet adhesion plays a major role in the blood clotting. Macrophage is a kind of immune cell and performs various functions such as migration, phagocytosis, secretion, antigen presentation and survival through precisely modulated adhesion, in living bodies. Although the molecular mechanism in macrophage adhesion is complex, dynamic and not yet fully understood, it is generally thought that the fewer amount of macrophage adhered onto the material surface, the immunological reaction or immunological rejection will decrease after the material has been planted into the living body. Therefore, it was definitely confirmed that the immobilization of PEG could render the membrane surface improved biocompatibility with blood components, which was mainly due to the excluded volume effect and dynamic motion of water-soluble PEG chains on the surface [64]. The reason why tethering PEG400 showed best performance could be attributed to the balance between PEG immobilization density and hydrophilic chain segments; in detail, lower-molecular-weight PEG grafted membrane had the higher immobilization density but the shorter PEG chain with less hydrophilic $–CH_2–CH_2–O–$ segment brought about relatively low hydrophilicity on membrane surface. The effect of the MA content was also studied. Results indicated that, the increase of MA content resulted in increasing the immobilization density of PEG, which improved the surface hydrophilicity and blood compatibility significantly. Nevertheless, the increasing tethering density caused the coverage of polymers on the surface, which decreased the porosity and pore size, leading to the lower permeability of pure water and BSA solution. Therefore, the middle content of MA in the copolymer was thought as the most suitable.

Other biocompatible macromolecules such as heparin or insulin could also be immobilized on the PANCMA membrane surface to improve hydrophilicity and hemocompatibility [52]. Briefly, the immobilization process involved the amination of

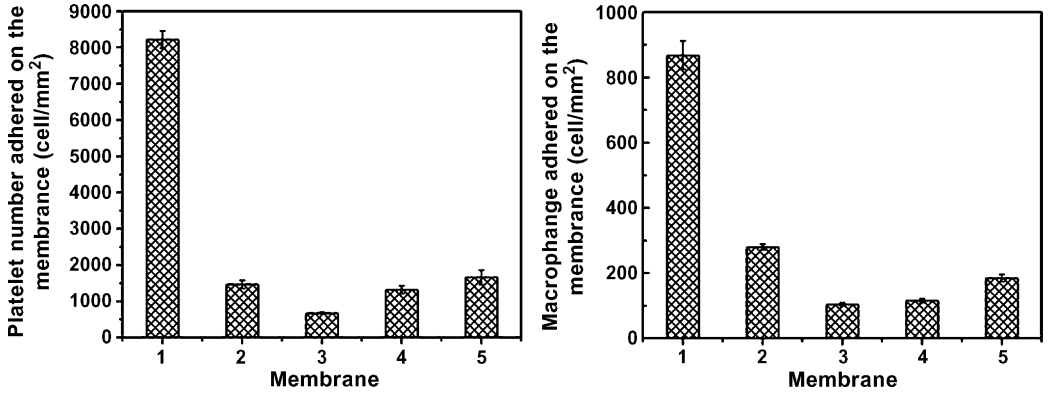

Fig. 6. Dependence of the number of adherent plasma platelet and macrophage on the molecular weight of the immobilized PEG. (1) PANCMA; (2) PANCMA-*g*-PEG200; (3) PANCMA-*g*-PEG400; (4) PANCMA-*g*-PEG600; (5) PANCMA-*g*-PEG1000.

