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Hemodialysis membrane coated with a polymer having a hydrophilic blood-contacting layer can enhance diffusional performance

Masashi Tagaya[1], Saki Nagoshi[2], Morihiro Matsuda[3], Shunsuke Takahashi[4], Shinya Okano[1], Kazunobu Hara[1]

[1] Department of Medical Engineering, National Hospital Organization Kure Medical Center and Chugoku Cancer Center, Hiroshima - Japan
[2] Department of Clinical Laboratory, National Hospital Organization Kure Medical Center and Chugoku Cancer Center, Hiroshima - Japan
[3] Division of Preventive Medicine, Institution for Clinical Research, National Hospital Organization Kure Medical Center and Chugoku Cancer Center, Hiroshima - Japan
[4] Department of Nephrology, National Hospital Organization Kure Medical Center and Chugoku Cancer Center, Hiroshima - Japan

ABSTRACT

Purpose: Currently, the foreign surfaces of various extracorporeal circulation devices are coated with a biocompatible polymer coating agent (BPA), which creates a hydrophilic blood-contacting layer to reduce thrombogenicity, while the membranes in hemodialyzers are not. We aimed to clarify other side effects of BPA-coated membranes by examining the diffusion performance in in vitro experiments.

Methods: We used a polyethersulfone membrane (sieving coefficient of albumin is ≤0.01) coated with BPA product, SEC-1™ (Toyobo), in a hemodialyzer. To estimate the diffusion rates of a wide range of molecules, 2 L of saline containing vancomycin, lysozyme, and albumin were recirculated in the circuit configured with a hemodialyzer, and dialyzed continuously using water. The concentrations of sodium, vancomycin, lysozyme, and albumin were measured every 5 minutes for 30 minutes and compared in experiments with BPA-coated (n = 4) and BPA-noncoated (n = 4) membranes.

Results: The removal rates of sodium and vancomycin after 5 minutes of dialysis (n = 24) were significantly higher in BPA-coated than noncoated membranes, while those of lysozyme and albumin were not significantly different. The removal rates of sodium and vancomycin after 30 minutes of dialysis (n = 4) were significantly higher, and those of lysozyme were significantly lower in BPA-coated than noncoated membranes, while those of albumin were not significantly different.

Conclusions: The preliminary study suggests that BPA-coated membranes enhanced the diffusion rate of molecules with low and middle molecular weight without affecting the sieving coefficient of albumin. Thus, BPA coating can enhance the dialysis performance of membranes.

Keywords: Albumin, Copolymer, Diffusion, Hydrophilic layer, Polymer membrane, Sieving coefficient

Introduction

Hemodialysis serves to compensate for impaired renal function in patients with end-stage renal disease. A hemodialyzer consists of a membrane that corrects the levels of blood components mainly through diffusion (1-4). In order to correct blood component imbalances, patients undergo hemodialysis three times weekly for 3 to 4 hours per session (5, 6). The blood is therefore exposed to the circuit – including the hemodialyzer – for a long duration, which activates the whole coagulation cascade; hence, the administration of anticoagulants is required (7-9).

Currently, extracorporeal circulation circuits used in cardiopulmonary bypass surgeries are coated with biocompatible polymers to prevent adverse effects due to contact between the blood components and foreign surface of the circuits, although a similar effect has not been observed in hemodialysis. This biocompatible polymer coating agent (BPA) is composed of a hydrophobic backbone adherent to the surface, and a hydrophilic blood-contacting layer. The hydrophilic layer swells when blood contacts it, creating a water-filled layer that maintains protein conformation and prevents surface activation (10), and many studies have reported a reduction in the circuit's thrombogenecity, or

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Corresponding author:
Masashi Tagaya, PhD
Department of Medical Engineering
National Hospital Organization Kure Medical Center
and Chugoku Cancer Center
3-1, Aoyamacho Kure
737-0023 Hiroshima, Japan
tagayam@kure-nh.go.jp

reduction in the heparin dose needed to achieve adequate anticoagulation by BPA coating (11-15). In our previous study, a hemoconcentrator, which has similar function as the hemodialyzer in cardiopulmonary bypass circuit, was probatively coated with BPA and investigated as regards hydrokinetics. BPA coating resulted in an enhancement of the filtration rate (16). However, the filtration rate enhancement leads to the possibility that the sieving coefficient of substances that are targeted to be dialyzed are altered, and those were not examined.

