

Electronic Supplementary Information

Towards antibiofouling ultrafiltration membranes by blending silver containing surface modifying macromolecules

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Result section

Table 1 Characterization data of SMMs

SMM	[‡] Repeat unit (m)	Weight average molecular weight (M_w , Dalton)	Silver content (wt%)
DEG-SL	4.59 ± 0.15	2.28×10^3	9.47
DEG-SC	3.52 ± 0.10	2.23×10^3	9.68
[†] PEG-SL	3.87 ± 0.21	2.65×10^3	24.39
[†] PEG-SC	1.78 ± 0.17	2.08×10^3	31.17

[‡]Calculated by ¹H NMR of various peak area ratio; [†]PEG of typical molecular weight is 200, then the n value is 4.14

Table 2 The Ag concentration (wt%) at the surface and at the three cross-sectional positions of the studied membranes

Ag detection position		3-A	3-B	3-C	3-D	5-A
Surface		0.77	0.57	0.56	1.05	1.75
Cross-section	Top	0.36	0.50	0.25	0.26	0.58
	Middle	0.37	0.36	0.58	0.31	0.76
	Bottom	0.21	0.38	0.30	0.37	1.04

Table 3 XPS results (wt%) of some of the studied membranes surfaces

Membrane		Fluorine	Silver	Oxygen	Nitrogen
0-A	Top	58.27	0.00	1.32	0.17
	Bottom	58.17	0.00	1.19	0.27
3-A	Top	46.17	0.84	4.82	1.32
	Bottom	46.10	0.56	4.54	1.95
3-D	Top	50.03	0.91	3.97	1.42
5-A	Top	45.84	3.50	4.44	1.10

Table 4 Separation (%) data of the membranes

Separation factor	0-A	3-A	3-B	3-D	5-A
<i>E. coli</i>	100.00	100.00	99.97	99.98	99.98
Total carbon	9.78	8.43	7.79	22.61	12.28
Conductivity	0.58	0.94	0.77	6.56	2.46

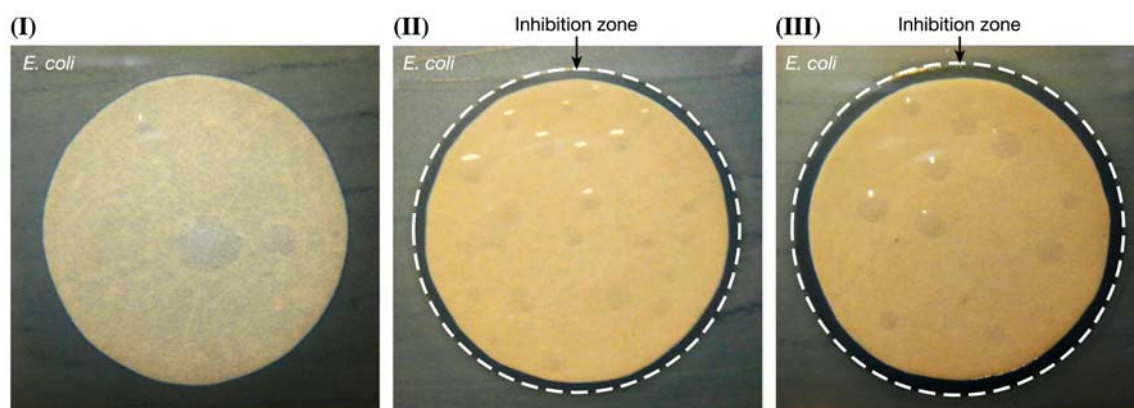


Fig. 1 Effect of SMM concentration in the casting solution on the IZ thickness: (I) 0 wt% (0-A), (II) 3 wt% (3-A), (III) 5 wt% (5-A). PVDF concentration in the casting solution, 18 wt%; “as-cast” membrane treated under condition - A (RT and 21% RH without air exposure time).

