



Characterization of fouling in a hollow-fiber membrane and control of anti-fouling properties by surface-reorganization

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Doctoral Dissertation

**Characterization of fouling
in a hollow-fiber membrane and control of
anti-fouling properties by surface-reorganization**

水処理用中空糸膜ファウリングの特性評価と膜表面構造
の制御によるファウリングの抑制

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Chapter I

Introduction

I.1 MEMBRANE PROCESS

I.1.1 Separation processes using membrane systems

Membrane separation processes have become valuable tools for the separation of molecular mixtures since the last century. They become the key components in artificial organs and in devices for the controlled release of active agents, or in energy conversion and storage systems [1-2]. Membrane systems have numerous industrial applications and provide many advantages: They offer appreciable energy savings; they are environmentally benign; the technology is clean and easy to operate; they replace conventional processes like filtration, distillation, and ion exchange; they produce high-quality products; and they offer greater flexibility in system design. Pressure-driven processes such as ultrafiltration, nanofiltration, and microfiltration are firstly established and various applications have been commercialized in the fields of pharmaceutical and biotechnology. Recently, various approaches of characterizing, controlling and preventing membrane fouling have been developed. The development of tailored membranes, fouling prevention, and optimization of chemical cleaning will ensure a high level of membrane process performance.

I.1.1.a Classification of membranes

Membranes can be classified by nature into biological or synthetic membranes first, and they differ completely with each other both in structure and functionality. This study focuses on synthetic membranes, and they can be subdivided into organic (polymeric or liquid) and inorganic (ceramic, metal) membranes.

According to the development of membrane processes, they can be divided into the first generation membrane processes such as microfiltration (MF), ultrafiltration (UF), nanofiltration (NF), reverse osmosis (RO), forward osmosis (FO), electrodialysis (ED), membrane electrolysis (ME) and diffusion dialysis (DD), and the second generation membrane processes such as gas separation (GS), vapor permeation (VP), pervaporation (PV), membrane distillation (MD), membrane contactors (MC) and carrier mediated processes [3]. The application range of membrane processes is summarized in Fig. I. 1. In industry, microfiltration (MF), ultrafiltration (UF), nanofiltration (NF), reverse osmosis (RO) and forward osmosis (FO) are mainly used in water treatments.

particle size	atomic/ ionic range	low molecular range	high molecular range	micro particle range	macro particle range
micrometer		0.001	0.01	0.1	1.0
nanometer		1.0	10	100	1000
molecular weight	100 200	1000	100,000 500,000		10,000
solutes	aqueous salt		colloidal silica		yeast cells
	metal ion		virus	bacteria	
	sugar		proteins		
		microsolutes			
membrane separation process	electrodialysis				
	diffusion dialysis				
	reverse osmosis				
	nanofiltration				
	gas separation		ultrafiltration		
	pervaporation			microfiltration	
		dialysis			

Fig. I. 1 Application range of various membrane processes.

I.1.1.b Applications of membrane systems

Membranes have developed from a research topic to a mature industrial separation technology in the last 40 years. It is an outstanding method of choice for separation and concentration of different components of a macromolecular mixture of solutions, especially for those components sensitive to high temperatures and chemical treating.

Various membrane processes are now used in numerous diverse fields extending from production of potable water [4] and separation of gases [5], to chemical (including petrochemicals) and medical [6] and food industries [7]. Among the applications based on membrane systems, I would like to mention two current examples.

Strong development and growth of membrane technology can be observed in the purification of wastewater. As a result of rapid industrial development, an increasing amount of wastewater is produced worldwide, in which wastewater contaminants may cause severe health and environmental problems. A process that uses both a biological stage and a membrane module, known as the membrane bioreactor (MBR) process, was developed (Fig. I. 2) [1]. The bioreactor and membrane module each have a specific function: (1) the biological degradation of organic pollution is carried out in the bioreactor by adapted microorganisms and (2) the separation of microorganisms from the treated wastewater is performed by the membrane module. The membranes constitute a physical barrier for all suspended solids, and therefore enable not only the recycling of the activated sludge in the bioreactor but also the production of a permeate that does not contain suspended matter, bacteria, or viruses.

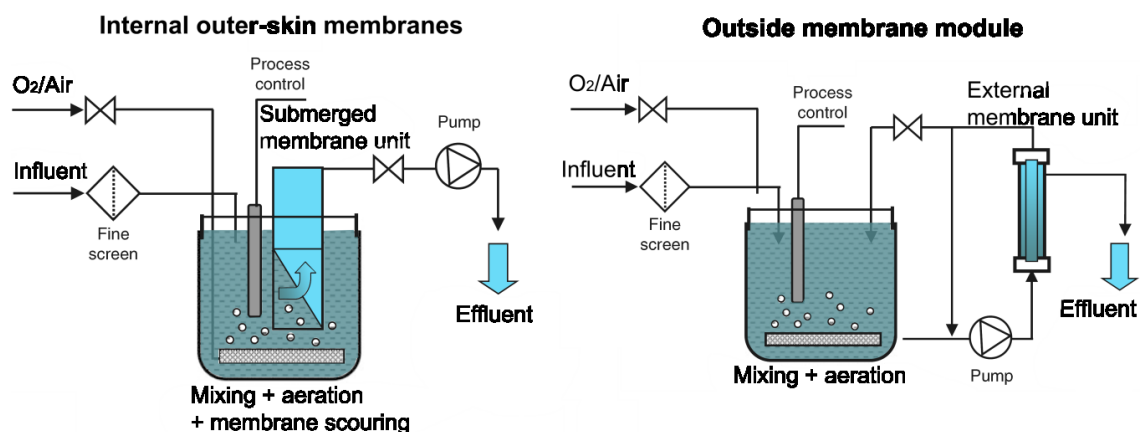


Fig. I. 2 An example of membrane bioreactors configurations [1].

Biocatalytic membrane reactors (Fig. I. 3) with ceramic membrane elements were developed to use the catalytic action of enzymes for agro-food, pharmaceutical, biomedical industries, and even for wastewater treatment [8]. In the agro-food sector, they could be used for: reducing the viscosity of juices by hydrolyzing pectins, reducing the lactose content in milk and whey by its conversion into digestible sugar, the treatment of musts and wines by the conversion of polyphenolic compounds and anthocyanins, and the removal of peroxides from dairy products. In the pharmaceuticals sector, these reactors could be implemented for the production of amino acids, antibiotics, anti-inflammatories, anti-cancer drugs, vitamins, etc. For water and wastewater treatments, biocatalytic membrane reactors are of interest for converting highly hazardous contaminants, which cannot be destroyed or removed by conventional treatments.

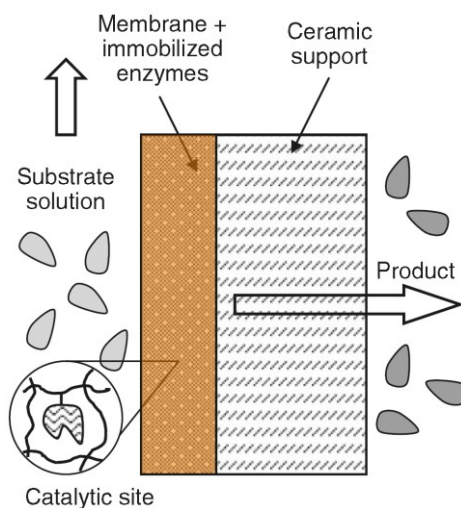


Fig. I. 3 Concepts of biocatalytic membrane reactors with immobilized enzymes.

Drug delivery systems using membranes are clarified to externally regulated and self-regulated delivery systems [9]. Some examples of achievements are developing drug release modulation based on a diffusive-controlled mechanism [10], isolating of particular structures from the external environment, inverting artificial implants

biocompatible [11], allowing drug release (by membrane dissolution) under particular environmental conditions, simulating the permeation properties of natural tissues [12].

I.1.1.c Design of module and process

Membranes are manufactured as flat sheets, capillaries, or in tubular shapes and are applied in various module configurations. To meet different requirements, the following membrane modules are designed for different applications: (a) the plate and frame module; (b) the spiral wound module; (c) the tubular membrane module; (d) the capillary membrane module; and (e) the hollow fiber membrane module.

Although a membrane is the heart of the membrane contactor technology, internal design dictates how the two phases flow inside the contactor and how the hydrodynamics in each phase is important. As the rate of mass transfer is directly dependent on the mass transfer coefficients in each of the phases, which in turn is dependent on the internal hydrodynamics [1].

I would like to give an example of the development of designing hollow-fiber membrane modules. Designs followed two categories: (1) the primary fluid being treated flows through the lumen of the hollow fibers and (2) the primary fluid being treated flows on the outside of the hollow fibers. Designers also consider keeping the flow direction of fluid in each phase horizontal along the axis of the membrane. Most membrane contactors at earlier ages have contactor houses with cylindrical shapes with tube-in-shell configuration, where the primary fluid flow on the lumen side from one end of the fiber to the other and the other fluid on the lumen side on the shell side in parallel direction (Fig. I. 4, left). The main drawback of the design is that nonuniform spacing of hollow fibers may result in poor flow distribution of flow channeling on the shell side. A significant improvement is the transverse-flow design, where the primary

fluid flows on outside of the hollow-fiber membrane at a transverse direction to the fiber axis, while the other fluid flow on lumen side of the hollow fiber. This design significantly improves the mass transfer efficiency [13]. However, it still needs improving, when scales the device up.

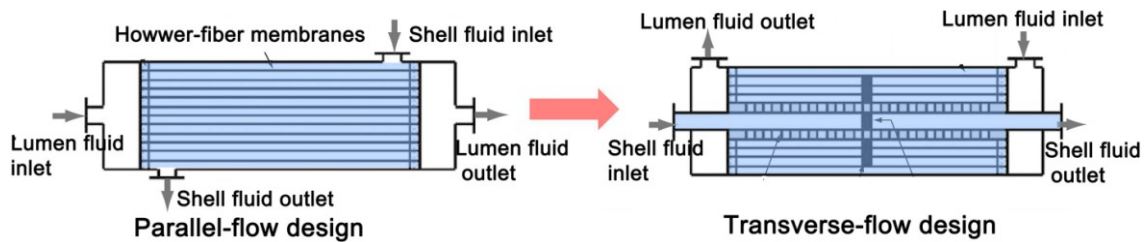


Fig. I. 4 Examples of designs of module devices for hollow-fiber membranes.

I.1.2 Water treatment by ultrafiltration (UF) and microfiltration (MF)

Microfiltration (MF) and ultrafiltration (UF) membranes have pore-size ranging from several nanometers to several micrometers (Fig. I. 1), and the filtration separation uses pressure difference as the driving force. It has the following advantages: (1) separation measure is a low-energy consumption type; (2) target to be separated is scarcely denatured or decomposed due to separation under mild conditions.

As mentioned in section I.1.1.b, the industrial use of MF/UF has progressed in various industrial fields, and its use has been further widely expanded in this decade. A major factor is that the MF/UF technology has been used as a clarification procedure in water supply and sewerage fields, where sand filtration had been conventionally used. An important trigger behind the recent surge in MF/UF growth was the development of hollow-fiber MF/UF membranes. Applying backwashing automatically with permeate to hollow-fiber membranes reduced extensive cleaning procedures.

In general, MF/UF membranes were characterized by measuring pore-size (MF) or molecular weight cutoff (MWCO) (UF), porosity, water permeability, surface

hydrophobicity (Contact angle), surface charge (zeta potential), and surface roughness (atomic force microscopy).

Although MF/UF systems have been presented as effective for removal of particles, turbidity, and bacteria/cysts without using disinfectants, neither MF nor UF can guarantee high water quality. In order to provide high water quality economically, MF/UF commonly combines other processes, such as adsorption, coagulation, or tight membranes (NF, RO), to form an integrated membrane system (IMS) (Fig. I. 5) [14]. MF/UF plays the role of controlling membrane fouling, while NF/RO removes a wide range of contaminants in IMS. If the IMS is used for treatment of groundwater, the objectives including precursor removal of disinfection by-products, color removal, removal of synthetic organic chemicals, and inorganics removal can be achieved. For treatment of surface water, the membrane plays an additional role of disinfection.

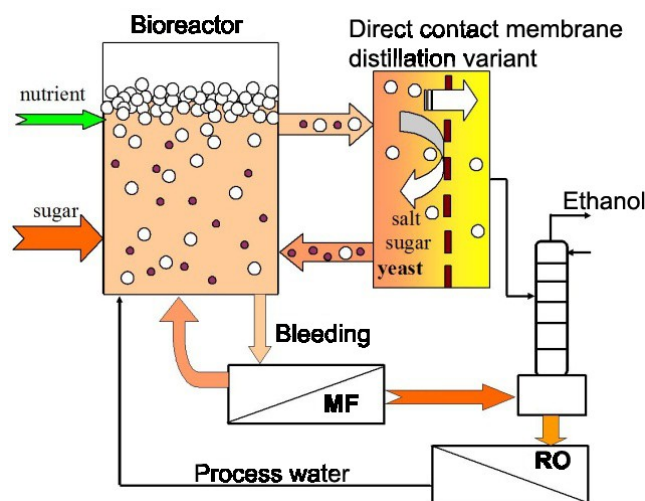


Fig. I. 5 An example of IMS for continuous ethanol production with effluents treatment [14].

In addition, in order to improve the treating efficiency of UF/MF systems, prefiltration for UF/MF systems is extensively applied. Pre-coagulation by adding coagulants and rapid mixing, such as FeCl_3 or FeSO_4 , alum, polyaluminum chloride, is

performed to increase the size of suspended and colloidal particle in feed waters [15]. A sequence process is commonly applied to settle out the flocs formed after adding coagulants, such as sedimentation and adsorption. A similar prefiltration called pre-flocculation is also used [16]. Other prefiltration methods, such as magnetic ion exchange [17] and ozonation [18] were also reported.

I.1.3 Transport in membranes

A membrane is essentially defined as a barrier that separates two phases (Fig. I. 6) and selectively restricts the transport of various chemicals. It can be differed in structures, materials, charges, etc. The membrane thickness can be as thin as hundreds of μm or as thick as several mm. Transport through a membrane can take place by convection or by diffusion of individual molecules, or it can be induced by an electric field (electrical potential difference) or concentration, pressure or temperature gradient (chemical potential difference).

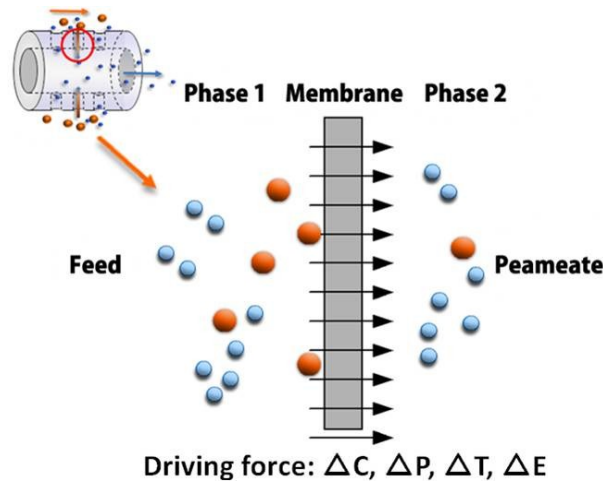


Fig. I. 6 Schematic representation of a two-phase system separated by a membrane.

The basic forms of transport through a membrane for a single component are summarized in Fig. I. 7 [19], although multi-component systems cannot be simply described due to the multi-interactions between components.

In passive transport, components or particles are transferred from a high potential to a low potential. A proportionality factor (A) was defined to determine how fast the component is transported through the membrane, or how small the resistance is exerted by the membrane as a diffusion medium. Besides of a simple diffusion process, another form of passive transport is “facilitated” or “carrier-mediate” transport. In this process, there is a carrier functioning in enhancing the transport, which interacts specially with single or multiple components in the feed solution and results in an increase in transport. In this case, transferring through a membrane against their chemical potential gradient is also possible.

Active transport processes are mainly process in living cell membranes, where the energy is provided by ATP. In these biological systems, specific and usually complex carriers are functioned.

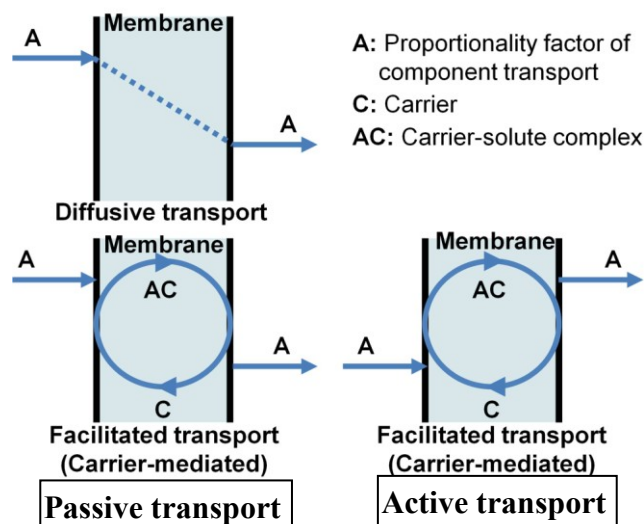


Fig. I. 7 Schematics of basic forms of passive transport and active transport and active transport.

Driving force is determined by the gradient in potential, or approximately by the difference in potential (ΔX , Fig. I. 6) divided by the membrane thickness (l).

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$$\text{Driving force} = \frac{\Delta X}{1} [\text{N/mol}] \quad (1)$$

The main potential difference are chemical potential difference ($\Delta\mu$) and electrical potential difference (ΔF), electrochemical potential is the sum of them ($\Delta\mu + \Delta F$).

Most transport processes take place because of a chemical potential difference $\Delta\mu$ under a constant temperature (T), while only in electrodialysis or other related processes an electrical potential difference works for transport. The chemical potential difference can be subdivided into a difference in composition and in pressure.

$$\Delta\mu = RT \ln a_i + V_i \Delta P \quad (2)$$

The composition contribution, which is given in terms of activities a_i , is equal to the product of RT and the logarithm of the composition. RT is equal to 2500 J/mol. The pressure contribution is equal to the product of the molar volume (V_i) and the difference in transmembrane pressure (ΔP). The molar volume of liquids is small, e.g. V (water) is $1.8 \times 10^{-5} \text{ m}^3/\text{mol}$ and V (organic solvent with MW 100 g/mol and density 1 g/ml) is $10^{-4} \text{ m}^3/\text{mol}$, meaning that applying ΔP over 50 bar would only provides $\Delta\mu$ 100 J/mol (water).

Transport processes through membranes cannot be considered as thermodynamic equilibrium processes, and thus only irreversible processes can be described thermodynamically. When providing driving force to an irreversible membrane transport process, the free energy is dissipated continuously and entropy is produced.

Transport occurs through the pores in porous membranes rather than the dense matrix and structure parameters, such as pore-size distribution, porosity and pore dimensions are important and have been taken into account in any model developed. On the other

hand, in dense membranes a molecule can only permeate if it dissolves in the membrane, whose solubility is determined by the affinity between the polymer (of a membrane) and the molecule with low molecular weight.

I.2 PREPARATION OF POLYMERIC MEMBRANES

A number of techniques are developed to prepare synthetic membranes (Table I.1) [1], some of which, such as sintering, stretching, track-etching (often polycarbonate), and phase inversion, are available for the preparing polymeric membrane. On the other hand, some techniques, such as template leaching, sol-gel process are only used for preparing inorganic membranes. This chapter is focus on methodology for polymer materials.

Table I.1 Several methods for preparing porous membranes.

Preparation processes		Applicable material examples	UF	MF	Properties
Phase separation	Nonsolvent-induced phase separation (NIPS)	polysulfone polyethersulfone polyacrylonitrile cellulose derivatives polyvinylidene fluoride	○	○	Asymmetric structures High permeability High porosity
	Thermally induced phase separation (TIPS)	Polyethylene polypropylene polyvinylidene fluoride		○	Generally high strength High porosity
Stretching (semicrystalline)		polyethylene polypropylene polytetrafluoroethylene		○	Slit-like pores
Nucleation track		polycarbonate, polyester		○	Uniform and straight pores
Sintered (particles)		Ceramics polymers	○	○	

I.2.1 Description of preparation methods

With sintering, stretching and track-etching, only porous membranes are prepared, while phase inversion can be used for preparing open-celled structures as well as dense structures. Composite membranes, which generally consist of a top separation layer

with thickness of several hundreds of nm and a porous support layer (probably of different materials), are usually prepared by multiple methods. The top separation layer can be prepared by coating, grafting, plasma polymerization, interfacial polymerization, in-situ polymerization [19].

Sintering technique is a simple method for preparing only microfiltration membranes with a general low porosity (approximately 10-20%). It is applied by compressing a powder consisting of particles of a given size and sintering at elevated temperatures. Polymers, which have high chemical and thermal resistance, are suitable to be used in sintering, such as polytetrafluoroethylene, polyethylene and polypropylene.

Stretching is only applied to semi-crystalline polymeric materials, such as polytetrafluoroethylene, polyethylene and polypropylene. Microfiltration membranes with pore-size between 0.1 μm and 3 μm and porosity up to 90% can be prepared.

Track-etching is able to prepare membranes with simplest pore geometry (i. e. parallel cylindrically shaped pores, uniform pore dimension). Pore-size of the membranes prepared by track-etching ranges from 0.02 μm to 10 μm , and surface porosity is generally low (approximately 10%).

Phase inversion is the most commercially available membrane preparation method, which is a very versatile technique allowing most kinds of morphologies to be prepared (See section I.2.2). Generally, the resultant membranes have a dense skin layer that is integrally bonded in series with a thick porous substructure. The skin and the substructure are composed of the same material. The skin layer, which contains the effective separation layer, is one of the key elements in determining membrane flux and separation factor for liquid separation, as well as permeance and selectivity for gas separation.

I.2.2 Phase inversion

Phase inversion is a process whereby a polymer is transformed in a controlled manner from a liquid to a solid state. The process of solidification is very often initiated by the transition from one liquid state into two liquids (liquid-liquid demixing). At a certain state during demixing, one of the liquid phases (the high polymer concentration phase) solidifies to form a solid matrix. The following phase inversion processes can be distinguished [19]: (1) precipitation from the vapor phase (vapor-induced phase separation (VIPS)); (2) precipitation by controlled evaporation; (3) thermal precipitation (thermally induced phase separation (TIPS)); and (4) immersion precipitation (liquid-induced phase separation). (1), (2) and (4) are called nonsolvent-induced phase separation (NIPS). The final morphology of the fibers and membranes obtained will vary greatly, depending on the properties of the materials and the process conditions.

1.2.2.a Basic principles of membrane formation by phase inversion

The system of the polymer systems are described by the following state functions [19]: internal energy (U), enthalpy (H), entropy (S) and free enthalpy (G). In a homogeneous solution, the free enthalpy of mixing (ΔG_m) at temperature T is given by:

$$\Delta G_m = \Delta H_m - T\Delta S_m \quad (3)$$

where ΔH_m is the enthalpy of mixing and ΔS_m is the entropy of mixing. When $\Delta G_m < 0$, two components (polymer/solvent) will mix spontaneously. Since ΔS_m is small for polymeric systems, solubility is determined by the ΔH_m .

When introducing an important solubility parameter δ , ΔH_m can be described as:

$$\Delta H_m = V_m (\delta_1 - \delta_2)^2 v_1 v_2 \quad (4)$$

$$\delta = [\Delta E/V]^{1/2} \quad (5)$$

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where V_m is the molar volume of the solution. δ_1, δ_2 represent the solubility parameters of two components, and v_1, v_2 represent the volume fractions. The solubility parameter was defined as the square root of the cohesive energy density ($\Delta E/V$), where ΔE is the energy of vaporization and V is the molar volume. When $\delta_1 \approx \delta_2$, ΔH_m approaches zero; a polymer and a solvent are miscible (ΔS_m is always positive).

When introducing the chemical potentials (μ), G_m can be given by the sum of the chemical potentials (the partial free enthalpy) per mole:

$$G_m = x_1\mu_1 + x_2\mu_2 \quad (6) \text{ (See Fig. I. 8)}$$

Where mixing n_1 mol polymer and n_2 mol solvent is described as:

$$\Delta G_m = n_1\Delta\mu_1 + n_2\Delta\mu_2 \quad (7)$$

The entropy of mixing can be expressed in volume fractions (ϕ):

$$\Delta S_m = -R (n_1 \ln\phi_1 + n_2 \ln\phi_2) \quad (8)$$

where R is the ideal gas constant. When $\Delta H_m = 0$, ΔG_m is given by:

$$\Delta G_m = RT(n_1 \ln\phi_1 + n_2 \ln\phi_2 + n_1\phi_1\chi) \quad (9)$$

χ is considered to be an excess parameter containing all non-ideality, and the chemical potentials change ($\Delta\mu$) can be expressed as:

$$\Delta\mu_1 = \mu_1 - \mu_1^0 = \left(\frac{\partial\Delta G_m}{\partial n_1}\right)_{P,T,n_2} = RT(\ln\phi_1 - (1 - \frac{V_1}{V_2})\phi_2 + \chi\phi_2^2) \quad (10)$$

$$\text{and } \Delta\mu_2 = \mu_2 - \mu_2^0 = \left(\frac{\partial\Delta G_m}{\partial n_1}\right)_{P,T,n_1} = RT(\ln\phi_2 - (1 - \frac{V_1}{V_2})\phi_1 + \chi\phi_1^2) \quad (11)$$

Fig. I. 8 shows ΔG_m versus ϕ curve at temperature T_1 and T_2 . ΔG_m exhibits an upward bend between ϕ^I and ϕ^{II} at T_1 . These two points on the tangent have the same derivative. Between them, there are always two points (e. g. ϕ_1 and ϕ_2) having the same chemical potentials (Fig. I. 8, left). Increasing the temperature will bring ϕ_1 and ϕ_2 closer, until eventually joining at one point, which is called critical point (at a) (at temperature

T_2 , Fig. I. 8, right). Plotting the locus of the minima in a ΔG_m versus ϕ gives a binodal curve (at different temperatures), and the locus of the inflection points is called the spinodal (Fig. I. 9).

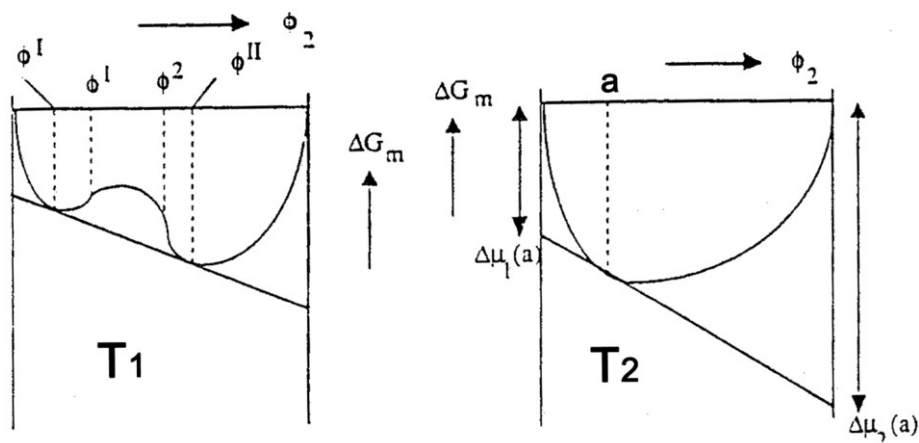


Fig. I. 8 Free energy of mixing as a function of composition for a binary mixture. ($T_1 < T_2$)

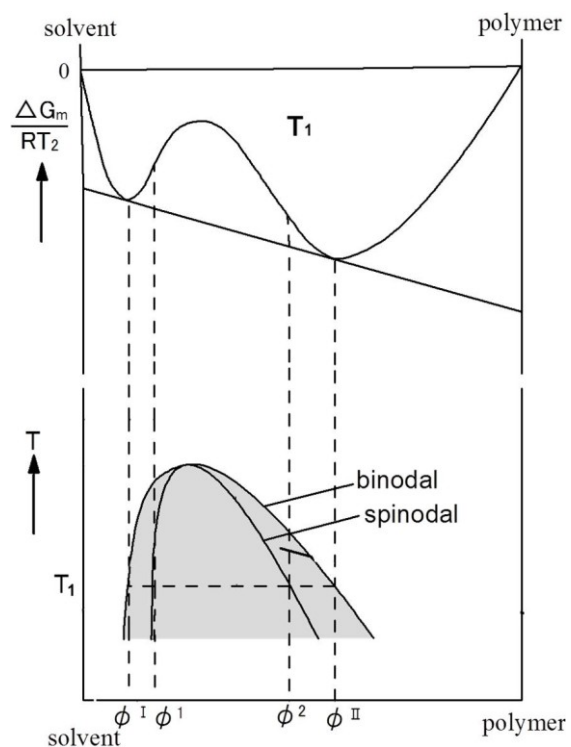


Fig. I. 9 Temperature-composition phase diagram for a binary polymer-solvent system.

I.2.2.b TIPS process

Thermally induced phase separation (TIPS) procedure is the simplest method to prepare porous membranes with pore-sizes of 0.01–1 μm [20]. The polymers for preparing a membrane by TIPS method can be crystalline or glassy polymers (See Table I. 1). The high temperature of the initial polymer solutions allows the use of high concentration of the polymers; therefore the membranes with high mechanical strength are usually obtained.

TIPS is induced from cooling a homogeneous polymer solution (using a high-boiling point solvent) at elevated temperatures. During the decrease of temperature, phase separation and solidification occurred. Solid–liquid (S-L) TIPS occurs when the polymer solidifies first, often by crystallization [21, 22]. Liquid–liquid (L-L) TIPS occurs when the polymer and diluent separate into two liquid phases before the polymer-rich phase solidifies (liquid-liquid demixing). Liquid–solid (L-S) TIPS occurs when the diluent crystallizes inside of a polymer-rich liquid matrix. Upon further cooling to solidify the polymer and subsequent extraction of the solvent, the nature of the resulting morphology depends upon the crystal structure and kinetics of crystal growth [21]. Upon extraction of the diluent, liquid–liquid TIPS often yields the cellular structure [23].

A binary phase diagram for TIPS in a polymer-solvent system is shown in Fig. I. 10. At a high temperature, the solution is homogeneous. During decreasing the temperature, the solution will separate into a polymer rich phase and a polymer lean phase first (L-L phase separation). As the temperature becomes lower, solidification of the crystallization along the crystallization line or glass transition of the polymer takes place. The binodal line shows the L-L demixing gap, and this region is bounded by the

spinodal line. The point that two curves coinciding with each other is the critical point (See section I.1.2.2.a). A membrane with good performances usually is prepared at the concentration higher than that of the critical point. The major factor determining whether S-L or L-L phase separation occurs involving a semi-crystalline polymer is (e. g. polyethylene, polypropylene, and aliphatic polyamides) the miscibility of the system, which is quantified as the interactions parameter of the polymer-solvent system. If the interaction between polymer-solvent is strong (small interaction parameter), S-L phase separation via polymer crystallization takes place, otherwise, the blend becomes unstable and undergoes L-L phase separation.

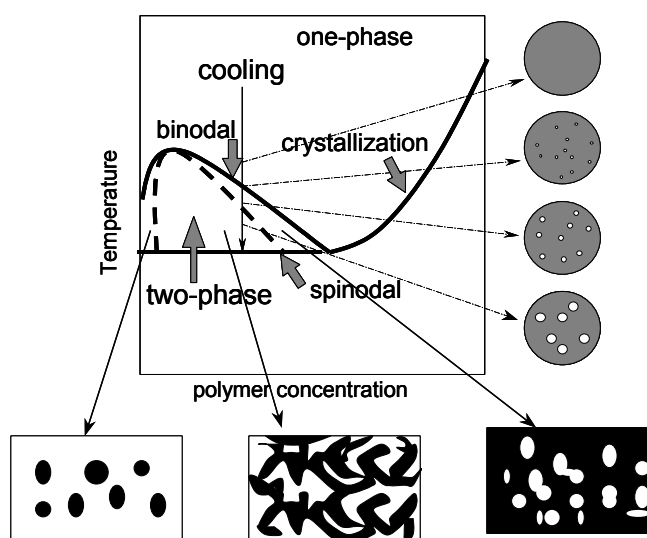


Fig. I. 10 Binary phase diagram for TIPS phase separation.

I.2.1.c NIPS process

Immersion precipitation (liquid-induced phase separation (LIPS)) is the first to be commercially explored and is one of the most popular membrane formation methods. Most of industrially produced membranes today are prepared by immersion precipitation. In contrast, the precipitation from the vapor phase (vapor-induced phase separation (VIPS)) is mostly applied as an assistant procedure in fabricating porous

membranes by LIPS. A NIPS process is a simple and cheap process allowing the preparation process at low temperature. The applicable polymeric materials are of broad range.

At least three components are used in a NIPS process [24]: (a polymer, a solvent and a nonsolvent, where the solvent and nonsolvent must be miscible with each other. Immersion precipitation can be regarded as a combination of nonsolvent penetration and solvent elimination. The simplest progress of preparing a membrane by NIPS can be described as follows:

(1) A desired shape is produced using a homogeneous polymer solution (flat-sheet film or tubular fiber) containing a polymer and a solvent; (2) A non-solvent is introduced into the system to exchange with the solvent in the polymer solution and to induce phase separation, and the porous structures of the polymer are prepared; (3) The solvent and nonsolvent in the membrane is removed by extracting or by other methods.

Regarding the NIPS process as an isothermal process, a ternary system can be plotted in a triangle (Fig. I. 11). Three corners represent the pure polymer, solvent, and nonsolvent. L-L phase separation occurs when a large amount of nonsolvent is added (Fig. I. 11, the path from area I to II), while S-L occurs when the concentration of the polymer is high concentration (Fig. I. 11, the path from area I to III).

It should be noted that the phase separation rate of a NIPS process is much slower than that of a TIPS process [25], which cause ununiformity phase separation at different positions of a membrane prepared by NIPS method and in turns of the membrane structures. At the surface of the membrane, where the polymer contacts with the nonsolvent first, liquid-liquid demixing starts immediately (less than 1 s) after immersion and becomes the top layer. All compositions directly beneath the top layer

can remain in the one-phase region (Fig. I. 11, area I) and still miscible. After a longer time interval, compositions beneath the top layer will cross the binodal line and demixing will start [19, 26]. As a result, a membrane prepared by NIPS method commonly has a top layer denser than the other parts. It should be noted that the top layer cannot form across the whole area of the membrane in most cases, which will allow a big amount of nonsolvent permeate into the polymer solution system. Eventually, this part around the nonsolvent will form large void spaces called macrovoids, which greatly affects mechanical strength and water permeability of a membrane. The asymmetric structures of a membrane prepared by NIPS can be controlled to an extent, by controlling the polymer-solvent-nonsolvent systems, evaporation of solvents (before immersion), temperature differences between polymer solution and nonsolvent (controlling the mass transfer rate), and etc.

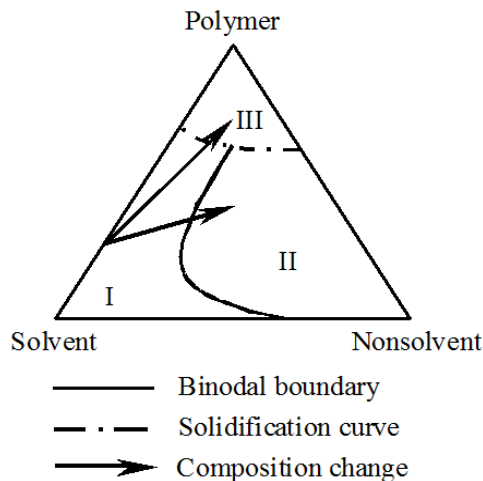


Fig. I. 11 Ternary phase diagram for NIPS process. I: homogeneous solution region; II: two-phase region; III: solidification region.

I.3 MEMBRANE FOULING

During a membrane filtration process, the membrane performances, especially the flux through the membranes, decrease over time. Such a behavior is mainly due to

membrane fouling. Membrane fouling is defined as the reversible deposition of retained particles, colloids, emulsions, suspensions, macromolecules, salts etc. on or in the membrane [27-30]. This phenomenon includes adsorption, pore blocking (using a porous membrane), precipitation and cake formation. Among all types of membranes, porous membranes (ultrafiltration, microfiltration) are mostly susceptible to fouling.

I.3.1 Fouling modes and main foulants

Fouling is a very complex phenomenon, and also a difficult problem to solve for membrane systems. To theoretically describe membrane fouling, the researchers generally divided fouling into three modes by foulant size and membrane pore-size: pore narrowing, pore plugging, and cake layer formation (Fig. I. 12).

When the foulant size in the feed solutions is much smaller than the membrane pore-size, foulants can absorb onto the surface or inside the pores of the membrane during filtration in most of the cases, which will narrow or constrict the membrane pores. The extent of pore narrowing is highly dependent upon the morphology of the membrane. In contrast, if the foulant size is almost the same with pore-size of the membrane, the foulants tend to block the pores. The modes of pore narrowing and pore plugging are also called pore clogging, and they both cause the decrease in membrane pore-size and porosity.

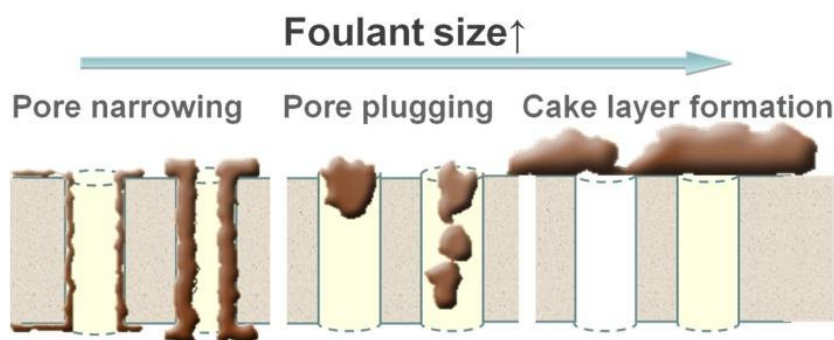


Fig. I. 12 Three membrane fouling modes.

The foulants in the feed solutions with the size larger than membrane pore-size tend to deposit onto the surface to form a cake layer. As a result of cake formation, permeation resistance increases significantly.