If BPA-coated membranes within a hemodialyzer have strong biocompatibility, being able to prevent thrombogenicity with coating of all surfaces of the hemodialysis circuit, anticoagulant-free dialysis may be possible, although investigations for markers of coagulation will be required. However, before examining the effect on anticoagulation, the performance shift of diffusion should be clarified first. In the present study, we aimed to clarify the diffusion performance of BPA-coated membranes in in vitro experiments to progress the investigation of BPA-coated membrane for clinical application.

Materials and methods

Experiments were performed to estimate diffusion rate alterations of a wide range molecules in a BPA-coated membrane by comparing diffusion rates between a coated and noncoated membrane under identical conditions.

Materials

Eight polymer membrane columns (Hemo Crystal; Mera), which had the same lot number, were employed as hemodialyzers in this study. Hemo Crystal, with 1.1 m^2 of membranes, is used as hemoconcentrator in cardiopulmonary bypass surgery, but can function as a high-flux hemodialyzer because sieving coefficients of beta-2-microglobulin and albumin are 0.6 and ≤0.01, respectively (17). Four of these were coated with a BPA product (SEC-1™; Toyobo): a coating agent composed of a copolymer comprised of hydrophobic alkyl-acrylate, hydrophilic polyethylene-glycol-acrylate, and water repellent silicon (silicone-methacrylate).

To estimate the variance of diffusion rates among a wide range of molecules, categorized as low (≤0.5 kDa), middle (0.5-5 kDa), and large molecular weight (≥0.5 kDa), in a BPA-coated membrane, we employed the following substances as the target solutes: sodium (23 Da), vancomycin (1485.87 Da) (18), lysozyme (14 kDa), and albumin (approximately 65 kDa).

Methodology

Two liters of the controlled solution, composed of saline, 120 mg of vancomycin (vancomycin hydrochloride; Kobayashikako), 120 mg of lysozyme (lysozyme chloride refined from egg white; Nacalai Tesque), and 25 g of albumin (Albuminar®-25; CSL Behring), were prepared as the experimental solution. The experimental circuit was configured with a hemodialyzer, a roller pump, polyvinyl chloride tubes, a sampling port, and inlet and outlet chambers. The controlled solution was maintained at approximately 36°C, with continuous agitation, and was conveyed to the experimental circuit, which was primed with saline, and recirculated at 300 mL/min for several minutes to mix the saline-primed experimental circuit with the controlled solution in the experimental circuit (Fig. 1). After mixing, the controlled solution was sampled at the sampling port to measure the baseline concentrations (C_0) of sodium, vancomycin, lysozyme, and albumin. Thereafter, the controlled solution was made to flow inside the membranes within the hemodialyzer at a flow of 100 mL/min, and reverse osmosis (RO) water was convected outside the membranes at 100 mL/min, that is, the recirculated controlled solution was dialyzed by RO water. While being continuously dialyzed, 5 mL of the controlled solution was sampled after 5, 10, 15, 20, 25, 30 minutes at the sampling port to measure the concentrations of the solutes (C_t; t = 5, 10, 15, 20, 25, 30). For sodium, the concentration was measured using ion selective electrode (ABL 800; Radiometer); for vancomycin, latex agglutination turbidimetry (Nanopia® TDM Vancomycin; Sekisui Medical) was used; for lysozyme, nephelometry, as previously described by Reitamo et al (19), was used; and for albumin, the modified bromocresol purple method (Iatoro ALB; LSI Medience) was used.

Similar experiments were performed with 4 BPA-coated hemodialyzers and 4 noncoated hemodialyzers. The circuit and the controlled solution were changed for each experiment.