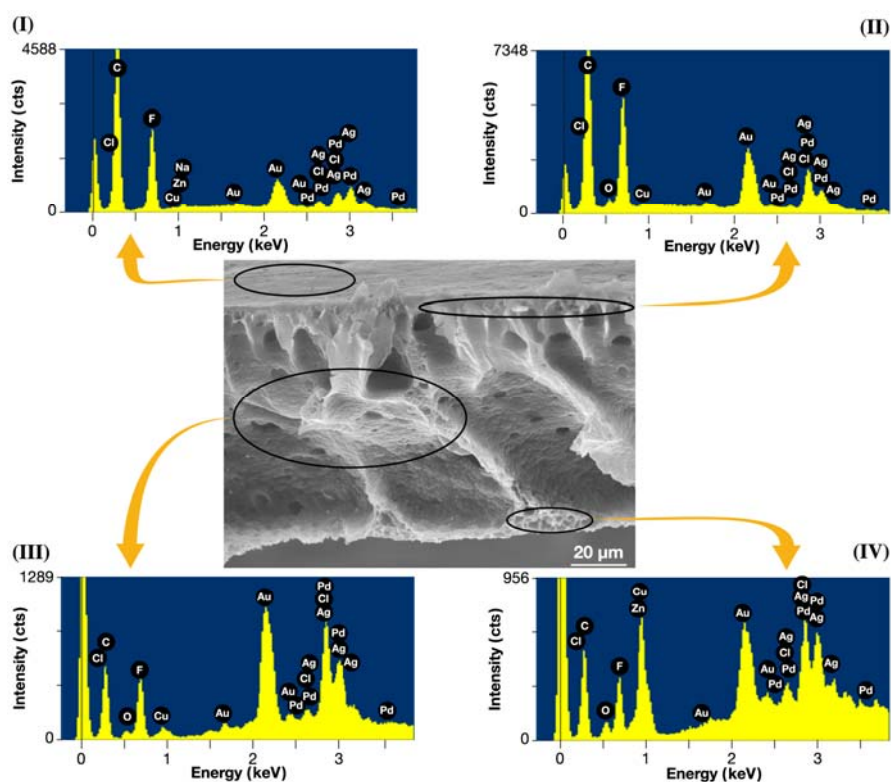


Fig. 2 EDS measurements on the cross-section of the membrane “5-A”: (I) top surfaces, (II) cross-section at near the top, (III) cross-section at the middle, and (IV) cross-section at near the bottom. The Ag concentration at different locations of the membrane cross-section demonstrates insignificant changes from the bottom to the top and they are lower than the surface Ag concentrations, indicating the migration of SMM to the surface during the cast film treatment period.