Although three modes are classified for membrane fouling, the actual fouling during filtration is very difficult to be distinguished accurately by particle-size, due to the complex foulant-foulant interactions or the complex porous structures of membranes. For example, the foulants with small size tend to form aggregates and cannot pass through the membrane pores in some cases. It is also possible that small foulants who have strong interactions with membrane materials may result in only surface deposition.

The foulants are typically colloidal materials of one sort or another and these properties and interaction with the membrane dominate fouling/cleaning processes. Colloids are defined as fine suspended particles in the size range of a few nanometres to a few micrometers.

The foulants are classified on the basis of fouling materials [31, 32]:

(1) Inorganic fouling due to deposition on membrane surface of inorganic scales (mainly BaSO_4 , CaSO_4 , CaCO_3); (2) Organic fouling due to natural organic material (NOM) found in the process stream (humic acids, protein and carbohydrate); (3) biofouling due to microbial attachment to membrane surfaces followed thereafter by their growth and multiplication in presence of adequate supply of nutrients in the pretreated feed or nutrients that deposited on membrane surfaces.

This thesis mainly focuses on organic fouling; several model foulants were used to study membrane fouling. Fouling caused by humic acid was studied in Chapter II. The degradation of organic matter, such as plants, in the soil produces a mixture of complex macromolecules called humic acid. These complex molecules have polymeric phenolic

structures with the ability to chelate metals. It is recommended that humic acids should be removed from process water before membrane filtration by complexation (i.e., flocculation/coagulation). Protein fouling is discussed in Chapters II, III and IV. The theory to classify membrane fouling by particle size seemed not quite suitable for studying protein fouling, due to the labile nature of protein and the strong interactions between protein molecules and most polymeric materials for membranes. Although commonly only trace amounts of protein exist in feed solutions (both in drinking water and wastewater), it still cause a big problem. In particular, extracellular polymeric substances, such as polysaccharides and amino sugars were found to play an important role in fouling. The colloidal fractions gave a high flux decline due to pore blockage.

I.3.2 Factors affecting membrane fouling

Membrane fouling is affected by the foulant, membrane properties, operation conditions and membrane module design, and etc.

The nature of foulants has large effects on membrane fouling. The foulants of inorganic or organic substances, the lability of the materials, the states of aggregates and shapes, the hydrophobicity of the materials, the net charge and so on will dominate the membrane fouling. Especially, there are various kinds of foulants existing in different water resources, which might have complex interactions and are almost impossible to be identified in industrial water treatments using membrane filtration. The substances that might cause a fouling problem in the water are also variable due to the natural or artificial reasons. The environments of the water resources may also affect membrane fouling and should be paid more attention. For example, the pH, the ion environment

(See Chapter II), the temperature and viscosity have significant effects on membrane fouling.

The membrane materials and morphologies, especially the surface structures, also greatly affect the membrane fouling. In order to avoid the attachments of most hydrophobic foulants, increasing the membrane hydrophilicity, as well as controlling the morphologies of membrane surfaces, becomes the most important subject in membrane engineering [33, 34].

The operation procedures, including the design of the module device for membranes, are also very important for controlling the membrane fouling. For example, to increase the flow velocity, or to keep the flow direction horizontal with the axis of membrane setting direction, the flow will have the potential to sweep the foulant deposits off the membrane surfaces and decrease the fouling. Recently, there are reports describing special devices in membrane modules for reducing the foulant deposition (e.g. a shaken device or a helical shaped module).

I.3.3 Analytical strategies of fouling and reduction of fouling

I.3.3.a Characterization of membrane fouling

Fouling resistance of a membrane has been studied for decades. The basic method is to perform filtration experiments using different pairs of model foulants and model membranes. For the characterization of fouling, the flux decline during filtration, the rejection variation of model foulants, permeance resistance and others are measured.

For studying the membrane after fouling, several techniques for analyzing the properties of a film are used for analyzing the surface of fouled membranes. Spectroscopy (e. g. scanning electron microscopy (SEM)) is commonly used to observe

the morphology change of the membranes after foulant deposition. It was believed that the smoothness of the membrane surface can influence the morphology of the fouling layer. Atomic force microscopy (AFM) technique is also employed to measure the surface roughness [35]. Fourier transform infrared (FT-IR) spectroscopy is used to provide insight into the chemical nature of deposits on membranes [36]. Deposits on a membrane surface can also be analyzed using SEM in combination with energy dispersive x-ray (EDS) combined with a microanalysis system permitting quantitative determination of elements [37]. Nuclear magnetic resonance (NMR) technique can also be employed to quantitatively measure the foulant layer [38].

Recently, some developed techniques, such as confocal laser scanning microscopy (CLSM) [39], fluorescence excitation emission matrix (EEM) [40], are used to visualize and characterize membrane fouling. P. Bacchin et al. developed a microfluidic filtration device made of polydimethylsiloxane, which allowed in-situ clogging observation at the pore scale during filtration [41].

I.3.3.b Cleaning and prevention of fouling

Proper pretreatments can prevent membrane fouling effectively. The common pretreatments include: heat treating, ionic or pH adjusting, adding chelating agents, chlorinating, cleaning using active carbon, chemical treating and prefiltrating. Simple procedures sometimes can be very effective. For example, pH adjustment before filtration of solutions containing proteins is very important, because severe protein fouling occurs at the pH value corresponding to the isoelectric point of protein, due to the neutral net charge of proteins. As people know that microfiltration is commonly used as a prefiltration method for RO systems and that flocculation can also be used as pretreatment for RO systems. A flocculator is able to remove suspended solids, organics,

and phosphorus from water [42]. Several types of coagulation systems are also very effective pretreatments for preventing membrane fouling.

In any industrial membrane processes, periodical cleaning is carried out to restore membrane performances. The cleaning methods can be divided to physical cleaning and chemical cleaning. Physical cleaning methods include for example: hydrodynamic forward or reverse flushing, permeate back pressure, air spurge and automatic sponge ball cleaning. These methods depend on a mechanical treatment to dislodge and remove foulants from the fouled membranes safely, although usually complex design and control of equipment is demanded. For a hollow-fiber membrane, backwashing periodically, often with chemical cleaning agents, is effective and easy to use (See Chapter II and III).

I.1.4 PURPOSE OF THIS STUDY

This dissertation focuses on microfiltration (MF) and ultrafiltration (UF) polymeric hollow-fiber membranes for water treatment. The main aim of the dissertation is to study the membrane fouling phenomena, including exploring the fouling mechanism, developing techniques to restrain fouling, and cleaning fouled membranes.

The purpose of the first part including Chapter II and III is to better understand the organic membrane fouling during filtration. The fouling phenomenon has been studied for decades and there still remain the uncertainties for MF and UF systems, because there are multiple factors affecting the fouling during filtration and also complexity in the fouling mechanism. In Chapter II, I studied the multiple effects of ionic environments on the fouling caused by model foulants of humic acid and several proteins and studied the cleaning after filtration.

Chapter I

Chapter III is the direct visualization of the internal fouling caused by model protein foulants and a model polysaccharide foulant using fluorescent microscopy, in order to provide the insights for fouling mechanism.

Chapter IV is to prepare a surface-reorganized hollow-fiber membrane using two types of phase separation and dip-coating. This chapter aims to control the pore-size, to obtain high mechanical strength, and to functionalize the membrane surface, which leads to high water permeability, high separation performance and high anti-fouling properties.

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Chapter II

Effect of ionic environments on fouling of hollow-fiber ultrafiltration membrane

II.1 INTRODUCTION

Ultrafiltration (UF) hollow-fiber membranes are widely used for water purification and solution clarification in the food industry. During the past few decades, membrane performances have been greatly improved [1-2]. ultrafiltration membranes still inevitably suffer from fouling, possibly because of strong interactions between various substances and the membranes.

Natural organic matter (NOM) is considered to be a major membrane foulant in drinking water production. Several mechanisms for NOM fouling, including size exclusion and electrostatic repulsion, have been proposed [3]. For example, Mallevalle et al. [4] showed that organic matter, including humic acid, serves as a "cement" for inorganic clays in the cake layer formed on a membrane surface. Lahoussine-Turcaud et al. [5] found that the flux decline during UF operation with Seine River water was primarily caused by the deposition of natural organic macromolecules, in particular humic substances, on membrane surfaces.

Protein fouling in particular is considered as a serious problem, but is a poorly understood phenomenon during membrane processes [6-9]. The labile nature and complex structures of protein molecules present additional challenges in fouling characterization. Protein denaturation and protein–protein interactions also increase the difficulties faced in understanding protein fouling [10-12]. To date, various types of

proteins have been used to explore the protein fouling of membranes [13-15]. These reports describe irreversible and severe fouling, even when the pore size of a membrane is much larger than the protein molecule [16], indicating that protein–protein and membrane-protein interactions play a role in the fouling.

There are multiple inorganic and organic compounds present in the aqueous process solutions that are subjected to membrane filtration in industry [17]. However, there have been only a limited number of reports on the effects of the mutual interactions of different solutes on membrane fouling. In this study, we aimed to investigate the effects of metal ions and metal colloids on the fouling of hollow-fiber UF membranes caused by a type of NOM humic acid and various proteins. We used polyethersulfone (PES) hollow-fiber UF membranes with various molecular weight cut-offs (MWCOs). The important mutual interactions between the various solutes were investigated.

II.2 EXPERIMENTAL

II.2.1 Materials

Commercial PES hollow-fiber membranes with MWCOs of 10, 150, and 300 kDa were purchased from Daicel Chemical Industries, Osaka, Japan; the original pure water permeabilities of the membranes were 160 ± 50 , 560 ± 50 and 850 ± 50 L/(m²·h·atm), respectively.

Humic acid, EDTA (Ethylenediaminetetraacetic sodium), and Bradford reagent for protein were purchased from Aldrich Chemical Co. (Milwaukee, WI). Triethanolamine-HCl was purchased from Tokyo Chemical Industry Co. (Tokyo, Japan). All other chemicals were purchased from Wako Pure Chemical Industries, Japan). The water used was high-quality deionized water (DI water, $>15 \text{ M}\Omega \text{ cm}^{-1}$) produced by an

Elix-5 system (Millipore).

II.2.2 Membrane filtration and characterization

Humic acid was used as a typical NOM foulant in filtration experiments. Fresh humic acid solutions (50 ppm) were prepared by dissolving humic acid in 0.5 mM NaHCO₃ solution, which was used without further purification. In the experiments using with salts or EDTA, salts or EDTA were dissolved in homogeneous humic acid solutions to match given concentrations of metal ions. The pH of the solutions was adjusted to 6.75 ± 0.05 using small amounts of 0.1 M HCl or NaOH solution.

Three types of proteins were used as model protein foulants in filtration experiments, respectively. Bovine serum albumin (BSA, MW 67000 Da), lysozyme (from chicken egg white, MW 14800 Da), and casein (from bovine milk, MW 19000–25000 Da) were used. Solutions containing BSA (500 ppm) or lysozyme (100 ppm) were prepared by dissolving weighed proteins in 50 mM Tris-HCl buffer solution (pH 7) and stirring for 1 h at 25°C to obtain homogeneous solutions. A casein solution was prepared by dissolving weighed casein in a 50 mM triethanolamine-HCl/NaOH buffer solution (pH 7) and stirring for 3 h at 25°C to obtain a homogeneous solution; the triethanolamine-HCl/NaOH buffer was used because it does not interfere with protein microassays performed using the Bradford method. To prepare protein solutions containing metal ions, inorganic salts (FeCl₃ or CaCl₂) were first dissolved in 0.1 M hydrochloric acid solutions to obtain a given concentration of metal ions, and small volumes of these solutions (approximately 0.05 vol% of the volume of the protein solutions) were then added to the protein solutions, to obtain the desired final concentrations of metal ions. To prepare a protein solution with a cheating agent EDTA

or a reducing agent dithiothreitol (DTT), weighted EDTA or DTT was dissolved to the solutions. The solutions were stirred for 3 h to ensure that no precipitation occurred, and that the pH met the desired value. The protein solutions were used for filtration experiments immediately after their preparation, without storage or purification. The concentrations of metal ions used in the present study were chosen to be very low (up to 1 mM), to minimize the effects of the ionic strength on the solubility of the proteins, and to mimic realistic possible metal concentrations in industrial water treatment processes.

A PES membrane with a MWCO of 150 kDa was used to filter a BSA solution (500 ppm), and a PES membrane with a MWCO of 10 kDa was used to filter a lysozyme solution (100 ppm), because of the difference in the molecular weights of the proteins. Since casein tends to form stable micelles with sizes above 100 nm [18], a solution containing 5 ppm casein was filtered through a PES membrane with a MWCO of 300 kDa, which is more than 10 times larger than the size of a single casein molecule.

DI water was permeated through PES hollow fiber membranes for 10 min at a flow rate of 15 ml/min at 0.5 atm (the total permeated volume was approximately 30 ml for a membrane with a MWCO of 300 kDa) before use, to remove the coating and wetting agents from the membranes. Filtration experiments were performed at room temperature, using a laboratory-scale apparatus developed in our laboratory (Fig. II. 1) [19]. The apparatus consisted of a minimodule device that contained a single hollow fiber membrane 10.6 cm in length, with an outer diameter of approximately 1.3 mm and an inner diameter of approximately 1.0 mm. The transmembrane pressure was controlled by nitrogen gas, via a back-pressure valve. Each filtration experiment was carried out by forcing a feed solution to permeate from the outside to the inside of the hollow fiber membrane at a flow rate of 15 ml min⁻¹ at 0.5 atm transmembrane

pressure; under these conditions, the flow velocity of pure water in the module was approximately 0.04 m/s for a membrane with a MWCO of 300 kDa. Prior to filtration experiments, DI water was passed through the membrane to measure the pure water permeability J_0 . The filtration of a solution containing a model foulants was carried out to measure the variation in flux of a protein solution with time, J .

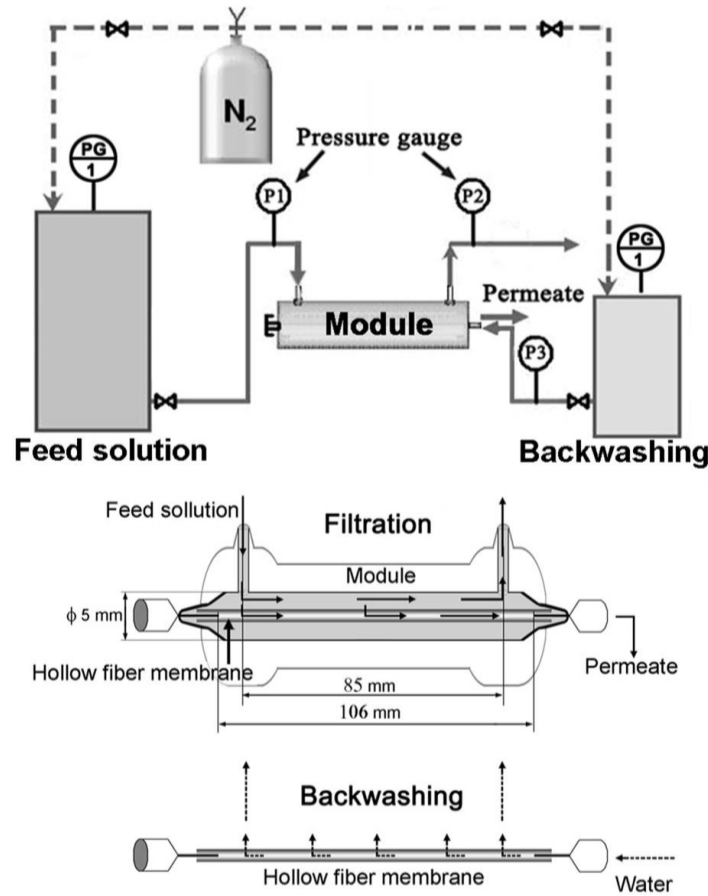


Fig. II. 1 Schematic diagram of minimodule laboratory scale apparatus for membrane filtration and backwashing.

The rejection, R , of a model foulant by the membrane was defined as follows;

$$R = \frac{1 - C_p}{C_0} \times 100 \text{ [\%]},$$

where C_p is the concentration of the foulant in the permeate, and C_0 is the concentration of the foulant in the initial feed solution. The humic acid concentration

was measured from the absorbance at 254 nm using a UV spectrophotometer (GE-Healthcare) [20]. The concentrations of BSA and lysozyme were determined by measuring the absorbance at 280 nm, and that of casein was determined using a Bradford method microassay.

For the backwashing of the fouled membranes, the membranes that were filtrated using protein solutions for 1 h were backwashed for 1 min using a flow rate of 15 ml/min, at a transmembrane pressure of 1 atm, using DI water (Fig. II. 2). The restorable flux decline was defined as reversible flux decline (RF), the non-restorable flux decline as irreversible flux decline (IF), and the sum of both as the total flux decline (TF). RF, IF and TF are illustrated in Fig.2.

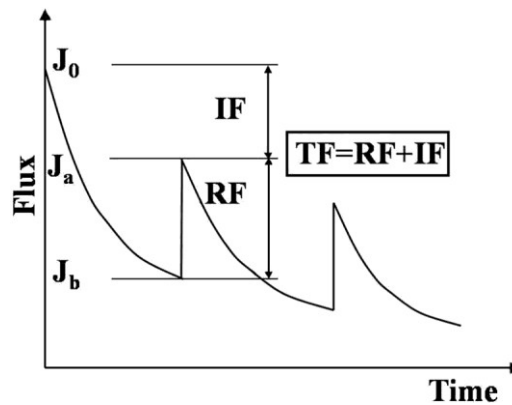


Fig. II. 2 Model profile of permeate flux in relation to irreversible and reversible fouling, through backwashing.

$$RF = J_a/J_0 - J_b/J_0 \quad (1)$$

$$IF = 1 - J_a/J_0 \quad (2)$$

$$TF = RF + IF = 1 - J_b/J_0 \quad (3)$$

where J_a and J_b indicate permeate flux after and before backwashing, respectively, and J_0 indicates the initial permeate flux. The backwashing efficiency (nRF) and normalized irreversible (nIF) flux decline were calculated by the following equations:

$$nRF = RF/TF = (J_a - J_b)/(J_0 - J_b) \quad (4)$$

$$nIF = IF/TF = (J_0 - J_a)/(J_0 - J_b) \quad (5)$$

The sum of nRF and nIF is unity.

The fractionation method developed by Aiken [21] was used to determine the apparent molecular weight distributions of foulants. Humic acid samples were fractionated using three ultrafiltration centrifugal filters (Amicon Ultra-4 centrifugal Filter Devices, Millipore) with nominal molecular weight cutoffs 10, 50 and 100 kDa, and two cellulose microfiltration membranes with pore sizes 0.1 and 0.45 μm (DURAPORE membrane filters, Millipore). The apparent molecular weight distributions were calculated from the difference in the humic acid concentrations between adjacent filtrate fractions.

To obtain dry membranes, the hollow fiber membranes were frozen in liquid nitrogen and dried with a freeze dryer (FD-1000, EYELA, Tokyo). The dry membranes were fractured in liquid nitrogen and sputter coated with Au/Pd for field emission-scanning electron microscope (FE-SEM) observation, while samples for EDS (energy dispersive X-ray spectrometry) analysis were sputter coated with carbon. The cross-sections and the surfaces of the hollow fiber membranes were examined using FE-SEM (JSM-7500F JEOL, Tokyo). Elemental compositions of the fouled layers were determined by EDS.

Metal contents in humic acid were measured using the inductive coupled plasma (ICP) technique (ICP-AES, SII, Tokyo). The samples for ICP measurements were prepared by dissolving humic acid in DI water without using buffer solutions.

To determine the size of the colloids in solution, dynamic light scattering (DLS) measurements were carried out at room temperature, using an ELSZ-2 instrument

(Otsuka Electronics Co. Ltd, Osaka, Japan).

II.3 RESULTS AND DISCUSSION

II.3.1 Effect of metal ions and metal colloids on humic acid fouling

Some multivalent metal ions can form a complex with, primarily, the carboxylic groups of humic acid, resulting in larger aggregates of humic acid in the deposit on a flat membrane surface [20]. However, the fouling of a hollow fiber membrane might be different from that of a flat membrane, because of the fluid flow lateral to the surface of the hollow fiber membrane. To study the effects of various metal ions on humic acid fouling of a hollow fiber membrane, cross-flow filtration of 50 ppm humic acid solutions using a polyethersulfone hollow fiber membrane at 0.5 atm were carried out for 2 h with periodic backwashing (1 min) after 1 h filtration. The relative flux (J/J_0) and rejection (R) data are shown in Figs. II. 3a and 3b, respectively.

In each case, at least two experiments were carried out to confirm reproducibility. The initial flux of water was $850 \pm 50 \text{ L (m}^2\cdot\text{h}\cdot\text{atm)}^{-1}$. The flux of humic acid solution (no additive, solid circles in Fig. II. 3a) decreased with time due to the membrane fouling. The flux decline was more rapid in the presence of additive metal ions (0.025 mM) than in the case of pure humic acid solution. In particular, the presence of FeCl_3 immediately reduced the flux to nearly one fifth within 1 h. The addition of Ca^{2+} reduced the flux more rapidly than without metal ions, but less rapidly than the addition of FeCl_3 . The presence of Mg^{2+} and Na^+ also resulted in rapid decline of the flux, however in both cases the rate of decrease was smaller than for FeCl_3 . Thus all of the metal ions that were added enhanced humic acid fouling during ultrafiltration using the hollow fiber membrane. These results and reported data suggest that membrane

fouling was accelerated by formation of large aggregates of humic acid due to the addition of metal ions. Multivalent metal ions tend to form complexes with carboxylic acid groups to produce intermolecular bonds between organic compounds [20, 22]. This is one of the possible reasons for the presence of FeCl_3 remarkably accelerating humic acid fouling. Na^+ , on the other hand, is a monovalent cation that does not have a strong ability to combine with carboxylic groups [22]. However, the presence of the monovalent cation would increase ionic strength, and thus decrease the repulsive force between negatively charged humic acid molecules, resulting in compaction of the humic acid deposit. Furthermore, the similar backwashing efficiency in all cases implies that the metal ions did not affect the interaction between the humic acid deposit and the membrane material.

The rejection of humic acid in the presence of FeCl_3 was higher at the beginning of filtration than that without metal ions, and the rejection rapidly increased with filtration time (Fig. II. 3b). After backwashing the rejection decreased to the initial level. These results suggest that large aggregates of humic acid initially formed in the feed solution due to FeCl_3 and that fouling occurred mainly on the membrane surface. That is the reason that the rejection easily recovered simply by backwashing. In the case of humic acid solution with Ca^{2+} , the rejection started from a low value ($\sim 15\%$) and suddenly increased to more than 60%. This indicates that the humic acid aggregates with Ca^{2+} were not as large as with FeCl_3 . Backwashing also decreased the rejection to the initial value. Although the addition of FeCl_3 and Ca^{2+} promoted membrane fouling, the rejection properties of the membrane could be easily restored by backwashing, probably due to the large cake layer of humic acid (discussed later) on the membrane surface that affected the rejection and was easily detached by backwashing. The

presence of Mg^{2+} and Na^{+} also resulted in slightly higher rejection than in the absence of metal ions. The difference in the fouling phenomena for different metal ions might be due to the different ability and different manner of metal ions to form a complex with humic acid. It should be noted that backwashing did not recover the flux completely in any cases, because irreversible fouling occurred [19].

In special, FeCl_3 itself supposed to cause fouling (See section II. 3.5 and 3.6). Iron should have existed in three states in the solution: oxide-hydroxide colloids, oxide colloids, and low concentrations of solubilized Fe^{3+} (nanomolar level). Although the iron was expected to exist as iron colloids (oxide-hydroxide and oxide colloids), DLS measurements could not detect colloidal nano- or microparticles, probably because of their low concentration.

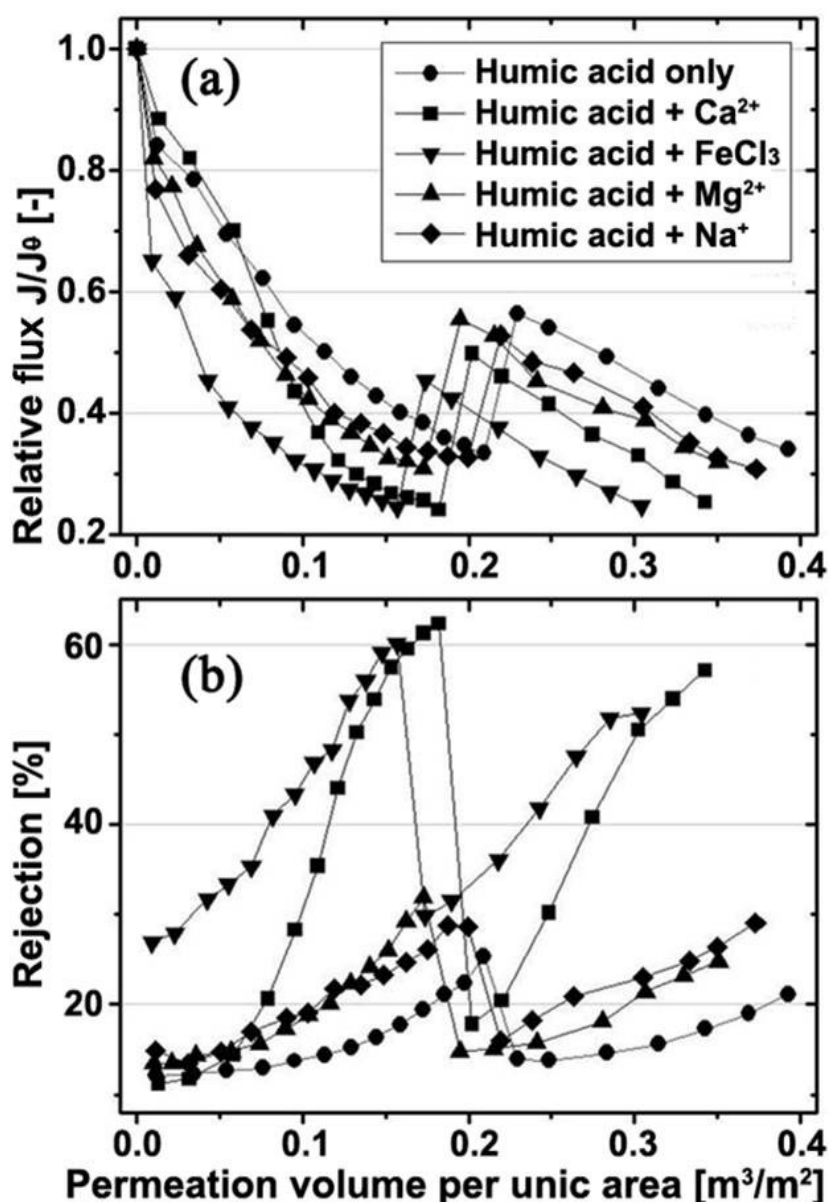


Fig. II. 3 Effect of various additive metal ions (0.025 mM) on the water flux and the rejection during filtration. (a) Relative flux J/J_0 , and (b) rejection.

Figure II. 4 shows the effect of Ca^{2+} concentration on humic acid fouling. The flux decline became steeper as the Ca^{2+} concentration increased from 0.025 mM to 0.5 mM, and the rejection of humic acid also increased with the increase of Ca^{2+} concentration. The difference of the initial rejections of the solutions with added Ca^{2+} is a reflection of the differences in physical size of the humic acid aggregates formed in the presence

of Ca^{2+} . By increasing the Ca^{2+} concentration backwashing also became less effective for flux recovery. This implies that the interaction between the membrane material and humic acids also became stronger with increasing concentration of Ca^{2+} . These results suggest that the concentration of metal ions is a critical factor for humic acid fouling, and that high Ca^{2+} concentration makes the fouling more irreversible.

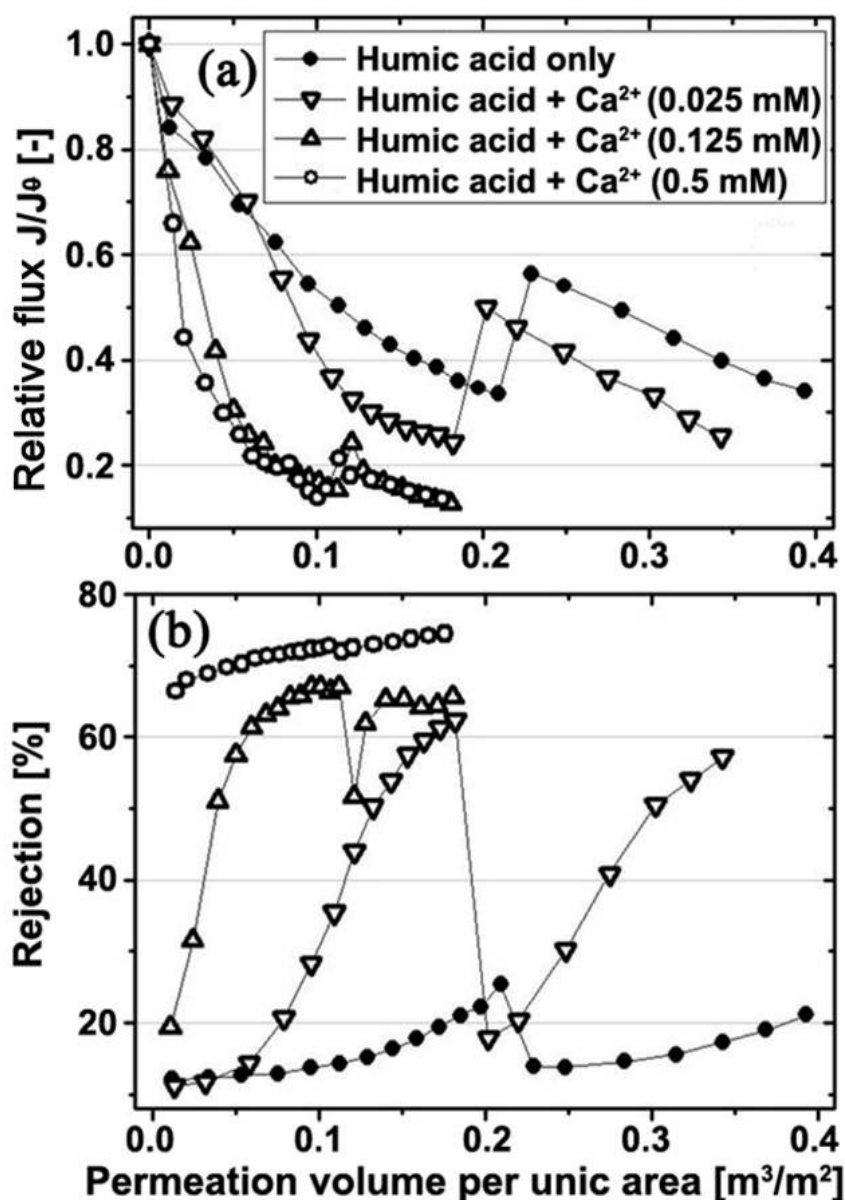


Fig. II. 4 Effect of different Ca^{2+} concentrations on the water flux and the rejection during filtration. (a) Relative flux J/J_0 , and (b) rejection.

Figure II. 5 shows photographic and FE-SEM images of a clean membrane and membranes fouled with humic acid (50 ppm, with and without additives, for 2 h permeation). The membrane fouled with humic acid became yellowish (Fig. II. 5-2(a)) and the membrane fouled with humic acid plus Ca^{2+} became dark brown (Fig. II. 5-3(a)), while the original clean membrane was white (Fig. II. 5-1(a)). The fouling promoted by Ca^{2+} could be observed with the naked eye, and FE-SEM observation also revealed the deposits of foulants on the membrane. There were foulant deposits (cake layer) 1 μm thick on the outer surface after the pure humic acid fouling, but the foulant deposit did not cover the whole outer surface under those experimental conditions. In the case of humic acid fouling in the presence of Ca^{2+} , a 5 μm thick cake layer spread over the membrane surface. Thus the presence of Ca^{2+} strongly expanded the foulant cake layer.

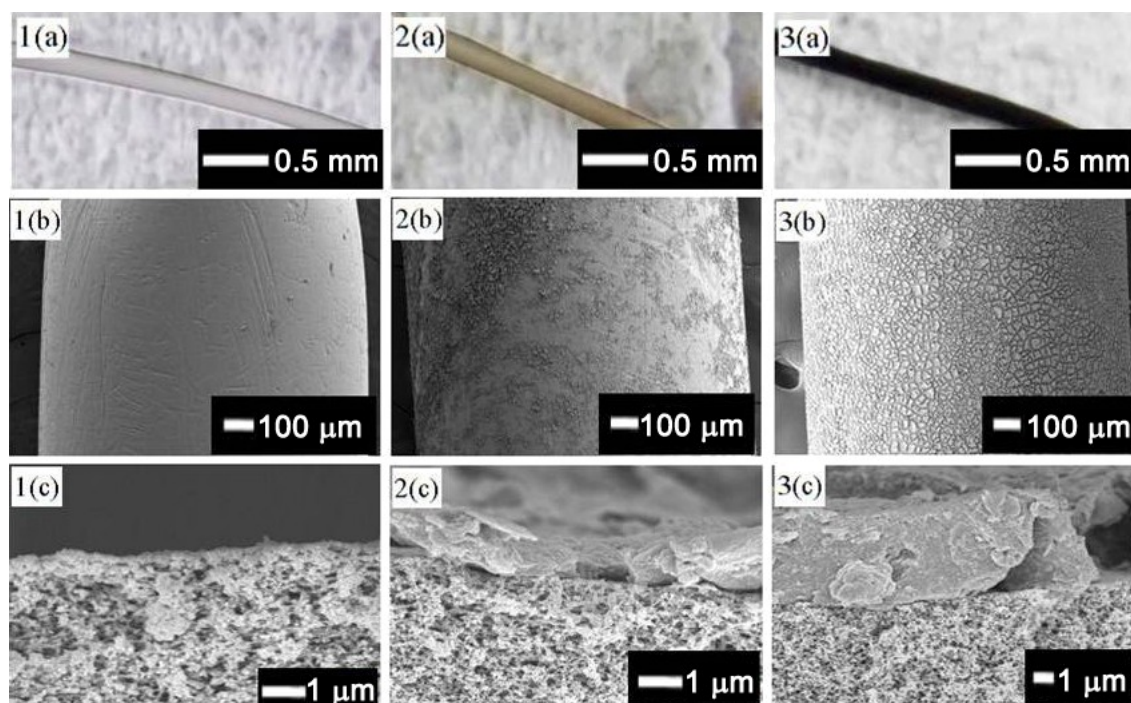


Fig. II. 5 Digital and FE-SEM images of the whole (a), the outer surface (b) and the cross section (c) of the membranes. (1) Membrane before use, (2) membrane fouled by a humic acid solution, and (3) membrane fouled by a humic acid solution with 0.5 mM Ca^{2+} .

II.3.2 Metal ions and metal colloids in humic acid and fouling layer

The metal content of the humic acid used in this study was measured using ICP-AES. The results shown in Table 1 confirmed the presence of various metals in the humic acid, which could cause the initial aggregation of humic acid and affect the humic acid fouling to some extent in the absence of added metal ions.

Table II. 1 Metal contents of humic acid.

	Ca	Fe	Mg	Al	Na	Cu
Amount in humic acid (wt%)	2	0.4	0.3	1.1	0.5	<0.01

Elemental analysis of the cake layer on the fouled membrane was carried out by EDS. Figure. II. 6 shows the presence of Ca, Fe and Mg in the surface of the cake layer, which is consistent with the original metal composition in the humic acid.

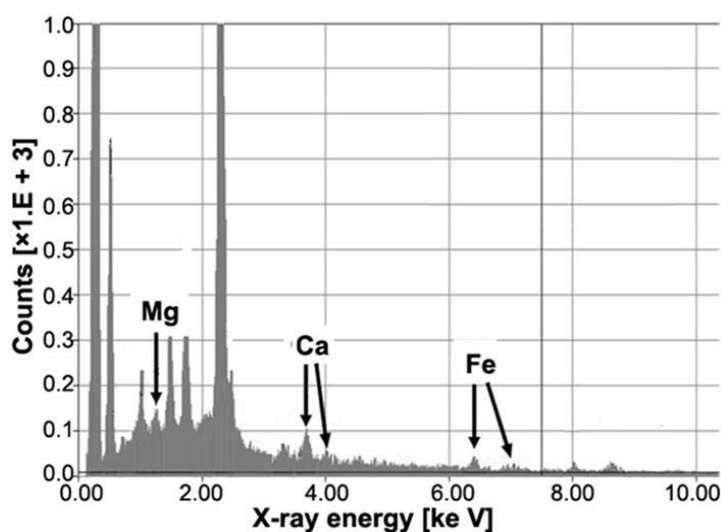


Fig. II. 6 EDS spectra of a membrane fouled by a humic acid solution. The x axis corresponds to x-ray energy (keV) and the y axis to counts.

According to the above investigations, metal ions seem to play important roles in the humic acid fouling. In the practical water purification process, metal ions present in raw water would enhance NOM fouling by complexing with humic substances. Contamination of metal ions leached from plant equipment should also be avoided.

II.3.3 Effects of EDTA on humic acid fouling in the presence of metal ions and metal colloids

The usefulness of EDTA in preventing fouling could arise because of its role as a chelating agent [23, 24], i.e. its ability to sequester metal ions such as Ca^{2+} and FeCl_3 . After complexation with EDTA, metal ions were expected to have reduced reactivity to humic acid.

Figure II. 7 shows the effect of EDTA on the relative f (J/J_0) and the rejection (R) of 50 ppm humic acid, with and without Ca^{2+} , for 2 h permeation. Addition of EDTA to the humic acid solution without Ca^{2+} slightly improved the permeability of the humic acid solution (open circles in Fig. 7), probably due to chelation of the metal ions originally present in the humic acid. The addition of EDTA to the humic acid solution with Ca^{2+} markedly improved the permeability. The color of the fouled membrane was yellowish in the presence of Ca^{2+} and EDTA, which was similar to that of the membrane fouled with humic acid only. However, EDTA did not restore the permeability of the solution containing Ca^{2+} to that of the pure humic acid solution. Although EDTA readily forms a complex with Ca^{2+} ion, there would be competitive complex formation between Ca^{2+} –EDTA and Ca^{2+} –humic acid. That is one possible major reason why the addition of EDTA did not entirely restore the permeability of the solution containing Ca^{2+} . The rejection measurements revealed that the addition of EDTA to the Ca^{2+} –humic acid solution decomposed the Ca^{2+} –humic acid complex, resulting in a rejection profile similar to that of the humic acid solution. It should be noted that there was also a difference in the rejection profile between the Ca^{2+} –humic acid solution with EDTA and the humic acid solution. This difference also indicates that a small proportion of humic acid formed a complex with Ca^{2+} even in the presence

of EDTA.