Statistical analysis

Diffusion rate alterations were assessed by comparing the ratio of the concentrations of sodium, vancomycin, lysozyme, and albumin after to before 5 minutes and 30 minutes of dialysis ($C_t/C_{t\text{-}5}$; C_{30}/C_0, respectively) between BPA-coated and noncoated hemodialyzers. All variables were compared using Student's t-test by Microsoft Excel software. The significance level was set at $\alpha = 0.05$.

Results

The percentage of the ratios of the remaining concentrations of sodium, vancomycin, lysozyme, and albumin after 5 minutes of dialysis ($100*C_t/C_{t\text{-}5}$; n = 24) were averaged in both BPA-coated and noncoated hemodialyzers, as shown Table I. The values of sodium and vancomycin were significantly lower in the BPA-coated than in the noncoated hemodialyzer, while those of lysozyme and albumin were not significantly different. Furthermore, percentages of the remaining concentrations obtained for the BPA-coated and the noncoated hemodialyzers, at each time point ($100*C_t/C_0$), were averaged, and were graphically represented as trends of percentages over time for each solute as shown in Figure 2. Figure 2 demonstrates that the percentages after 30-min of dialysis ($100*C_{30}/C_0$; n = 4) for sodium and vancomycin were significantly lower (means ± standard deviations were 34.6 ± 1.00 vs. 41.3 ± 0.85; $p<0.05$, and 50.5 ± 1.13 vs. 57.5 ± 0.93; $p<0.05$, respectively), and those of lysozyme were significantly higher (67.8 ± 2.62 vs. 57.9 ± 3.48; $p<0.05$) in the BPA-coated than in the noncoated hemodialyzers, respectively; while those of albumin were not significantly different (90.9 ± 0.00 vs. 95.8 ± 4.81; $p = 0.08$).

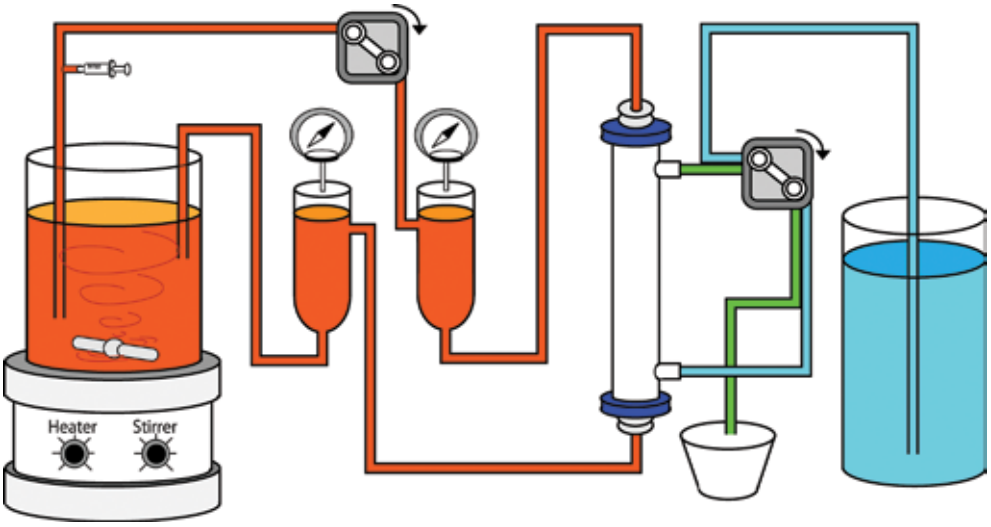

Fig. 1 - Experimental circuit. Circuit pathway of controlled solution is shown in brown. Reverse osmosis (RO) water in the plastic reservoir was delivered to the hemodialyzer (shown in blue) by a roller pump, flowed outside the membranes, and discharged from the hemodialyzer (shown in green) by the same roller pump in order to equalize both delivered and discharged RO water. The sampling port was configured prior to the inlet chamber.