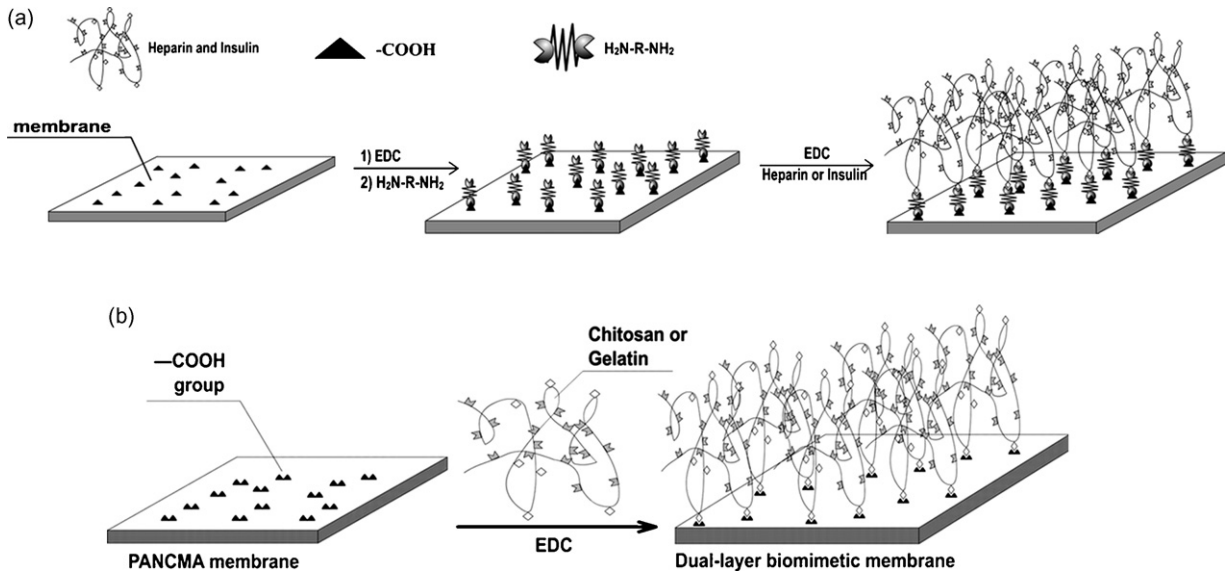

Fig. 7. Schematic representation for the immobilization of (a) heparin/or insulin and (b) chitosan/ or gelatin onto the PANCMA membrane.

–COOH contained membrane surface and then the immobilization of heparin/ or insulin previously activated with EDC on the amino-bound membrane surface (Fig. 7(a)). Heparin is referred to as an excellent anticoagulant and insulin can also reduce the aggregation of platelets; therefore, immobilizing the two biomacromolecules was expected to improve the surface properties. Results indicated the bound heparin/or insulin formed a thin layer covering on the porous surface, the hydrophilicity plus hemocompatibility were markedly improved (the number of adherent platelet and macrophage decreased significantly) and tethering heparin showed more efficient improvement than insulin. Chitosan or gelatin was also immobilized on the PANCMA membrane surface using EDC coupling (Fig. 7(b) [53]) and it was also observed that the number of adherent platelet and macrophage decreased.

4.2.2. Anchoring of phospholipid moieties onto the PANCHEMA membrane [54–56]

The concept of anchoring is inspired by the mimicry of biomembranes mainly constructed of neutral phospholipids and phosphorylcholines. Phosphorylcholine is the headgroup of phosphatidylcholine, the major component of the extracellular side of cell membranes. The fact that proteins in the bloodstream do not foul onto the surface of cells shows that the outer surface of the cell walls, comprising PC groups, is biocompatible [65,66]. Accordingly, phosphorylcholines materials have the potential to reduce protein adsorption significantly, since their hydrated surfaces are able to interact with proteins without inducing three-dimensional conformational changes, which only result in the reversible adsorption of protein. Surface modifications with phospholipids analogous polymers (PAPs) and phospholipid moieties were extensively studied to improve the biocompatibility for various biomaterials in recent years [67–70]. It is recognized that just a small amount of phospholipid moieties can improve the surface biocompatibility of materials, because the zwitterionic nature of the phospholipid moiety, combined with its ability to bind water tightly around itself, gives these materials remarkable biocompatibility. Therefore, it would be rather meaningful to perform the investigation of anchoring phospholipid moieties onto the PANCHEMA membrane. The introduction process mainly involved the reaction of hydroxyl groups on the HEMA-contained membrane surface with 2-chloro-2-oxo-1,3,2-dioxaphospholane (COP) followed by the ring-opening reaction of COP with trimethylamine (Fig. 8). As a result, for the phospholipid moieties modifying PANCHEMA membranes, the hydrophilicity and the permeation of protein (bovine serum albumin) solution were greatly enhanced, the protein adsorption and the fouling were obviously weakened, and much less platelet and macrophage adhesion occurred. Until now, how the phospholipids interact with proteins to achieve the biocompatibility is not entirely understood. However, it is generally recognized that the chemical and electronic configuration of the zwitterionic moieties structures and the association with a large amount of water allow reversible interaction with biomacromolecules that approach the surface. By controlling the adsorption of biomacromolecules and the formation of any mediating layers, cellular adhesion can also be controlled. By these basic principles, biofouling is governed on any scale. With regard to the PANCHEMA membrane surface anchored by phospholipid moieties, the hemocompatibility could be easily tailored by adjusting the content of HEMA and thus phospholipid moieties (Fig. 9).