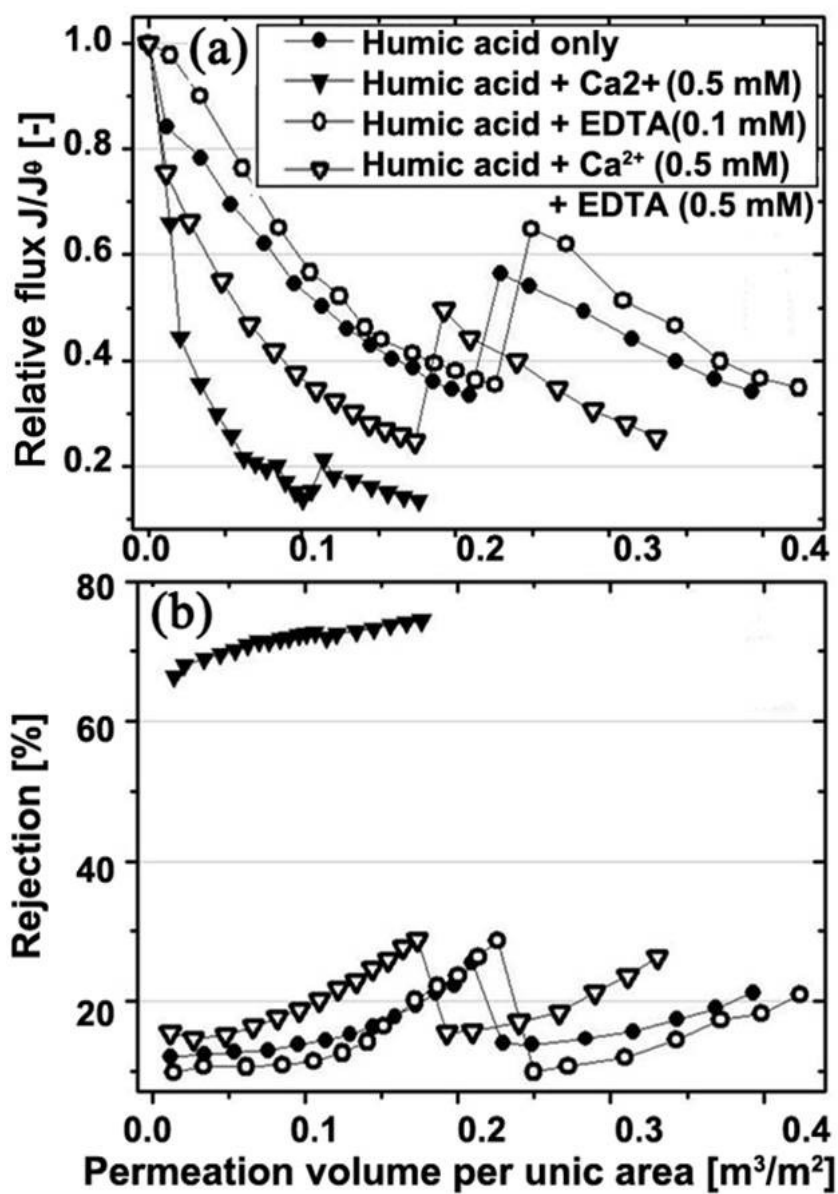


Fig. II. 7 Effect of EDTA on humic acid fouling with and without Ca²⁺ during filtration. (a) Relative flux J/J_0 , and (b) rejection.

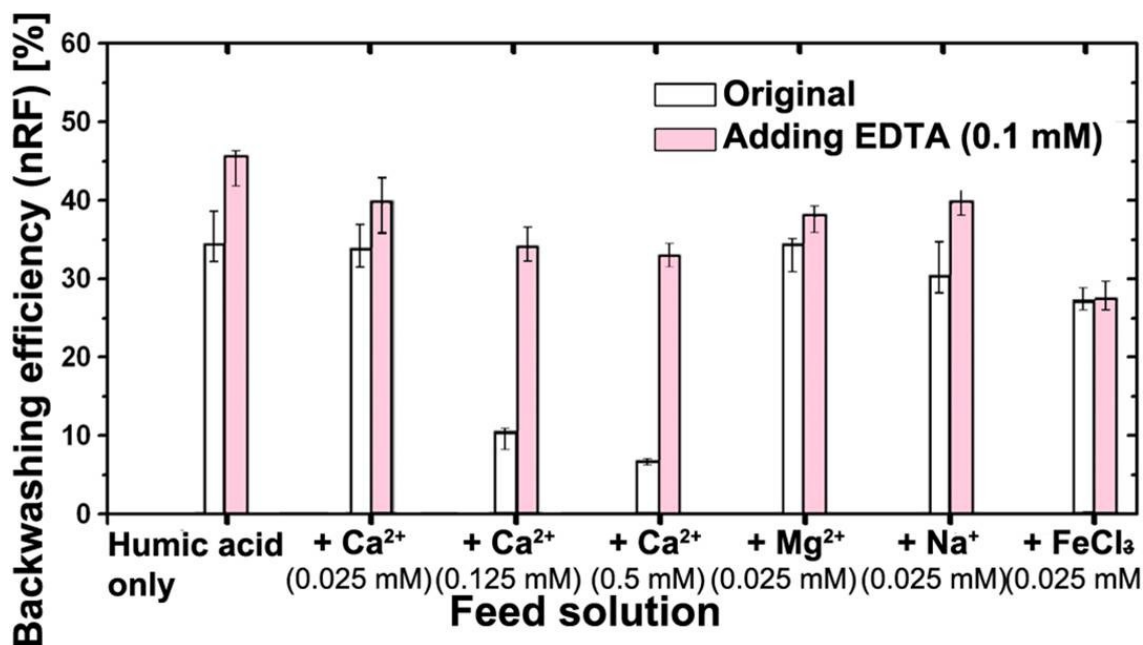


Fig. II. 8 Backwashing efficiency (nRF) of membranes fouled by humic acids with various additives. Backwashing was carried out using distilled water for 1 min, after 1h filtration.

Backwashing is one of the practical methods to recover a fouled membrane, and backwashing with DI water was carried out for membranes fouled under various conditions. In the present study, the recovery of flux induced by backwashing was considered as reversible fouling, and the unrecovered flux was regarded as irreversible fouling.[17]. The backwashing efficiency (nRF) under various filtration conditions is summarized in Fig. II. 8. The nRF values for the humic acid solution with 0.025 mM metal ions were almost the same as for the pure humic acid solution, which has two possible implications. One is that the compactivity of the cake layer on the membrane surface was similar; the other is that metal ions used at this concentration did not affect the interaction between the humic acid aggregate and the membrane material. The presence of Ca²⁺ at high concentration (0.5 mM) drastically decreased nRF. The high Ca²⁺ concentration might contribute to the connection between the humic acid aggregates in the cake layer, promoting irreversible fouling. The addition of EDTA to

the feed solutions, except for the case of humic acid with FeCl_3 , increased the nRF. That effect was possibly due to EDTA making the fouling layer less compacted and hence much easier to wash off by backwashing.

In the above discussion, we attributed the enhanced fouling to large aggregates of humic acid due to the additive metal ions. To determine the aggregation of humic acid, we measured the apparent molecular weight distribution (MW) of pure humic acid and humic acid with additives (Fig. II. 9). In relation to the MW distribution of pure humic acid (Fig. II. 9-a), the fractions of small (<10 kDa) and large (>0.1 μm) molecules were under 10% (based on the absorbance). The major fractions of MW between 10 kDa and 0.1 μm contributed more than 80%. After adding 0.025 mM Ca^{2+} (Fig. II. 9-b), only the fraction between 10 and 50 kDa decreased, and the large MW (0.1-0.45 μm) fraction increased. This means that aggregation of humic acid was enhanced by adding Ca^{2+} .

The addition of EDTA to a humic acid solution (Fig. II. 9-c) decreased the fraction with MW between 100 kDa and 0.1 μm , while the number of molecules between 50 and 100 kDa increased. In the case of a humic acid solution with Ca^{2+} , the effect of EDTA (Fig. II. 9-d) was more obvious. The fractions between 0.1 μm and 0.45 μm and between 100 kDa and 0.1 μm shifted to those between 50 and 100 kDa, and 10 and 50 kDa. The decrease of the apparent MW of humic acid on addition of EDTA was due to inhibition of the interaction between the humic acid molecules and metal ions. The initial aggregation and metal ion-induced aggregation of humic acid are also of great importance in membrane fouling. That is one of the major reasons why the addition of EDTA was effective in inhibiting membrane fouling.

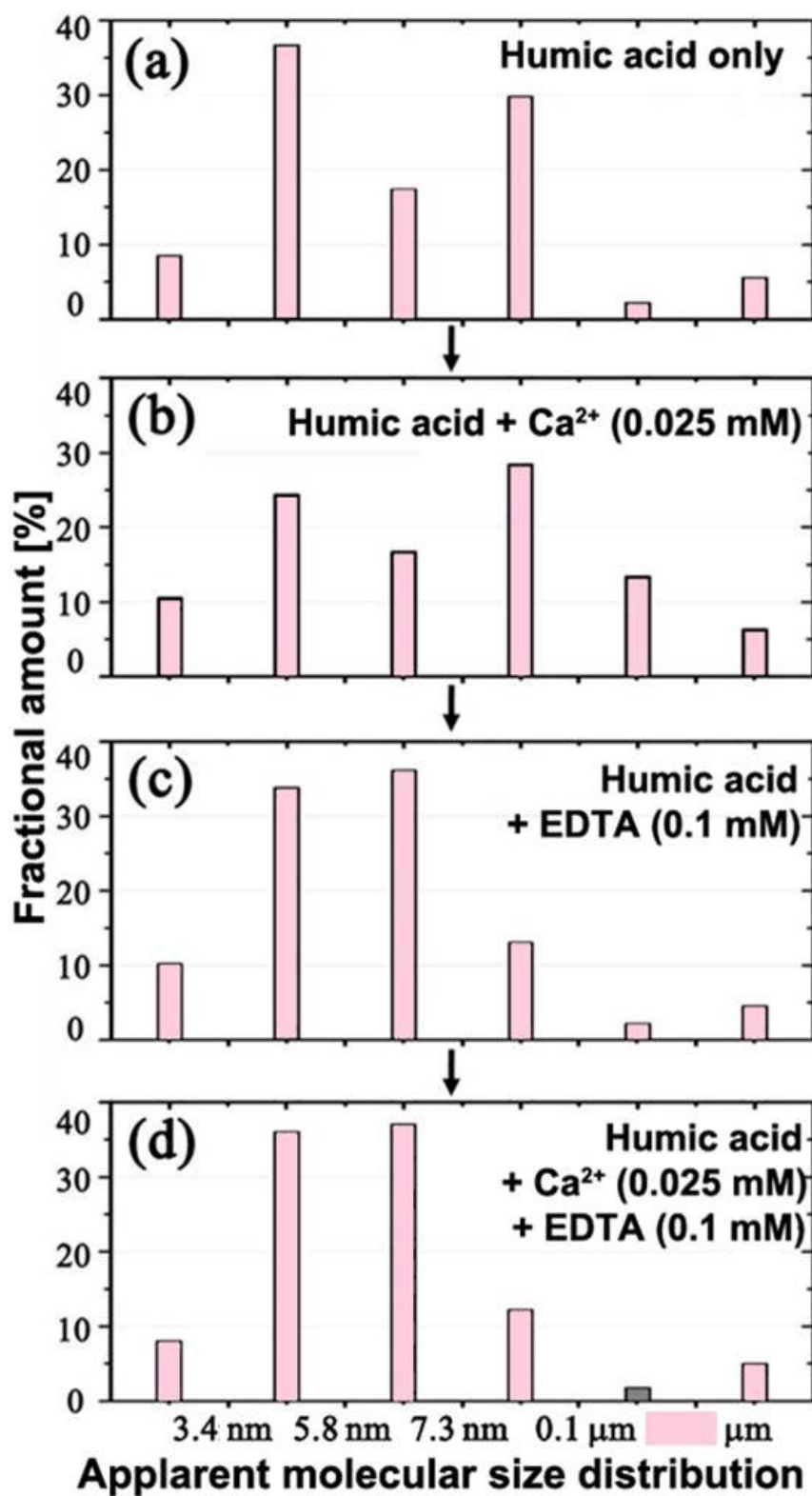


Fig. II. 9 Apparent molecular weight distribution for humic acid only, and humic acid with additives. (a) Humic acid only; (b-d) humic acid with (b) 0.025 mM Ca²⁺, (c) 0.1 mM EDTA, and (d) 0.025 mM Ca²⁺ and 0.1 mM EDTA.

II.3.4 Effect of metal ions on protein fouling

To study the effects of multivalent metal ions (or metal colloids) on the protein fouling of hollow-fiber membranes, the cross-flow filtration of protein solutions using PES hollow-fiber membranes was carried out. The model protein foulants used had different characteristics in terms of their molecular weight, pI, and aggregation. When the pore size of a membrane is comparable to the molecular size of a protein foulant, the effects of the solution environment are likely to be felt. Hollow-fiber membranes with different MWCOs were used for the filtration experiments, which assessed different kinds of proteins. The relative flux (J/J_0) and rejection (R) of the proteins are shown in Fig. II. 10.

The relative flux of the BSA solution without metal ions decreased with time (Fig. II. 10-1a), and the BSA rejection was less than 10% (Fig. II. 10-1b). The major causes of BSA fouling are typically considered to be the adsorption and deposition of BSA on membranes [25]; BSA aggregates also have a negative impact on protein fouling [10]. BSA is “a metal-transport protein”. It binds several types of multivalent metal ions, using the two deprotonated amides and imidazole at its N-terminus [26]. Indeed, Carr revealed the binding of Ca^{2+} to BSA at neutral pH [27]. There is therefore the potential that adding multivalent metal ions could affect the BSA fouling process. However, Fig. 2-1a shows that the addition of Ca^{2+} did not produce a decline in the relative flux, and the rejection also remained at a similar low level (less than 10%), consistent with a previous report [28]. These results suggested that the BSA molecules only interacted with Ca^{2+} , which induced neither a conformational change nor aggregation in the BSA molecules, and therefore did not influence the fouling.

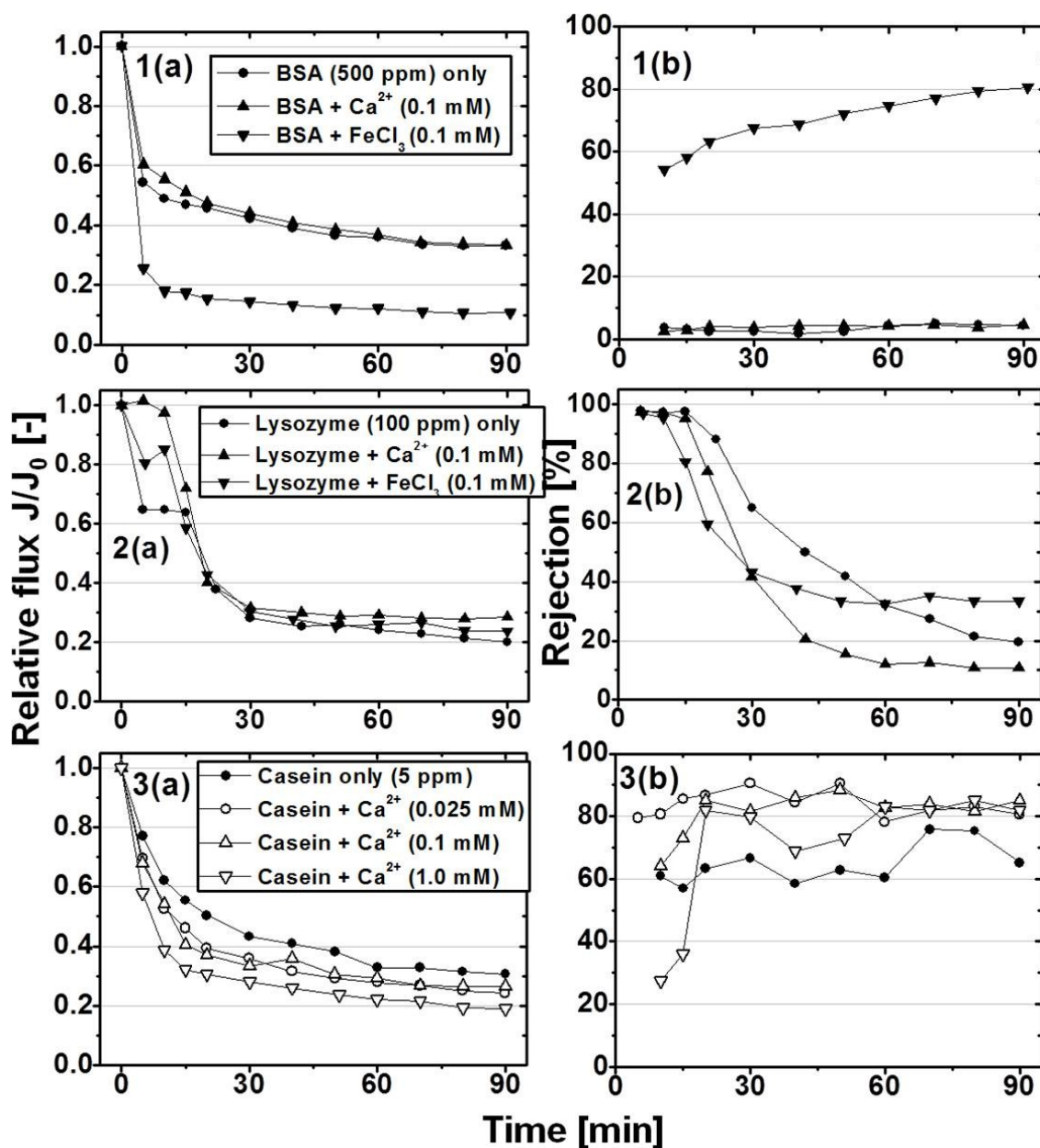


Fig. II. 10 Effects of Ca^{2+} (0.1 mM) and FeCl_3 on the relative flux (a), and protein rejection (b) during filtration. A BSA (500 ppm) solution was filtered using a PES membrane with a MWCO of 150 kDa (1), a 100 ppm lysozyme solution was filtered using a membrane with a MWCO of 10 kDa (2), and a 5 ppm casein solution was filtered using a membrane with a MWCO of 300 kDa (3).

In contrast, a BSA solution at pH 7 containing 0.1 mM FeCl_3 exhibited significant membrane fouling. Iron should have existed in three states in this solution: oxide-hydroxide colloids, oxide colloids, and very low concentrations of solubilized

Fe^{3+} (nanomolar level). Although the majority of the iron was expected to exist as iron colloids (oxide-hydroxide and oxide colloids) in the present study, DLS measurements of the BSA-free buffer containing FeCl_3 failed to detect colloidal nano- or microparticles, probably because of their low concentration. However, the iron colloids seemed to increase the BSA fouling (see section II. 3.5 and 3.6). The BSA rejection in the presence of FeCl_3 was much higher than that without FeCl_3 , which also indicated that the BSA and the iron colloids interacted. The addition of CaCl_2 or FeCl_3 did not cause any obvious turbidity or precipitation in the BSA solution.

For the cases using lysozyme solutions (Fig. II. 10-2), the relative flux remained high for the first 10 min of filtration, and then decreased markedly to approximately 0.25 and remained constant. The rejection of lysozyme remained high (almost 100%) for the first 15 min of filtration and then decreased suddenly, indicating that during the initial period of the filtration, lysozyme molecules were adsorbed onto the membrane. The decrease in the flux preceded that of the decrease in rejection, which also indicated pore blocking by lysozyme. The low rejection (~20%) of lysozyme showed that the intrinsic pore size of the membrane was large enough to allow most lysozyme molecules to pass through. There was an attractive interaction between the PES membrane surface (which was negatively charged) and free lysozyme molecules, because of their highly positive charge (the pI of lysozyme is 10.8); this induced the initial adsorption of lysozyme on the membrane. After the adsorption of the lysozyme, there was a repulsive interaction between the adsorbed and free lysozyme molecules, which prevented the multilayer adsorption of lysozyme on the membrane surface [29]. This may account for the plateau after the rapid decline in the relative flux.

The effects of the addition of CaCl_2 and FeCl_3 on the lysozyme filtration were

unclear (Fig. II. 10-2a). There was no obvious difference in the relative flux between the lysozyme solutions. The rejection profiles (Fig. II. 10-2b) showed that the addition of metal ions shortened the initial adsorption period. Burns and Zydney reported that Cl^- ions were adsorbed onto a positively charged membrane surface [30]. Under the experimental solution conditions used in the present study, the surface of the PES membrane was expected to be negatively charged [31]. Metal cations also engage in electrostatic interactions, and might have adsorbed on the negatively charged PES membrane surface; this was expected to affect the initial period of lysozyme adsorption. After the initial adsorption, the presence of the metal ions did not seem to affect the lysozyme fouling, indicating that there were only negligible interactions between the lysozyme and the metal ions under the present conditions.

Figure II. 10-3a shows that the casein solutions caused severe membrane fouling, although the casein concentration was very low (only 5 ppm), and the size of a single casein molecule was much smaller than the pore size of the PES membrane, which had an MWCO of 300 kDa. There was almost no decline in flux when a buffer containing Ca^{2+} but no casein was filtered through the PES membrane (data not shown). In solution, casein molecules form stable micelle-like aggregates over 100 nm in size. Casein molecules have hydrophilic heads and hydrophobic tails, and can adsorb on various polymeric surfaces [32]. These characteristics account for the significant fouling observed in the present study. It should be noted that under the present conditions, the casein solution was transparent. The addition of Ca^{2+} to the feed casein solution clearly increased the severity of the fouling, and the extent of the fouling increased with increasing Ca^{2+} concentration. The increase in fouling in the presence of Ca^{2+} might have been caused by the formation of larger aggregates of casein molecules

via the creation of Ca^{2+} bridges between the casein molecules, and by Ca^{2+} bridges between the casein in solution and the casein adsorbed on the membrane surface [33]. Ca^{2+} might also have formed a bridge between the negatively charged PES membrane and the casein molecules. Figure II. 10–3b shows that the presence of Ca^{2+} increased casein rejection, indicating a reduction in the fraction of single casein molecules (that did not form micelles) in solution. Because adding FeCl_3 (even at a concentration of only 0.01 mM) to a casein solution caused obvious white precipitation within several minutes, UF experiments were not conducted in the presence of FeCl_3 .

II.3.5 Effect of the FeCl_3 concentration on BSA fouling

To study the effect of FeCl_3 on the BSA fouling, a series of filtration experiments using BSA solutions with various concentrations of FeCl_3 were carried out. First, solutions containing only FeCl_3 were used in filtration to reveal the fouling caused by FeCl_3 . A FeCl_3 -only solution (0.05 mM) did not exhibit a noticeable decline in relative flux during filtration, while a 0.1 mM FeCl_3 solution showed an obvious decline in flux (Fig. II. 11a). The results indicate that fouling was caused by iron colloids, although the 0.1 mM FeCl_3 solution was not turbid to the naked eye. However, the decline in flux (0.1 mM FeCl_3 solution) was much slower than that observed in the BSA solution.

Figure II. 11a shows that FeCl_3 -accelerated BSA fouling occurred even at a very low concentration of FeCl_3 (0.01 mM). As the FeCl_3 concentration increased, the decline became more marked. At 0.05 mM FeCl_3 /BSA, the relative flux at 90 min was 0.12, whereas the values for a BSA solution and a FeCl_3 solution were 0.33 and 0.87, respectively. The total permeated volume of a 0.1 mM FeCl_3 /BSA solution was only 25% that of a BSA solution after 90 min of filtration. There have been several reports

showing that Fe^{3+} and iron colloids interact with BSA [34-36]. In the present study, the addition of FeCl_3 to the BSA solutions caused the aggregation of BSA, or the formation of BSA/iron colloids complexes, resulting in severe membrane fouling. The BSA rejection increased with increasing Fe^{3+} concentration (Fig. II. 11b). The high BSA rejection (60%) for a membrane with a MWCO of 150 kDa implied that either the BSA aggregates or a BSA/iron colloid complex were rejected by the membrane.

The membrane surfaces fouled with BSA were observed using FE-SEM (Fig. II. 12a-d), and the topography of the Fe on the surfaces was investigated using EDS (Fig. II. 12e & f). Open pores were observed on the outer surface of a clean PES membrane with a MWCO of 150 kDa (Fig. II. 12a). After filtration of the BSA solution, blockage of the pores was clearly observed, and most of the surface appeared to be covered by foulants, while only a few of the pores remained open (Fig. II. 12b). Interestingly, large amounts of deposits were formed on the surface after the filtration of a 0.1 mM FeCl_3 solution (Fig. II. 12c); the deposits were apparently composed of iron colloids, which likely caused the decline in the flux seen in Fig. 3a. For the filtration of a BSA solution with 0.1 mM FeCl_3 , the whole surface of the membrane was covered by deposits, which were packed more densely and thickly than in other cases (Fig. II. 12d).

The outer surfaces of the fouled membranes were examined using EDS. The analysis revealed the presence of Fe on the surfaces of membranes fouled with a FeCl_3 solution (Fig. II. 12e) and with a BSA/ FeCl_3 solution (Fig. II. 12f). The EDS measurements showed that the foulants observed on the membrane surfaces contained Fe. Compared with the amount of Fe on the surface fouled by the 0.1 mM FeCl_3 solution, the low amounts of Fe on the surface fouled by the BSA/ FeCl_3 solution implied that BSA molecules surrounded the iron colloids in the solution, and the BSA/iron colloids

complexes were then deposited on the membrane surface.

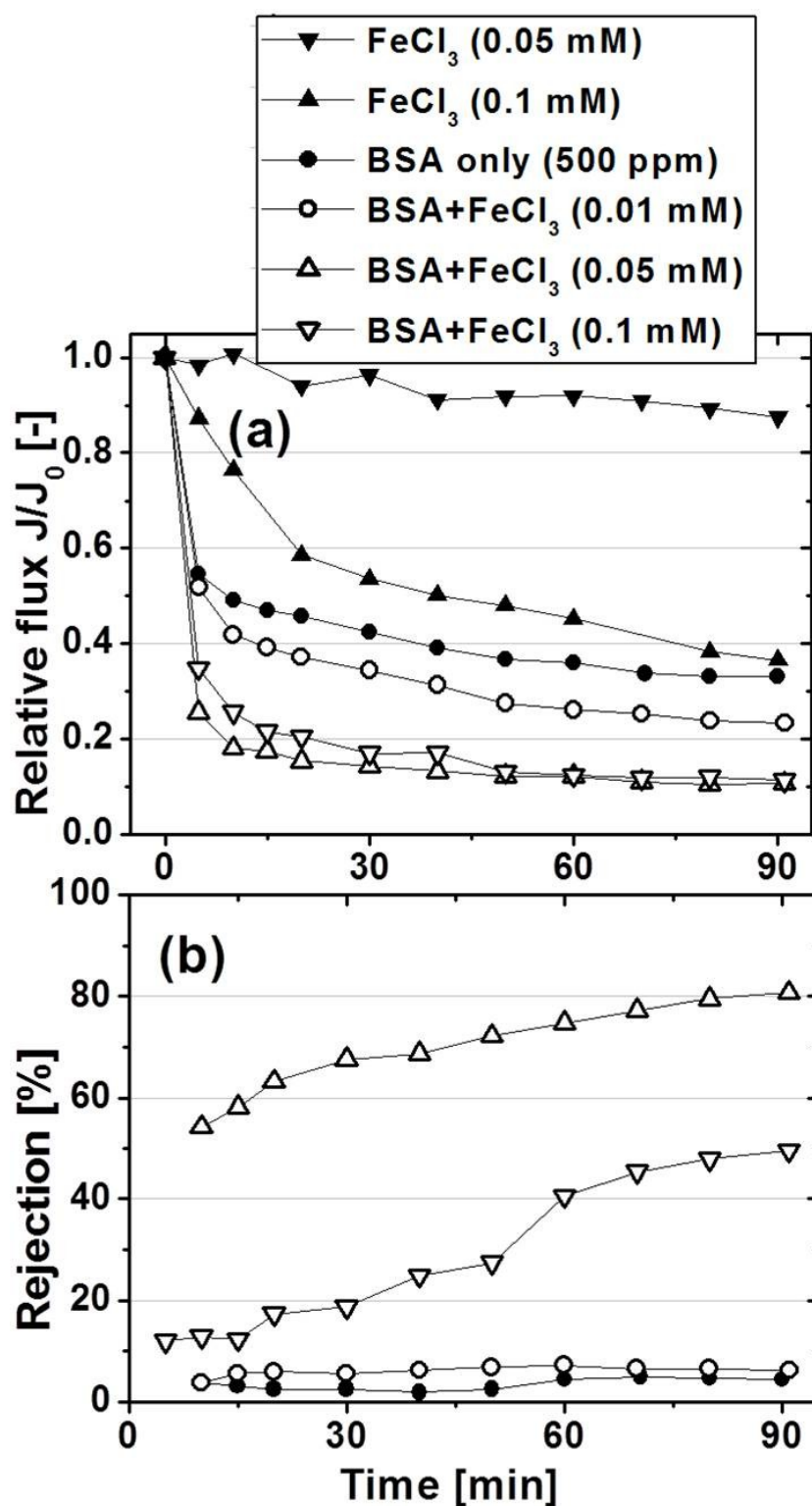


Fig. II. 11 Effect of the FeCl_3 concentration on the relative flux and the rejection of BSA during filtration. A BSA (500 ppm) solution was filtered through a PES membrane with a MWCO of 150 kDa.

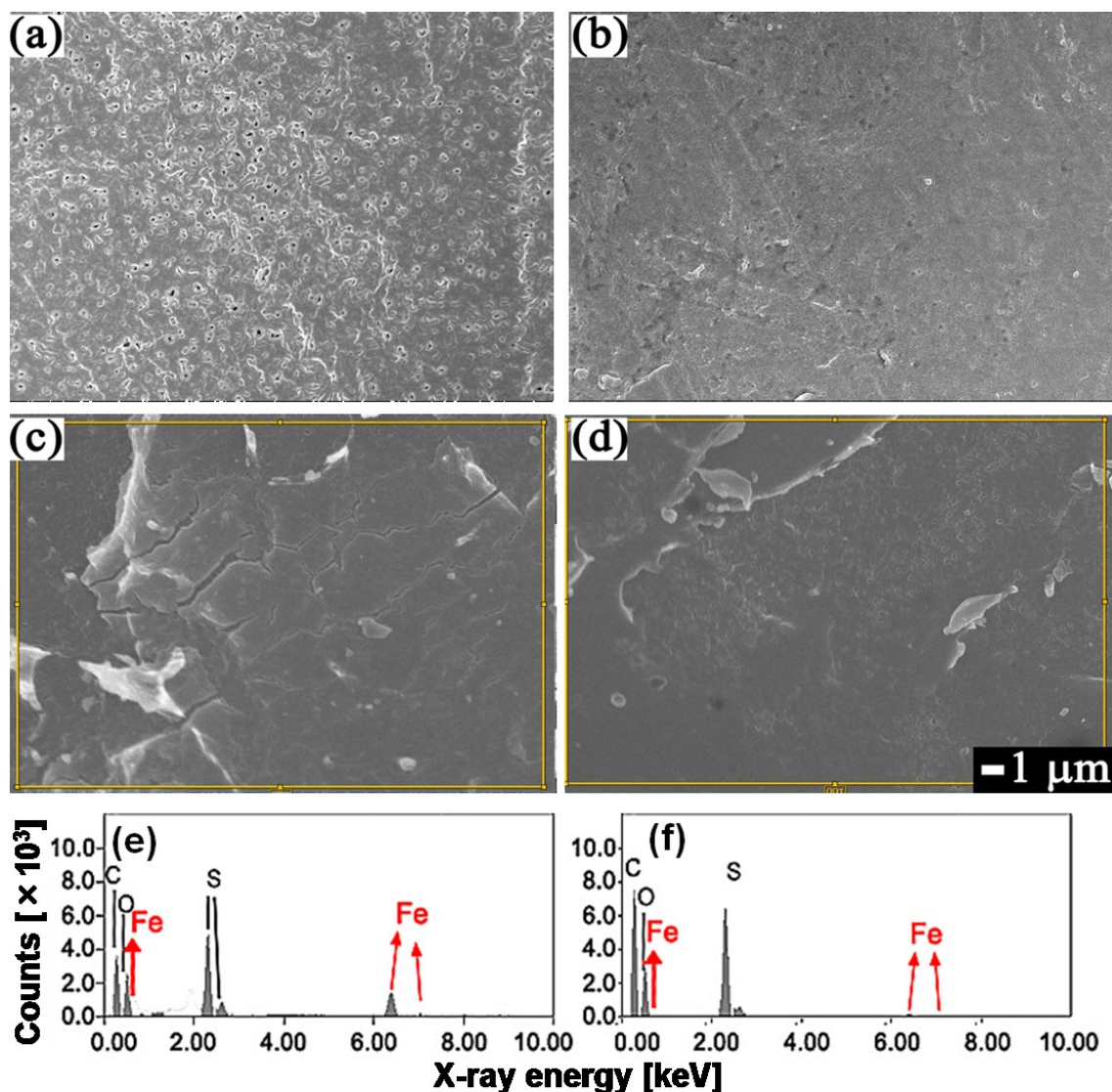


Fig. II. 12 FE-SEM and EDS analyses of the outer surfaces of PES membranes with a MWCO of 150 kDa. The outer surfaces of a membrane before use (a), fouled by a 500 ppm BSA solution (b), fouled by a 0.1 mM FeCl_3 solution (c), and fouled by a 500 ppm BSA solution with 0.1 mM FeCl_3 (d). EDS spectra of the surfaces fouled by a 0.1 mM FeCl_3 solution (e), and fouled by a 500 ppm BSA solution 0.1 mM of added FeCl_3 (f). Filtration was carried out for 1.5 h at 0.5 atm to prepare the fouled membranes. The orange lines in Fig. 3c and d represent the areas of the EDS measurements.

II.3.6 DLS measurements of aggregates or colloids in feed solutions

The above results suggested the presence of protein aggregates and metal colloids along with the proteins in the feed solution. DLS measurements were carried out to

determine the size distributions of the protein aggregates or metal colloids in the feed solutions [37]. Because Ca^{2+} is soluble in aqueous solutions at neutral pH, the DLS measurements did not detect any colloidal substances in the Ca^{2+} solutions. The measurement on the casein solutions (5 ppm) without any Ca^{2+} showed that the casein micelles in the solution had a relatively broad size distribution that ranged from 50–150 nm (Fig. II. 13), in agreement with the literature [18]. The addition of Ca^{2+} increased the size of the casein aggregates. The presence of 1 mM Ca^{2+} produced large aggregates, 400 nm in diameter, and broadened the size distribution.

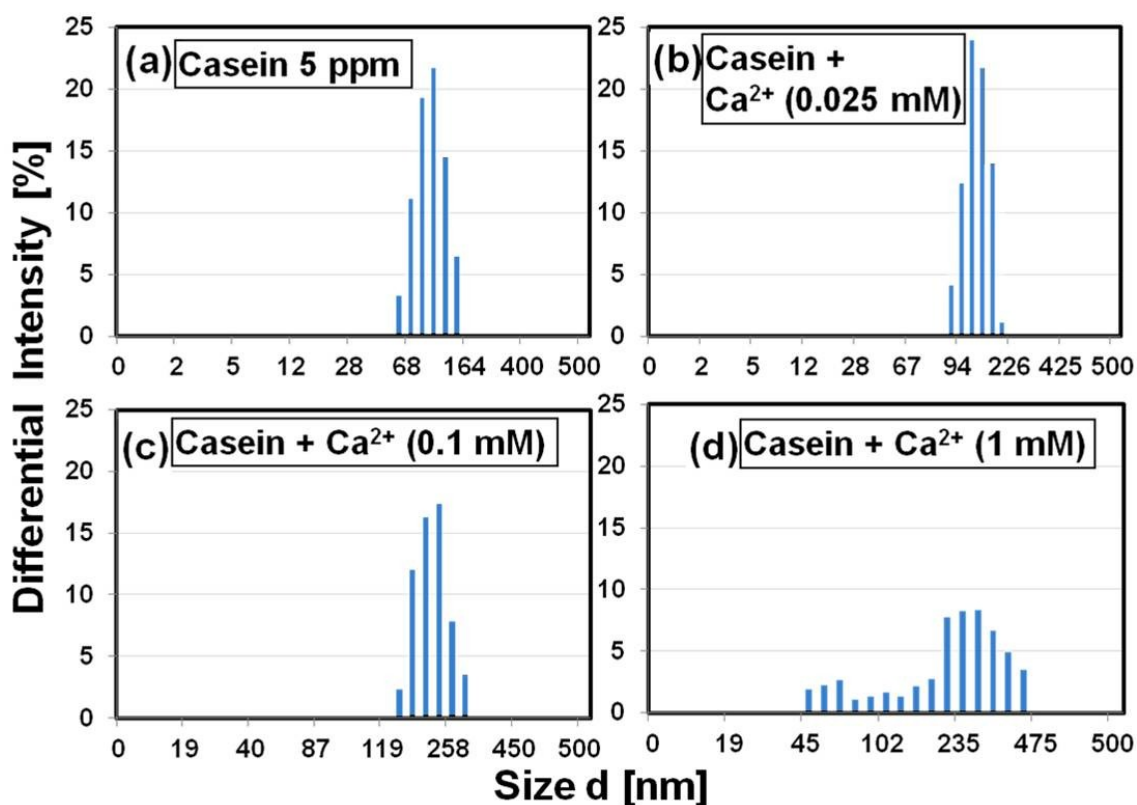


Fig. II. 13 Dynamic light scattering (DLS) measurements of 5 ppm casein in solutions with and without Ca^{2+} .