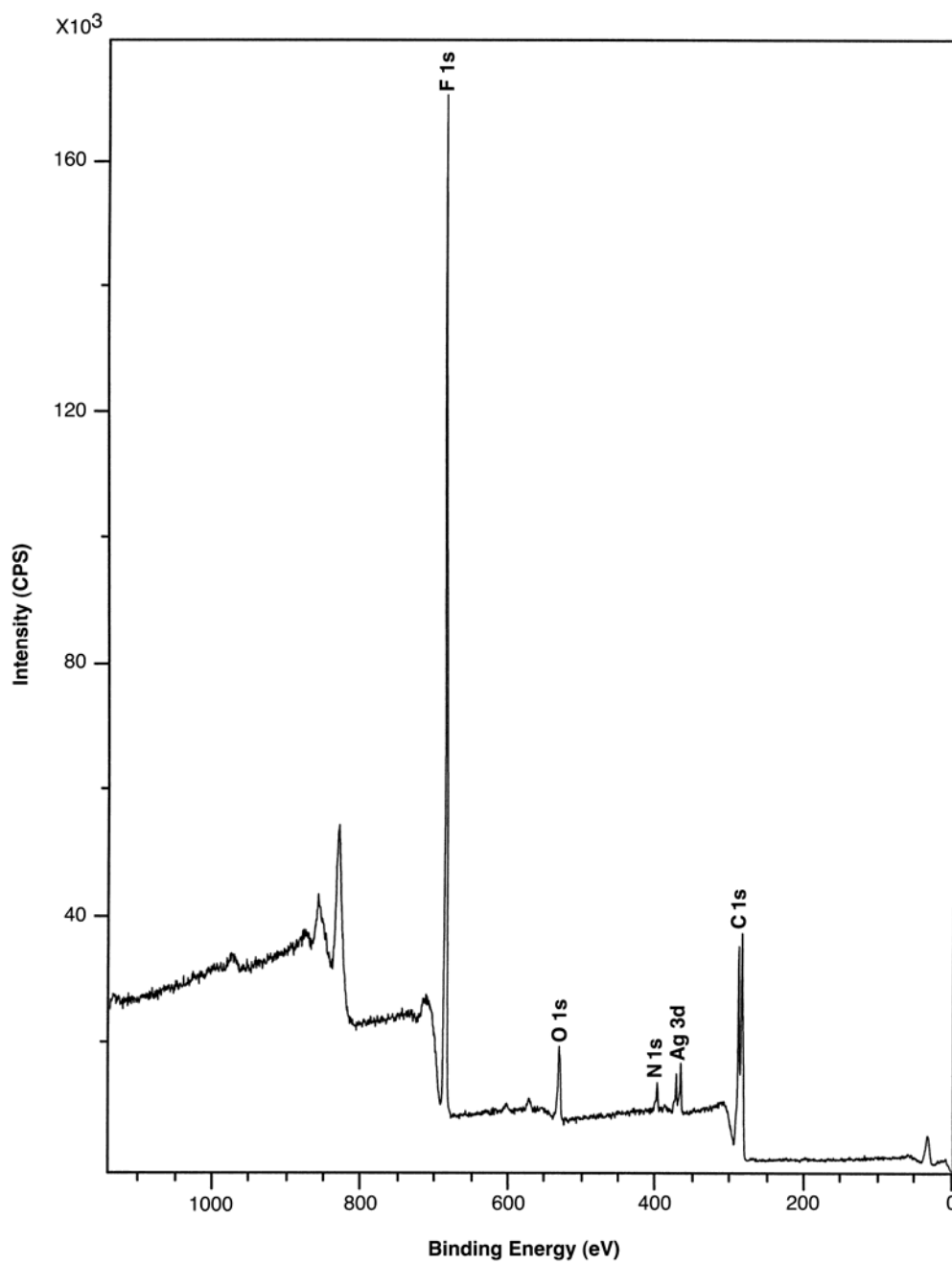


Fig. 3 (I) Typical XPS survey spectrum of membrane of 5-A at top surface.

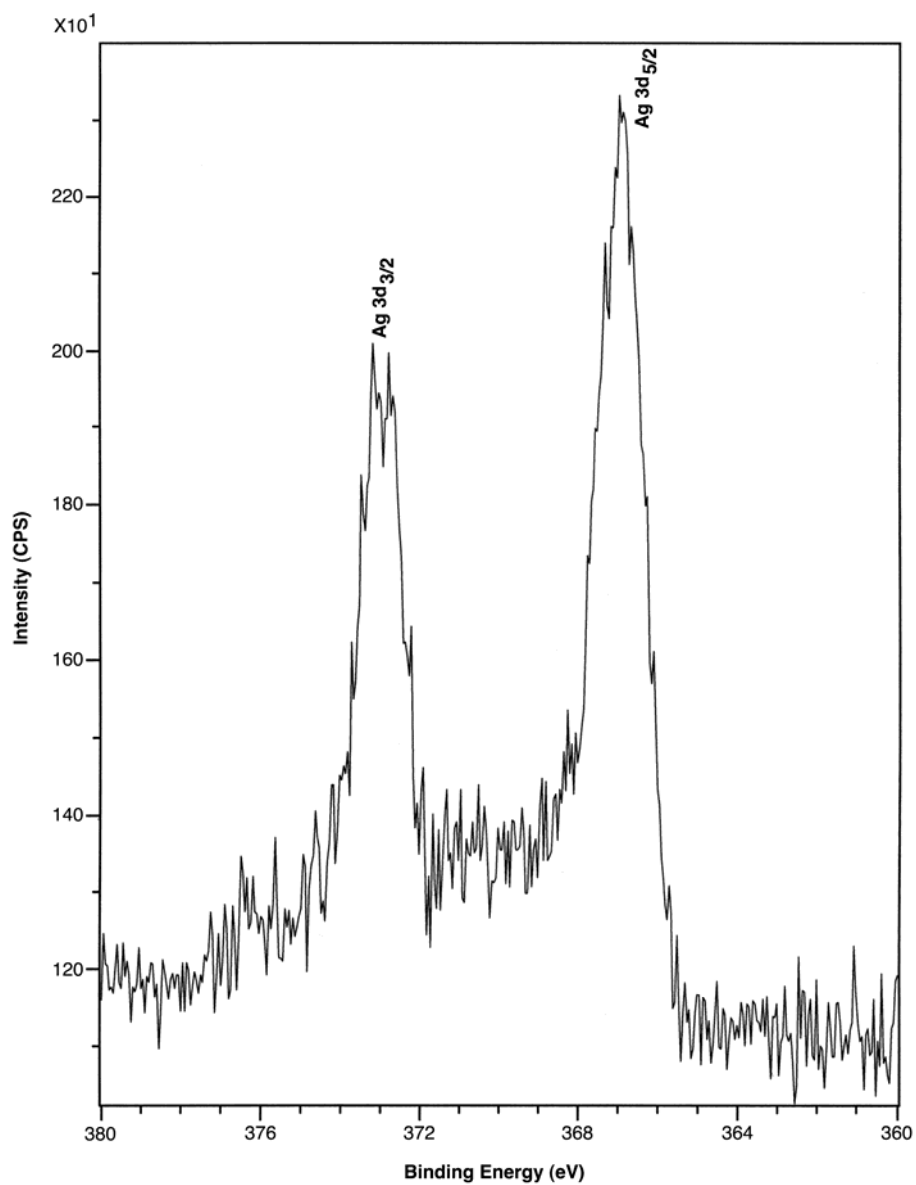


Fig. 3 (II) XPS spectrum at Ag 3d_{3/2} and Ag 3d_{5/2} signals of membrane of 5-A at top surface.