So far, for the purpose of surface immobilization, the reactive groups can be generated on the PAN-based membranes either by copolymerizing or hydrolysis. The two methods have each advantages and disadvantages. PAN and its various membranes are relatively more commercially available; however, the exposure to the base, especially NaOH, may more or less damage the membrane structure and thus the mechanical strength, and more at serious condition such as long treatment time and high base concentration. On the other hand, the copolymerization method can be used to control the reactive groups introduced

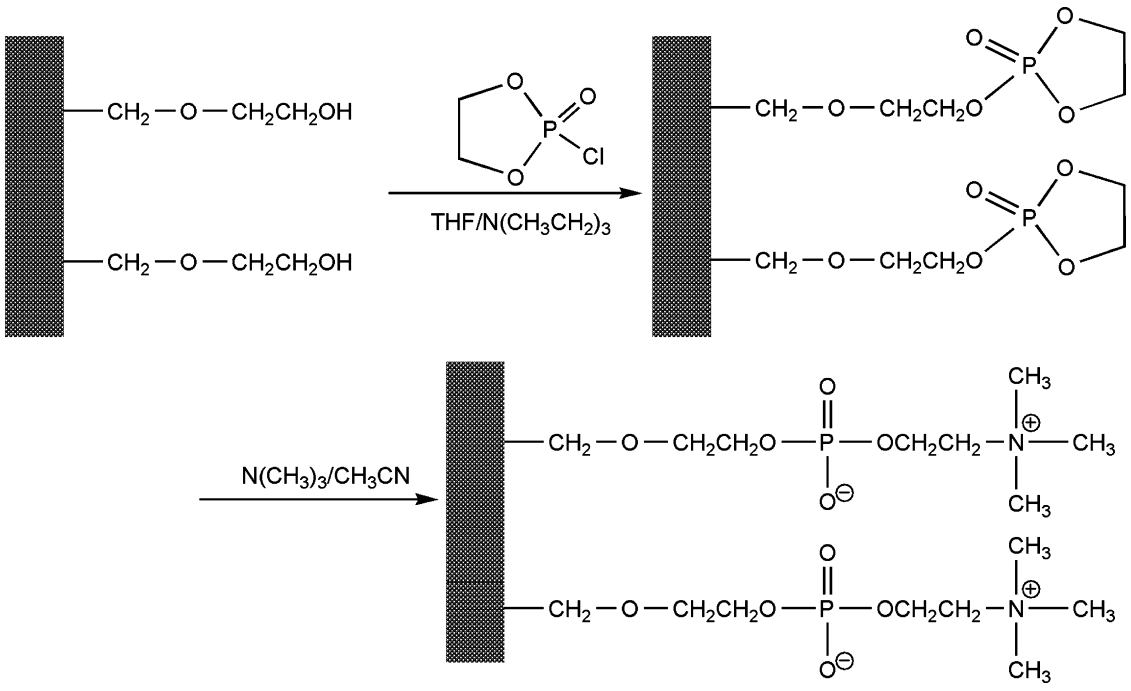

Fig. 8. Schematic representation for the surface modification of the PANHEMA membrane by the formation of phospholipids moieties.