Although previous reports [38] (and our FE-SEM observations) indicated the presence of iron colloids in FeCl_3 solutions, these colloids were not observed in our DLS measurements, probably because of the low concentrations of FeCl_3 . BSA

solutions containing FeCl_3 were also measured using DLS (Fig. II. 14). A main peak at approximately 8 nm was observed in the BSA solution without FeCl_3 , which was in agreement with the molecular size of a single BSA molecule [39]. A small peak at approximately 40 nm was interpreted as indicating the presence of BSA aggregates, which was also consistent with a previous report [10]. The addition of FeCl_3 to the BSA solution markedly shifted the size distribution to a larger range. As the FeCl_3 concentration increased, the fraction at 10 nm decreased and, coincidentally, a fraction at approximately 200 nm appeared, and the size distribution became broader. These results also suggested interactions between the BSA and the iron colloids, and explained the high levels of BSA rejection in the presence of FeCl_3 . A BSA fraction remained at approximately 20 nm with a concentration of 0.1 mM FeCl_3 ; this corresponded to the BSA that had not interacted because of the low concentrations of FeCl_3 . The fraction at approximately 20 nm accounted for the result showing that approximately 40% of the BSA passed through the membrane at 0.1 mM FeCl_3 (Fig. II. 11b).

The DLS measurements and the fouling results suggested that the addition of metal ions or metal colloids to a protein solution induced the aggregation of proteins, depending on the type of protein and metal ions. It is likely that the protein aggregates were deposited on the outer surfaces of the membranes, because of their large physical size compared with the membrane pores. Indeed, the cross-sectional observations using EDS showed that the Fe counts inside the fouled membrane were much lower than those measured on the outer surface (data not shown). The deposited protein aggregates formed a dense layer on the membranes, which was attributed to the increased fouling during the membrane filtration.

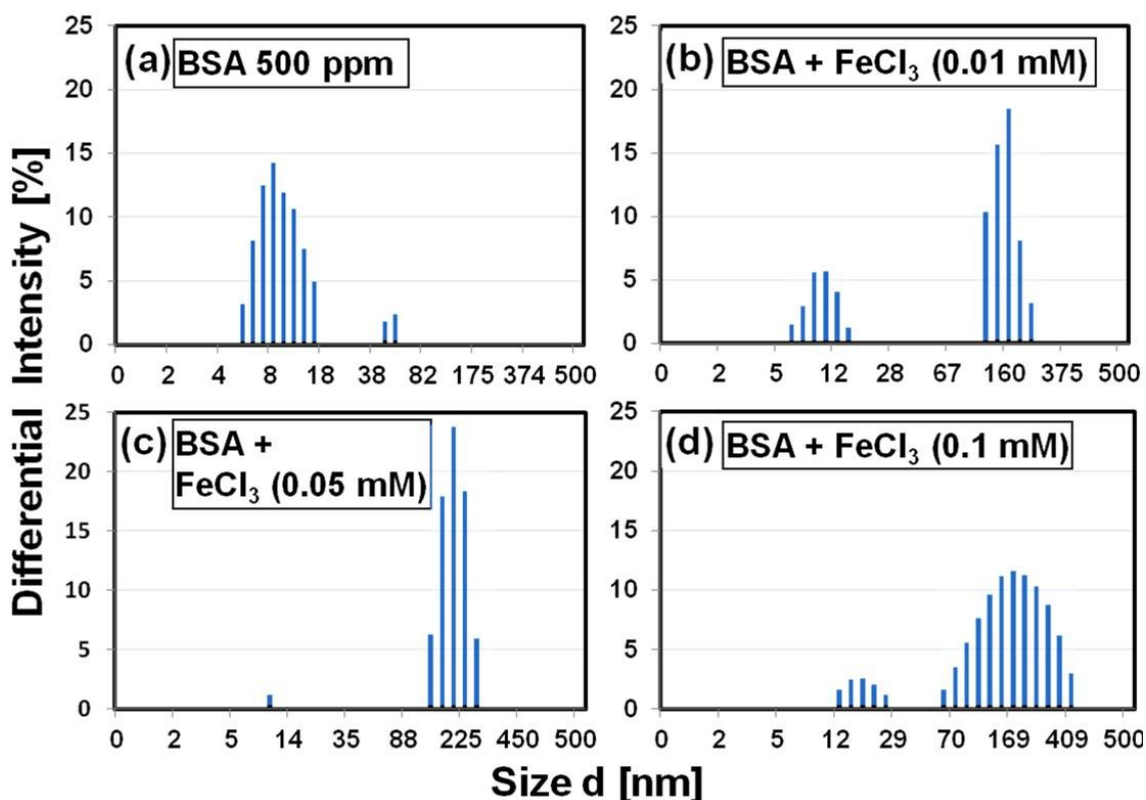


Fig. II. 14 Dynamic light scattering (DLS) measurements of 500 ppm BSA solutions with and without FeCl_3 .

II.3.7 Effects of other additives on the BSA- FeCl_3 fouling

We have revealed above that the addition of FeCl_3 increased the BSA fouling through the formation of large aggregates. We then tried to prevent the increase in fouling in the case of a FeCl_3 /BSA solution through the use of additive chemicals. A chelating agent, ethylenediaminetetraacetic acid (EDTA), was expected to eliminate the effects of Fe^{3+} via the complexation of Fe^{3+} . The addition of a reducing agent, dithiothreitol (DTT), was also studied, because it can be adsorbed on iron colloids, and might detach proteins from iron colloids [40].

Figure II. 15 shows the effects of EDTA (0.5 mM) and DTT (0.1 mM) on the relative flux of a 500 ppm BSA solution containing 0.1 mM FeCl_3 . The decline in the

flux across the membranes was slightly alleviated by the addition of EDTA. Although EDTA forms a strong complex with ferric ions, the effect of EDTA was not large, probably because of the very small fraction of ferric ions in the feed solution. EDTA also reacts with iron oxides [41]; however, the effect of EDTA in the present study was not obvious, perhaps because of the relatively strong interactions of the iron colloids with the BSA.

The addition of DTT was more effective than the addition of EDTA in recovering the flux. The decline in flux was suppressed by DTT, but was still more significant than that observed in the BSA solution. DTT is a thiol compound that can bind to many kinds of metals and metal oxides. In the present study, it is likely that DTT adsorbed on the iron colloids and detached BSA molecules, presumably preventing the formation of a dense layer of protein aggregates on the membrane surface. The fact that the levels of fouling were still higher than for a BSA solution might have been because of the additional fouling of the iron colloids themselves. In other words, the fouling observed in the presence of DTT was the sum of the BSA fouling and the fouling of the iron colloids.

Backwashing of the fouled membranes was carried out using deionized water for 1 min after 1 h of filtration. DI water was passed from the inside to the outside of the membrane, at 15 ml min^{-1} , under 1 atm of transmembrane pressure. However, only a slight recovery (less than 0.05 of $\Delta J/J_0$) of the relative flux was observed, for all cases. The low backwashing efficiency in the BSA solution with the additives implied that the foulants were strongly bound to the membrane surface via the BSA molecules, which was similar to the case of fouling with a BSA solution [42]. The backwashing efficiency was not improved in the presence of EDTA or DTT.

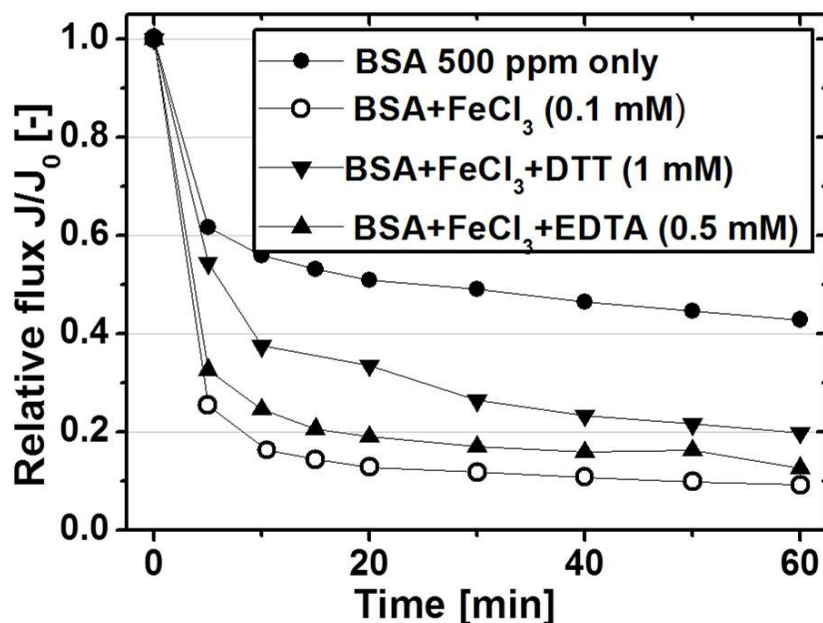


Fig. II. 15 Effects of EDTA and dithiothreitol (DTT) on the relative flux of a BSA solution containing FeCl_3 . The filtration conditions were the same with those used in Fig. II. 10.

II.4 CONCLUSIONS

The significant effects of ionic environment and metal colloids on fouling of hollow-fiber ultrafiltration membranes were studied. We revealed that the presence of metal ions and metal colloids, even at low concentrations, could affect fouling greatly during filtration.

Although there are reports on aggregation of humic acid induced by multivalent metal ions, there was still uncertainty about humic acid fouling of hollow fiber membranes. The presence of various metal ions accelerated humic fouling, even though the lateral fluid flow associated with the hollow-fiber membrane has the potential to sweep foulant deposits off the membrane surface. The humic acid aggregate size increased with addition of the metal ions, and the observed size was larger than the pore size of the membrane. The amount of humic acid deposition on the membrane surface increased to a remarkable extent in the presence of Ca^{2+} , indicating

that the foulant deposit could not be swept away by the lateral flow of the feed solution. Addition of the chelating agent EDTA inhibited, to a large extent, metal ion-induced aggregation of humic acid, resulting in inhibition of fouling.

The presence of metal ions and metal colloids increased casein and BSA fouling of hollow-fiber membranes, but did not affect lysozyme fouling. The increase in fouling depended on the type of metal ions and proteins. The protein aggregation induced by the metal ions and the protein-metal colloid interactions was related to the increased fouling.

The present study proposes the importance of mutual interactions amongst diverse solutes (even at low concentrations) in fouling and membrane processing. In membrane processes for water treatment and the food industry, metal ions and metal colloids are often present in raw water and feed solutions, albeit at low concentrations. The aqueous solutions that are treated using membrane filtration in real industrial applications do not contain only a single type of foulants, but multiple kinds of foulants. We must therefore pay close attention to the small amounts of metal ions and metal colloids that leach from plant equipment and are present in natural raw water.

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Chapter III

Visualization of fouling inside a hollow-fiber ultrafiltration membrane by fluorescent microscopy

III.1 INTRODUCTION

Membrane fouling remains the biggest problem for the ultrafiltration hollow-fiber membrane systems. Since the inception of widespread use of membrane technology in the food and other biotechnology industries, membrane filtration of solutions containing macromolecular species, such as proteins and polysaccharide, has received increasing attention.

There are numerous causes of protein fouling in membrane filtration [1]. The labile nature and complex structures of biological foulants, such as proteins, is still a poorly understood phenomenon during filtration processes. The strong and complex interactions between protein molecules and membrane or protein and protein molecules cause serious fouling problem in membrane processes. In addition, membrane fouling can be even caused by proteins with molecular weights much smaller than the molecular weight cut-off (MWCO) of a membrane.

Extracellular polymeric substances (EPS), known as macromolecular compounds commonly both in natural water and waste water, are reported to be responsible for membrane fouling, especially in MBR [2-4]. Polysaccharide is one of the major components of EPS and alginates are often used as a model polysaccharide for EPS fouling studies. Unlike protein-like foulants, interactions between polysaccharides and membrane materials were weak. Sweity et al. reported the QCM-D study on the interaction between polyvinylidene fluoride (PVDF) and EPS and demonstrated that

the EPS deposition on a PVDF film was easily removed by washing with an aqueous solution [5-6], indicating a difference in the fouling mechanism between protein- and polysaccharide-membrane fouling.

Although there have been a number of studies on membrane fouling, many of these focused on flux decline during filtration or deposition of foulants on an outer membrane surface [7-10]. However, fouling takes place not only on the outer surface but inside the porous membrane matrix. Foulants with diameters smaller than the mean membrane pore-size still cause internal fouling, and it is still unclear which part of a membrane determines the rejections of macromolecular species. There are several techniques (e.g. spectroscopy) for monitoring static adsorption on smooth or transparent surfaces [11], the deposition of foulant molecules inside a porous materials during filtration is much more difficult to characterize.

In this study, we tried and succeeded in the visualization of protein fouling and fouling of sodium alginate (SA) inside an ultrafiltration hollow-fiber membrane. We fluorescently labeled proteins and SA and used them as foulants during filtration through ultrafiltration hollow-fiber membranes. We observed the cross-sections of fouled membranes using a fluorescent microscope. The present study reveals the model foulant distribution inside a hollow-fiber membrane and also proposes that the internal fouling is important for improved understanding of the fouling of a hollow-fiber membrane. Fouling behaviors of model protein and polysaccharide substances inside membranes varied with the type of foulant type and membrane type.

III.2 EXPERIMENTAL

III.2.1 Materials

Commercial polyethersulfone (PES) hollow-fiber membranes of MWCOs 10, 30, 150 and 300 kDa were purchased from Daicel Chemical Industries, Japan.

Fluorescein isothiocyanate–bovine serum albumin (FITC-BSA), casein (from bovine milk) and lysozyme (from egg white) were used in the ultrafiltration experiments. The molecular weights of BSA and lysozyme are 67 and 14.8 kDa, respectively, while that of casein ranges from 19 to 25 kDa. Sodium alginate (molecular weight between 1.6×10^3 and 2.2×10^5 g/mol) was purchased from Wako Pure Chemical Industries (Osaka, Japan).

FITC-BSA (the modification degree was 7 mole–FITC per mole-BSA), FITC and polyvinylpyrrolidone (PVP, k-30) were obtained from Aldrich Chemical Co. (Milwaukee, USA). Rhodamine 123 was obtained from Biotium Inc. (Hayward, CA, USA). *N,N*-dimethyl-4-aminopyridine (DMAP) was purchased from Nacalai Tesque Inc. (Kyoto, Japan). All the other chemicals were purchased from Wako Pure Chemical Industries (Osaka, Japan).

The water used was high-quality deionized water (DI water, $>15 \text{ M}\Omega \text{ cm}^{-1}$) produced by an Elix-5 system (Millipore, Molsheim, France). For membrane preparation, poly(vinylidene fluoride) (PVDF) (Type 6020, MW 322 kDa) was purchased from Solvay Soiexis K.K (Tokyo, Japan).

III.2.2 Fluorescent modification of model foulants

FITC was used to modify casein and lysozyme [12]. Protein (0.1 g) was dissolved in 0.1 M $\text{NaHCO}_3/\text{Na}_2\text{CO}_3$ buffer (pH 9, 10 ml). A small aliquot of FITC in

dimethylformamide (1 ppm, 1 ml) was added to the protein solution and kept in the dark at 4°C for 6 h. The solution was dialyzed using a Spectra/Por® membrane (MWCO 15 kDa for casein and MWCO 3 kDa for lysozyme, Spectrum Laboratories, Inc., Rancho Dominguez, CA) for 2 days. Finally, FITC-protein was freeze-dried, and the obtained powder was stored at 4°C prior to use.

Labeling degrees of casein and lysozyme were calculated by measuring the amount of unreacted FITC, which was collected after dialysis. The FITC concentration was determined by measuring the fluorescent intensity using a fluorescent spectrometer (FR-8200, JASCO, Tokyo, Japan). The FITC-modification degrees were approximately 0.5 mole–FITC per mole–casein and 0.1 mole–FITC per mole–lysozyme.

A fluorescent tag, rhodamine 123, was used to modify SA as previously reported [13]. According to the manufacturer's information and the molecular weight of single sugar unit (approximately 200 g/mol), the maximal molecular weight of SA was calculated as 2×10^5 g/mol. For the calculation of the labeling degree of SA, the maximal molecular weight was used as the representative molecular weight of SA.

A small aliquot of rhodamine 123 in dimethylformamide (DMF) (2 mM, 0.5 ml) and DMAP (12.2 mg, 10 mM) were mixed with an aqueous solution of SA in 0.1 M NaHCO₃ (8.4, 9.5 ml). After stirring for 5 h, 1-ethyl-3-(3-dimethylaminopropyl) carbodiimide hydrochloride (38.3 mg, 20 mM) and *N*-hydroxysuccinimide (23.0 mg, 20 mM) was added and the mixture was stirred at 25 °C for 8 h. A dark red solution was obtained. The mixture was vacuum-dried and dissolved in Na₂CO₃ solution (0.1 M, 100 ml) at pH 9 and contacted with dichloromethane (100 ml) for 6 times to remove unreacted rhodamine 123. A clear red solution was obtained and dialyzed with a Spectra/Por® membrane (3 kDa) for two days. Finally, rhodamine–SA was freeze-dried

and resultant powder was stored at 4 °C prior to use.

The labeling degree of rhodamine-SA was calculated by measuring the fluorescent intensity of a rhodamine-SA solution (in DMF/Water 25/75 vol%) and pure rhodamine 123 solutions using a fluorescent spectrometer (FR-8200, JASCO, Tokyo, Japan). The rhodamine-labeling degree was approximately 2.6 mole– rhodamine per mole–SA (MW 200,000).

III. 2.3 Membrane preparation using TIPS and NIPS method

PVDF hollow-fiber membranes with different membrane structures (with or without a skin-layer) were prepared by a batch-type extruder (BA-0, Imoto Machinery Co., Kyoto, Japan) by the thermally-induced phase separation (TIPS) and nonsolvent-induced phase separation (NIPS) methods (Fig. III. 1) [14, 15].

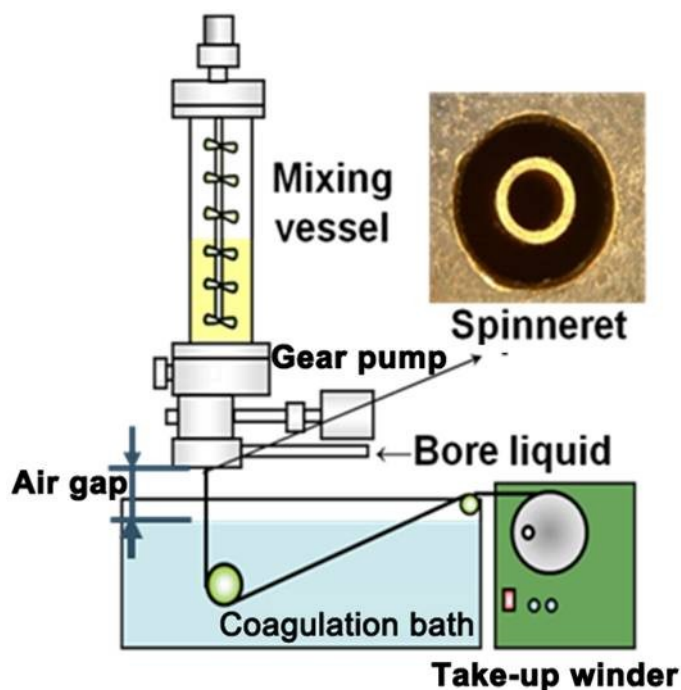


Fig. III. 1. Schematic diagram of the melt-spinning process for hollow-fiber membrane preparation.

For membrane preparation by the TIPS method, PVDF in a triacetin/glycerol solvent (PVDF/triacetin/glycerol: 30/60/10 wt%) was fed into a vessel and heated to 463 K. The mixture was stirred for 1 h to solubilize PVDF in the solvent and kept at this high temperature for another 1 h to remove bubbles. The homogeneous PVDF solution was fed to a spinneret by a gear pump under pressure with nitrogen gas. The spinneret consisted of an outer tube and an inner tube of diameter 1.58 mm and 0.83 mm, respectively. The PVDF solution was extruded from the outer orifice of the spinneret and a bore solution (triacetin) was introduced into the inner orifice to make the lumen of the hollow-fiber membrane. The extruded PVDF solution entered a water bath at 273 K to induce phase separation and solidification. The air gap between the spinneret and the bath was 2 cm. The solidified PVDF hollow-fiber was wound on a take-up winder at around 0.26 m/s. The bore solution and solvents remaining in the hollow-fiber membrane were extracted by immersing the membrane in ethanol and then keeping it in DI water for 3 days with stirring. The prepared membrane had a skin-layer on its outer surface. The membrane was kept in DI water until use.

For membrane preparation by the NIPS method, a homogeneous *N*-methyl-2-pyrrolidone (NMP) solution containing PVDF and PVP (PVDF/PVP/NMP: 17/3/80 wt%) was prepared by mixing them at 313 K for around 12 h and then holding the mixture for another 12 h at room temperature to remove bubbles. The solution was carefully placed into a vessel and heated to 313 K again. A hollow-fiber membrane was prepared by extruding the PVDF solution from the outer orifice of the coaxial spinneret to a coagulation bath (deionized water, $>15 \text{ M}\Omega \text{ cm}^{-1}$) at 293 K, inducing non-solvent phase separation in the PVDF solution. The air gap was adjusted to 5 cm. When extruding the PVDF solution from the spinneret, a bore solution (a mixture of NMP and

water; 20/80 vol%, 313 K) was simultaneously extruded from the inner orifice of the spinneret. A membrane with open pores on its outer surface was obtained and kept in DI water until use.

III.2.4 Membrane filtration

Cross flow filtration experiments were performed with a laboratory-scale apparatus [16]. The outer-pressured type of filtration experiment was carried out by permeating a feed solution from the outside to the inside of the hollow-fiber membrane. A minimodule device that contained a single hollow-fiber membrane was used (Fig. III. 2a), and the detail geometry of the device was shown in Fig. II. 1 in Chapter II. For an inner-pressured type of filtration experiments, a feed solution was permeated from the inside to the outside of a membrane (Fig. III. 2b), and length of hollow-fiber membrane was approximately 11 cm.

Feed solutions containing proteins were prepared by dissolving weighted fluorescently-labeled proteins in 0.05 M phosphate buffer solutions (pH 8) and stirring for 1 h at 25°C to obtain homogeneous solutions. Protein concentrations were 50 ppm for BSA, 20 ppm for casein and 100 ppm for lysozyme. These concentrations were adopted to cause similar flux declines (apparent fouling). Rhodamine-SA (50 ppm) for filtration experiments were prepared by dissolving weighted rhodamine-SA in 0.1 M NaHCO₃ buffer solution (pH 8.4) and stirring for 1 h at 25°C.

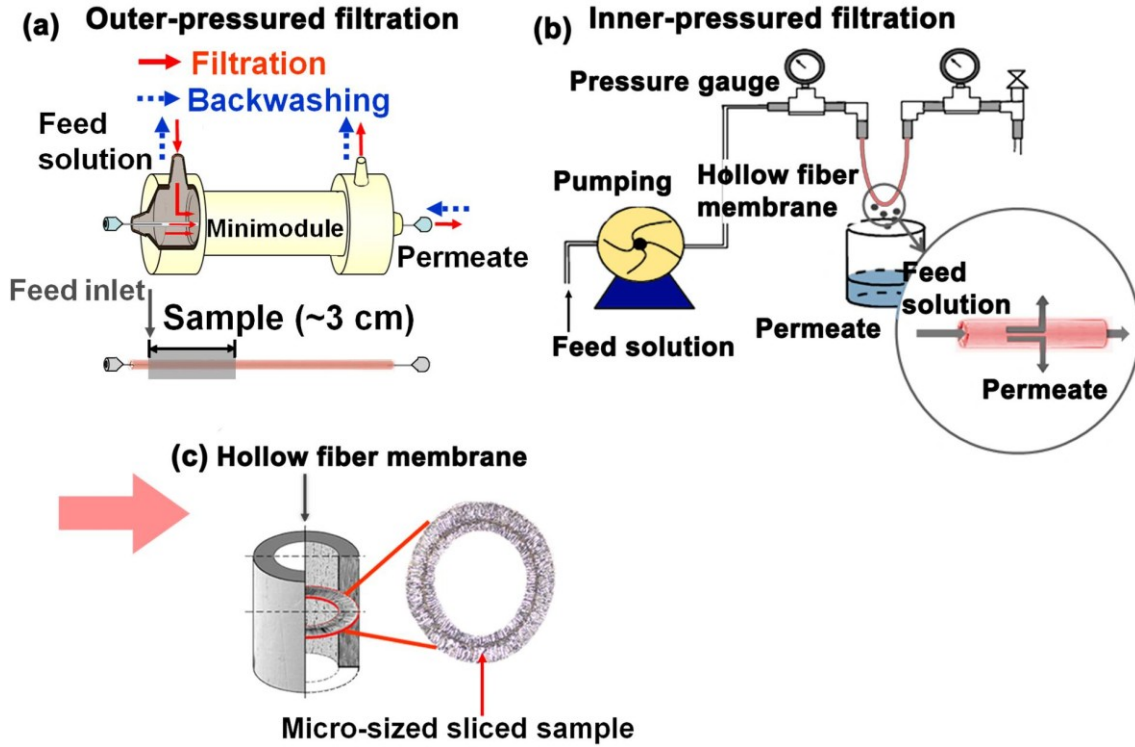


Fig. III. 2. Schematics of the laboratory-scale minimodule used for membrane filtration. Outer-pressured filtration (a) and inner-pressured filtration (b). Schematic illustration of the preparation of sliced membrane samples for internal fouling observation (c).

Each filtration experiment was carried out by permeating a feed solution through the hollow-fiber membrane at a flow rate of 15 ml/min at a transmembrane pressure of 0.5 atm. Prior to filtration experiments, DI water was passed through the membrane to measure the pure water permeability, J_0 . Filtration of a protein solution was then carried out to measure the permeability of a protein solution, J . The protein rejection, R , by a membrane was determined by measuring the absorbance at 280 nm of proteins in the permeate solution using a UV spectrophotometer.¹⁸ The rejection was defined as follows,

$$R = \left(1 - \frac{[\text{protein concentration in permeate}]}{[\text{protein concentration in feed}]} \right) \times 100 \quad [\%] \quad (1)$$

The rejection of fluorescently-labeled casein was determined by fluorescent

measurements.

The rejection of SA was determined by measuring the SA concentration in solutions using a total organic carbon analyzer (TOC-V CSH, Shimadzu Corp. Kyoto, Japan).² The rejection of SA was defined as follows,

$$R = \left(1 - \frac{C_p - C_b}{C_0 - C_b}\right) \times 100 \quad [\%] \quad (2)$$

where C_p is the carbon concentration in the SA permeate, and C_0 is the carbon concentration in the initial SA feed solution. C_b is the carbon concentration in 0.1 M NaHCO₃ buffer solution. After filtrating with a feed solution, DI water was finally passed through the membrane for 1 min under the same conditions, in order to eject a SA solution residue from the membrane prior to microscopic observation.

III. 2.5 Backwashing of membranes

For the backwashing of the fouled membranes, the membranes that were filtrated using solutions containing different model foulants were backwashed for 1 min at a flow rate of 15 ml/min at a transmembrane pressure of 1 atm (Fig. II. 2). The backwashing efficiency was defined as the recovery ratio of the flux, $\Delta J/J_0$, where ΔJ was the flux increment induced by backwashing (Described in chapter II, section II. 2.2).

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Backwashing was carried out using DI water or aqueous solutions containing various cleaning agents at room temperature, after 2 h filtration of protein solutions. DI water or aqueous solutions sodium hypochlorite (NaClO) (500 ppm), urea (8 M), sodium hydroxide (NaOH) (0.01 M), sodium lauryl sulfate (SDS) (1 wt%) (pH 6.2, and pH 10 adjusted by 0.1 M NaOH) was used as cleaning agents. NaClO was used as an oxidant

to degrade and dissolve fouled proteins, while urea at a concentration of 8 M was used as a chaotropic agent to denature and solubilize fouled proteins. NaOH of 0.01 M was used as a caustic agent to make protein more soluble. After backwashing with a cleaning agent, DI water was filtered through the membrane from inside to outside for several seconds to remove a cleaning agent.

For the backwashing of rhodamine-SA fouled membrane in an inner-pressure type of filtration experiment, the membrane was set into the minimodule device (for the outer-pressured type of filtration), and DI water was filtrated from outside to inside of the membrane.

III. 2.6 Microscopic observation of membrane fouling

The cross-sectional and surface morphology of a hollow-fiber membrane was observed by field emission–scanning electron microscopy (abbreviated as FE-SEM, JSM-7500F, JEOL Ltd., Tokyo, Japan) operating at an accelerating voltage of 5 kV. The samples were sputter-coated with Pt/Pd.

The internal fouling of a hollow-fiber membrane was visualized using an inverted fluorescent microscope (Olympus IX 81, Tokyo, Japan). To visualize the internal fouling of a hollow-fiber membrane, membrane samples around 3 cm from the inlet of a feed solution was collected after being fouled. For a membrane fouled by the inner-pressured type of filtration, samples for observing were collected approximate 3 cm from the terminus of the membrane (Fig. III. 2). The membrane samples (3 cm from the inlet of a feed solution for an outer-pressured type of filtration, or 3 cm from the terminus of an inner-pressured type of filtration) were broken in liquid nitrogen after filtration to obtain a short piece of a membrane, and freeze-dried.

For a protein fouled membrane sample, after freeze-drying, the sample was fixed using a mixture of EpoFix resin and a hardener (with a ratio of 25:1, Struers, Ballerup, Denmark) in an polyethylene capsule (Okenshoji, Tokyo, Japan), then cut to 10 μm in thickness by a microtome (EMS-150S, ERMA Inc., Tokyo) (Fig. III. 2c). For a SA fouled membrane sample, the sample was gently immersed into a polyallylamine hydrochloride (PAH) solution (10 wt%) for several seconds, which was a cationic polyelectrolyte, in order to obtain a thin protecting layer for the foulant against acrylic resin. For the case of the inner-pressured type of filtration, PAH solution was gently injected into the inner lumen of the membrane sample to obtain the protecting layer without permeation. After drying in room temperature, the sample was fixed using a mixture of acrylic resin (with a ratio in weight of solid/liquid 3/2) (ClaroCit, Struers, Ballerup, Denmark) in a polyethylene embedding capsule (Polysciences, Inc. Warrington, USA), and then cut to 50 μm in thickness by a microtome.

The above procedure was carried out while avoiding exposure to light to prevent fluorescence fading. The sliced sample was observed using an inverted fluorescent microscope with a fixed fluorescent intensity. The protein fouled membrane samples were observed at exposure time of 500 ms with sensitivity of ISO 200, while the SA fouled membrane samples were observed at exposure time of 50 ms with sensitivity of ISO 200. Under such conditions for observation, a PES or PVDF membrane before use presented a completely dark image (no fluorescence). Each filtration experiment was carried out at least twice to confirm the reproducibility in fluorescent observation, relative flux and rejection. Image J (NIH, Version 1.46) was used to characterize the fluorescence distribution of the whole cross section of the membranes.

III.3 RESULTS AND DISCUSSION

III.3.1 Fouling behaviors of protein through PES membranes of different pore-size

To visualize protein fouling behaviors inside membranes, we employed commercial PES hollow-fiber membranes with different pore sizes (MWCO 30, 150 and 300 kDa) and prepared membranes fouled with FITC-BSA by passing FITC-BSA solutions (50 ppm) through the membranes. The relative flux (J/J_0) and the rejection of FITC-BSA (R) are shown in Fig. III. 3a and b. In each case, we observed a gradual decrease in the relative flux and an increase in the rejection, meaning that fouling occurred. According to the relative flux, the degree of fouling increased as the MWCO of the membrane decreased. The final relative flux decreased to 0.2 for a PES membrane of MWCO 30 kDa, to 0.4 for a PES membrane of MWCO 150 kDa, and to 0.5 for a PES membrane of MWCO 300 kDa after 2 h filtration. The rejection also increased as the MWCO of the membrane decreased. The rejection was approximately 90% for a membrane of MWCO 30 kDa, 40% for a membrane of MWCO 150 kDa, and 20% for a membrane of MWCO 300 kDa after 2 h filtration.

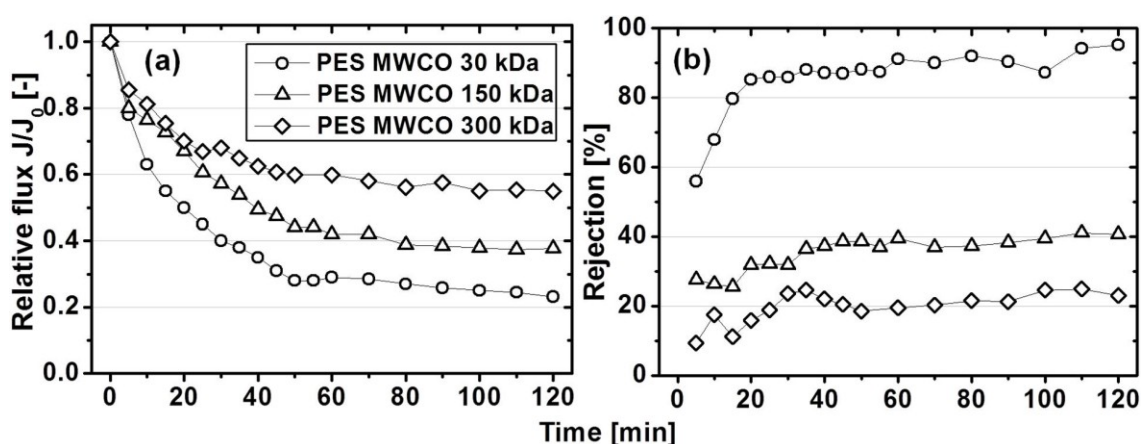


Fig. III. 3 Relative flux (a) and rejection (b) during the filtration of FITC-BSA solution through PES hollow-fiber membranes with different MWCOs.

SEM images show that the PES membranes with MWCOs of 30 and 150 kDa had

double-finger structures (Fig. III. 4a and d), while that with a MWCO of 300 kDa had a relatively homogeneous structure, without macrovoids (Fig. III. 4g). The cross-sections of the fouled membranes were observed by a fluorescent microscope (Figure 2b, e and h). All fouled membranes had particularly fluorescently-bright areas over their outer surfaces, which indicates the deposition of FITC-BSA and suggests that a cake layer of the foulants formed during filtration. Interestingly, we also observed FITC-BSA deposition near the inner surface of the membrane with MWCO 30 kDa (Fig. III. 4b). Deposition of a small amount of BSA was observed on a part of the inner surface of the PES membrane with MWCO 150 kDa. It should be noted that FITC-BSA molecules was not distributed throughout the membrane but accumulated preferentially at the area near the inner surface of the membrane of MWCO 30 kDa. Considering that the molecular weight of BSA is around 67 kDa, we had expected that BSA molecules would be rejected by the outer surface of the membrane. The observation of fluorescence, however, suggests that the BSA molecules were rejected mainly near the inner surface of the membrane, which could be due to the denser structure of the inner surface of the MWCO 30 kDa PES membrane. In the case of the membrane with MWCO 150 kDa, the accumulation of BSA was also distributed heterogeneously (Fig. III. 4e), which indicates the membrane structure was not homogeneous and that the membrane fouling occurred heterogeneously. On the other hand, a relatively homogeneous accumulation of BSA was observed all over the membrane with MWCO 300 kDa (Fig. III. 4h). Given that the pore size of this membrane was much larger than a single BSA molecule, it is natural that BSA molecules could infiltrate the membrane and permeate throughout the membrane. As a result, BSA molecules were adsorbed onto the whole membrane. There are several reports on BSA aggregation and on the membrane fouling affected by

protein aggregates [17-19]. The FITC-BSA used would also contain the BSA aggregates and also affect the geometry of BSA deposit in a fouled membrane. To avoid the effect of the aggregates, the fouling investigations need to be carried out using a solution containing only monomeric protein (see later).