Experimental section

Materials

Poly(vinylidene fluoride) (PVDF) with a weight average molecular weight of 254 kD was supplied by Elf Autochem Canada Inc. (Oakville, ON, Canada). Diethylene glycol (DEG, 99%) 4,4'-methylene bis(phenyl isocyanate) (4,4'-diphenylmethane diisocyanate, MDI, purity 98%), N, N-dimethylacetamide (DMAc, anhydrous 99.8%), N-methyl-2-pyrrolidinone (NMP, anhydrous 99.5%), poly(ethylene glycol) (PEG, typical average molecular weight 200 Daltons), silver lactate (SL, 97%), and silver citrate hydrate (SC) were received from Sigma-Aldrich Inc. (St. Louis, MO, USA) and were used without further purification. Sodium chloride (NaCl, ACS reagent grade of purity 99.99%) was received from Fisher Scientific Company (Fair Lawn, NJ, USA). For the microbial part of the experiments, *Escherichia coli* (American Type Culture Collection, ATCC # 33456) was obtained from ATCC, Manassas, VA, USA. Luria-Bertani (LB) broth and LB agar, both purchased from Sigma-Aldrich, Inc. (St. Louis, MO, USA), were used for the preparation of liquid and solid media. Polyester non-woven fabric was supplied by Osmonics Inc., Minnetonka, MN, USA.

SMMs synthesis

Surface modifying macromolecules (SMMs) were synthesized by two-step condensation polymerization. First step was the reaction between 4,4'-diphenylmethane diisocyanate (MDI) and diol, and then the urethane pre-polymer was end-capped using silver material containing alcohol group to finalize the reaction and to deposit Ag to the SMMs for antibiofouling membrane development. The molar ratio of the MDI, diol and Ag-material was 3, 2 and 2, respectively. The solvent, *N,N*-dimethyl acetamide (DMAc), was degassed to remove the trace of water for more than 3 days. Details of the synthesis conditions are shown in **Fig. 4**. After the reaction, the SMMs were precipitated and washed with distilled water (DW). The SMMs were then dried at 50 °C over 3 days before use. All the process of Ag containing SMMs synthesis was performed in dark to avoid the light effect on silver nanoparticles formation.

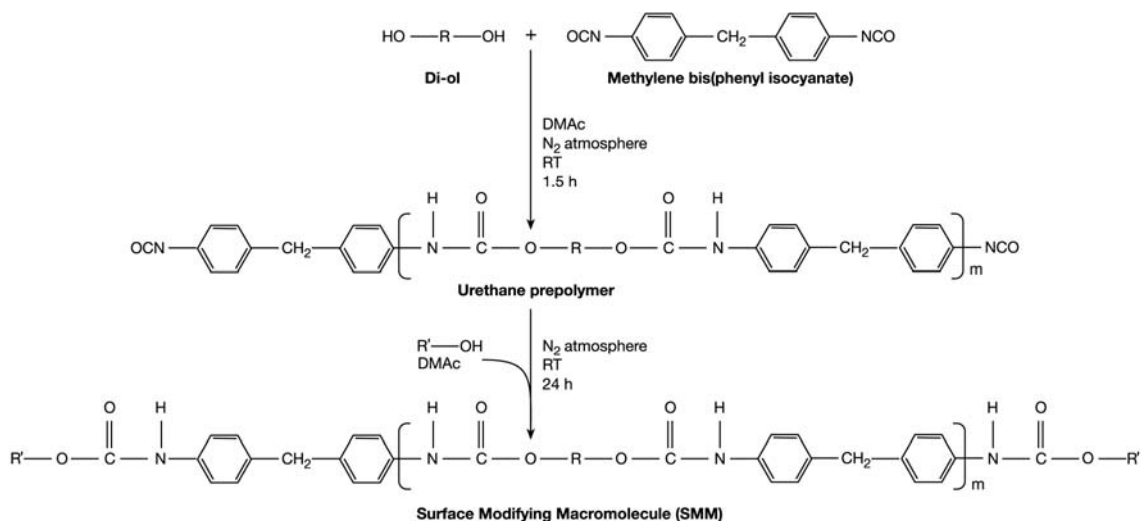


Fig. 4 The process of SMM synthesis.

SMMs characterization

The chemical name of the SMMs is poly(4,4'-diphenylenemethylene ethyleneurethane) with both ends capped by silver lactate or silver citrate. The chemical structure of the SMMs used in this study is shown in **Fig. 4**. The solubility of the SMMs was studied on nine different solvents at room temperature. The SMMs were soluble in DMAc, NMP, dimethylformamide, dimethylsulfoxide, and tetrahydrofuran, whereas SMMs were insoluble in acetone, cyclohexane, n-hexane, and toluene.