TABLE I - Remaining ratios of sodium, vancomycin, lysozyme, and albumin, after 5 minutes of dialysis

Target solutes	BPA-Coated (n = 24)	Noncoated (n = 24)	p value
Sodium (%)	83.8 ± 0.61	86.3 ± 0.53	<0.05
Vancomycin (%)	89.3 ± 3.78	91.2 ± 3.09	<0.05
Lysozyme (%)	94.0 ± 4.48	91.5 ± 5.90	0.22
Albumin (%)	98.5 ± 4.48	99.4 ± 5.90	0.54

Ratios (C_t/C_{t-5}) were averaged for 6 time points in the 4 BPA-coated and the 4 noncoated hemodialyzers (n = 24), represented as means ± standard deviations.

Discussion

To estimate diffusion rate alterations of a wide range of molecules in BPA-coated membranes, we compared diffusion rates of sodium, vancomycin, lysozyme, and albumin between both BPA-coated and noncoated membranes, in which the sieving coefficient of albumin is ≤0.01, under identical conditions. In accordance with these comparative results, BPA coating resulted in increased sodium and vancomycin removal, and no differences in albumin removal, indicating that the diffusion rates of molecules with low and middle molecular weight were specifically enhanced without affecting the sieving coefficient of albumin by BPA coating; that is to say, diffusion rates of the molecules that can easily pass through the membrane pore, are solely enhanced by BPA coating. This change is expected to be favorable for membrane technology.

We previously reported that BPA coating of membranes can enhance the filtration rate without physical alteration of the membranes, and concluded that the decrement of drag resulting from water-filled layers on BPA contributed to this enhancement (10, 16, 20). Similarly, the following laws can explain the reason why decrement of drag results in molecular transfer enhancement. Based on Fick's laws of diffusion, diffusion flux (J) is directly proportional to concentration (C) and is inversely proportional to distance (x). Furthermore, based on the Stokes-Einstein equation, the molecular diffusion coefficient (D) is directly proportional to temperature (T) and mobility (B). The mobility is inversely proportional to viscosity (μ) and molecular radius (a). Those two laws indicate that the diffusion flux is increased proportionally with the decrement of viscosity.

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

$$D = KTB = \frac{kT}{6\pi\mu a}$$

[k = Boltzmann constant]

Moreover, according to Newton's law of friction, frictional stress (τ) is directly proportional to viscosity.

$$\tau = \mu\frac{\partial U}{\partial y}$$

[U = relative velocity, y = diameter of pathway]

Restating the expression of Newton's law of friction differently, the reduction of friction stress induces the decrement of viscosity. The diffusion flux is therefore increased by the greater diffusion coefficient resulting from the decrement of viscosity. Considering these interpretations, the water-filled layer on BPA, which decreases drag in the fluid pathway, can reduce the frictional stress, leading to a higher diffusion performance for low- and middle-molecular-weight molecules, such as sodium and vancomycin. Similarly, larger molecular weight reduces molecular mobility, counteracting the effect of reduced frictional stress; thus, diffusion performance is not enhanced in molecules with large molecular weight such as albumin. Considering the above factors, enhanced diffusion