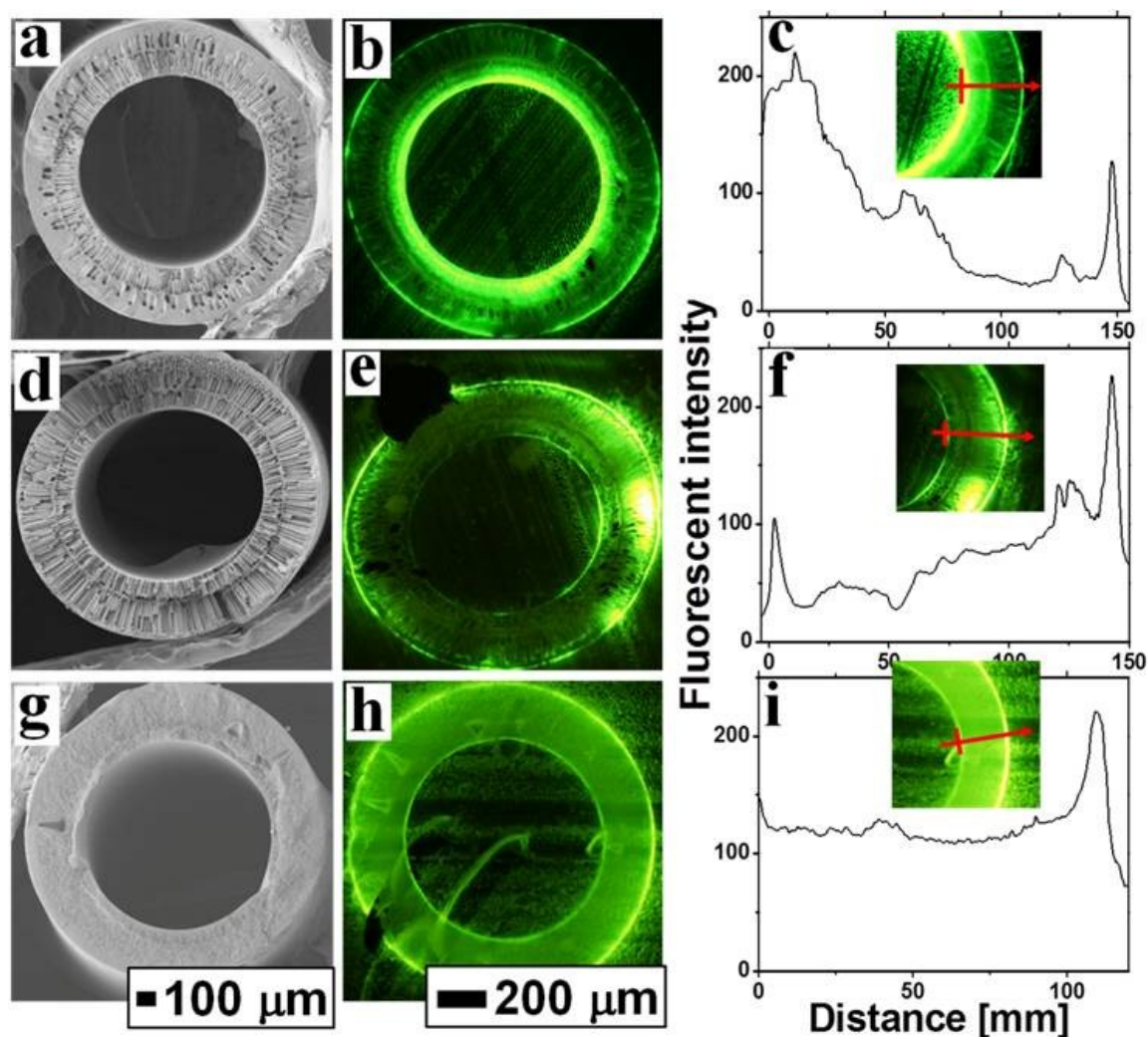


Fig. III. 4 FE-SEM and fluorescent microscope images of the whole and magnified cross-sections, and linear fluorescence distribution of the magnified cross-sections of PES hollow-fiber membranes. FE-SEM images are the cross-sections of unused membranes (a, d and g). Fluorescent images are those after the filtration of 50 ppm FITC-BSA solution for 2 h (b, e and h). (c, f and i) Fluorescence distribution along the red line across the cross-section (inset). The horizontal axis indicates the distance from inner surface. (a-c) PES membrane with MWCO 30 kDa, (d-f) MWCO 150 kDa and (g-i) MWCO 300 kDa.

Image J (an image analysis software) was employed to characterize the fluorescence distribution along a red line (inset), which crossed the whole section of membrane (Fig. III. 4c, f and i). The horizontal axes of Fig. III. 4c, f and i start from the inner surface of the membrane. In Fig. III. 4c, there are two sharp peaks of fluorescence indicating the deposition close to the membrane surfaces. Remarkable BSA accumulation near the inner surface was observed for the fouled PES membrane with MWCO 30 kDa. The inner part (distance 0-70 μm) of the cross section exhibits higher fluorescence than the outer part. The center of the cross section had slightly high fluorescence, which means that the center of the double-finger structure had a slightly denser structure comparing to the other parts of the membrane. In the case of PES membrane with MWCO 150 kDa (Fig. III. 4f), the BSA accumulation on the outer and inner surfaces was also larger than that of the other parts of the membrane. In Fig. III. 4i (PES membrane with MWCO 300 kDa), there are a plateau and a sharp peak in the fluorescence distribution. The plateau in the fluorescence distribution inside the membrane indicates a homogeneous structure of the membrane inside. A sharp peak of fluorescence suggests the relatively dense structure on the outer surface, which allowed BSA accumulation. These fluorescence profiles are in an agreement with the SEM observations.

Variations in filtration time were examined using a PES membrane with MWCO 30 kDa (Fig. III. 5). These images suggest that BSA fouling occurred in the following manner. In the beginning, BSA was adsorbed onto the outer surface of the membrane and formed an obvious bright layer within a very short filtration time (1 min, Fig. III. 5a). BSA fouling then took a second step to deposit onto the inner surface, while the area inside the membrane remained dark (Fig. III. 5b). After 25 min filtration, fluorescence all over the membrane indicates that BSA accumulated all over the

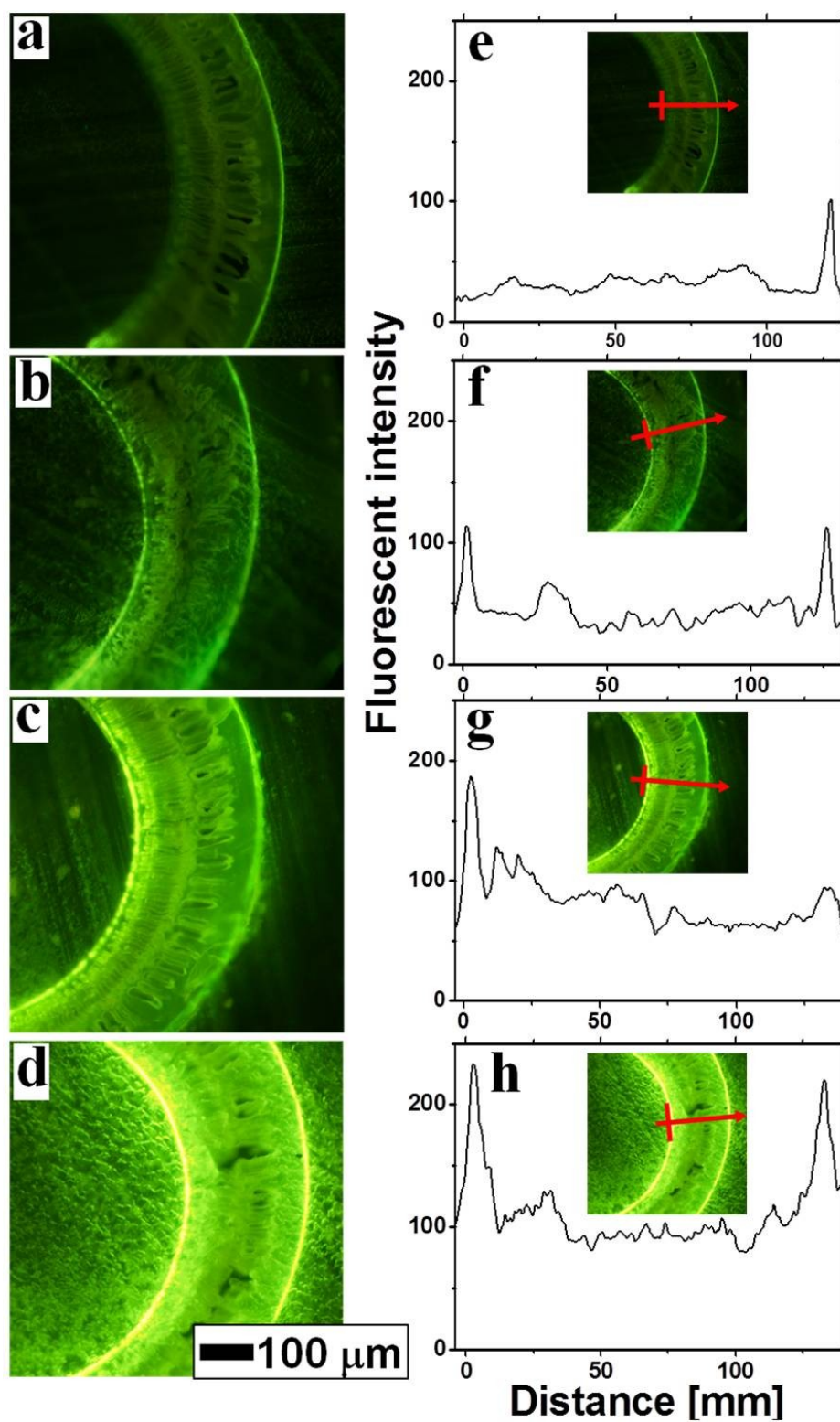


Fig. III. 5 Fluorescent microscope images and linear fluorescence distribution of magnified cross-sections of PES membranes with MWCO 30 kDa after the filtration of 50 ppm FITC-BSA solution for 1 min (a, e), 10 min (b, f), 25 min (c, g), 60 min (d, h).

membrane, while the BSA deposition on the outer and inner surfaces grew both wider and brighter (Fig. III. 5c). After the water flux declined and approached a stable state (Fig. III. 3) at filtration time 60 min, there was more BSA deposited on both surfaces, indicating that the fouling layer became denser than that initially observed (Fig. III. 5d). The fluorescent microscopic observation of a cross section of a hollow-fiber membrane reveals the time-dependent fouling geometry. From the fluorescence distribution along the lines in the cross section of membranes (Fig. III. 5e, f, g and h), we also confirmed time-dependent BSA deposition on the surfaces and the accumulation inside of the membranes. Fluorescence intensity of the whole cross-section increased with the filtration time.

III. 3.2 Fouling behaviors of different types of proteins

We then investigated the internal fouling with different types of proteins. Similar to the above investigations, aqueous solutions containing FITC-labeled casein (20 ppm) and FITC-labeled lysozyme (100 ppm) were passed through PES membranes of MWCO 300 and 10 kDa, respectively, to prepare protein-fouled membranes. In this part, PES membranes with different pore sizes for the two types of proteins and different protein concentrations were chosen to obtain a similar fouling degree after 2 h filtration in terms of the final relative flux and of the shape of the permeability decline. The decline in permeability of the two protein solutions is shown in Fig. III. 6. The relative flux (J/J_0) of the two solutions decreased similarly to that of a FITC-BSA solution, although there was a small time lag to start to decrease the relative flux in a lysozyme solution. The rejection (R) was 99% for FITC-labeled casein and 5% for FITC-labeled lysozyme, respectively, after 2 h filtration. The membrane manufacturer would

determine the MWCO of the membrane using non-protein molecules, which might be a possible reason for the extremely low rejection of lysozyme. Although proteins usually become hydrophobic due to the FITC modification, the degrees of FITC labeling were set at very low and did not affect the protein nature much in the present study. The filtration experiments of non-labeled BSA and non-labeled casein were also carried out under the same conditions. We could confirm that the FITC-modification did not affect the flux decline and the rejection of these proteins

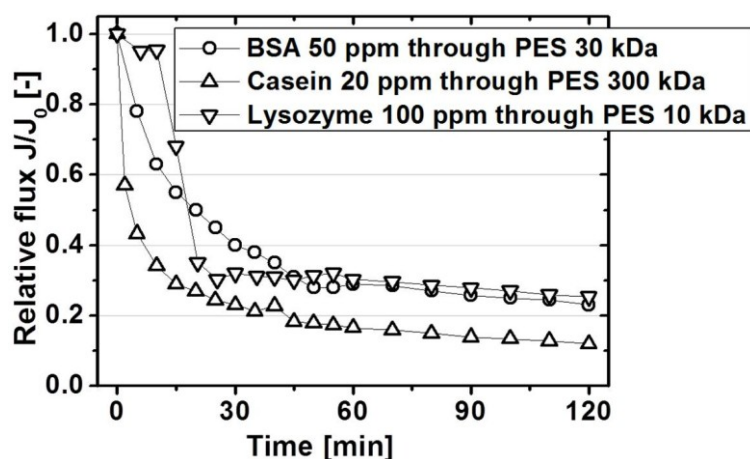


Fig. III. 6. Relative flux during the filtration of aqueous solutions containing different proteins. A FITC-BSA (50 ppm) solution was filtered through a PES membrane of MWCO 30 kDa, a FITC-labeled casein (20 ppm) solution was filtered through a PES membrane of MWCO 300 kDa, and a FITC-labeled lysozyme (100 ppm) solution was filtered through a PES membrane of MWCO 10 kDa for 2 h.

The membranes fouled with these proteins were observed by a fluorescent microscope (Fig. III. 7). Interestingly, different types of proteins showed remarkably different protein deposition inside the membranes. Casein was located only on the outer surface of the membrane (Fig. III. 7b). Casein molecules usually form micelles in an aqueous buffer [20, 21]. This explains why the membrane with MWCO 300 kDa, much larger than the molecular weights of casein (19–25 kDa), showed a high rejection. The fluorescence observation and the high rejection mean that the casein micelles were

much larger than the pore size of the membrane outer-surface. Casein aggregates prefer to be adsorbed onto a hydrophobic surface as a monolayer, of which the hydrophobic part adsorbed onto the surface with the hydrophilic-tail exposing to the solution [22]. Our result also indicates that casein formed thin layer on the outer surface of a skin-layer-free membrane (PES membrane with MWCO 300 kDa).

On the other hand, lysozyme was distributed throughout the membrane (Fig. III. 7d). Lysozyme is monomeric protein in solution and the molecular weight of lysozyme is 14 kDa, which is comparable to the MWCO of the membrane used. Lysozyme molecules entered the pores of the membrane and were homogeneously distributed inside the membrane. The lysozyme molecules would be adsorbed on the membrane matrix. After the adsorption, the membrane pore would be enough large for lysozyme molecules to pass through the membrane. That is why the rejection of lysozyme was only 5% after 60 min filtration. It should be noted that there was a bright area close to the inner surface, meaning a marked deposition of lysozyme. This result is similar to that of FITC-BSA fouling inside the MWCO 30 kDa membrane. These results indicate that the MWCO 10 and 30 kDa membranes had relatively dense layers close to the inner surface, which would determine the MWCO of the membranes.

It is difficult to fully remove residual free and unreacted FITC molecules after fluorescent modification. There might be a very small amount of FITC molecules in the feed solution. We carried out the filtration experiment of a 26 μ M FITC solution (hydrolyzed FITC) using a PES membrane with MWCO 30 kDa for 2 h, which concentration was 5 times higher than the FITC-moiety concentration in a FITC-BSA solution. Although hydrolyzed FITC molecules were also adsorbed to the membrane during filtration (Fig. III. 8), the fluorescence image was homogeneous and much darker

than that of the FITC-proteins, despite the free FITC concentration being much higher than those of FITC-proteins. The estimated concentration of free FITC molecules (unreacted and unremoved by dialysis) was very low. The effect of free FITC molecules would be eliminated in the present study.

Fouling behaviors inside membranes were quite different between proteins, even though the permeability declines were similar to each other. The present methodology revealed that the fouled part in a membrane depended on the kinds of proteins and membranes. These observations provide a novel clue to understanding membrane fouling.

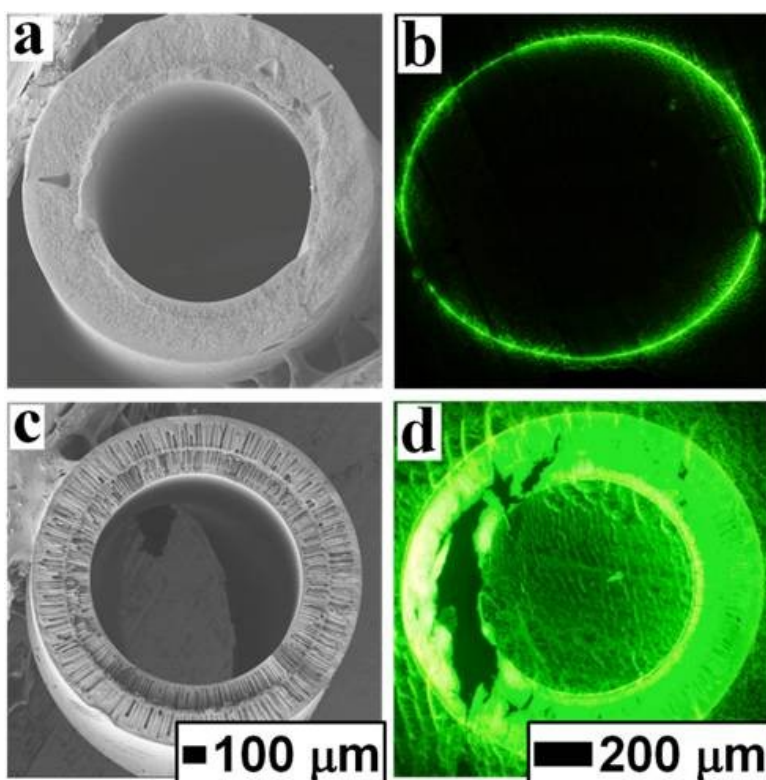


Fig. III. 7. FE-SEM and fluorescent microscope images of the PES membranes. FE-SEM images are the cross-sections of unused membranes (a and c). Fluorescent images are the cross-sections of the membranes after the filtration of a 20 ppm FITC-labeled casein solution (b) and a 100 ppm FITC-labeled lysozyme solution (d) for 2 h. (a and b) PES membrane of MWCO 300 kDa. (c and d) PES membrane of MWCO 10 kDa.

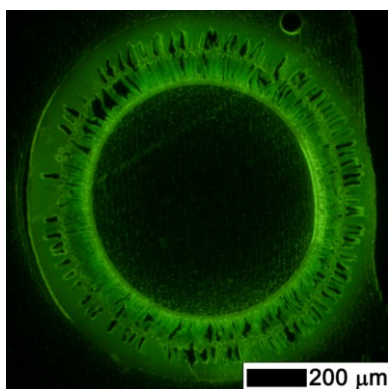


Fig. III. 8. Fluorescent microscope image of cross-section of the FITC-filtrated PES membrane with MWCO 30 kDa. A solution containing 26 μM FITC, preincubated for 12 h to hydrolyze FITC, was permeated through the membrane for 2 h.

III. 3.3 Effect of a skin layer of a membrane on internal BSA-fouling

To validate the present methodology, we prepared two types of PVDF membranes by the TIPS and NIPS methods. If a hollow-fiber membrane was prepared with a skin layer at its outer surface, we anticipated that protein molecules would be rejected at the skin layer. We prepared PVDF hollow-fiber membranes with and without a skin layer but with similar initial water permeabilities. PVDF was chosen as a model membrane material owing to its low fluorescence intensity and to the capability to prepare hollow-fiber membranes by different phase-separation methods. A membrane prepared by the TIPS method that had a skin-layer at its outer surface and exhibited an initial water permeability of 263 $\text{L}/(\text{m}^2 \cdot \text{h} \cdot \text{atm})$. A membrane without a skin layer was prepared in the presence of a pore-opener by the NIPS method and exhibited an initial permeability of 371 $\text{L}/(\text{m}^2 \cdot \text{h} \cdot \text{atm})$. The filtration experiments were carried out by pressuring a feed solution from the outside of a hollow-fiber membrane. These two membranes were fouled with FITC-BSA under the same filtration conditions as above and then subjected to fluorescence observation.

FE-SEM images of the two PVDF membranes are shown in Fig. III. 9a-c and e-g.

The membrane prepared by the TIPS method consisted of a typical homogenous sponge structure and a surface skin-layer. On the other hand, a membrane with large macrovoid structures near the outer surface was prepared by the NIPS method (Fig. III. 9e and f). The membrane had visible pores on the outer surface due to the addition of a pore-opener polymer (PVP) during the membrane preparation (Fig. III. 9g). A membrane prepared with PVP, prior to FITC-BSA fouling, did not exhibit fluorescence under the present fluorescent intensity.

After the filtration of a FITC-BSA solution for 2 h, the relative flux decreased to 0.3 for the skin-layered membrane and to 0.4 for the skin-layer-free membrane. The rejections of FITC-BSA were 100% and 18%, respectively. The fluorescent cross-sectional images of the fouled membranes are shown in Fig. III. 9d and h. We observed different fouling behaviors by the two types of the membranes, although the differences in the water permeability and the flux decline between the membranes were not large (final J/J_0 values were 0.43 and 0.27, respectively). FITC-BSA was located only at the outer surface of the membrane with the skin layer. In Fig. III. 9d, the dark region at the inner part of the membrane shows that the skin layer rejected most of the FITC-BSA molecules. Indeed, the membrane with a skin layer exhibited 100% rejection for FITC-BSA. On the other hand, fluorescence was homogeneously distributed throughout the membrane without the skin layer and macrovoids were not fluorescently-dyed. This result means that FITC-BSA molecules entered the membrane pores and were adsorbed onto the inside of the membrane matrix. BSA molecules are usually adsorbed strongly on a PVDF surface. The BSA rejection of the membrane without a skin layer was only 18%. These can explain the entire distribution of FITC-BSA inside the membrane without localization of fluorescence (Fig. III. 9h),

which is similar to the result of lysozyme (Fig. III. 7d). These investigations suggest that the present methodology provides useful information on the protein fouling inside the hollow-fiber membranes with different membrane structures.

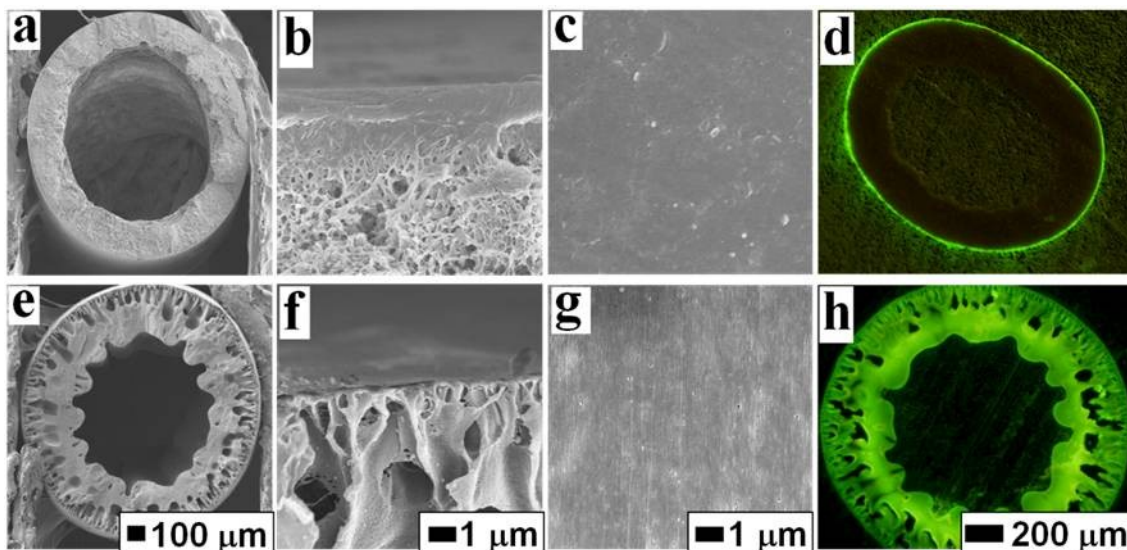


Fig. III. 9 FE-SEM and fluorescent microscope images of PVDF hollow-fiber membranes prepared by the TIPS and NIPS methods. Cross-sections (a and e), magnified cross-sections (b and f) near the outer surfaces and outer surfaces (c and g) of PVDF hollow-fiber membranes. (d and h) Fluorescent microscope images of cross-sections of PVDF membranes after filtration of 50 ppm FITC-BSA solution for 2 h. (a–d) Membrane with a surface skin layer prepared by the TIPS method, and (e–h) membrane without a skin layer prepared by the NIPS method.

III. 3.4 Effect of various cleaning agents on backwashing of BSA-fouled membranes

To restore the water flux of membranes after protein filtration, various chemicals or their combinations are practically employed as cleaning agents in backward washing. Many reports discussed washing efficiency but there was nevertheless a lack of information on which part of a fouled membrane was cleaned. We prepared PES membranes (MWCO 30 kDa) fouled with FITC-BSA, then backwashed the fouled

membranes with various cleaning agents, and carried out observation of the cross-sections of the membranes.

The backwashing efficiency changed greatly with the cleaning agents (Fig. III. 10). Briefly, the backwashing efficiency was defined as the degree of the recovered permeability compared with the fouling-reduced permeability (See chapter II, section II. 2.2). Backwashing by pure water had no effect on recovering permeability, indicating a strong interaction between FITC-BSA and the membrane material (PES), which is consistent with a previous report [23]. Amongst these cleaning agents, urea and sodium hypochlorite (NaClO) showed high backwashing efficiencies. NaClO is an oxidant that is normally used in industry for cleaning organic foulants during water treatment [24].

Urea (8 M) is widely used as a chaotropic agent to denature and solubilize protein molecules in an aqueous solution by disrupting intra- and inter-molecular non-covalent forces such as hydrogen bonds, van der Waals forces and hydrophobic interactions [25]. Previous studies reported that the protein fouled on a membrane was denatured presumably owing to interaction with the membrane material [26, 27]. That is a possible reason why recovering from protein fouling is difficult. In the present study, 8 M urea worked effectively as a cleaning agent, probably owing to the detachment and solubilization of fouled BSA from the membrane.

Sodium hydroxide is also frequently used as a cleaning agent for protein adhesion [28], as a so-called "caustic agent" that cleaves the hydrogen bonds in a protein molecule to make proteins more soluble in water. Backwashing using anionic surfactant (sodium dodecyl sulfate, SDS) at neutral and alkaline pH is also a practical way to clean a fouled membrane [29]. In the present study, these cleaning agents also recovered the permeability to some extent.

The fluorescent observations of the fouled PES membrane (MWCO 30 kDa) after backwashing are shown in Fig. III. 10. Compared with that of the fouled membrane without backwashing (Fig. III. 11a), the fluorescent geometry of the cross-sectional image changed with the cleaning agents. Backwashing with high efficiency resulted in discolored images, especially at the outer surfaces or close to the inner surfaces (Fig. III. 11b-d). Backwashing with low efficiency resulted in relatively bright images (Fig. III. 11e-f), meaning that protein remained inside a membrane. In all cases, FITC-BSA remained after backwashing. These results agreed with the fact that the backwashing efficiency did not reach at 100% in any case.

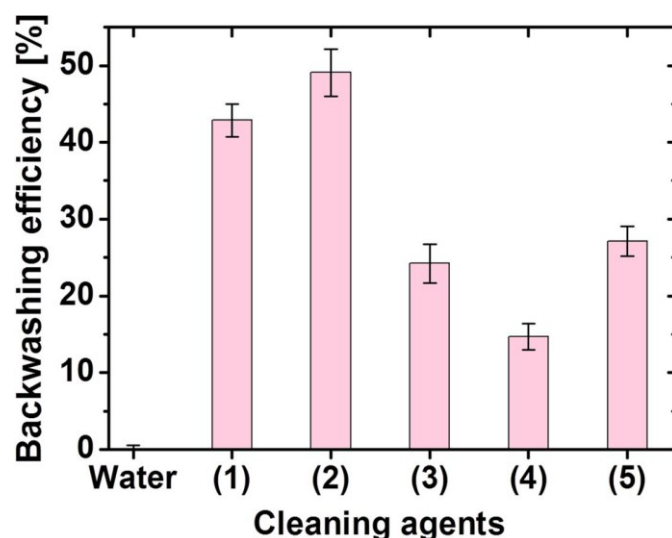


Fig. III. 10. Backwashing efficiency of BSA-fouled membrane using various cleaning agents: (1) NaClO 500 ppm, (2) urea 8 M, (3) NaOH 0.01 M, (4) SDS 1 wt% (pH 6.2), and (5) SDS 1 wt% (pH 10).

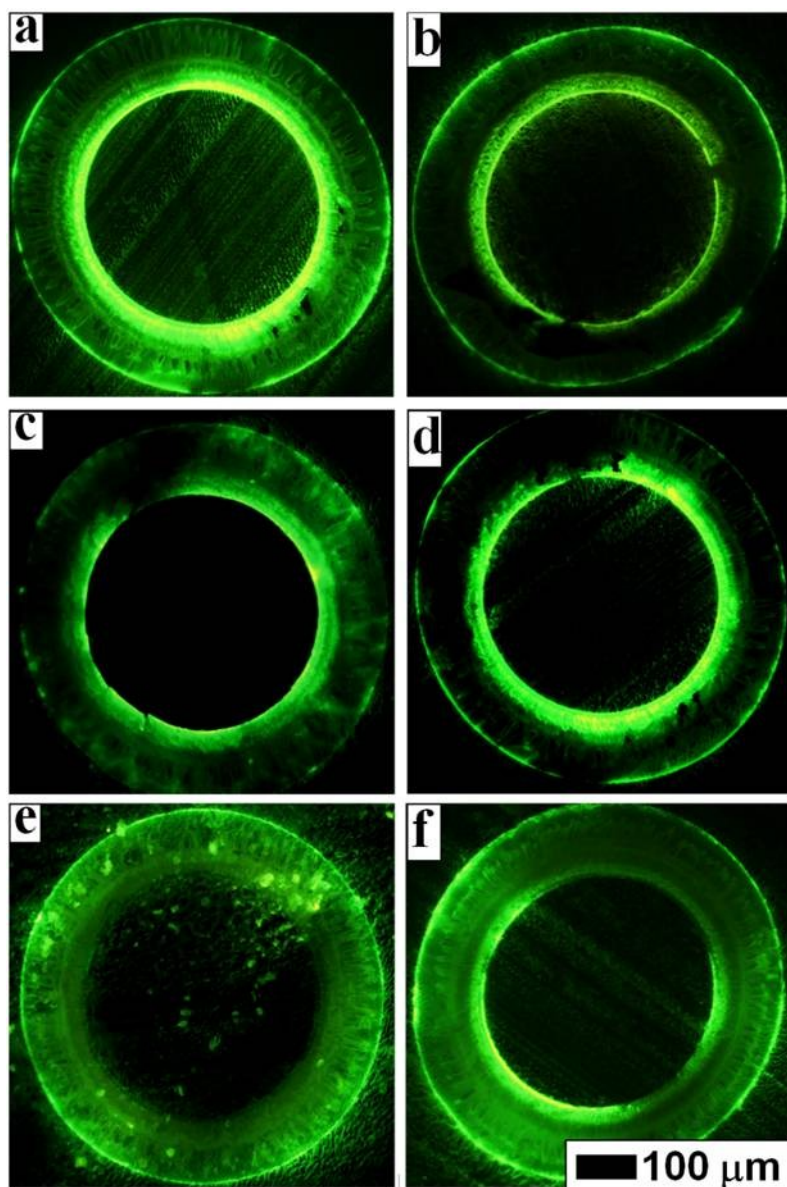


Fig. III. 11 Fluorescent microscope images of cross-sections of FITC-BSA-fouled and backwashed PES membranes with MWCO 30 kDa. (a) Fouled membrane, (b) backwashed using NaClO 500 ppm, (c) urea 8 M, (d) NaOH 0.01 M, (e) SDS 1 wt% (pH 6.2), and (f) SDS 1 wt% at pH 10.

III. 3.5 Fouling behaviors of SA through membranes with different pore-size

To visualize the fouling behaviors of SA in the hollow-fiber membranes, a rhodamine-SA solution (50 ppm) was filtrated through PES membranes with MWCOs 30, 150, and 300 kDa from the outside to the inside of a hollow-fiber membrane, at 15

ml/min at 0.5 atm. The relative flux (J/J_0) during filtration decreased with time (Fig. III. 12a), indicating membrane fouling, and the very similar decline of flux was obtained among three different kinds of membranes. The final rejection (R) of rhodamine-SA reached at 81% for a PES membrane with MWCO 30 kDa, 73% for that with MWCO 150 kDa, and 38% for that with MWCO 300 kDa after 2 h filtration (Fig. III. 12b). The relative flux kept decreasing gradually during the whole 2 h filtration without any sudden increase, indicating that the fouling of SA on the three types of PES membranes progressed with gradual foulant accumulation on the membrane surface or inside the membrane.

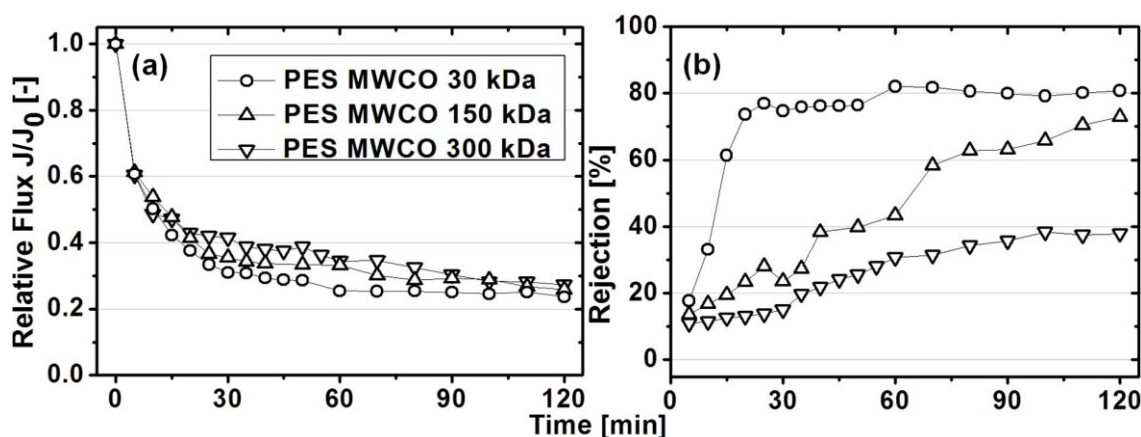


Fig. III. 12 Relative flux (a) and rejection (b) during the outer-pressured filtration of a rhodamine-SA solution through PES hollow-fiber membranes with different MWCOs.

The cross-sectional images of the membranes are shown in Fig. III. 13. The cross-sectional FE-SEM images show that PES membranes with MWCOs 30 kDa and 150 kDa had double-finger structures (Fig. III. 13a and d). While a membrane with MWCO 300 kDa seemed to have a homogeneous structure over the whole cross section (Fig. III. 13g), the magnified SEM image shows that there was a relatively loose structure in the middle of the membrane comparing to the region close to the membrane surfaces (inset of Fig. III. 13g).

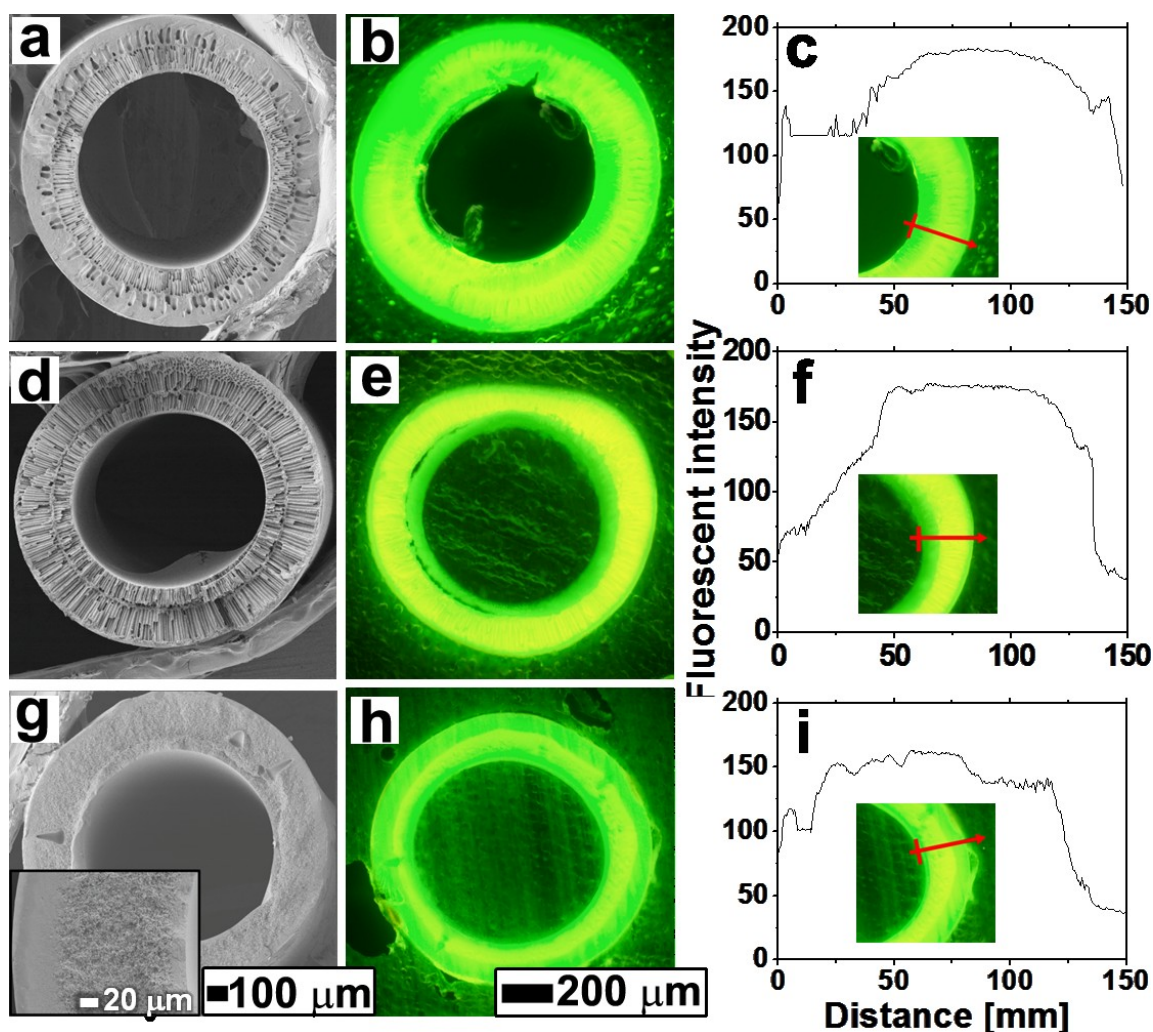


Fig. III. 13. FE-SEM and fluorescent microscope images of the cross-sections, and linear fluorescence distribution of the magnified cross-sections of PES hollow-fiber membranes. FE-SEM images are the cross-sections of unused membranes (a, d and g). Fluorescent images are those after the outer-pressured filtration of 50 ppm rhodamine-SA solution for 2 h (b, e and h). Fluorescence distribution along the red line across the cross-section in the inset (c, f and i). The horizontal axis indicates the distance from inner surface. (a-c) PES membrane with MWCO 30 kDa, (d-f) MWCO 150 kDa and (g-i) MWCO 300 kDa (inset of g is the magnified cross-section).

The fluorescent images of the fouled membranes exhibit different distribution of rhodamine-SA in the cross-section among the membranes (Fig. III. 13b, e and h). Fluorescence was observed over the whole cross-sections of all three types of membranes. Interestingly, there was no obvious foulant layer on the surfaces regardless

the surface morphologies, while the remarkable foulant deposition on a membrane surface was observed in the fouling of protein and humic acid (See Chapter II, section II. 3.1 and II. 3.5). Highly bright fluorescence was found at the middle of all three membranes, indicating large amounts of SA accumulation at these regions. It should be noted that a bare membrane did not exhibit fluorescence under the present observation conditions. It should be noted that we repeated more than three times the microscopic observations using different samples prepared under the same conditions and obtained similar results with small variations in the fluorescent images.