The structure of SMMs was identified by multinuclear NMR spectroscopy. A concentrated sample for NMR analysis was prepared by dissolving as much polymer as possible in DMF-d₇. All NMR data were collected on a Bruker AVANCE 500 NMR spectrometer. The ¹H NMR spectrum was acquired under quantitative conditions with 32 scans using a 30° pulse, 10 sec recycle delay and a 3.6 sec acquisition time. The proton decoupled ¹³C NMR spectrum was collected using 12268 scans, a 30° pulse, a 1 sec recycle delay and a 1 sec acquisition time. The ¹³C DEPT 135 spectrum was collected using 3361 scans, a 2 sec recycle delay and a 1 sec acquisition time. A ¹H - ¹H gradient COSY-45

spectrum was collected with 8 scans to determine connectivity within the molecule. A ^1H - ^{13}C HMQC spectrum was collected with 16 scans for each of 256 increments with a 170 ppm ^{13}C spectral window. The recycle delay was 1 sec. The acquisition time was 0.183 sec. A ^1H - ^{13}C HMBC spectrum was collected with 64 scans for each of 256 increments with a 230 ppm ^{13}C spectral window. The recycle delay was 1.5 sec. The acquisition time was 0.183 sec. The data were treated with a shifted sine squared weighting function prior to Fourier transform.

Particularly, chemical structure of DEG-SL is discussed. The ^1H , COSY, HMQC spectra are shown in **Fig. 5**. The peak assignment of ^1H spectrum is presented in **Table 5**. (1) The COSY spectrum indicates that there is spin-spin coupling between the CH protons at 5.08 ppm with CH_3 protons at 1.45 ppm. (2) Due to an absence of a correlation in the HMQC spectrum the NH protons can be assigned at 8.70 and 9.64 ppm. One NH is due to the end-capped urethane group and other is due to the repeat unit urethane group.

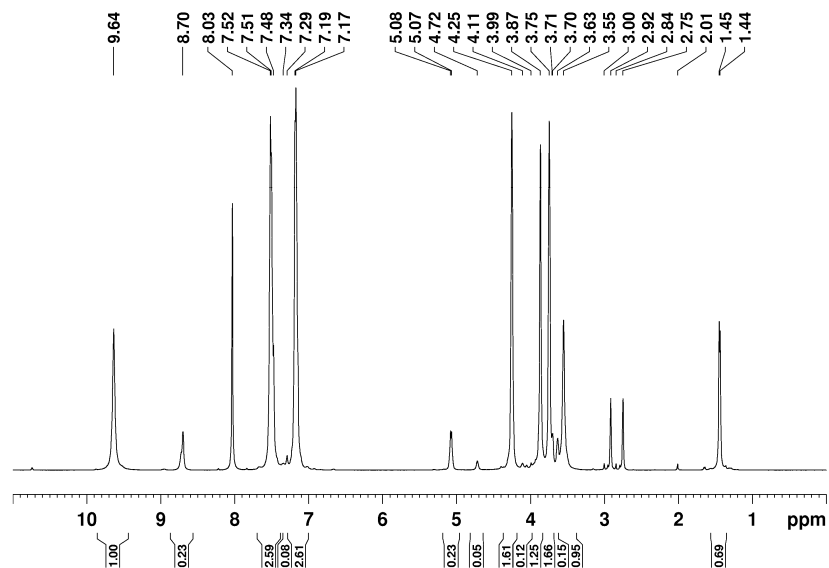


Fig. 5 (I) ^1H spectrum.

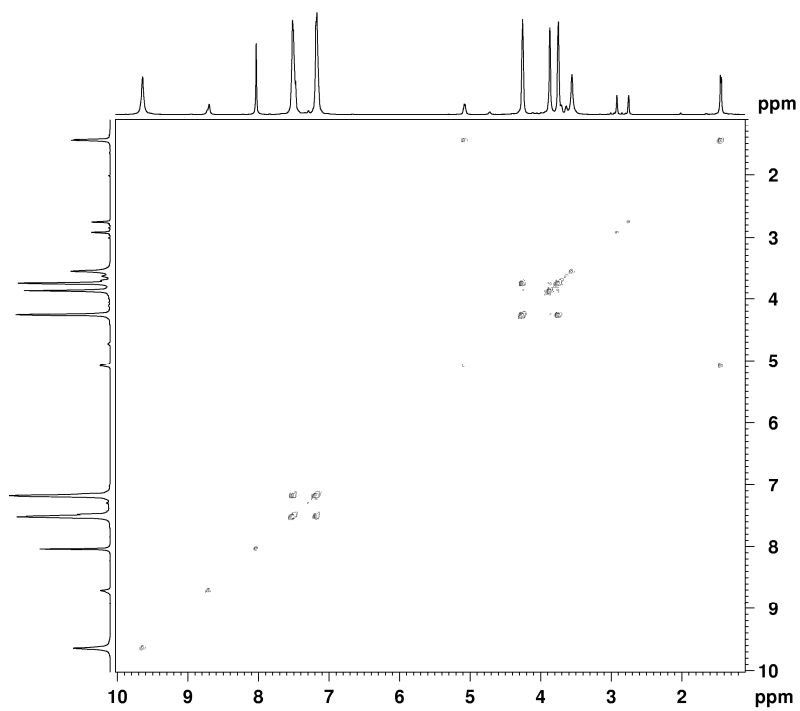


Fig. 5 (II) COSY spectrum.