more easily, maintain the mechanical strength and even better, because the comonomer segments may reduce the strong interaction between PAN chains and make them more flexible; however, the copolymer was usually synthesized in the lab and the monomers are usually not easy to obtain. Moreover, whatever hydrolysis or copolymerization for grafting-to method is advantageous over the mentioned graft-from method, over preventing waste of materials due to homopolymerization and crosslinking and more easily controlling graft density.

Functionalization of PAN-based membranes for the antifouling and blood-contacting purpose has been generally reviewed through the above paragraphs. A variety of modifications have been utilized to endow the membrane surface with the hydrophilicity and the inertness to protein adsorption followed by antifouling performance, some of which, however, are not available to the hemodialysis field. Collagen and chitosan as modifiers are the typical examples. To meet the request of the hemodialysis process, the mimicry of biological inert interface is concerned. In modifying PAN-based membranes, mimicking the surfaces of blood cells only ever involved the introduction of phospholipid moieties. Nevertheless, the attempt provided an alternative avenue to develop the PAN-based membranes towards the application to hemodialyzer, so the method of the mimicry in preparing such membrane is rather promising. Anyway, we can expect with confidence that the modifications of PAN-based membranes with so many excellent intrinsic properties have great potential to deepen and expand its influence in more biomedical fields.

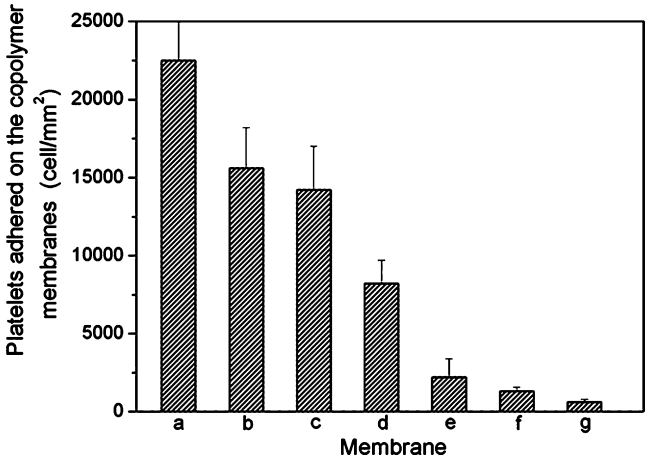

Fig. 9. Adhesion of platelet on PAN, PANCHEAM and PCANCP dense membrane surface. The HEMA mole fraction in the copolymer membrane is (a) 0%; (b) 6.4%; (c) 9.3%; (d) 17.8%. The mole fraction of phospholipid moiety on the copolymer membrane surface is (e) 6.09%; (f) 9.19%; (g) 17.1%.

5. PAN-based membrane surface for enzyme immobilization

Enzymes, proteins with high efficiency in catalysis and unparalleled selectivity, have been extensively realized in various fields, such as fine-chemical synthesis, pharmaceuticals, commodity catalysts in food processing and detergent applications, biosensors, bioremediation, polymerase chain reaction, protein digestion in proteomic analysis, and biofuel cells. And due to their biological character, enzymes are well known as “green catalysts”. However, some inevitable disadvantages of enzymes practically restrict their further development in large-scale operations, such as the short lifetime from the intrinsic instability and high cost from disable recycling. In practice, almost all industrial operations prefer the enzymes in immobilizing form, because they offer easy recycling, feasible continuous operations, and simple product purification. Among the large amount of sup-