The linear fluorescence distribution along a red line (inset), which crossed the membranes, was plotted in Fig. III. 13c, f and i. The figures also show the highly bright fluorescence at the middle of the membranes. For the cases of the membranes with MWCOs 30 and 150 kDa, the remarkable accumulation of rhodamine-SA was observed at the finger-structured areas, and there was the relatively high accumulation at the loose-structured area of the membrane with MWCO 300 kDa (inset of Fig. III. 13g). It seems that SA molecules preferred to be deposited at vacant spaces of the membranes during filtration. Since alginate is a highly anionic polyelectrolyte, there is repulsion between alginate molecules at neutral pH in the absence of multivalent cations. The repulsive force would prevent the compact deposition of rhodamine-SA inside the membrane. Consequently, rhodamine-SA would tend to accumulate in the vacant spaces (like “a pool”) inside the membrane.

For all the membranes, the rejection of rhodamine-SA was less than 80%, meaning that a portion of rhodamine-SA can permeate through the membranes. SA was natural polysaccharide and had a wide distribution of a molecular weight. That explains that rhodamine-SA reached at the inner surface of all the membranes.

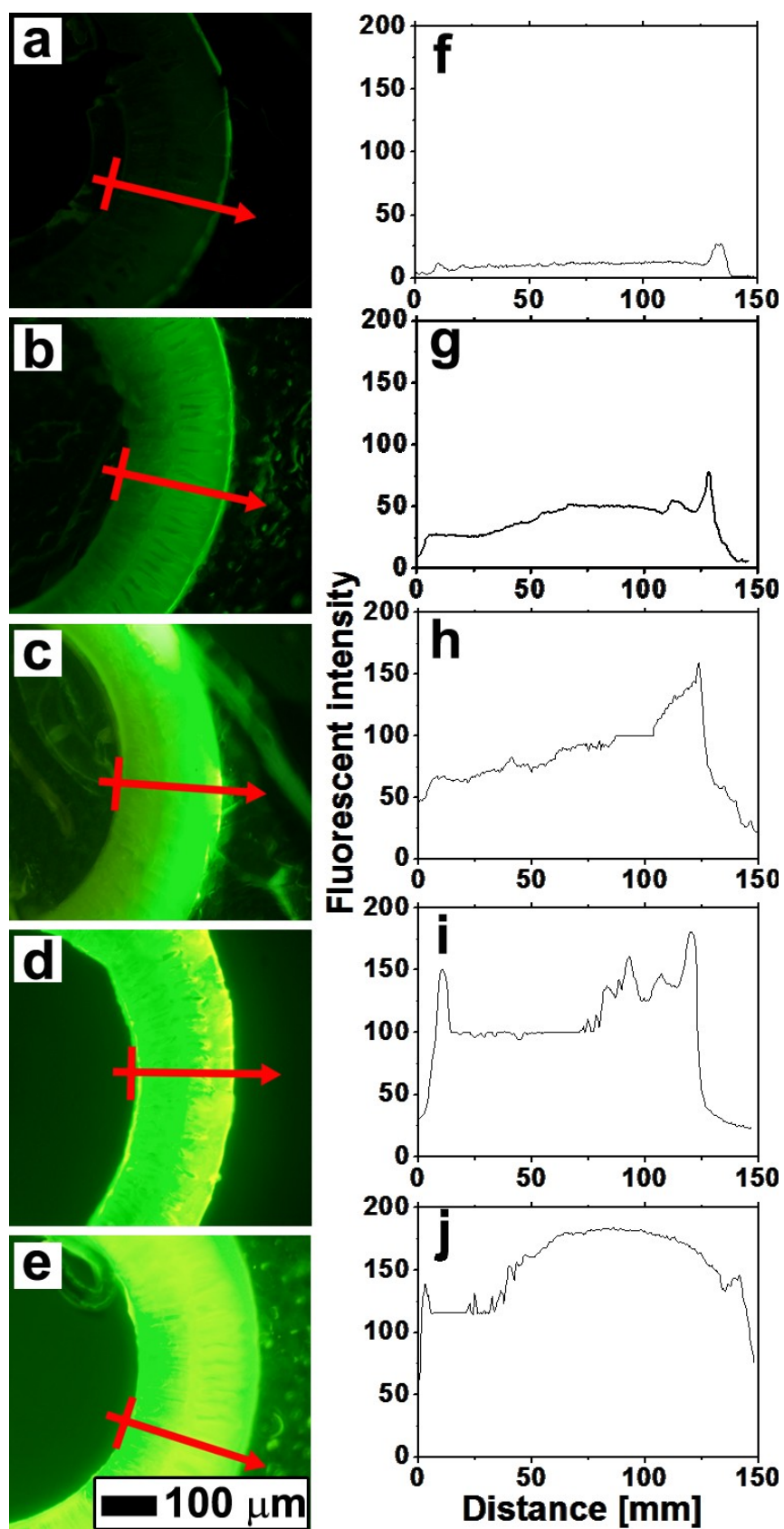


Fig. III. 14 Fluorescent microscope images and linear fluorescence distribution of

magnified cross-sections of PES membranes with MWCO 30 kDa (outer-pressured filtration). The filtration time of 50 ppm rhodamine-SA solution were 1 min (a, f), 10 min (b, g), 30 min (c, h), 60 min (d, i) and 120 min (e, j).

Accumulation behaviors of rhodamine-SA with the filtration time were examined by measuring the linear fluorescence distribution along the red lines in Fig. III. 14a-e. A gradual accumulating process of rhodamine-SA from the outside to the inside was observed and the fluorescence intensity became higher with the filtration time (Fig. III. 14f-j). In the beginning, the high fluorescence was observed close to the membrane outer surface. After 30 min, the fluorescence spread over the whole cross-sectional area of the membrane. After 1 h, the inside of the membrane started to get much brighter and, after 2 h, the region of the finger-structure (distance 40- 125 μm from the inner surface) became much brighter than the outer surface. Although the flux and rejection did not change so much from the filtration time of 1 h to 2 h (Fig. III. 12), rhodamine-SA continued to accumulate at vacant spaces inside the membrane.

III. 3.6 Effect of backwashing on the SA fouled membrane

As described in section III. 3.5, rhodamine-SA tended to accumulate at the vacant spaces inside a membrane during filtration (i.e. finger-structured areas). We then carried out backwashing for 1 min for the fouled membrane that was prepared by the filtration of a rhodamine-SA solution through a membrane (MWCO 300 kDa) for 1 h. The removal of the rhodamine-SA accumulation from the fouled membrane by backwashing with water was supposed to be easier than other foulants (e.g. protein and humic acid), because SA is very hydrophilic and interactions between them were weak. The backwashing efficiency for the fouled PES membrane was 31%. This backwashing efficiency indicates that a certain amount of the rhodamine-SA deposition

(not a large amount) was removed from the membrane.

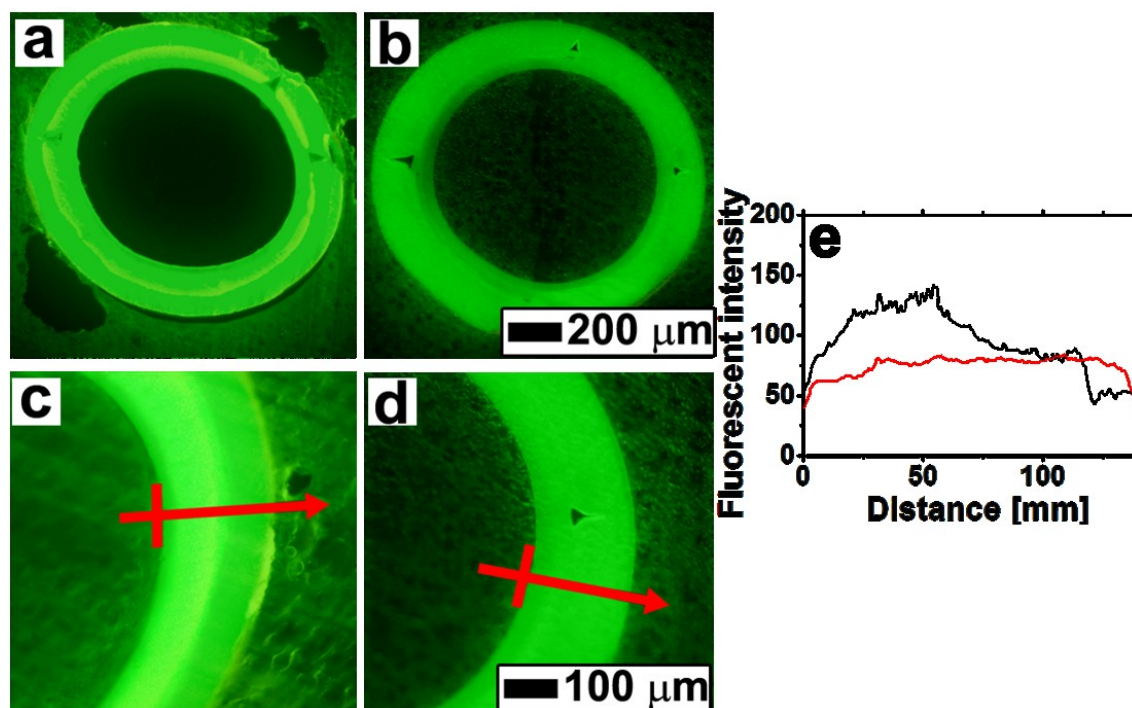


Fig. III. 15 Fluorescent microscope images of the whole (a, b) and magnified (c, d) cross-sections of PES membranes with MWCO 300 kDa, and linear fluorescence distribution of magnified cross-sections (e). A black line is the fluorescence of the fouled membrane before backwashing, a red line is that of the membrane after backwashing. (a, c) PES membranes fouled by 50 ppm rhodamine-SA (outer-pressured filtration for 1 h). (c, d) PES membrane fouled for 1 h and backwashed for 1 min.

Fig. III. 15. shows the fluorescent images of the fouled membrane (filtration for 1 h) and the backwashed membrane with MWCO 300 kDa (backwashing for 1 min). After backwashing, the whole cross section of the membrane exhibits apparently homogeneous fluorescence. Fig. III. 15e also exhibits the decrease and broadening of the fluorescence intensity in the membrane. These results indicate the removal of the remarkable deposition of rhodamine-SA from the middle of the membrane, which would result in the partial flux recovery by backwashing. Fig. III. 15b and e, however, shows that a portion of rhodamine-SA still remained adsorbed in the fouled membrane even after the backwashing, which agrees with the insufficient backwashing efficiency

(30 %).

The remaining rhodamine-SA after the backwashing indicates the interaction between the membrane material (PES) and rhodamine-SA. The surface of a PES membrane is negatively charged [30]. and alginate is also negatively charged due to its carboxylic groups at pH 8.4 (pKa of sodium alginate is generally between 3.4 and 4.4). Although there is the electrostatic repulsion between PES and alginate, a couple of research groups reported the attractive interaction between them [31, 32]. Several reports described that it is relatively easy to remove SA deposition from a membrane by washing a membrane with distilled water comparing to the deposition of proteins and humic substances [31, 33], suggesting a weak interaction between a membrane material and SA. These reports agree with the present results of backwashing.

III. 3.7 Filtration of protein through the membrane fouled with sodium alginate.

The above investigations demonstrate that the SA fouling resulted in the accumulation of SA in the middle of a hollow-fiber membrane, especially in the relatively loose structure. The membrane fouled with SA might exhibit separation characteristics different from intrinsic ones. In particular, the penetration of solutes through a membrane and the rejection of other types of solutes would be affected by the foulant inside a membrane. As we described in section III. 3.1, the fluorescent microscopic observation of cross-sections of hollow-fiber membranes revealed that bovine serum albumin (BSA) was deposited both onto the outer surface and near the inner surface inside of a PES membrane with MWCO 30 kDa during filtration, indicating the presence of the dense structure near the inner surface. The fluorescent microscopic observation can provide the information on the penetration of solutes,

which is related to the pore size of a membrane inside. We filtrated a non-labeled SA solution (50 ppm) through a PES membrane with MWCO 30 kDa for 2 h and then filtrated a fluorescently-labeled BSA solution (50 ppm FITC-BSA) through the membrane for 2 h. After the 4 h filtration, DI water was filtrated for 1 min to eject the feed solution residue from a membrane. Finally, the fouled membrane was sliced to 10 μm in thickness and observed by a fluorescent microscope (fluorescence exposure time 500 ms).

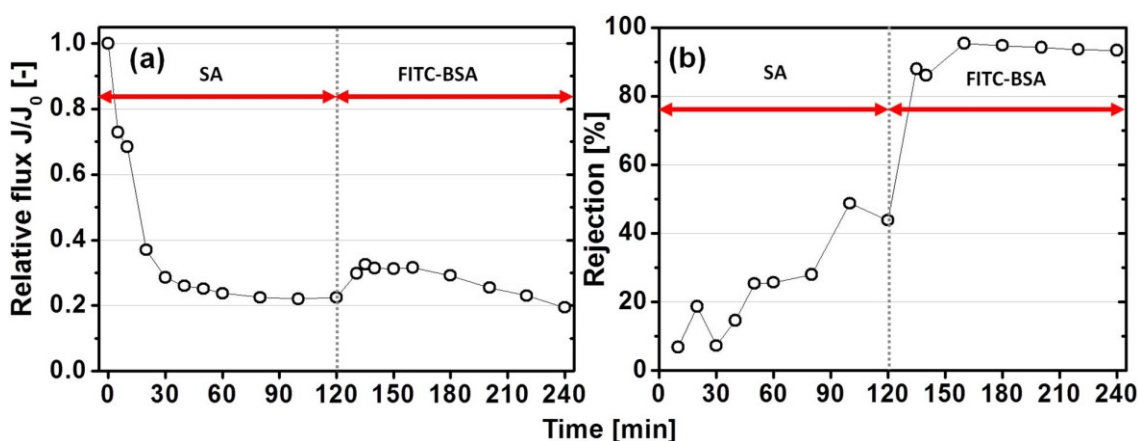


Fig III. 16 Relative flux (a) and rejection (b) during the sequential filtration of 50 ppm SA solution and of 50 ppm FITC-BSA solution through a PES hollow-fiber membrane with MWCO 30 kDa (outer-pressured filtration).

Relative flux (J/J_0) and rejection (R) during the 4 h filtration is shown in Fig. III. 16. In the first filtration using a SA solution, the relative flux of SA was very similar to that of a rhodamine-SA solution (Fig. III. 16a). The rejection was lower than that of a rhodamine-SA solution (Fig. III. 16b), implying that the fluorescent modification using rhodamine 123 increased the hydrophobicity of SA or cross-linked SA via an electrostatic interaction to produce SA of a higher molecular weight. When the feed solution was changed from a SA solution to a FITC-BSA solution, the relative flux increased a little (at 120 min in Fig. III. 16a) and then decreased gradually. The little increase of the flux would be due to the removal of the foulant (SA) by the FITC-BSA

solution. The rejection of FITC-BSA was slightly higher than that of the single filtration using only a FITC-BSA solution, which suggests that the SA fouling would affect the porosity and effective pore size of the membrane.

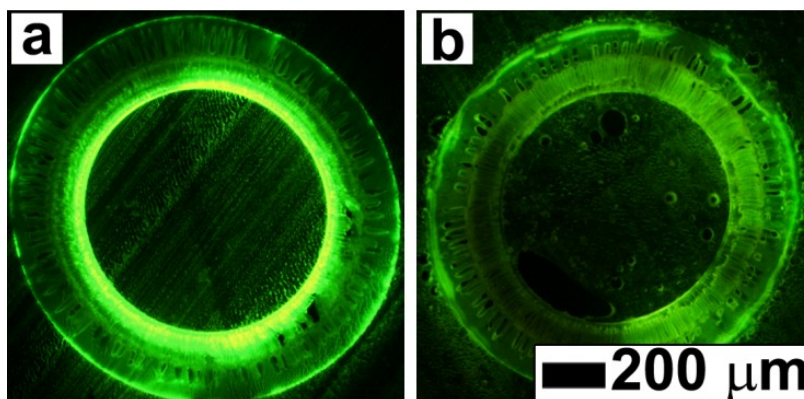


Fig. III. 17 Fluorescent microscope images of the cross-sections of fouled PES membranes with MWCO 30 kDa. (a) The membrane after the filtration of only a FITC-BSA solution (50 ppm for 2 h). (b) The membrane after the filtration of SA (50 ppm for 2 h) and FITC-BSA (50 ppm for 2 h).

The fluorescent microscopic images of the cross sections of fouled membranes are shown in Fig. III. 17. To our surprise, both the deposition amount and distribution area of FITC-BSA inside the membrane fouled with SA and FITC-BSA were less than those of the single filtration using only a FITC-BSA solution. In the absence of any initial SA fouling, the majority of the FITC-BSA was deposited near the inner surface of the membrane (Fig. III. 17a). In contrast, there was no significant deposition of FITC-BSA inside the membrane with initial SA fouling (Fig. III. 17b), and the inner finger-structure was dyed only slightly with FITC-BSA. These results suggested that the accumulation of SA inside the membrane prevented some of the FITC-BSA from penetrating into and adsorbing onto the membrane. The initial accumulation and adsorption of SA might narrow the pores of the membrane. Indeed, backwashing removed only a small amount of SA fouled inside a membrane, meaning stable

adsorption of SA onto a membrane matrix.

III. 3.8 SA Fouling behaviors of inner-pressured types of filtration

An inner-pressured type of filtration is also used in practical filtration processes. As we described above, the PES membranes used here were asymmetric membranes and had a dense layer close to the inner surface, which determined the rejection of solutes. The filtration types (inner- or outer-pressured) might affect membrane fouling and also accumulation of foulants inside a membrane.

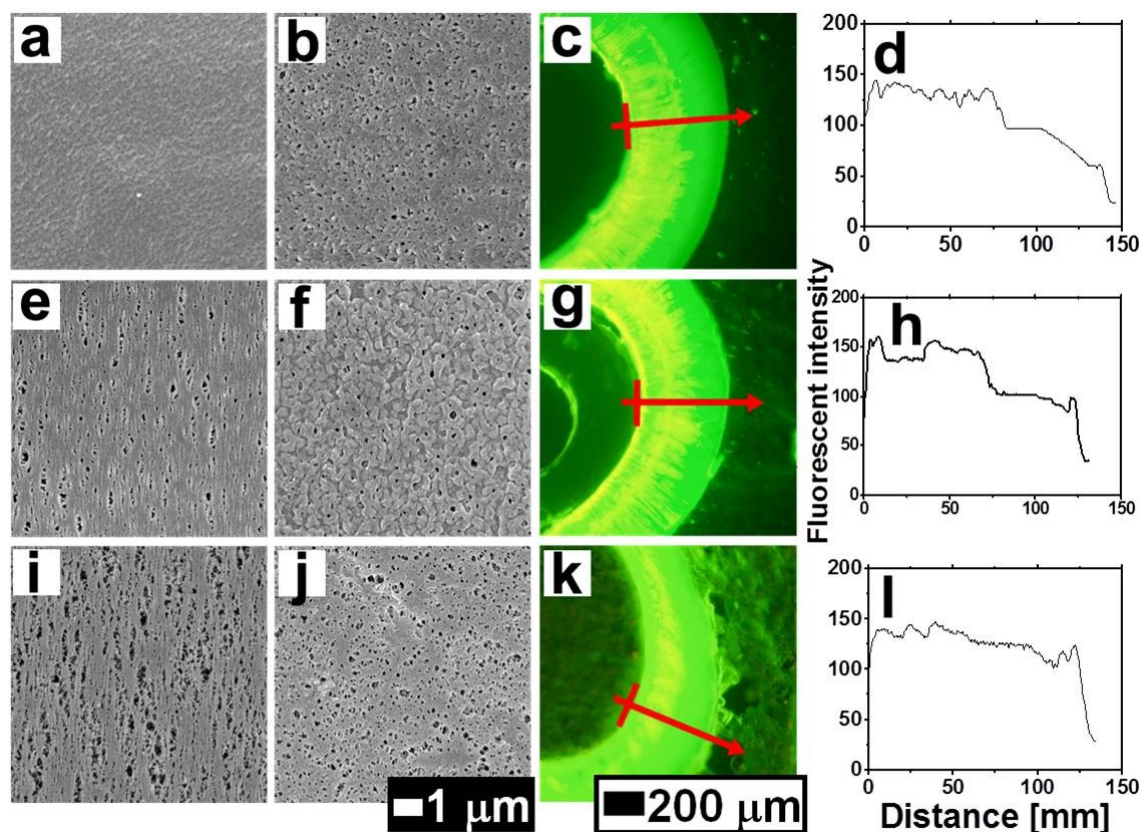


Fig. III. 18 FE-SEM and fluorescent microscope images of PES hollow-fiber membranes, and linear fluorescence distribution of the magnified cross-sections of PES hollow-fiber membrane. FE-SEM images of the inner surfaces (a, e, i) and the outer surfaces (b, f, j) of unused PES membranes. Fluorescent images are the cross-sections of membranes after the inner-pressured filtration of 50 ppm rhodamine-SA (c, g, k). (a-d) PES membrane with MWCO 30 kDa, (e-h) MWCO 150 kDa and (i-l) MWCO 300 kDa. Figs 7d, h, l show

fluorescence distribution along the red line in c, g and k, respectively. The horizontal axis indicates the distance from inner surface.

There was a dense layer at the inner surface of a PES membrane with a MWCO 30 kDa, while the pores of the outer surface were visible (Fig. III. 18a and b). Although submicron open pores were observed on the inner and outer surfaces of PES membranes with MWCOs of 150 kDa and 300 kDa (Fig III. 18e, f, i and j), the morphologies of the inner surfaces were apparently different from those of the outer surfaces.

Fig. III. 18c, g and k show the cross sections of PES membranes fouled with rhodamine-SA by the inner-pressured filtration. Compared with the outer-pressured filtration (Fig. III. 18), the inner-pressured filtration gave relatively low fluorescence intensity in the cross section of PES membrane with MWCO 30 kDa (Fig. III. 18c) and the bright fluorescence was observed close to the inner surface due the blockage of rhodamine-SA at the dense layer close to the inner surface. The higher fluorescent intensity was also confirmed from the fluorescence distribution profile (Fig. III. 18d, from distance around 0 to 50 mm).

The rhodamine-SA distribution inside the PES membrane with MWCO 150 kDa was much different between the filtration types of inner-pressure and outer-pressure. Interestingly, rhodamine-SA accumulated both at the inner surface and at the boundary between the outer and inner finger structures for the inner-pressured filtration (Fig. III. 18g). In Figure 7h, fluorescent intensity at these finger-structure regions (from distance 0 to 10 mm, and distance 30 to 70) was higher than the other regions. In the case of a PES membrane with MWCO 300 kDa (Fig. III. 18k), the inner-pressured filtration gave little larger amounts of accumulation of rhodamine-SA in the inner loose

structures, which was similar to that of the outer-pressured filtration. The fluorescent intensity in this region (from distance 30 to 50 mm) was also slightly higher than other regions (Fig. III. 18l).

It should be noted that the flux and rejection of rhodamine-SA at the inner-pressured filtration of 2 h were quite similar to those of the outer-pressured filtration for each membrane, and both flux and rejection reached a stable state at the end of the filtration (2 h). The difference in the permeation direction resulted in different accumulation of the foulant inside the membranes.

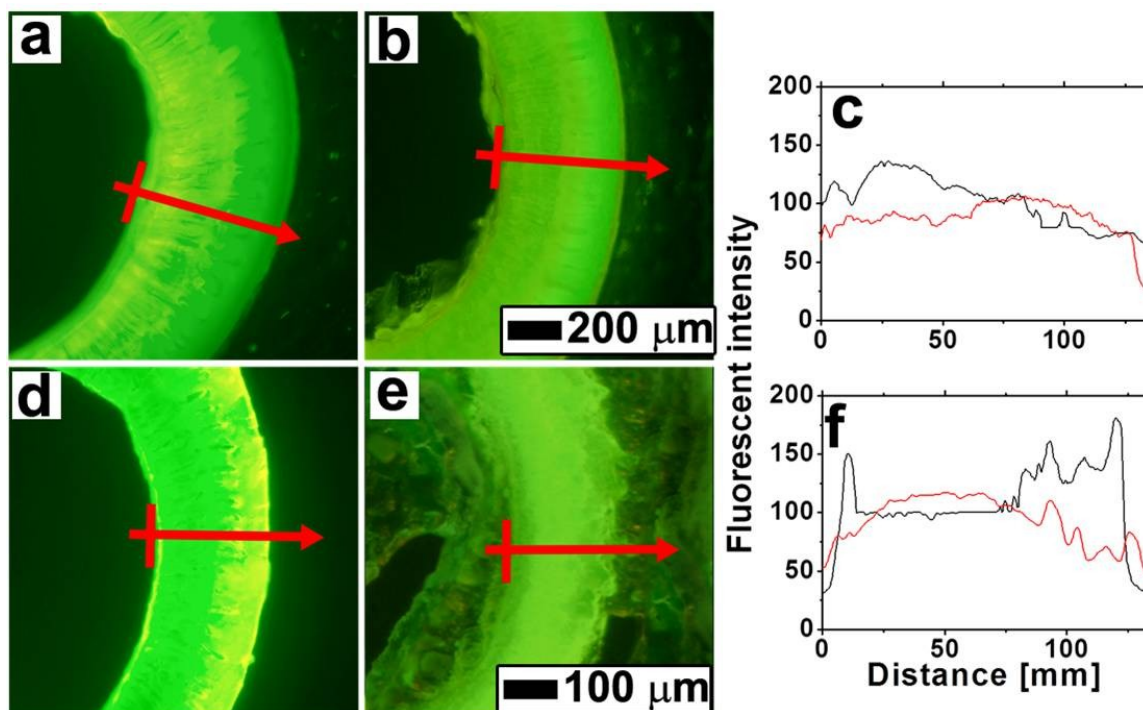


Fig. III. 19 Fluorescent microscope images of the cross-sections of PES membranes with MWCO 30 kDa and linear fluorescence distribution of the cross sections. (a) PES membrane fouled by 50 ppm rhodamine-SA by the inner-pressured filtration (for 1 h), (b) PES membrane fouled by the inner-pressured filtration for 1 h and backwashed (outer-pressured) for 1 min, (c) PES membrane fouled by 50 ppm rhodamine-SA by the outer-pressured filtration (for 1 h) and (d) PES membrane fouled by the outer-pressured filtration for 1 h and backwashed (inner-pressured) for 1 min. (e) Fluorescence distribution along the red lines across the cross-sections in a and b. (f) Fluorescence distribution along the red lines across the cross-sections in d and e. The horizontal axis

indicates the distance from inner surface. The black lines in c and f are the fluorescence of the fouled membrane before backwashing, and red lines are those of membranes after backwashing.

It is possible that the permeation direction affects membrane cleaning and a membrane life. Backwashing after a 1 h filtration was applied to the membranes fouled with different filtration directions and internal accumulation of rhodamine-SA in the membranes were visualized (Fig. III. 19). The backwashing efficiency for the membrane fouled with inner-pressured filtration was approximately 20%. The accumulation of rhodamine-SA near the inner surface was partially removed by backwashing (Fig. III. 19a-c). The backwashing efficiency for the membrane fouled with inner-pressured filtration was approximately 14%, which was lower than that of the inner-pressured filtration. However, it was also observed that the accumulation near the outer surface was partially removed by backwashing (Fig. III. 19d-f). The difference of the filtration direction affected fouling geometry due to the asymmetric structure of the membrane and also resulted in different backwashing efficiencies.

Above results suggest that a structure of a hollow-fiber membrane should be carefully designed for filtration processes and also that a filtration operation should be designed to a structure of a hollow-fiber membrane, in order to keep the membrane performance and to reduce the maintenance cost of the membrane module.

II.4 CONCLUSIONS

In this study, we succeeded in visualization and characterization of the internal fouling of a hollow-fiber membrane caused by model proteins and model extracellular polymeric substance sodium alginate (SA). The present methodology can reveal which part of a membrane determines the rejection of solutes and also which part of a fouled

membrane can be washed by backwashing. This study provided novel information on membrane fouling and aid understanding of membrane fouling.

Internal fouling behaviors of three model proteins were remarkably different, although there were similarities in the permeability decline amongst these proteins. We also prepared membranes with differing structures (with and without a skin-layer on the outer surface) and demonstrated that internal fouling behavior varied with a skin-layer, even though there were no big differences in water permeability or permeability decline during filtration. Finally, we used various cleaning agents in backwashing, and reveals that backwashing efficiency and which part can be cleaned during backwashing was very depended on properties of cleaning agents.

Three types of PES membranes with different MWCOs were filtrated with fluorescently-labeled SA solution, and SA fouling behaviors differed from the membrane pore sizes, structures, and filtration directions. The SA deposition inside a membrane was much different from the protein deposition. Furthermore, the SA fouling inside a membrane affected protein fouling.

The method we developed was simple to use and versatile. It requires only a fluorescent microscope and a sample-slicing technique. It actually can be applied to different types of foulants which can be fluorescently labeled. If the foulants in target raw water are identified, internal fouling will provide useful information for the rational design and rational selection of a membrane type. From the aspect of a membrane structure, surface modification of a membrane and design of a membrane structure could be also studied using a fluorescently-labeled chemical as a marker by this method.

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Chapter IV

Reorganization of the surface geometry of hollow-fiber membranes using dip-coating and vapor-induced phase separation

IV.1 INTRODUCTION

Phase separation of a polymer solution is a powerful approach to prepare a polymeric porous hollow-fiber membrane. Nonsolvent-induced phase separation (NIPS) is the major approach to commercially prepare polymeric membranes. It easily produces a porous membrane with macrovoids and with relative low mechanical strength [1]. Thermally induced phase separation (TIPS) is another approach to prepare a porous membrane without a macrovoid. The membrane without a macrovoid has a relatively homogeneous structure of an inner membrane matrix, which contributes to high mechanical strength of a membrane and the high water permeability. In many cases, a hollow-fiber membrane prepared via TIPS has a dense layer on its outer-surface and the dense layer is supposed to determine the separation characteristics of a membrane [2, 3].

FE-SEM images of an example membrane prepared using the NIPS method was shown in (Fig. IV. 1). It would be of great importance to prepare a hollow-fiber membrane having a controlled surface geometry and a homogeneous membrane matrix.

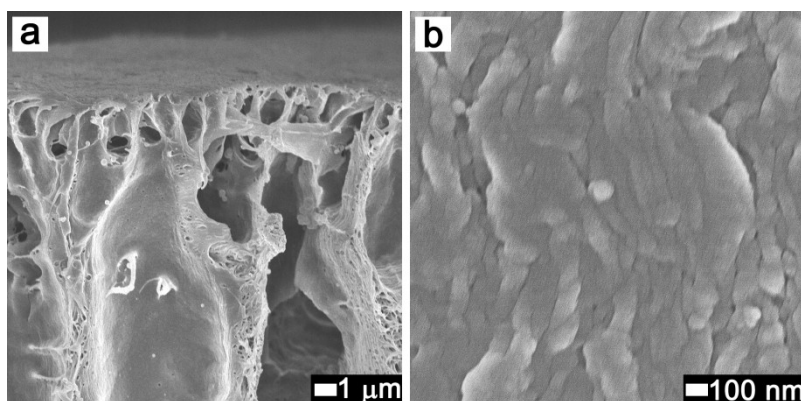


Fig. IV 1 FE-SEM images of the cross-section near the outer surface (a) and the outer surface (b) of a hollow-fiber membrane prepared using the NIPS method (PVDF/NMP 20/80 wt%).

Vapor-induced phase separation (VIPS) is classified as a NIPS process. VIPS is a very slow process compared with other phase separation processes due to the slow mass-transfer rate of a non-solvent vapor to a polymer solution [2]. There are only a limited number of reports describing membrane preparation via VIPS, although VIPS can produce membranes with various types of surface morphologies, especially with highly porous and ordered surface pattern [4]. These reports described mainly the preparation of flat membranes via VIPS. VIPS sometimes plays an assistant role in the preparation of hollow-fiber membranes via NIPS [4, 5]. Before a polymer solution extruded enters into a coagulation bath to induce the phase separation, controlled water vapor in an air gap (the space between an extruder spinneret and a coagulation bath) induces the partial phase separation of a polymer solution to help the formation of a membrane surface. However, the control of the surface geometry at the air gap was very limited due to the too short time for VIPS in the air gap.

Herein, I employed TIPS, dip-coating and VIPS to prepare a hollow-fiber membrane with controlled surface geometry and with a homogeneous inner membrane matrix, which led to high membrane performances. A PVDF hollow-fiber membrane was

prepared by the TIPS method, and the membrane was then dip-coated with another PVDF solution, followed by a VIPS treatment. The VIPS treatment produced a reorganized outer surface of the membrane with a controlled geometry, which facilitated as a new separation layer for a hollow-fiber membrane.

Using this approach, a series of hydrophilic synthesized copolymers poly(MMA-*r*-PEGMA) were also used in dip-coating to modify only the membrane surface in order to improve the hydrophilicity of the surface and the anti-fouling properties.

IV.2. EXPERIMENTAL

IV.2.1 Materials

Poly(vinylidene fluoride) (PVDF) (Type 6020, MW 322 kDa) was purchased from Solvay Soiexis K.K (Tokyo, Japan). Methyl methacrylate (MMA), 2-aminoethyl methacrylate hydrochloride (AEMA-HCl), fluorescein isothiocyanate (FITC) and humic acid were purchased from Aldrich Chemical Co. (Milwaukee, USA). Methyl methacrylate (MMA) and poly(ethylene glycol) methyl ether methacrylate (Mn 300 and 500) was also obtained from Aldrich Chemical Co. (Milwaukee, USA). Triethanolamine-HCl and 2,2'-azodiisobutyronitrile (AIBN) were purchased from Tokyo Chemical Industry Co. (Tokyo, Japan). Polystyrene nanospheres and coomassie plus (Bradford) protein assay were purchased from Thermo Scientific, (Fremont, USA). All other chemicals were purchased from Wako Pure Chemical Industries (Osaka, Japan). The water used was high-quality deionized water (DI water, $> 15 \text{ M}\Omega \text{ cm}^{-1}$) produced by an Elix-5 system (Millipore, Molsheim, France).

IV.2.2 Membrane preparation (TIPS)

A model PVDF hollow-fiber membrane was prepared using a batch-type extruder (BA-0, Imoto Machinery Co., Kyoto, Japan) by the thermally-induced phase separation (TIPS) method. A homogeneous PVDF solution (PVDF/diethyl phthalate (DEP) = 25/75 wt%) at 190 °C was extruded through a spinneret with an inner lumen made by a bore solution (DEP) into a water bath (20°C, quenching bath) to induce phase separation and solidification. The gap between the spinneret and the bath was 5 mm, and the membrane was wound on a take-up winder at approx. 0.26 m/s. Finally, the residual solvent in the prepared membrane was extracted using excess amounts of ethanol and DI water. The membranes were stored in DI water until use. The outer diameter of the membrane was approx. 1.0 mm and the inner diameter was approx. 0.6 mm. The detail experimental methods were described in Chapter III, section III. 2.3.

A blend hollow-fiber membrane composed of PVDF and a synthesized hydrophilic copolymer poly(MMA-*r*-PEGMA) (See section IV.2.5) was also prepared by the same procedure. The composition of the polymer solution for extruding was PVDF 24 wt% and poly(MMA-*r*-PEGMA) (M_n of PEGMA 300) (9:1 mole ratio) 2 wt% and 74 wt% DEP. Temperature for the extruding solution and a coagulation bath was 190°C and 20°C, respectively. All the other conditions were the same with those of a PVDF hollow-fiber membrane. The composition in the blend membrane prepared was analyzed by the elemental analysis.

IV.2.3 Surface reorganization using dip-coating and VIPS

For the reorganization of a surface of a PVDF hollow-fiber membrane, another PVDF solution was prepared for dip-coating. Dip-coating solutions were prepared by

dissolving weighted PVDF (or other polymers) into a mixture of an organic solvent and nonsolvent (for PVDF). A dimethylacetamide (DMAc)/ethanol (65/35 vol%) mixture was used for a dip-coating solution. . The PVDF solutions were stirred at 80°C for 2 h. A mixture of *N*-methylpyrrolidone (NMP)/toluene/ethanol (60/10/30 vol%) was used only for the dip-coating solution containing poly(MMA-*r*-PEGMA (M_n 300)) (7:3 mole ratio)/PVDF (0.5/0.5 wt%).

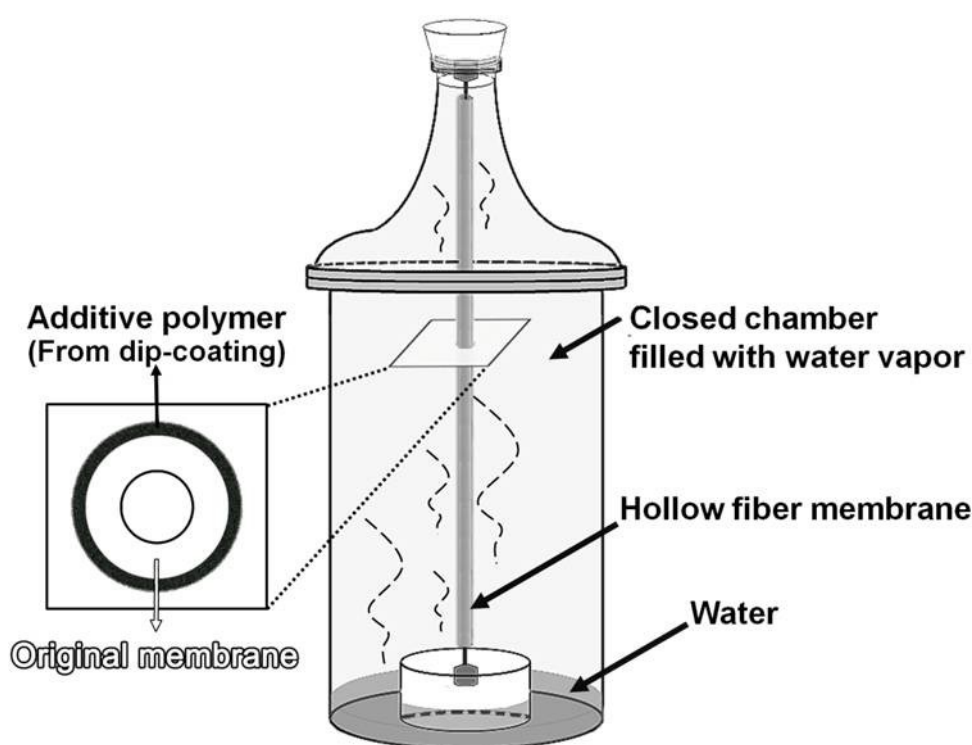


Fig. IV.2 Schematic of VIPS treatment set-up.