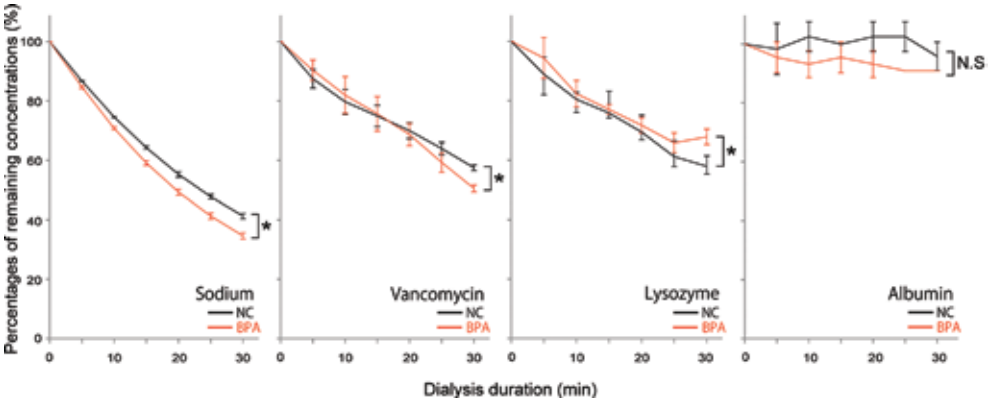

Fig. 2 - Changes in the concentration of sodium, vancomycin, lysozyme, and albumin with dialysis duration. The horizontal axis indicates dialysis duration (min), and the vertical axis indicates the percentages of remaining concentrations of each solute. The **red lines** connect the average values of the percentages of the ratio at each time point ($100*C_t/C_0$; n = 4) for the BPA-coated hemodialyzers, and the **black lines** connect those for the noncoated hemodialyzers. The error bars indicate the standard deviation.
NC = noncoated; BPA = BPA-coated; * $p<0.05$; N.S. = not significant.

rates in BPA-coated membrane can be attributed to drag decrement due to the water-filled layers on BPA.

Polymer membranes, which are called high-flux membranes, have been developed, and are currently widely used. The main focus of high-flux membrane development has been to sharpen the molecular weight cutoff of the membrane to maximize removal of low-molecular-weight proteins while minimizing the removal of albumin (21). Considering this concept, our finding that BPA coating can enhance the removal of low- and middle-molecular-weight solutes and while not affecting the sieving coefficient of albumin, is advantageous and can be considered a new improvement in membrane technologies.

On the other hand, the removal of lysozyme, which is a low-molecular-weight protein, was not maximized, which is amenable to further investigations. An explanation might be that noncoated membranes adsorb lysozyme, leading to deceptive enhancement of removal in noncoated membranes (22); thus, there is a possibility that essential diffusion rate enhancement occurred but was not observed in BPA-coated membranes. Furthermore, assessment of protein transfer is difficult since protein may undergo posttranslational modification with albumin (23). Therefore, for better clarification of the findings in the present study, additional investigations regarding low-molecular-weight proteins are needed.

In the field of cardiopulmonary bypass surgeries, polymers such as poly-2-methoxyethylacrylate (10, 12, 13) and 2-methacryloyloxyethyl phosphoryl choline (24-26), are used as BPA rather than the acrylic-monomers-copolymer that is used in this study. All of these BPAs can prevent thrombogenicity by an identical mechanism: the hydrophilic layer swells when blood contacts it, creating a water-filled layer that maintains protein conformation and prevents surface activation (10). Therefore, since all of these BPAs are expected to have an efficacy similar to that observed in this study, further studies with other BPAs could be justified. Furthermore, there are many reports about the favorable effect of BPAs on the reduction of cell adhesion, hemolysis, protein adhesion, and inflammatory markers (13, 24-26). Moreover, several reports on noncoated hemodialyzers showed that the exposure to polymer membranes has detrimental effects in terms of impaired platelet function, increased oxidative stress, and inflammation (27-30). BPA-coated membrane is therefore expected to improve hemodialysis biocompatibility, although further investigations are required.

Our investigation of BPA-coated membrane was based on the possibility of performing hemodialysis without anticoagulant administration. However, before investigating the relation to anticoagulation, if BPA coating proves to adversely affect the function of hemopurification, no further investigations would be pursued; thus, we first tried to assess the function of hemopurification in the present study. The present findings not only allow for the elimination of conceivable adverse effects by BPA coating, but also show that diffusion performance could be higher with the BPA-coated membrane. Therefore, further investigations for BPA-coated membranes are justified. We hope that our finding might support the clinical application of BPA-coated membranes.

Limitations

Vancomycin and lysozyme are adsorbed by several polymers (18, 22). There is a possibility that our results may have been affected by the adsorption, which is a major limitation of this study. Therefore, future studies with complete elimination of efficacy of adsorption should be conducted to confirm our findings.

Conclusions

The results of this preliminary study suggest that the use of a hemodialyzer with a BPA-coated membrane, in which the sieving coefficient of albumin is ≤0.01, can enhance the removal of low- and middle-molecular-weight solutes without affecting the sieving coefficient of albumin. Those aspects may occur secondary to the drag decrement of the fluid pathway induced by water-filled layers of BPA.