Table 1
Enzymes immobilized on PAN-based membranes

Enzyme (EC number)	Support[a]	Immobilization	Corresponding author	Ref
Amyloglucosidase (EC 3.2.1.3)	PAN membrane	I	M. Ulbricht	[71]
Amyloglucosidase (EC 3.2.1.3) *or* Invertase (EC 3.2.1.26)	PAN and PAN-AA (HA treating PAN) membrane	II, III	M. Ulbricht	[72]
Glucose oxidase (EC 1.1.3.4)	Copolymer (A) membrane treated by NaOH/HMDA and by HA	IV	T. Godjevargova	[73]
Glucose oxidase (EC 1.1.3.4)	Copolymer (A) membrane grafting DEAEM and AMPSA[(a),(b)]	V	T. Godjevargova	[74,75]
Glucose oxidase (EC 1.1.3.4)	Copolymer (A) membrane with various modifications	IV, V	T. Godjevargova	[76]
Urease (EC 3.5.1.5)	Copolymer (A) membrane grafting DEAEM and AMPSA[(a)]	V	T. Godjevargova	[77]
Glucose oxidase (EC 1.1.3.4)	Copolymer (B) membrane	IV, V	T. Godjevargova	[78]
Glucose oxidase (EC 1.1.3.4)	Copolymer (C) membrane	V	T. Godjevargova	[79]
Urease (EC 3.5.1.5)	Copolymer (A) with seven chemical modifications	IV	T. Godjevargova	[81,95]
Peroxidase (EC 1.11.1.7)	Hydrophilic PAN membrane	IV	M. Mateus	[82]
Glucose oxidase (EC 1.1.3.4) and Catalase (EC 1.11.1.6)	PAN by NaOH/H_2O_2 treatment	IV	T. Godjevargova	[83,84]
Glucose oxidase (EC 1.1.3.4) and Catalase (EC 1.11.1.6)	PAN by NaOH/H_2O_2 treatment	IV	T. Godjevargova	[83,84]
Glucose oxidase (EC 1.1.3.4)	PAN and copolymer (D) blending membrane	VI	N. Vasileva	[85]
Cellulase (EC 3.2.1.4)	Copolymer (E) membrane grafting AAm[(a)]	IV	J. Sheng	[86]
Urease (EC 3.5.1.5)	PAN membrane treated with NaOH/1,6-hexanediamine	IV	M.C. Yang	[87,88]
Cholesterol oxidase (EC 1.1.3.6)	PAN membrane treated with NaOH/1,6-hexanediamine	IV	M.C. Yang	[89,90]
Lipase (EC 3.1.1.3)	Copolymer (F) membrane	III	Z.K. Xu	[91,92]

[a] *Abbreviations*: HA, hydroxylamine; HMDA, hexamethylendiamine; DEAEM, 2-dimethylaminoethyl mathacrylate; AMPSA, 2-acryl-amido-2-methylpropanesulphonic acid; AA, acrylic acid; AAm, acrylamide. Copolymers: (A) poly(acrylonitrile-*co*-methylmethacrylate-*co*-sodium vinylsulphonate); (B) poly(acrylonitrile-co-glycidylmethacrylate); (C) poly(acrylonitrile-*co*-*N*-vinylimi-dazol); (D) poly(methyl methacrylate-*co*-dichlorophenylmaleimide); (E) poly(acrylonitrile-*co*-methylmethacrylate-*co*-sodium methylpropylenesulfonate); (F) poly(acrylonitrile-*co*-maleic acid). Grafting initiator: (a) Fe^{2+}/H_2O_2; (b) Fe^{2+}/radiation. Immobilization techniques: (I) covalent binding of enzyme to the membrane treated by $NH_2OH{\cdot}HCl$/DPPA-Et_3N solution; (II) adsorption of enzymes on and in the membrane followed by glutaraldehyde crosslinking; (III) covalent binding of enzyme to the membrane activated by carbodiimide; (IV) covalent binding of enzyme to the membrane activated by gluataraldehyde; (V) adsorption of enzymes on the membrane; (VI) direct covalent binding of enzyme to the membrane.

ports, PAN-based membranes can offer some special promises as cradle for enzyme immobilization, which is mainly because:

(1) As a separation tool, the membrane with immobilized enzyme can integrate both efficient functions (biocatalysis of enzyme and separation of membrane) in one structure.
(2) Reactive groups can be easily generated on the membrane surface for the covalent immobilization of enzymes.
(3) The PAN membrane surface can be facilely tailored to adjust the microenvironment for enzyme catalysis.