A PVDF hollow-fiber membrane of approx. 25 cm in length, whose both ends were tied up not to allow a dip-coating solution to enter into the lumen, was dipped in a dip-coating solution for approx. 10 s and was slowly taken up by hand. The dip-coated membrane was set in a closed chamber saturated with a water vapor at controlled given temperature to induce VIPS of the dip-coated layer (Fig. IV. 2). The transfer time between coating and putting a membrane in a chamber was kept less than 15 s. After

the VIPS treatment, the membrane was immersed into DI water for several minutes to thoroughly solidify the surface. The water was then kept stirring for several hours for extracting organic solvents from the membrane.

IV.2.4 Schematic of VIPS treatment set-up.

A fluorescent copolymer, poly(methyl methacrylate-co-2-aminoethylmethacrylate)-fluorescein isothiocyanate) (poly(MMA-*r*-AEMA-FITC) (98:2 molar ratio), was synthesized for visualizing the polymer layer dip-coated on a membrane surface (Fig. IV. 3). The method for observing the cross-section of the surface-reorganized PVDF membrane was the same as that described in Chapter III, section III.2.6.

A random copolymer (poly(MMA-*r*-AEMA·HCl)) composed of methyl methacrylate (MMA, 98 mol%) and 2-aminoethylmethacrylate (AEMA, 2 mol%) was synthesized via free radical polymerization [6]. MMA (147 mmol, 14.7 g), AEMA·HCl (3.0 mmol, 0.5 g), and AIBN (38.3 mg, 0.2 mmol) were dissolved in 20 g of 2-propanol/water mixture (4:1 weight ratio) in a 50 ml glass vial. The mixture was purged with nitrogen gas for 30 min first, and polymerization was performed in the closed vial at 60°C for 6 h. The produced copolymer was precipitated by adding excess water, and dried under vacuum at 50°C overnight. The yield was 60%.

Poly(MMA-*r*-AEMA·HCl) (1 g, approximately 1.5 mmol), Et₃N (3 g, 30 mmol) was dissolved in 20 ml chloroform in a screw-cap glass vial, and the solution was kept stirring at room temperature for 30 min. FITC (0.5 g, 3 mmol) was added into the solution and the solution was kept stirring for 4 h to conjugate FITC with the amino groups of poly(MMA-*r*-AEMA). The produced fluorescent copolymer was precipitated

in excess methanol, redissolved in 10 ml chloroform, and reprecipitated in an excess amount of *n*-hexane. The product was freeze-dried overnight and stored in dark until use.

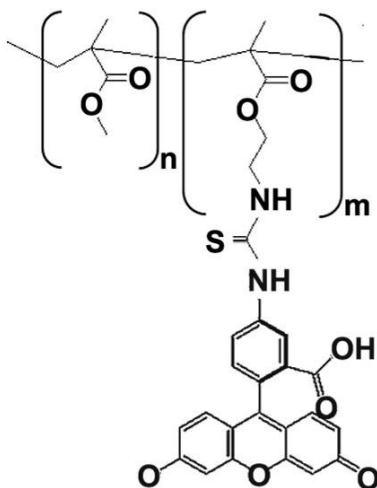


Fig. IV. 3 Chemical structure of the synthesized fluorescent copolymer.

The reaction product in each step was characterized by ^1H NMR (300 M Hz; in chloroform-*d*, Varian Gemini 300, Varian, Palo Alto, CA, USA), FT-IR (Fourier transform infrared spectroscopy, Bruker Optics, ALPHA with Ge, Billerica, MA, USA), and GPC (Gel permeation chromatography, Jasco LC2000 plus, Tokyo, Japan) equipped with an $80 \times 300 \text{ mm column} \times 2$ (ShowaDenko K. K., Tokyo, Japan) and a RI detector (Jasco RI2031 plus) at 40°C . Chloroform was used as an elution solvent, and PMMA molecular weight standards were used for a standard curve. The monomer and FITC compositions in the final product, poly(MMA-*r*-AEMA-FITC), was determined by elemental analysis (Systems Engineering Inc., Tokyo, Japan).

NMR and IR assignments and elemental analysis for poly(MMA-*r*-AEMA-FITC): ^1H NMR chemical shifts at 3.6 ppm ($-\text{CO}_2-\text{CH}_3$, $-\text{CO}_2-\text{CH}_2-\text{CH}_2-$), 1.9–1.81 ppm (C- CH_2-), 1.25–0.85 ppm (C- CH_3) indicate the MMA and AEMA moieties. The FT-IR measurement shows the absorbance at 1729 and 1148 cm^{-1} , suggesting the C-O and

C=O bonds in the copolymer. GPC revealed that M_n of the copolymer was 2.7×10^5 and that M_w/M_n was 1.19. The elemental analysis suggested that the molar ratio of MMA/AEMA was 55:1.

IV.2.5 Synthesis of hydrophilic copolymer

A series of random hydrophilic copolymers, composed of methyl methacrylate (MMA) and poly(ethylene glycol)methyl ether methacrylate (PEGMA), were synthesized for partial modification of a PVDF membrane surface (Fig. IV. 4).

The random copolymers were synthesized via free radical polymerization. The compositions of MMA and PEGMA moieties were shown in Table IV. 1.

Table IV. 1 Synthesis of 4 types of poly(MMA-*r*-PEGMA).

Copolymer	MMA	PEGMA		MMA : PEGMA (mole ratio)
		M_n 300	M_n 500	
1	9.0 g /90 mmol	3 g /10 mmol	--	9 : 1
2	7.5 g /75 mmol	7.5 g /25 mmol	--	7.5 : 2.5
3	7.0 g /70 mmol	9 g /30 mmol	--	7 : 3
4	9.0 g /90 mmol	--	5 g /10 mmol	9 : 1

Toluene 50 ml was used as a solvent, and AIBN (32.84 mg, 0.2 mmol) was used as an initiator for each case. The mixture was dissolved in a 100 ml glass vial, and purged with nitrogen gas for 30 min before polymerization. The polymerization was performed in the closed vial at 60°C for 6 h. The produced copolymer was precipitated by adding excess hexane, and dried under vacuum at 50°C overnight. The yield was approx. 90%.

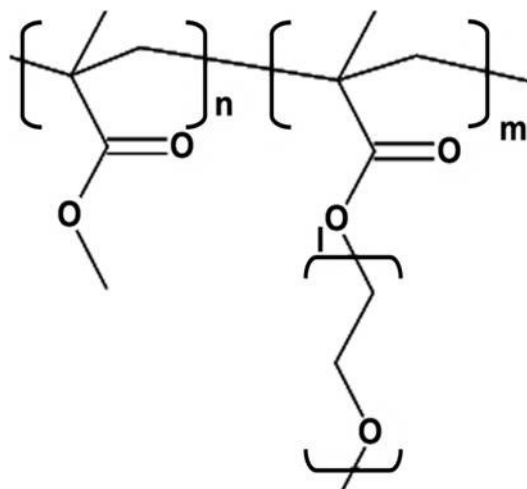


Fig. IV. 4 Chemical structure of the synthesized hydrophilic copolymer.

GPC analysis revealed that M_n was 9.7×10^4 for poly(MMA-*r*-PEGMA) (M_n of PEGMA 300) (MMA:PEGMA 9:1 mole ratio), M_w/M_n was 1.62; M_n was 1.1×10^5 for poly(MMA-*r*-PEGMA) (M_n of PEGMA 500) (MMA:PEGMA 9:1 mole ratio), M_w/M_n was 1.68; M_n was 1.2×10^5 for poly(MMA-*r*-PEGMA) (M_n of PEGMA 300)) (MMA: PEGMA 7.5:2.5 mole ratio), and M_w/M_n was 1.70; M_n was 1.3×10^5 for poly(MMA-*r*-PEGMA) (M_n of PEGMA 300) (MMA: PEGMA 7:3 mole ratio), M_w/M_n was 1.45. The produced copolymers were characterized and confirmed by the FTIR measurement, ^1H NMR, and elemental analysis.

IV.2.6 Characterization of surface reorganized membranes

DI water filtration was performed with a laboratory-scale apparatus developed in our laboratory at room temperature to determine the pure water permeability of a membrane (See Chapter II, Fig. II. 1). The apparatus consisted of a minimodule device that contained a single hollow-fiber membrane, 10.6 cm in length, with an outer diameter of approx. 1.0 mm and an inner diameter of approx. 0.6 mm. DI water was permeated from the outside to the inside of a hollow-fiber membrane at a flow rate of

15 ml/min at a transmembrane pressure of 0.5 atm (Chapter II, Fig. II. 1). The transmembrane pressure was controlled by nitrogen gas. Pure water permeability was measured after 5 min filtration of DI water.

The pore-size of the membrane was evaluated by filtrating solutions containing standard polystyrene nanospheres with diameter of 80 nm, 100 nm, 200 nm and 300 nm, respectively. The feed solutions containing polystyrene nanospheres were prepared by dispersing standard nanospheres stock solutions into buffer solutions containing Triton-X (0.1 wt%). The polystyrene nanospheres with the diameter above 100 nm were dispersed at the stock solution/buffer ratio of 1/300 vol%. The polystyrene nanospheres with the diameter less than 100 nm were dispersed at the ratio of 1/100 vol%.

An aqueous solution containing styrene nanospheres was filtered through a membrane from the outside to the inside at 15 ml/min at 0.5 atm. Each filtration experiment was carried out for 20 min to reach a stable permeation state. The rejection of the nanospheres, R_{np} , by a membrane was determined by measuring the absorbance at 380 nm in the permeate solution using a UV spectrophotometer (GE Healthcare Japan, Tokyo, Japan). The rejection was defined as follows,

$$R_{np} = \left(1 - \frac{[\text{Absorbance in permeate}]}{[\text{Absorbance in feed}]} \right) \times 100 \quad [\%] \quad (1)$$

The pore-size of a membrane was defined as the size of the diameter of nanospheres when the rejection reached at 90%. Each experiment was repeated for 3 times to confirm the reproducibility.

The tensile strength of a hollow-fiber membrane was measured with a tensile strength tester (AGS-J, Shimadzu Co., Kyoto, Japan). A membrane was fixed

vertically using two pairs of tweezers. The sample was extended at a constant rate of 50 mm/min until broken.

The cross-sectional and surface morphology of a hollow-fiber membrane was observed by a field emission-scanning electron microscope (FE-SEM, JSM-7500F; JEOL Ltd., Tokyo, Japan) operating at an accelerating voltage of 5 kV. Samples were sputter-coated with Pt/Pd.

The surface hydrophilicity was determined by measuring the contact angle of an air bubble θ , while immersing the membrane samples in DI water, using a contact angle meter (Drop Master 300, Kyowa science. co., LTD., Niiza, Japan). The higher the air bubble contact angle is, the higher hydrophilic the membrane surface is.

PVDF membrane surfaces were analyzed by FT-IR and X-ray photoelectron spectroscopy (XPS). The membrane samples for XPS analysis were freeze-dried and the elemental composition of the outer surfaces were measured using PHI X-tool (ULVAC-PHI, Incorporated., Chigasaki, Japan). XPS measurements were carried out using an Al K α source operated at 24 W. Photoelectrons were detected with a hemispherical analyser at a pass energy of 280 eV at 45° take-off angle. The data was analyzed using the MultiPak software.

IV.2.7 Membrane filtration and cross-sectional visualization

Humic acid, bovine serum albumin (BSA, MW 67,000) and casein (from bovine milk, MW 19,000–25,000) were used in the filtration experiments as model foulants. A humic acid solution (50 ppm, pH 6.75 ± 0.05) was prepared by dissolving weighted humic acid in a 0.5 mM NaHCO₃ solution and stirring for 1 h, and the pH of the solution was adjusted by adding 0.1 M HCl solution. A BSA solution (1000 ppm, pH

7) was prepared by dissolving weighed BSA in a 0.05 M phosphate buffer solution (pH 7) and stirring for 1 h at 25 °C. Casein solutions (10 or 20 ppm) were prepared by dissolving weighed casein in a 50 mM triethanolamine–HCl/NaOH buffer solution (pH 7.5 or 7) and stirring for 3 h at 25 °C; the triethanolamine HCl/NaOH buffer was used because it does not interfere with protein microassays performed using the Bradford method.

In order to perform fluorescent microscopic observation of the membrane fouling, fluorescently-labeled casein (FITC-casein) was also used as a foulant in filtration experiments. The fluorescent modification of casein was described in Chapter III, section III.2.2. The degree of FITC modification was approx. 0.5 mol of FITC/mol of casein.

Filtration experiments were performed with the apparatus, which was used for measuring the pure water permeability, using aqueous solutions containing different foulants at room temperature. Each filtration experiment was carried out by permeating a feed solution from the outside to the inside of a hollow-fiber membrane at a flow rate of 15 ml/min at a transmembrane pressure of 0.5 atm (Chapter II, Fig. II.1). Prior to filtration experiments, DI water was passed through a membrane to measure the pure water permeability, J_0 . Filtration of a feed solution was then carried out to measure the permeability, J . The rejection of foulants (without fluorescent modification), R , by a membrane was determined by measuring the absorbance of the permeate and feed solutions using a UV spectrophotometer. Wavelengths of absorbance measurement were 256 nm for humic acid solutions and 280 nm for protein solutions. The rejection of FITC-casein was determined by fluorescent intensity measurement using a fluorescent spectrometer (FR-8200, JASCO, Tokyo, Japan).

To visualize the internal fouling of the membrane fouled with FITC-casein, the cross-section of a hollow-fiber membrane at the position of approx. 3 cm from the inlet of a feed solution was sliced and observed using a fluorescent microscope (Olympus IX 81) (Chapter III, Fig. III. 2c). The fluorescent intensity was set at an exposure time of 100 ms with a sensitivity of ISO 200, under which conditions a PVDF membrane before use exhibited no fluorescence. The detail experimental methods were described in Chapter III, section III.2.6.

IV.3 RESULTS AND DISCUSSION

IV.3.1 Effects of the dip-coating (using a PVDF solution) and VIPS on membrane surface morphologies

The PVDF hollow-fiber membrane prepared by the TIPS method had a spherulite structure on the outer surface and in the inside without a macrovoid (Fig. IV. 5a, b and c). Fig. IV. 5b shows the homogeneous inner structure. There observed pores with various sizes (from submicrometers to few micrometers in diameter) between spherulites of approx. 10 μm in diameter, meaning that the porosity of the surface was not high. FE-SEM observations of the outer-surface and the cross-section indicate that the porosity inside the membrane was higher than that of the outer surface. These morphological differences between the surface and the inside of the membrane can be attributed to the evaporation of a solvent in the air gap during the membrane preparation via TIPS, which resulted in the relatively high PVDF concentration at the surface of the PVDF solution extruded from a spinneret. The membrane prepared exhibited high initial water permeability ($1500 \text{ L}/(\text{m}^2 \cdot \text{h} \cdot \text{atm})$) and had high mechanical strength (tensile strength was approx. 3 MPa).

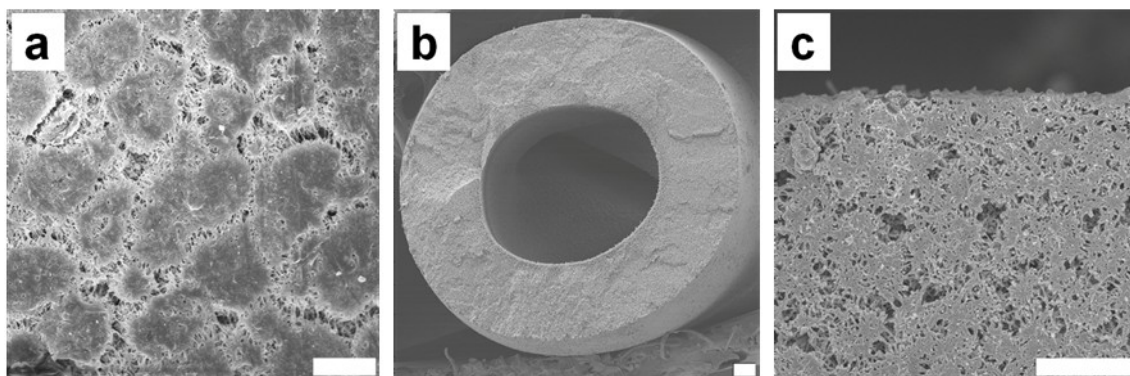


Fig. IV. 5 FE-SEM images of the outer surface (a) and the cross-section (b, c) of the untreated PVDF membrane. (b) The whole cross-section. (c) The magnified cross-section near the outer surface of the membrane. Scale bars in (a, c) represent 10 μm , and a scale bar in (b) represents 100 μm .

The dip-coating was carried out using a mixture of DMAc and ethanol (65/35 vol%) as a solvent for PVDF. Since DMAc is a good solvent to solubilize PVDF [7], the use of DMAc alone as a solvent for dip-coating resulted in the rapid dissolution of a PVDF hollow-fiber membrane. I found that the addition of 35 vol% ethanol to DMAc can solubilize PVDF at a few wt% and that the DMAc/ethanol solution containing PVDF at a few wt% did not dissolve a PVDF membrane entirely.

DMAc/ethanol solutions containing 0.5 wt%, 1.5 wt% and 2 wt% PVDF were used to dip-coat the membrane at 80°C, and this was followed by the VIPS treatment of the coated PVDF solution for 10 min at 80 °C. FE-SEM observations show that mesh-like structures were formed on the outer surfaces after the dip-coating and the VIPS treatment (Fig. IV. 6a-c). The amounts of PVDF immobilized on the original membrane surface by the dip-coating increased when increasing the PVDF concentration in dip-coating. The immobilization amounts were 0.02 mg/cm² for a 0.5 wt% PVDF dip-coating solution, 0.06 mg/cm² for a 1.5 wt% dip-coating solution, and 0.09 mg/cm² for a 2 wt% dip-coating solution. The mesh-like layers newly formed on

the surfaces were not peeled off (Fig. IV. 6d), indicating the stable adhesion of the newly formed PVDF layer on the original membrane.

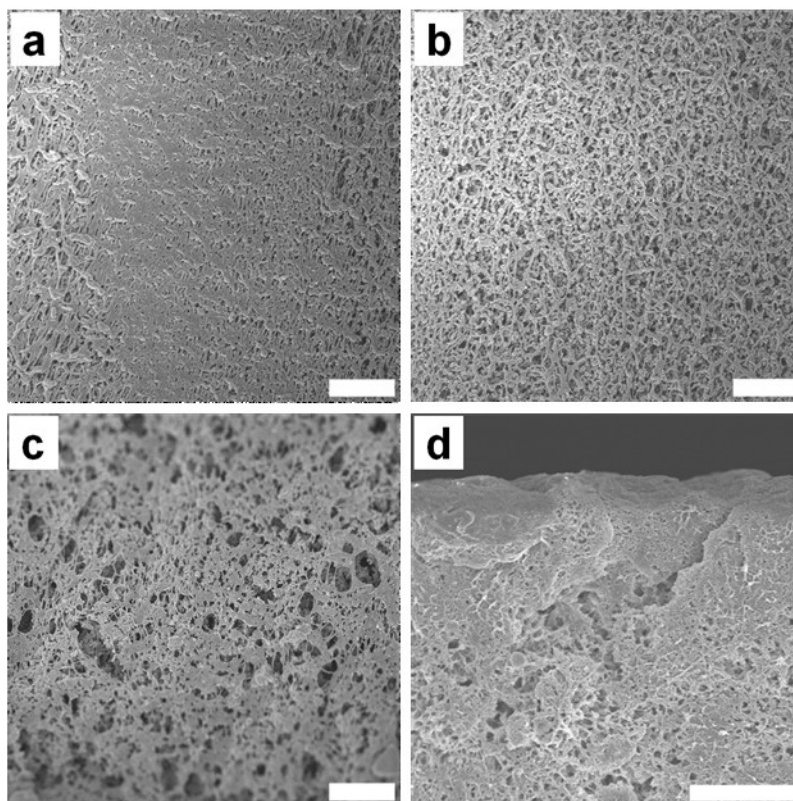


Fig. IV. 6 FE-SEM images of the outer surfaces (a-c) and cross section (d) of surface-reorganized PVDF membranes. The membranes were prepared by dip-coating and VIPS. Dip-coating solutions were PVDF solutions (in DMAc/ethanol = 65/35 vol%) with the concentrations of (a) 0.5 wt%, (b, d) 1.5 wt% and (c) 2 wt%. Temperature for dip-coating and VIPS was 80 °C. VIPS time was 10 min. Scale bars represent 10 μ m.

Temperature for dip-coating affects the solubility of PVDF in the solvent mixture and temperature for VIPS also affects the vapor pressure of water. The geometry of the membrane surface newly formed in the present method would be influenced by temperature for dip-coating and VIPS. Dip-coating using a 0.5 wt% PVDF solution at 25 °C, 40 °C, 60 °C, 80 °C was performed, and this was followed by the VIPS treatment for 10 min at each temperature. In Fig. IV. 7, the surface became more uniform when increasing the temperature. At low temperature (25 °C), nubby

deposition was observed on the surface and the original spherulite structures still seemed remained (Fig. IV. 7a), while the mesh-like layer completely covered the surface at 80 °C (Fig. IV. 7d). Increasing the temperature will increase the solubility of PVDF in mixed DMAc/ethanol (65/35 vol%). Dip-coating using only a mixture of DMAc/ethanol 65/35 vol% (PVDF 0 wt%) failed to obtain an uniform membrane surface, which means that the presence of PVDF at a low concentration in dip-coating solution was important to prepare a homogeneous surface.

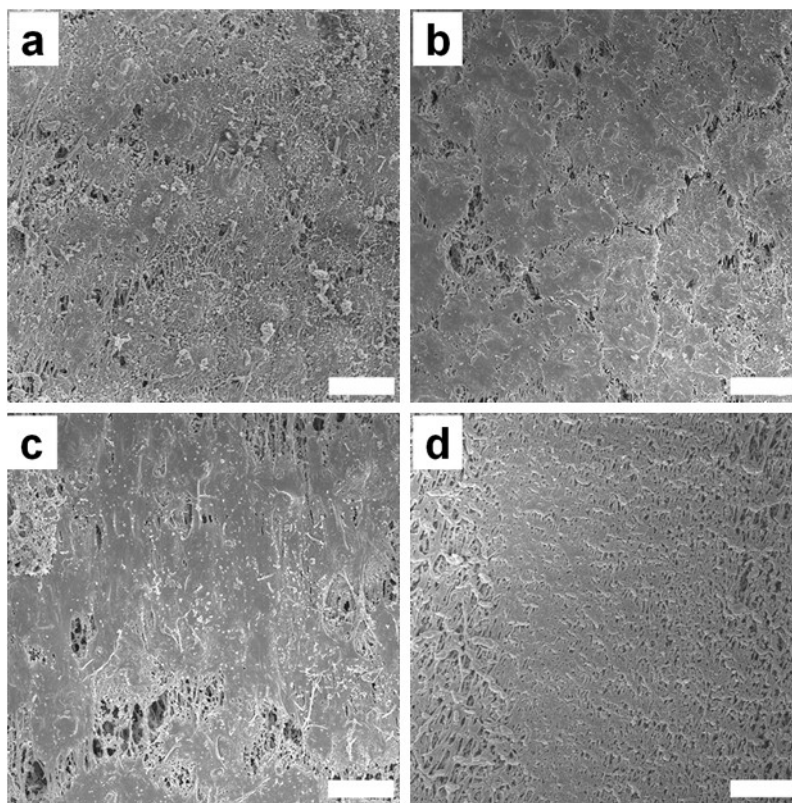


Fig. IV. 7 FE-SEM images of the outer surfaces of the surface-reorganized membranes. The membranes were prepared by dip-coating with a 0.5 wt% PVDF solution (in DMAc/ethanol = 65/35 vol%) and by subsequent VIPS. Operation temperature for dip-coating and VIPS was (a) 25 °C, (b) 40 °C, (c) 60 °C, and (d) 80 °C. VIPS time was 10 min. Scale bars represent 10 μm .

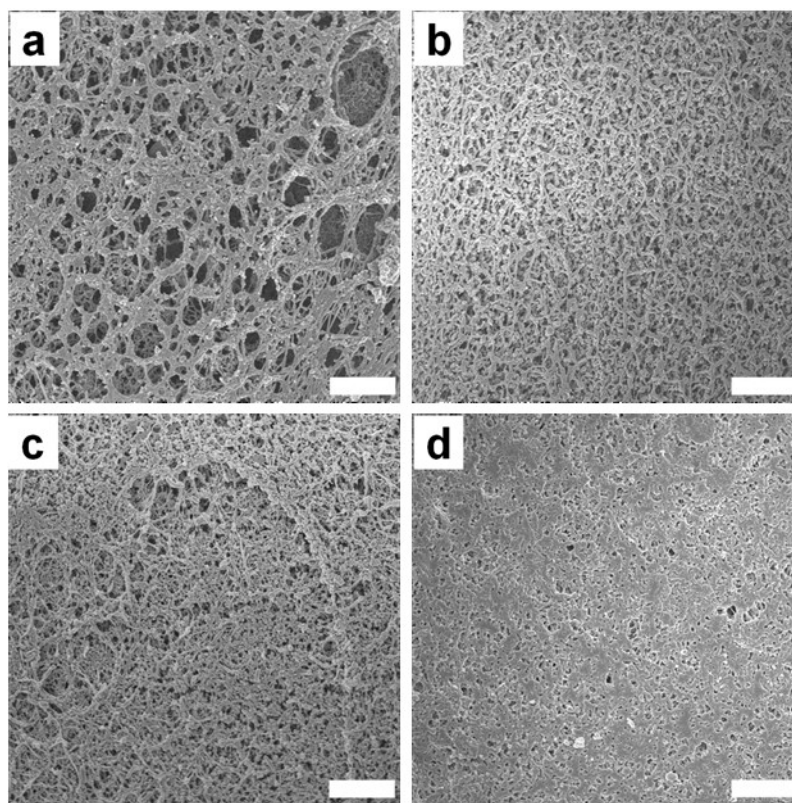


Fig. IV. 8 FE-SEM images of the outer surfaces of the surface-reorganized membranes. The membranes were prepared by dip-coating with a 1.5 wt% PVDF solution (in DMAc/ethanol = 65/35 vol%) and by subsequent VIPS. Operation temperature for dip-coating and VIPS was 80°C. VIPS time was (a) 1 min, (b) 10 min, (c) 30 min, and (d) 60 min, respectively. Scale bars represent 10 μm .

As stated above, the mass transfer rate of a non-solvent in a VIPS process is very slow. It is possible that the duration for VIPS (10 min) was not sufficient to complete the whole surface reorganization via VIPS. The VIPS-treating time was varied to study the effect on the membrane structures. Dip-coating using a 1.5 wt% PVDF solution at 80 °C was performed and then the VIPS treatment was performed for 1 min, 10 min, 30 min and 60 min at 80 °C. In Fig. IV. 8, FE-SEM images show that the membrane surface became denser as the VIPS time was longer (See section IV. 3.2).

Figs. IV. 6-8 suggest that the membrane surface was reorganized in the following manners. When a PVDF solution was dip-coated onto a membrane surface, the surface

of the substrate membrane began to be dissolved partially. In the VIPS process, water vapor induced the phase separation of a PVDF solution and partially solidified the PVDF coated on the membrane. The subsequent immersion of the membrane into water completed the phase separation and the solidification of PVDF on the original membrane, resulting in the formation of the new porous layer on the original membrane surface.

IV.3.2 Effects of the dip-coating (using a PVDF solution) and VIPS on the membrane properties

In general, high water permeability, controllable separation properties and sufficient mechanical strength are required for a hollow-fiber membrane for the practical use [8]. I examined the water permeability, pore-size and tensile strength of the surface-reorganized PVDF membranes.

The initial water permeability of the untreated PVDF membrane and the surface-reorganized membranes was shown in Fig. IV. 9. The untreated PVDF membrane had a high permeability of approx. 1500 L/(m²·h·atm) (black bar). The permeability of the surface-reorganized membranes prepared using 0.5 wt%, 1.5 wt% and 2 wt% PVDF solutions (at 80°C, VIPS treatment for 10 min) was approx. 1200, 1400 and 1400 L/(m²·h·atm), respectively. Dip-coating using a solution with a lower PVDF concentration resulted in lower permeability, which agreed with the surface density in FE-SEM images (Fig. IV. 6). This would be because a dip-coating solution containing PVDF at a low concentration was likely to dissolve the original surface and to form denser structures. The use of a 3 wt% PVDF solution under the same conditions resulted in the formation of a new thick layer that peeled from the original

membrane surface, probably due to the phase separation-induced formation of a solvent-rich phase between the newly-formed PVDF layer and the surface of the original membrane.

Temperature for VIPS had a small effect on the permeability while temperature affected the surface geometry of the membrane (Fig. IV. 7). When the temperature was raised from 25 °C to 60 °C, the permeability decreased slightly and it reached down to 1200 L/(m²·h·atm) (a 20% drop) at 80°C. These differences might have been due to the changes in the PVDF solubility and the mass transfer rate at different temperatures. Increasing the VIPS time decreased the permeability sharply, which agreed with the pore-sizes data shown in Fig. IV. 8. The smaller the pores, the lower the permeability.

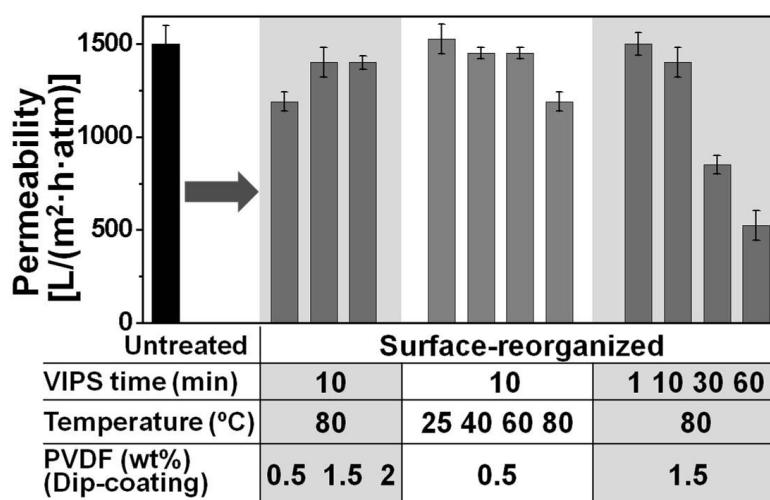


Fig. IV. 9 Initial water permeability of the untreated (black bar) and the surface-reorganized PVDF membranes.

The pore-sizes of the membranes were estimated by measuring the rejection of polystyrene nanospheres (Fig. IV. 10). The untreated PVDF membrane had the pore-size a little smaller than 300 nm. Variation in the PVDF concentration used in the dip-coating treatment and the duration of the VIPS treatment had only small effects on the membrane pore-size, although there were remarkable differences in morphology of

the membrane surfaces and also in the water permeability. The temperature (25-80 °C) used in the VIPS treatment affected the pore-sizes of the membranes. For instance, VIPS performed at 25 °C resulted in 66% rejection of 200 nm nanospheres and in 96% rejection of 300 nm nanospheres, meaning that the pore-size was between 200 nm and 300 nm. As the temperature increased, the rejection of 200 nm nanospheres increased and it reached 95% at 80°C, meaning that the pore-size of the membrane produced at 80 °C was smaller than 200 nm. As the VIPS time was longer, the rejection of 200 nm nanospheres was slightly higher, indicating that the increase of the VIPS time slightly decreased the pore-size.

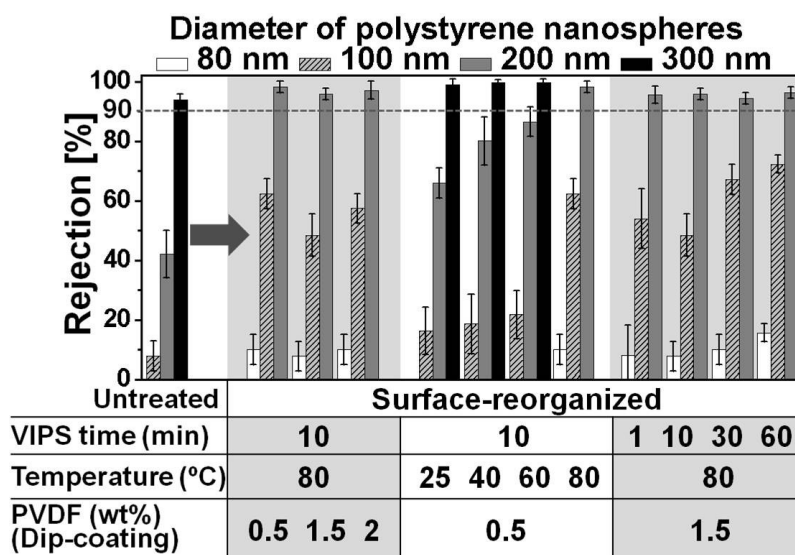


Fig. IV. 10 Rejection of standard polystyrene nanospheres for the untreated and the surface-reorganized PVDF membranes.

Another important property of a hollow-fiber membrane is its mechanical strength. The tensile strength of the PVDF membranes was measured (Fig. IV. 11). The tensile strength of the untreated PVDF membrane was approx. 3 MPa (black bar), while the dip-coating and the VIPS treatment slightly decreased the tensile strength of the membranes (grey bars), except for the membrane of VIPS for 60 min at 80 °C. The

membrane that was VIPS-treated for 60 min (and dip-coated using a 1.5 wt% PVDF solution at 80 °C) had a tensile strength of less than 2 MPa. The long VIPS time and high temperature might accelerate the dissolution of the original membrane surface, resulting in a low tensile strength and low permeability (Fig. IV. 9). The tensile strengths of the different parts (upper part, middle part and bottom part of the membranes set in the VIPS chamber) of the membranes (dip-coated and VIPS treated under varying conditions) were examined, and the similar strengths were obtained.

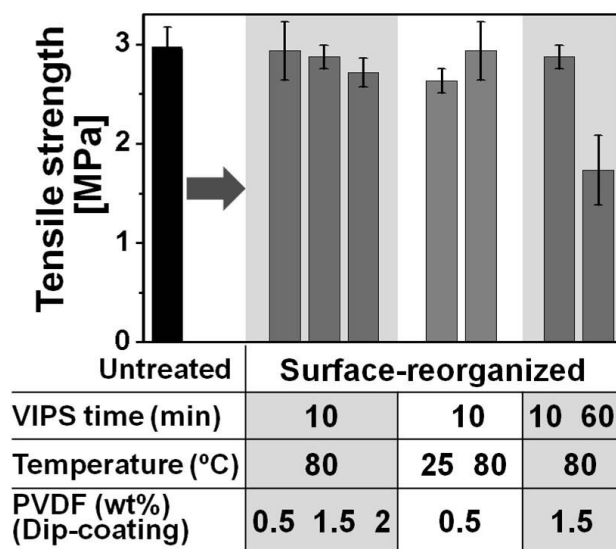


Fig. IV. 11 Tensile strength of the untreated (black bar) and the surface-reorganized PVDF membranes.

IV.3.3 Identification of a new surface-layer formed by dip-coating and VIPS treatment

FE-SEM observations show that variations in the dip-coating and the VIPS treatments brought large difference in the morphology of the membrane surface. However, the FE-SEM observation did not provide any evidence that the new PVDF layer observed on the membrane surface was derived from the dip-coating solution, and not from the original membrane. In an attempt to confirm this, I synthesized the

fluorescently-labeled copolymer poly(MMA-*r*-AEMA-FITC). Polymethacrylate is highly compatible with PVDF. A DMAc/ethanol (65/35 vol%) solution containing PVDF/poly(MMA-*r*-AEMA-FITC) (1.5/0.5 wt%) was used to dip-coat a membrane surface at 80 °C, and the VIPS treatment was performed for 10 min at 80 °C. The cross-section of the dip-coated membrane was observed using a fluorescent microscope. In Fig. IV. 12, there was a bright thin fluorescent layer (less than 10 μm in thickness) formed on the membrane surface and negligible fluorescence inside the membrane. These results indicate that the fluorescently-labeled copolymer and the PVDF, which was derived from the dip-coating solution, successfully formed a thin layer on the membrane surface in the present method, and also that the polymers in the dip-coating solution did not infiltrate into the membrane, suggesting little effect on the internal structure of the membrane.

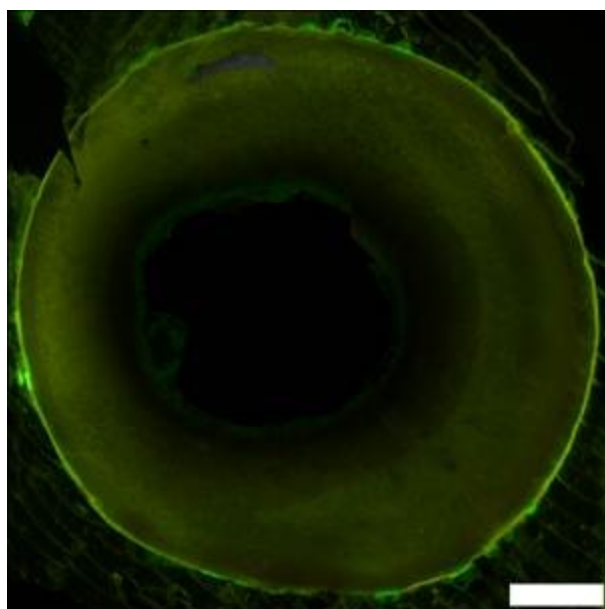


Fig. IV. 12 Fluorescent images of the cross section of the surface-reorganized membrane. The surface-reorganized membrane was prepared by dip-coating with a DMAc/ethanol mixture (65/35 vol%) containing 1.5 wt% PVDF and 0.5 wt% poly(MMA-*r*-AEMA-FITC) at 80 °C, and by subsequent VIPS for 60 min. Sale bar represents 200 μm.