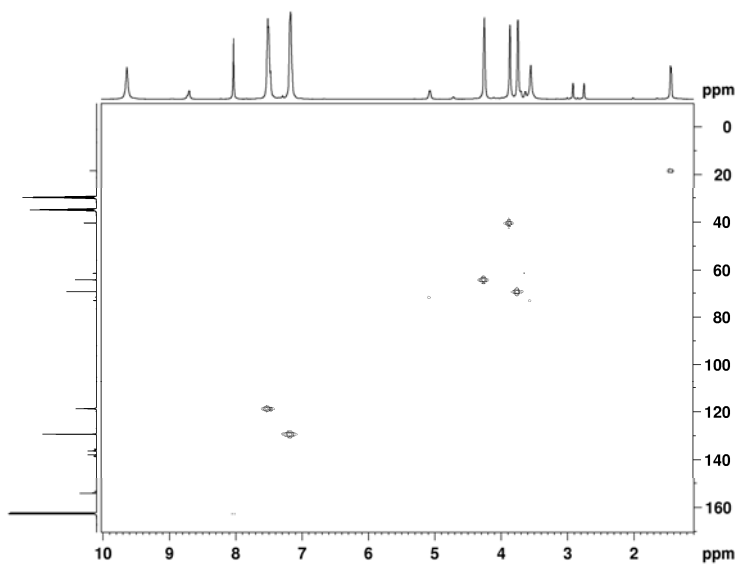


Fig. 5 (III) HMQC spectrum.

Table 5 The assignments of ^1H NMR characteristic peaks of polyurethane having silver lactate as both ends groups

Assignment	δ_{H} (ppm)
CH_3 hydrogen (SL)	1.44
CH_2 (DEG) adjacent to the ether bond	3.55, 3.71
CH_2 between the phenyl groups of MDI	3.87
CH_2 (DEG) adjacent to the urethane bond	4.25
CH hydrogen (SL)	5.08
Aromatic hydrogen (MDI)	7.17 - 7.52
Urethane NH	8.70, 9.64

2.75, 2.92, and 8.03 ppm peaks due to DMF- d_7 , $(\text{CD}_3)_2\text{N-CDO}$

The determination of the exact number of repeating unit is possible through precise integration of ^1H NMR. The peak at 1.44 represents six CH_3 (SL) protons of peak area 0.69 and the peak at 5.07 represents two CH (SL) protons of peak area 0.23. The average peak area per unit proton for both two cases is 0.115. There are many ways to calculate the repeat unit value, however, three ways are presented herewith: (a) The peaks at 7.17-7.52 ppm represent $8m+8$ aromatic protons. Therefore, $(8m+8) = (2.59+0.08+2.61)/0.115 = 45.91$, so the m is equal to 4.74. (b) The peak at 3.87 ppm represents the $2m+2$ CH_2 bridging protons between the aromatic rings. Therefore, $(2m+2) = 1.25/0.115$, so $m = 4.43$. (c) The peak at 3.55, 3.71 and 4.25 ppm represent the $8m$ CH_2 protons of DEG. Therefore, $8m = (0.95+1.66+1.61)/0.115$, so $m = 4.59$. Therefore, the average repeating unit (m) number is 4.59 ± 0.15 .

Membrane preparation

PVDF was used as the base polymer with a concentration of 18 wt% in N-methyl-2-pyrrolidinone. A certain amount of SMMs was added into the base polymer solution. All casting solutions were degassed before use. Membranes were prepared by the phase

inversion method.¹ The PVDF solution was cast on a polyester non-woven fabric (Osmonics Inc., Minnetonka, MN, USA). Then, the as-cast film was treated under various conditions before it was immersed into the coagulation bath containing 4 °C distilled water.

Scanning electron microscopy (SEM) and energy dispersive spectrometer (EDS) measurements of membranes

The morphology and the silver content of the membranes were determined by SEM and EDS, respectively. The cross-sectional area as well as the top surface of the dry membranes was observed by the SEM. A model Tescan Vega-II XMU VPSEM, Tescan USA Inc., Cranberry Twp., PA, USA, was used to take SEM photographs. For the elemental analysis, Oxford Inca Energy 250X EDS was used. For both investigations, secondary electron detector was used. The membrane sample was mounted on a chair-shaped sample holder for both surface and cross-sectional observation. For cross-sectional observation, the membrane was prepared by fracturing cryogenically by immersing in liquid nitrogen. The samples were coated under vacuum with a thin layer of 60% gold and 40% palladium in a sputter system (Hummer VII, Anatech, Springfield, VA, USA).