In addition, the intrinsic excellent properties (stability to organic solvents, etc.) also make PAN-based membranes a kind of qualified candidate for enzyme immobilization and catalysis in various surroundings.

An overview of enzymes immobilized on PAN-based membranes, reported in the literatures over the last decade [71–92], is presented in Table 1. It implies that PAN-based membranes have attracted much interest as supports for the immobilization of such biocatalysts. However, there usually exist many problems, and one of them is the encountered activity loss due to the nonspecific interaction between the enzyme and the surface. It is generally recognized that a biocompatible surface can reduce the interaction because the biomimetic surface is capable of preserving the natural conformation of enzymes. Inspired by the concept, our previous work [93,94] tethered biomacromolecules such as chitosan or gelatin on the membrane surface for covalently immobilizing lipase. Chitosan or gelatin firstly covalently attached on the membrane surface using EDC activation and subsequently lipase was immobilized on the biolayers with glutaraldehyde coupling. Compared with the enzymes on nascent membranes, the ones on the modified membranes showed enhanced activity and stability in the pH, thermal and reused conditions.

The enzyme-immobilized membranes have showed wide applications in separation and purification. For example, Yang et al. [87–90] used urease-immobilized membrane as dialyzer for the removal of urea and cholesterol oxidase immobilized membrane for determination and clearance of cholesterol. However, the applications are still on elementary trial only in the laboratory, and the interplay in the biocatalytic system is needed to verify. Even, great hopes are being because the support, PAN-based membranes, is justified to be excellent.

Concluding remarks and perspectives

Polyacrylonitrile, a commercial available material, has been widely applied in the preparation of separation membranes. These membranes have received much attention in the fields of water treatment, pervaporation and supports for other (bio-) macromolecules. However, in the aspect of hemodialysis and protein-involved filtration, the application of PAN-based membranes is limited for its relatively low hydrophilicity and biocompatibility. To overcome these obstacles, surface engineerings such as hydrolyzation, “grafting to” and “grafting from” have mainly been used to modify the PAN-based mem-

branes. The membranes after the above treatment showed much improved resistance to the adsorption of protein or blood component (platelet, macrophage). Especially, the concept of biomimicry, i.e. mimicking the outer surface of the cells, was introduced. In this case, the hemocompatibility of the PAN membrane surface was improved a lot. PAN-based membranes have more superiority over other materials when subject to biomimicry modifications. One of the advantages is that its polymer derivatives possess reactive groups favoring anchoring other biocompatible molecules. In addition to the dialysis and filtration, one of the most important applications of the PAN membrane is for attaching enzyme. Such enzyme-membrane can simultaneously realize the functions of catalysis and separation and turn out to be a bioactive membrane. The modification methods previously described to improve surface biocompatibility can also enhance the activity and stability of immobilized enzyme, i.e. biomimicry is beneficial to the surface attaching enzymes.

Because of the unique characteristics of PAN, the surface engineerings of the membranes seem rather potential in improving its present use and widening its further applications. However, until now, there still exist two problems. Firstly, the modification methods are relatively expensive and complex. Therefore, to make the modified membranes more available, simplifying the modification procedure is necessary. For example, by copolymerizing with other biocompatible monomers with, the PAN-based membranes showed rather strong resistance to protein and platelet adhesion. Secondly, the mechanism involving the interaction between the modified surface and proteins (enzymes, etc.) is still obscure and needed to verify.

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