IV.3.4 Effects of the dip-coating (using a PVDF solution) and VIPS on membrane fouling

From the above investigations, the dip-coating of the hollow-fiber membrane with a 1.5 wt% PVDF solution at 80 °C and the VIPS treatment performed for 10 min decreased the pore-size from approx. 300 nm to 200 nm, while keeping high permeability and high tensile strength. In addition, the resultant mesh-like structure provided the pores over the surface homogeneously, compared with those of the original surface. Such changes might have some effects on fouling of a hollow fiber membrane during filtration. I employed humic acid, BSA and casein, as model foulants, to test the fouling properties of the membranes.

The flux (J) and rejection (R) of different foulants through the untreated and surface-reorganized membranes during filtration were shown in Fig. IV. 13. Humic acid, a type of natural organic matter, was reported to be one of main foulants in filtration and easily form cake layer on the membrane surface [9]. The flux (J) of the surface-reorganized membrane after 2 h filtration was approx. 310 L/(m²·h), which was 2 times higher than that of the untreated PVDF membrane (Fig. IV. 13-1a), while the rejection (around 20%) was quite similar to each other (Fig. IV. 13-1b). It should be noted that both the membranes were composed of only PVDF and had similar initial water flux.

There was also a difference in the water flux when filtering a BSA solution through these membranes (Fig. IV. 13-2a). Although a remarkable flux decline in the beginning of filtration was observed for both of the membranes, the surface-reorganized membrane showed the stable flux from 10 min to 2 h. The flux of the untreated membrane kept gradual decreasing after the initial flux decline. After 80 min filtration,

the flux of the surface-reorganized membrane was higher than that of the untreated membrane. The BSA rejection was lower than 10% for both the membranes (Fig. IV. 13-2b), indicating that most of BSA molecules passed through the membranes.

In the case of a casein solution, a remarkable flux decline was also observed in the beginning of the filtration (Fig. IV. 13-3). After 30 min of filtration, the flux of the treated membrane was higher than that of the untreated membrane. At 120 min, the flux of the surface-reorganized membrane was 2 times higher than that of the untreated membrane (Fig. IV. 13-3a). Interestingly, the rejection of casein was very different between the two types of the membranes. The rejection of the surface-reorganized membrane was almost 100%, while that of the untreated membrane was approx. 40% after 2 h of filtration (Fig. IV. 13-3b). Although the molecular size of casein was much smaller than the pore-sizes of both membranes, casein molecules easily form micelles with a size of over 100 nm in a solution [10]. The formation of casein micelles would account for the high rejection observed for the surface-reorganized membrane. The casein micelles were larger than the pores of the surface-reorganized membrane than but smaller than those of the untreated membrane.

To demonstrate that the newly-formed layer facilitated as a separation layer in the surface-reorganized membranes, FITC-labeled casein was prepared and used in filtration experiments (See Chapter III). Feed solutions (FITC-casein 10 ppm in 50 mM triethanolamine-HCl/NaOH buffer, pH 7.5) were filtered through the untreated and surface-reorganized membranes for 2 h under the conditions that were the same as those used in the experiments for the non-labeled casein solutions. The membranes fouled with FITC-casein were sampled and sliced, followed by fluorescent microscopic observation. Fluorescent images of the whole cross-sections of

membranes fouled with FITC-casein are shown in Fig. IV. 14. The fluorescence was observed over the whole cross-section of the untreated membrane, which indicates casein adsorption inside the untreated membrane (Fig. IV. 14a). In contrast, only a narrow fluorescently-dyed area near the outer surface was observed in the surface-reorganized membrane (Fig. IV. 14b). The penetration results of FITC-labeled casein were in agreement with the rejection data described above, and proved that the newly-formed surface acted as a separation layer in the surface-reorganized membrane. The improved anti-fouling properties of the surface-reorganized membrane would be attributed to the newly-formed surface. The present results suggest the possibility that the surface geometry of a hollow-fiber membranes plays an important role in the membrane fouling.

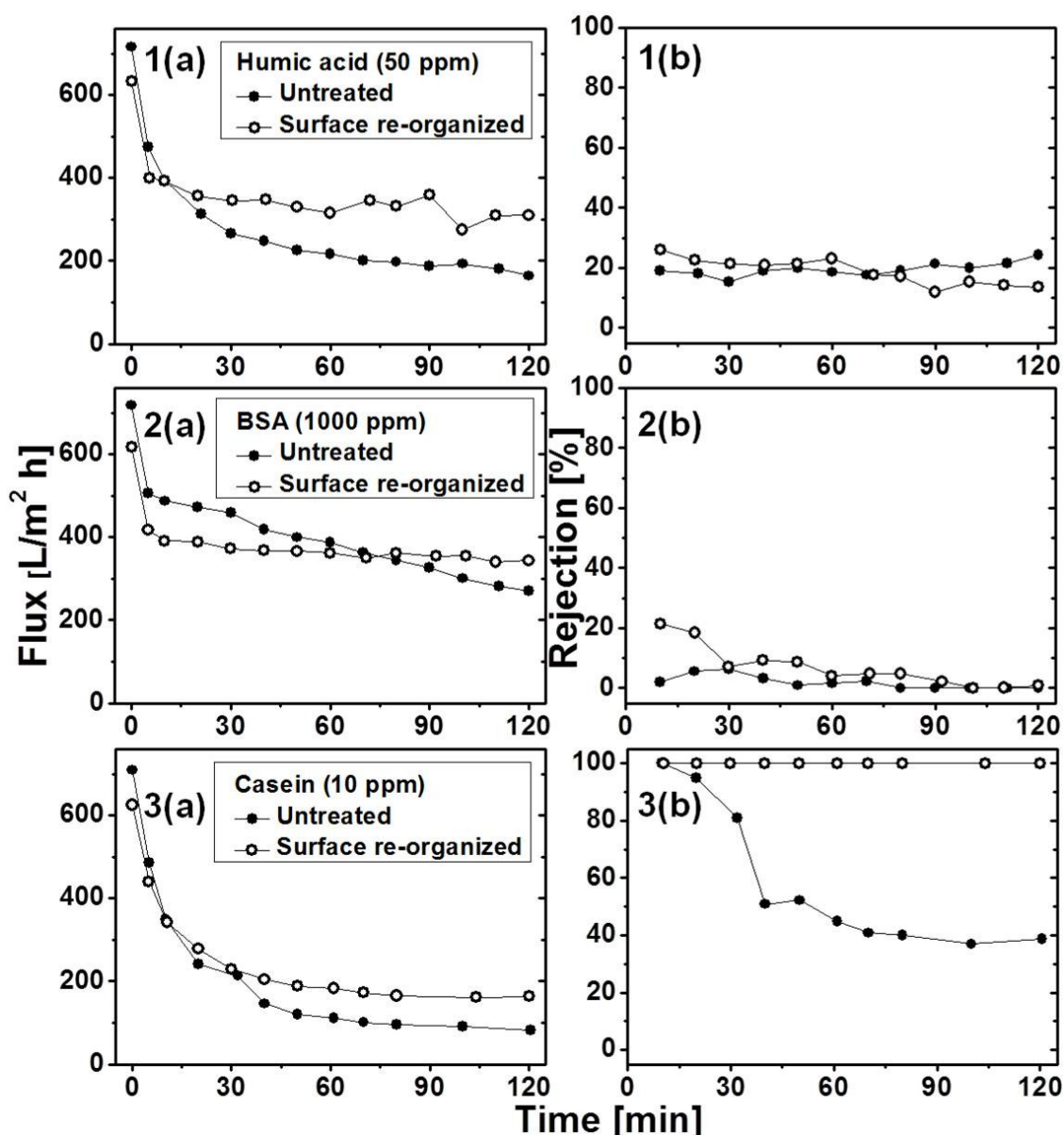


Fig. IV. 13 Flux (a) and rejection (b) of the filtration of foulant-containing solutions through the untreated and the surface-reorganized PVDF membranes. Filtration tests were performed at flow rate 0.5 ml/min under 0.5 atm for 2 h. The surface-reorganized membranes were prepared by dip-coating with a 1.5 wt% PVDF solution at 80 °C and by subsequent VIPS for 10 min. Foulants were (1) humic acid (50 ppm), (2) BSA (1000 ppm) and (3) casein (10 ppm).

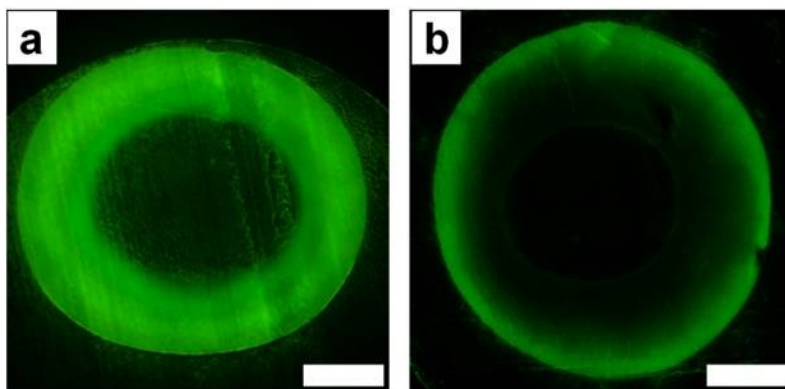


Fig. IV. 14 Fluorescent microscopic images of cross sections of PVDF membranes fouled with FITC-casein. (a) Untreated membrane, and (b) the surface-reorganized membrane prepared by dip-coating with a 1.5 wt% PVDF solution at 80 °C and by subsequent VIPS for 10 min. FITC-casein solutions (10 ppm) were filtrated through membranes at a flow rate of 0.5 ml/min under 0.5 atm transmembrane pressure for 2 h and then DI water was filtrated for 1 min. Sale bars represent 200 μm .

IV.3.5 Effects of surface-reorganization (using a hydrophilic polymer) on membrane properties

The above investigations demonstrated that the present method is able to immobilize other polymers on the outer surface of a hollow-fiber membrane. The use of functional polymers as the dip-coating polymer can provide a membrane surface a secondary function. I then employed a hydrophilic polymer as the dip-coating polymer to modify the outer surface of a hollow-fiber membrane. PVDF membranes for water purification still suffer for severe fouling during filtration, because of the high hydrophobicity resulting in strong hydrophobic interactions between membranes and many foulants. Hydrophilic modification of PVDF hollow-fiber membranes has been studied for decades, mainly in order to improve the anti-fouling properties of the membranes [11-13]. However, the most challenging issue remains how to uniformly modify a membrane with hydrophilic materials and to control the pore-size at the same time.

A series of random hydrophilic copolymers composed of the hydrophilic moieties (PEGMA) and the moieties adhesive to membrane surfaces (MMA) were synthesized and used in dip-coating. I aimed to prepare a PVDF hollow-fiber membrane having a hydrophilic outer-surface and high separation performances. Another important point was not to alter the inner matrix (the hydrophobic part) of the substrate membrane to avoid an increase in the flow resistance during filtration.

Table IV. 2 Dip-coating and VIPS conditions for the surface modification of PVDF hollow-fiber membranes. The VIPS treatment was carried out for 10 min.

Entry	poly(MMA- <i>r</i> -PEGMA) M _n of MMA:PEGMA PEGMA (mole ratio)		Polymer solution for dip-coating	Nonsolvent (in VIPS)	Temperature (°C)
a	300	9:1	PVDF/Copolymer 1/1wt%	ethanol	70
b			Copolymer 2 wt%	ethanol	70
c		7.5:2.5	Copolymer 2 wt%	ethanol	70
d		7:3	PVDF/Copolymer 0.5/0.5wt%*	water	80
e	500	9:1	Copolymer 2 wt%	ethanol	70

* A mixture of *N*-methylpyrrolidone (NMP)/toluene/ethanol (60/10/30 vol%) was used as a solvent.

The dip-coating and VIPS treatments using different copolymers were performed under the conditions shown in Table IV. 2. FE-SEM images of the membrane surfaces after the surface modification show that mesh-like surfaces were uniformly formed on the outer surfaces (Fig. IV. 15-1), covering the original spherulites (Fig. IV. 5a). The cross-sectional images show that the newly-formed layers adhered to the original membrane well without peeling off (Fig. IV. 15-2).

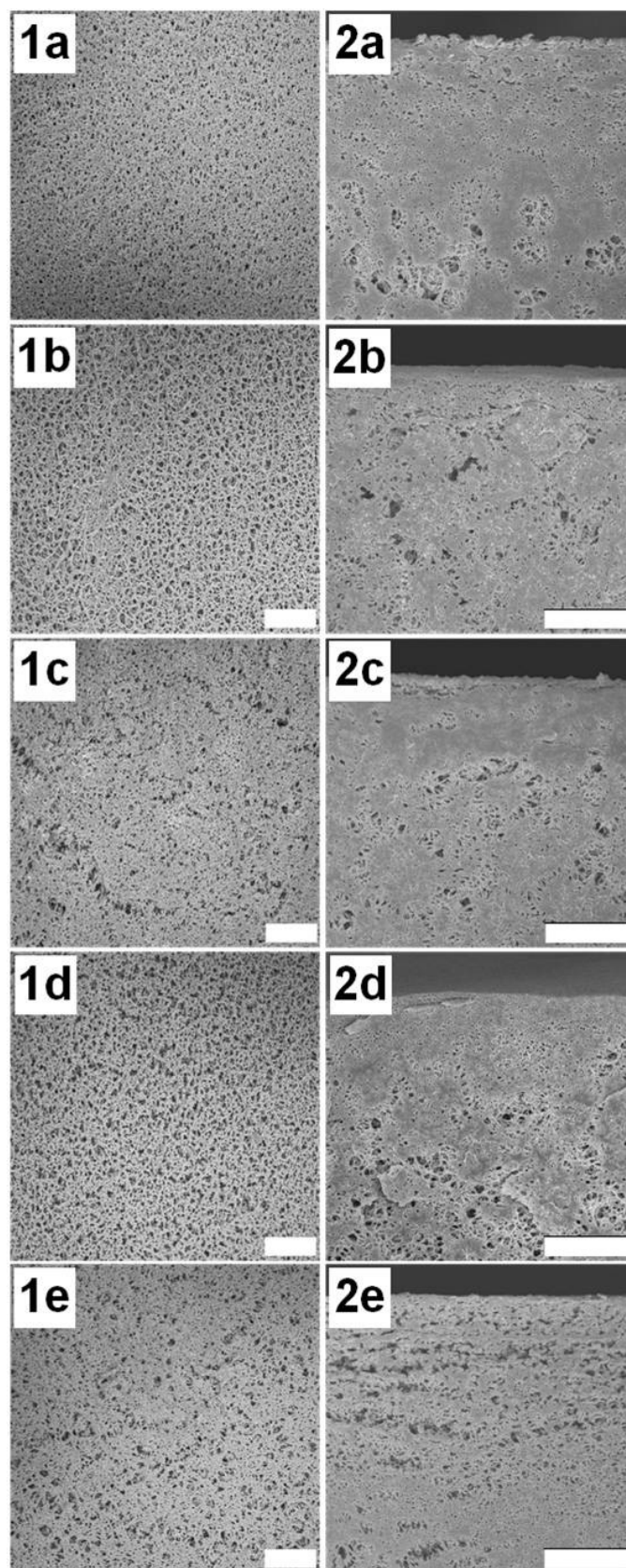


Fig. IV. 15 FE-SEM images of the outer surfaces (1) and the cross-sections (2) of and the

surface-modified PVDF hollow-fiber membranes. The membranes were modified by the dip-coating and by the subsequent VIPS treatment. (a)-(e) indicate those listed in Table IV. 2. Scale bars represent 10 μm .

The initial water permeability of the untreated PVDF membrane and the surface-modified membranes were shown in Fig. IV. 16. Compared with the untreated PVDF membrane ($1500 \text{ L}/(\text{m}^2 \cdot \text{h} \cdot \text{atm})$, black bar), the surface-modified membranes of (a), (b), (d) and (e) had the initial permeability of approx. $1100 \text{ L}/(\text{m}^2 \cdot \text{h} \cdot \text{atm})$, while the surface-modified membrane of (c) had the approximately $1000 \text{ L}/(\text{m}^2 \cdot \text{h} \cdot \text{atm})$. The decline in the initial permeability by the surface modification was only 27-33% in all the cases. The modification by the surface modification did not cause a severe decrease in the permeability.

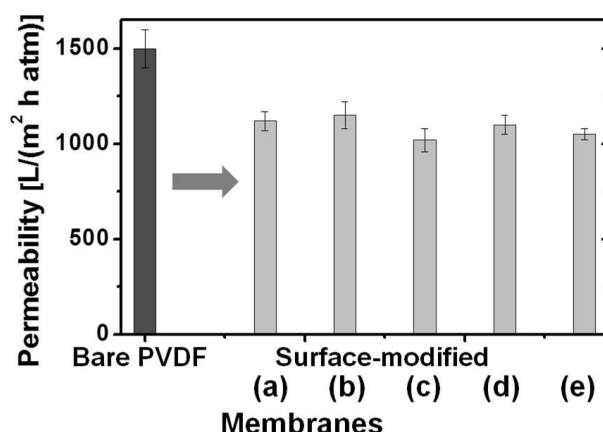


Fig. IV. 16 Initial water permeability of the untreated (black bar) and the surface-modified PVDF membranes. The membranes were modified by the dip-coating and by the subsequent VIPS treatment. (a)-(e) indicate those listed in Table IV. 2.

The rejection of the standard polystyrene nanospheres by the membranes is shown in Fig. IV. 17. The rejection of 300 nm nanospheres by the untreated PVDF membrane indicates the pore-size of a little smaller than 300 nm. The surface-modified membranes exhibited the rejection of 200 nm nanospheres which was higher than that by the untreated membrane. These results suggest that the pore-size of the

surface-modified membrane (a) was a little smaller than 200 nm, while the pore-sizes of membranes (b)-(e) were in between 200 nm and 300 nm. The rejection of 200 nm nanospheres by membranes (b)-(e) exhibited 60-80%, whereas that by the untreated membrane was 40%. These results indicate that the pore-size distribution of the membranes became narrower after the surface modification. The variations in the pore-size among membranes (a)-(e) would be due to the difference in the phase separation behavior of dip-coated copolymers.

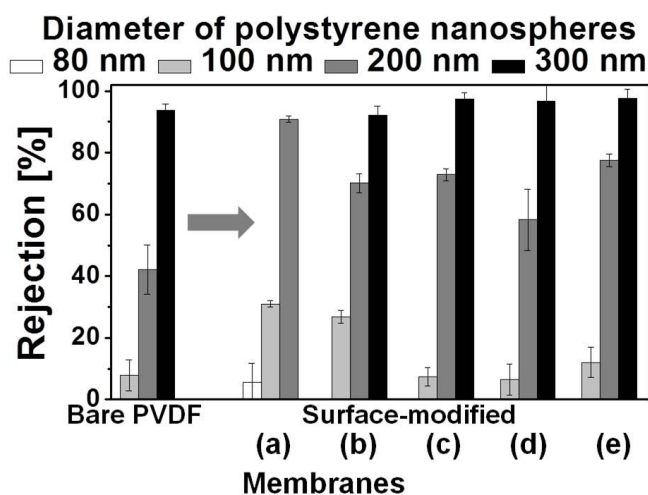


Fig. IV. 17 Rejection of standard polystyrene nanospheres for the untreated and the surface-modified PVDF membranes. The membranes were modified by dip-coating and by subsequent VIPS (Table IV. 2).

The air-bubble contact angles of the untreated and the surface-reorganized membranes are shown in Fig. IV. 18. The air-bubble contact angle θ for the untreated PVDF membrane surface was approx. 125° (black bar). The surfaces of membranes (a)-(e) exhibited air-bubble contact angle values higher than that for the untreated membrane, meaning that the surfaces became more hydrophilic by the surface modification. The air bubble contact angles of membranes (b) and (e) were approx. 137° and 140° , respectively. The copolymers used in the dip-coating for these two types membranes had the same hydrophilic moiety mole content (10 mol%) but had different

lengths of the hydrophilic chains (PEG chains). The results indicate that the use of the copolymer with longer hydrophilic chains in the dip-coating increased the hydrophilicity of the membrane surface. The membrane (a) exhibited the air bubble contact angle of approx. 133°, which was lower than that for the membrane (b). The content of the amphiphilic copolymer in the dip-coating solution affected the hydrophilicity of the membrane surface.

Poly(MMA-*r*-PEGMA) (M_n of PEGMA 300) (MMA: PEGMA 7:3 mole ratio) had the highest content of the hydrophilic moiety (30 mol%) among the copolymers tested. The use of this copolymer alone in the dip-coating failed to obtain a uniform membrane surface (data not shown), probably due to the low solidification rate of the copolymer. To solve this problem, a mixture of the copolymer and PVDF was dissolved in a mixture of NMP/toluene/ethanol (60/10/30 vol%) and used in the dip-coating. Water was used as vapor in the VIPS treatment to increase the phase separation rate.

Although there was a difference in the mole ratios of the hydrophilic moieties of the copolymers between membranes (b) and (c), no remarkable difference in the air bubble-contact angle was observed.

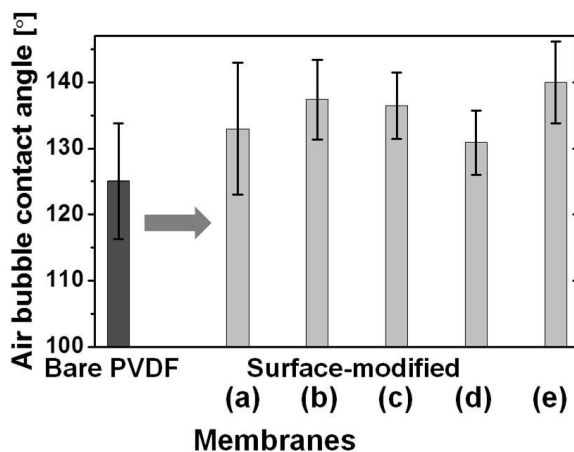


Fig. IV. 18 Air bubble contact angle of untreated PVDF membrane surface (black bar)

and the surfaces of modified PVDF membranes (a-e). The membranes were modified by dip-coating and by subsequent VIPS (Table IV. 2).

IV. 3. 6 Effects of the hydrophilic surface-modification on membrane fouling

I adopted casein (20 ppm, pH 7) as a model foulant to test anti-fouling properties of the surface-modified membranes. Aqueous solutions containing casein (20 ppm, pH 7) was filtrated through the untreated and surface-modified membranes. The flux (J) and rejection (R) through the membranes are shown in Fig. IV. 19. The decline of the flux during filtration was restrained by the hydrophilic surface-modification, and the final flux (at 120 min) of the surface-modified membranes was twice or three times higher than that of the untreated membrane (Fig. IV. 19a). The rejection of the membrane was also affected by the surface modification (Fig. IV. 19b). The final rejection of the untreated membrane was approximately 60%, while that of the surface-modified membranes was over 85%. Although the flux of the surface-modified membranes was very similar to each other, the variation in the rejection was observed.

The flux declines indicate that the surface modification using the hydrophilic copolymer improved the anti-fouling property of the PVDF hollow-fiber membrane.

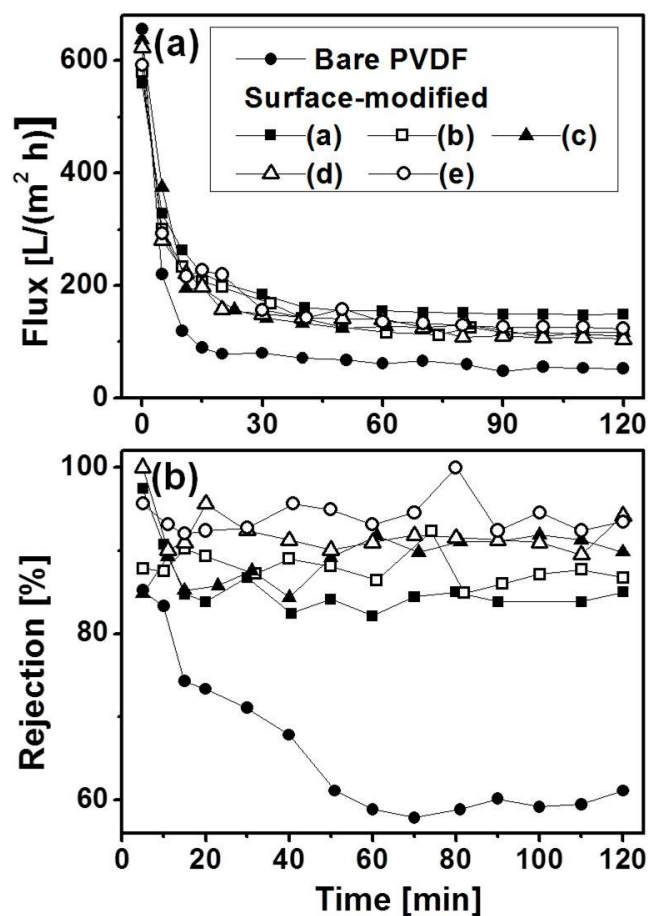


Fig. IV. 19 Flux (a) and rejection (b) of the filtration of casein solutions (20 ppm, pH 7) through the untreated and the surface-modified PVDF membranes. Filtration tests were performed at a flow rate of 0.5 ml/min under 0.5 atm for 2 h. The membrane surfaces were modified by the dip-coating and by the subsequent VIPS treatment (Table IV. 2).

IV. 3. 7 Analysis of the surfaces of the untreated and the surface-modified membranes.

FT-IR measurements were carried out to analyze the surfaces of membranes (Fig. IV. 20). The absorbance at 1729 cm^{-1} and 1276 cm^{-1} suggests the C=O bonds in the copolymer on the surface-modified membrane (red line). The absorbance at 1148 cm^{-1} suggests the C-O bonds of the PEGMA moiety in the copolymer. These bands observed indicate that the copolymer was successfully immobilized on the membrane surface by the present method.

The membrane surfaces were also analyzed by XPS. Fig. IV. 21 illustrates the XPS wide scan spectrum of the PVDF membrane surfaces. Compared with the surface of the membrane prepared using PVDF alone via TIPS (Fig. IV. 21a), O1s peak had higher intensity and F1s peak had lower intensity for the surface of the PVDF membrane modified by the dip-coating and by the subsequent VIPS treatment (Table IV. 2, the entry a) (Fig. IV. 21b). The high O1s peak and low F1s peak would be due to the dip-coated copolymer. The XPS results also indicate that the copolymer was successfully immobilized on the membrane surface by the present method.

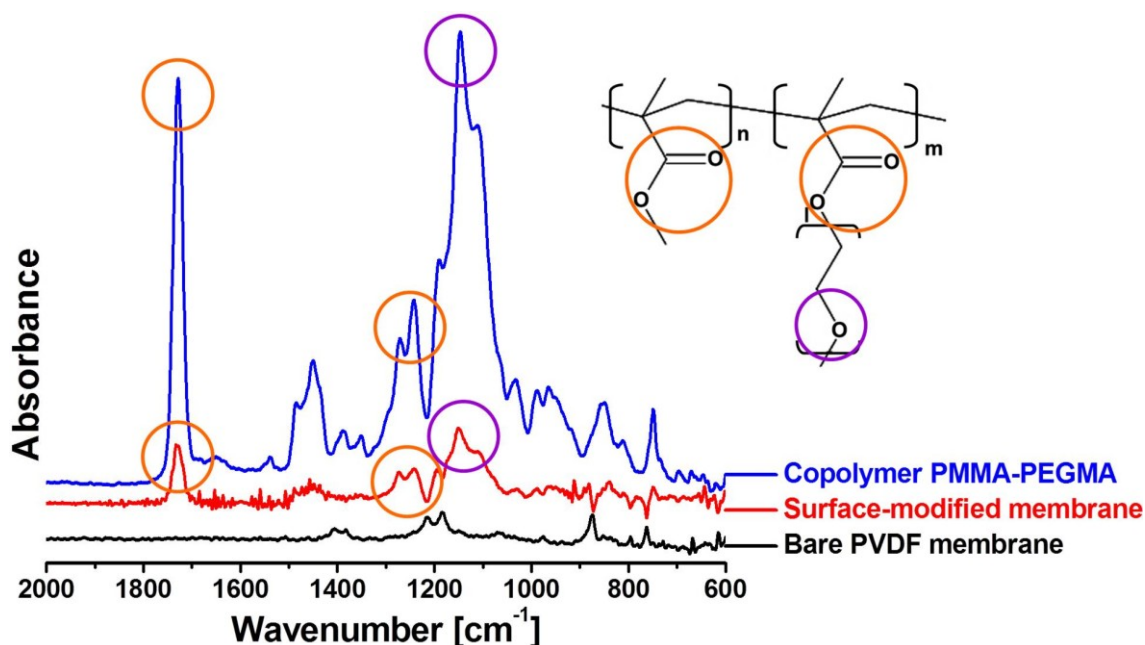


Fig. IV. 20 FT-IR spectra of copolymer poly(MMA-*r*-PEGMA) (M_n of PEGMA 300) (MMA: PEGMA 9:1 mole ratio) (blue line), and the outer surfaces of the PVDF membranes. The membranes are: the membrane prepared using PVDF alone via TIPS (black line); the membrane modified by the dip-coating and by the subsequent VIPS treatment (Table IV. 2, conditions a) (red line)

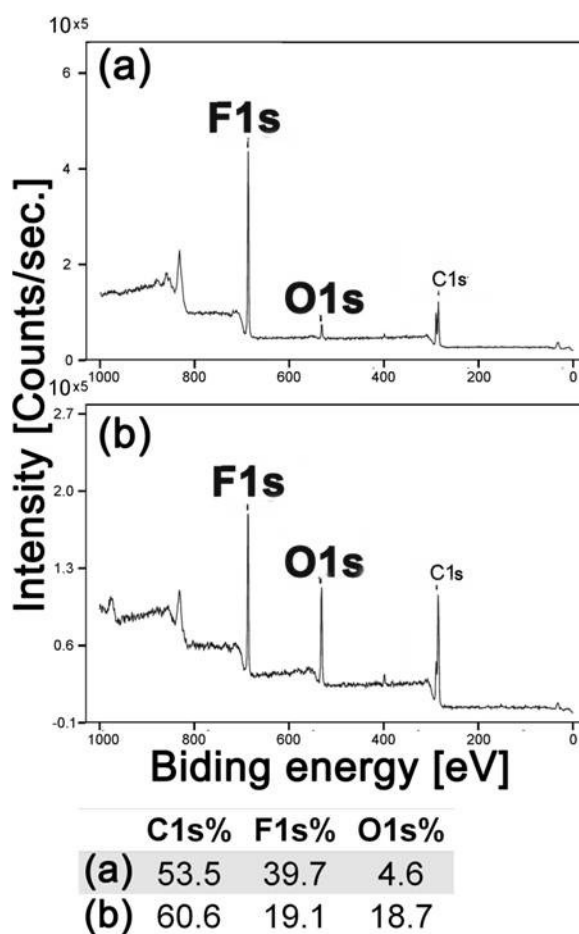


Fig. IV. 21 XPS wide scan spectrum of the outer surfaces of the PVDF membranes. The membranes are: (a) The membrane prepared using PVDF alone via TIPS. (b) The membrane modified by dip-coating and by subsequent VIPS (Table IV. 2, condition a). The table represents the XPS atom% for C1s, F1s and O1s.

II.4 CONCLUSIONS

In this study, I employed simple strategies of dip-coating and VIPS to reorganize the outer surfaces of PVDF hollow-fiber membranes prepared by the TIPS method. Highly-porous mesh-like structures were successfully prepared on the surface of the original PVDF membranes using another PVDF solution in the dip-coating. The present method can alter the surface morphologies and the separation performances of a PVDF hollow-fiber membrane. The reorganized surface acted as a new separation

layer and the homogeneous matrix inside the membrane contributed to the mechanical strength. The combination of TIPS, dip-coating and VIPS produced the hollow-fiber membrane with highly-porous surface that had high water permeability, controlled pore-size and high mechanical strength. The surface-reorganized membrane also exhibited the improved anti-fouling properties compared with the original PVDF membrane.

The present method can be extended to the chemical modification of the outer surface of a PVDF hollow-fiber membrane with other kinds of functional polymers. A hydrophilic copolymer, poly(MMA-*r*-PEGMA), was synthesized and successfully immobilized on the membrane surface to produce a porous hydrophilic surface. The membrane modified with the hydrophilic copolymer exhibited better anti-fouling property compared with a original PVDF hollow-fiber membrane.

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Chapter V

Conclusion

Fouling phenomena in microfiltration (MF) and ultrafiltration (UF) hollow-fiber membrane separation systems was studied from several different angles in this dissertation. I examined fouling mechanism and cleaning techniques after fouling in the first part of this work (Chapter II and III). I developed membrane preparation techniques for preparing a membrane with better anti-fouling properties in the second part of this study (Chapter IV).

In Chapter II, the significant effects of ionic environment in fouling of a polyethersulfone (PES) UF hollow-fiber membrane caused by humic acid and proteins were studied. Although a number of studies have been reported organic fouling of UF membrane systems, they mainly focused on single model foulants and failed to report the multiple influences for organic fouling due to multiple substances in natural raw water. My investigations revealed that the presence of various metal ions and metal colloids, even at low concentrations, could affect fouling caused by humic acid and proteins greatly during filtration. The presence of metal ions and metal colloids accelerated humic acid fouling of the hollow fiber ultrafiltration membrane. The humic acid aggregate size increased with addition of the metal ions, and the observed size was larger than the pore size of the membrane. The amount of humic acid deposition on the membrane surface increased to a remarkable extent in the presence of Ca^{2+} , indicating that the foulant deposit could not be swept away by the lateral flow of the feed solution. Addition of the chelating agent EDTA inhibited, to a large extent, metal ion-induced aggregation of humic acid, resulting in inhibition of fouling. For protein

fouling, the presence of metal ions and metal colloids at low concentrations increased casein and BSA fouling of hollow-fiber ultrafiltration membranes, but did not affect lysozyme fouling. The increase in fouling depended on the type of metal ions and proteins. The protein aggregation induced by the metal ions and the protein–metal colloid interactions was related to the increased fouling. The lateral fluid flow associated with the hollow-fiber membranes in a cross-flow filtration system, who has the potential to sweep foulant deposits off the membrane surface failed to prevent foulant deposition. The results of the present study proposes the importance of monitoring and controlling metal ion concentrations in the source water, and the necessity of preventing metal ion contamination from plant apparatus to reduce fouling. Backwashing fouled membranes using a chelating agent solution might be an effective strategy for restoring membrane permeability.

In Chapter III, a methodology for visualizing the internal fouling of a hollow-fiber membrane using fluorescent microscopy was developed. Fouling caused by proteins and sodium alginate (SA) was studied. Membrane fouling was usually characterized by the flux decline with time, rejection variations of the foulant, deposition state on the membrane surfaces during filtration. Although several recent studies provided information of foulant depositions inside of a membrane pores at a limit depth, there were no reports providing the internal fouling of the whole hollow fiber membrane with complex internal structures during filtration. In this study, I successfully visualized internal fouling of three different fluorescently-labeled proteins and one type of fluorescently-labeled polysaccharide sodium alginate. Internal fouling behaviors of three model proteins were remarkably different, although there were similarities in the permeability decline amongst these proteins. The present

methodology can reveal which part of a membrane determines the rejection of solutes and also which part of a fouled membrane can be washed by backwashing using different types of cleaning agents. I also prepared membranes with differing structures (with and without a skin-layer on the outer surface) and demonstrated that internal fouling behavior varied with a skin-layer, even though there were no noticeable differences in water permeability or permeability decline during filtration between two types of membranes. Fouling of SA through three types of PES membranes with different MWCOs was also visualized. SA fouling behaviors differed from the membrane pore sizes and structures. The SA deposition inside a membrane was much different from the protein deposition. The SA accumulation inside a membrane affected protein fouling. This study on internal fouling provided novel information on membrane fouling and aid understanding of membrane fouling.

In chapter IV, a two-step process was developed to prepared a asymmetric poly(vinlidend fluoride) (PVDF) hollow-fiber membrane, mainly for achieving better anti-fouling properties for the membrane. In the first step, a PVDF hollow-fiber membrane with high permeability and high mechanical strength was prepared via thermally-induced phase separation (TIPS); in the second step, dip-coating and vapor-induced phase separation (VIPS) was performed to reorganize the outer surface of the membrane, and a new non-peeled separation layer with uniform mesh-like structures and precisely controlled pore-size was prepared. In the surface reorganization process, the membrane surface morphologies and separation performances was able to be controlled. By using blend PVDF and a synthesized copolymer poly(MMA-r-AEMA-FITC) in dip-coating, a uniform new layer from dip-coating was successfully confirmed. Under optimized conditions of surface

organization, a surface-reorganized membrane with controlled pore-size was prepared without losing the water permeability and the mechanical strength. The better anti-fouling properties of the membrane were successfully achieved. The methodology was also able to modify the membrane surface with other kinds of functional polymers. A hydrophilic copolymer, poly(MMA-*r*-PEGMA), was synthesized and successfully used to provide the membrane surface porous hydrophilic thin separation layer.

List of publications:

Yan Hao, Akihito Moriya, Tatsuo Maruyama, Yoshikake Ohmukai, Hideto Matsuyama, Effect of metal ions on humic acid fouling of hollow fiber ultrafiltration membrane, *J. Membr. Sci.* **2011**, 376, 247.

Yan Hao, Akihito Moriya, Yoshikage Ohmukai, Hideto Matsuyama, Tatsuo Maruyama, Effect of metal ions on the protein fouling of hollow-fiber ultrafiltration membranes, *Sep. Purif. Technol.* **2013**, 111, 137.

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