X-ray Photoelectron Spectroscopy (XPS)

The elemental composition at the surface of membrane coupon was determined by X-ray photoelectron spectroscopy (XPS, Kratos Axis HS X-ray photoelectron spectrometer, Manchester, UK). Each randomly taken membrane was cut into samples of 1 cm² from the membrane sheet. Monochromatized Al K_α X-radiation was used for excitation and a 180° hemispherical analyzer with a three channel detector was employed. The X-ray gun was operated at 15 kV and 20 mA. The pressure in the analyzer chamber was 1.33×10⁻⁴ to 1.33×10⁻⁵ Pa. The size of the analyzed area was about 1 mm². All the membrane surfaces were analyzed for specific element content at take-off angle (θ) of 0° which corresponded to the X-ray penetration depths of 6.3 nm.

E. coli preparation

The *Escherichia coli* (*E. coli*) was grown in a liquid Luria-Bertani (LB) media at a temperature of 37 °C and at a shaking speed of 150 rpm using a shaking incubator (Model:

Lab-Line Orbit Environ-Shaker, Lab-Line Instruments Inc., Melrose park, IL, USA). The *E. coli* solution was freshly prepared for each experiment and used within 12 h.

Halo test

The halo test was used to appraise the sterilization effect of membrane on *E. coli* growth. This technique is actually designed for evaluation of pharmaceutical products named as ‘single disk methods.’² A paper disk soaked with an antibiotic is placed on the target bacteria inoculated solid media. The media plate with the disk is incubated under the desired growth condition of the bacteria. The antibiotic diffuses into the media, and inhibits the bacteria growth or kills the bacteria. The effect of the antibiotic appeared as a clear ring around the disk. Moreover, the intensity of the antibiotic can be evaluated by measuring the thickness of the ring. Due to the appearance of the clear ring around the disk, this technique is also called ‘halo test’. It is also used in other fields of research, especially in anti-biofouling membrane development.³

In the present investigation, solid media of nutrient (LB) agar (called agar plate, hereafter) was prepared. The membrane was cut into a 2 cm diameter disk. *E. coli* was inoculated on an agar plate, and the membrane disk was placed. The prepared agar plate was incubated at 37 °C for 5 h. All the experiments were done under bacteria-free environment. The thickness of the inhibition zone that appeared around the membrane disk was then measured.

Surface resistance (SR) test

There are several methods to measure the amount of biomass in different ways; i.e., chemically (adenosine triphosphate and deoxyribonucleic acid analysis, etc.), microscopically (direct count microscopy, epifluorescence, etc.) and physically (dry weight method, wet weight method, turbidimetry, etc.).⁴ The turbidimetric method is widely used for bacterial cell measurement,⁵ and the experimental procedure is as follows: A membrane was inserted at the bottom of the test cell and a certain amount of *E. coli* inoculated LB broth was loaded. The test cell was incubated at 37 °C and 150 rpm for 5 h. All the

experiments were done under bacteria-free environment. The absorbance, called optical density (OD) at a wavelength of 600 nm (absorbance cuvette length: 1 cm) of the LB broth was measured at 0 and 5 h incubation period using a UV-Visible spectrophotometer (Model: Helios Gamma, Thermo electron corporation, Hemel Hempstead, Hertfordshire, England, UK). The specific growth rate, μ , was calculated by

$$\mu = \frac{\ln(OD/OD_0)}{t}$$

The *E. coli* solution grown for 5 h together with various membranes shown in **Fig. 6**.

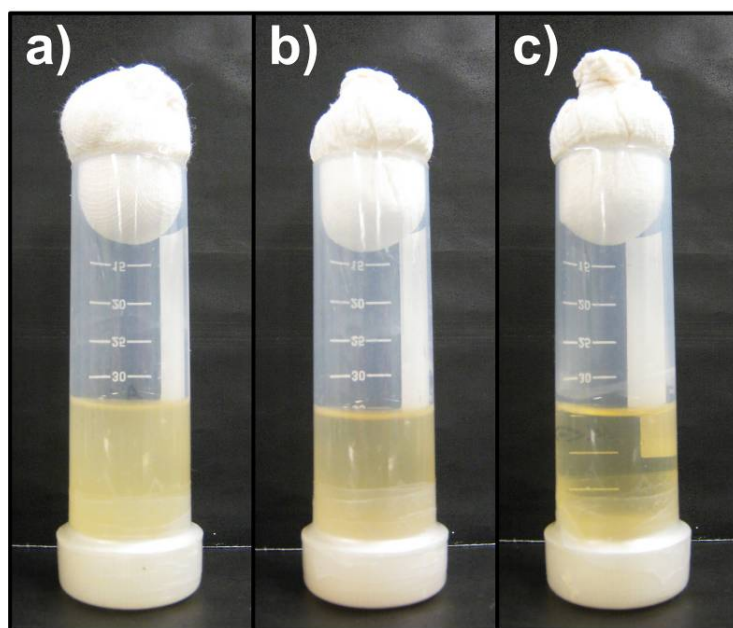


Fig. 6 The pictures of SR test after growing the *E. coli* solution for 5 h. The membranes-prepared with three different concentrations of SMM, (a) 0 wt% which is control membrane ‘0-A’, (b) 3 wt% ‘3-A’ and (c) 5 wt% ‘5-A’. The casting solution contained 18 wt% PVDF. The “as-cast” membrane treated under the condition -A (i.e. room temperature and 21% RH without air exposure).

Filtration test

The filtration experiments were conducted using a laboratory-scale filtration system consisting of a reservoir, a pump, a temperature controller, two pressure regulators, and five filtration cells connected in series (**Fig. 7**).⁶ The effective membrane area was 20.4 cm². The details of the design of the cell are given elsewhere.⁷ The flow in the cross-flow cells was assumed as turbulent. The experiment was operated at 25 °C. Membranes were inserted into the cleaned filtration cells and pre-compacted with DW at 80 psig for 1 h, followed by circulation of DW at 50 psig for 15 h. Pure water permeation rates were then determined. Synthetic waste water (SWW) was circulated in the set-up for 30 min and then *E. coli* was inoculated at the concentration of 10⁶ cells/L in SWW. The *E. coli* containing SWW was re-circulated at 50 psig for 25 h. Flux of each membrane was measured every 1 h.

The synthetic waste water (SWW) used above consisted of 1.16 mM sodium citrate, 0.94 mM ammonium chloride, 0.45 mM potassium phosphate, 0.5 mM calcium chloride, 0.5 mM sodium bicarbonate, 2.0 mM sodium chloride and 0.6 mM magnesium sulfate dissolved in DW. 1 mL of LB media was added to 1 L of SWW.

Membrane filtration set-up must be cleaned before the filtration experiment begins for the sterilization of the set-up and for the elimination of trace organic matters that remain inside the set-up eliminated during the cleaning process.⁸ Cleaning was done by recirculation of (1) 0.5% sodium hypochlorite for 2 h, (2) DW for 30 min, (3) 2 mM sodium dodecylbenzene sulfonate (SDS) for 1 h at the pH of 11, (4) DW for 30 min, (5) 95% ethanol for 1 h, (6) DW for 30 min.

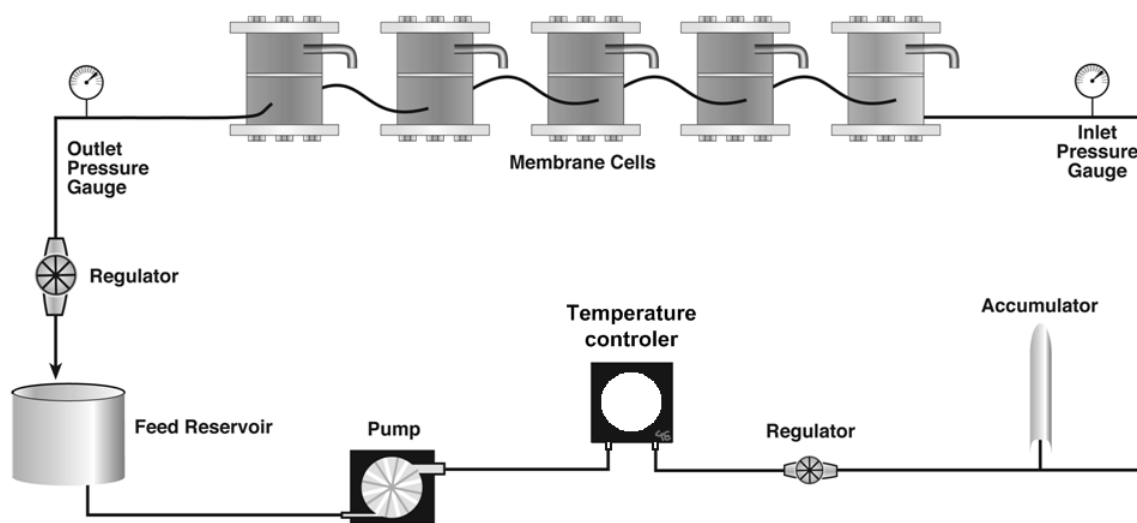


Fig. 7 The filtration set-up used for biofouling experiment.

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