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Disclosures

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Conflict of interest: There are no conflicts of interest to declare.

References

1. Clark WR. Hemodialyzer membranes and configurations: a historical perspective. Semin Dial. 2000;13(5):309-311.
2. Eloot S, De Vos JY, Hombrouckx R, Verdonck P. Diffusive clearance of small and middle-sized molecules in combined dialyzer flow configurations. Int J Artif Organs. 2004;27(3):205-213.
3. Tagaya M, Matsuda M, Yakehiro M, Izutani H. Prospects for using a hemoconcentrator as an alternative hemodialysis method in cardiopulmonary bypass surgeries. Perfusion. 2014;29(2):117-123.
4. Tagaya M, Matsuda M, Yakehiro M, Izutani H. Features of an alternative hemodialysis method using a hemoconcentrator during cardiopulmonary bypass surgeries. Perfusion. 2015;30(4):318-322.
5. Buoncristiani U. Fifteen years of clinical experience with daily haemodialysis. Nephrol Dial Transplant. 1998;13(Suppl 6):148-151.
6. Eknoyan G, Beck GJ, Cheung AK, et al. Hemodialysis (HEMO) Study Group. Effect of dialysis dose and membrane flux in maintenance hemodialysis. N Engl J Med. 2002;347(25):2010-2019.
7. Koutsikos D, Fourtounas C, Kapetanaki A, et al. A cross-over study of a new low molecular weight heparin (Logiparin) in hemodialysis. Int J Artif Organs. 1996;19(8):467-471.
8. Opatrný K, Bouda M, Jr, Kohoutková L, Vít L, Sefrna F. A clinical study to assess the effect of heparin in dialyzer rinsing solutions. Int J Artif Organs. 1997;20(2):112-118.
9. Maruyama Y, Yoshida H, Uchino S, et al. Nafamostat mesilate as an anticoagulant during continuous veno-venous hemodialysis: a three-year retrospective cohort study. Int J Artif Organs. 2011;34(7):571-576.
10. Ask A, Holt D, Smith L. In vivo comparison study of FDA-approved surface-modifying additives and poly-2-methoxyethylacrylate circuit surfaces coatings during cardiopulmonary bypass. J Extra Corpor Technol. 2006;38(1):27-32.
11. Zimmermann AK, Weber N, Aebert H, Ziemer G, Wendel HP. Effect of biopassive and bioactive surface-coatings on the hemocompatibility of membrane oxygenators. J Biomed Mater Res B Appl Biomater. 2007;80B(2):433-439.
12. Yoshizaki T, Tabuchi N, van Oeveren W, Shibamiya A, Koyama T, Sunamori M. PMEA polymer-coated PVC tubing maintains antithrombogenic properties during in vitro whole blood circulation. Int J Artif Organs. 2005;28(8):834-840.
13. Suzuki Y, Daitoku K, Minakawa M, Fukui K, Fukuda I. Poly-2-methoxyethylacrylate-coated bypass circuits reduce activation of coagulation system and inflammatory response in congenital cardiac surgery. J Artif Organs. 2008;11(3):111-116.
14. Fukui T, Nishida H, Takanashi S. Biocompatibility of cardiopulmonary bypass circuit with new polymer Senko E-Ternal Coating™. Perfusion. 2015;30(7):572-579.
15. Ranucci M, Pazzaglia A, Isgrò G, et al. Closed, phosphorylcholine-coated circuit and reduction of systemic heparinization for cardiopulmonary bypass: the intraoperative ECMO concept. Int J Artif Organs. 2002;25(9):875-881.
16. Tagaya M, Takahashi S, Matsuda M, Takasaki T, Hamaishi M, Hara K. Prospects for clinical applications of polymer-coated haemoconcentrator on extracorporeal circuit in cardiopulmonary bypass surgeries. Int J Artif Organs. 2016;39(8):415-420.
17. Pharmaceuticals and Medical Devices Agency. Product information. http://www.info.pmda.go.jp/downfiles/md/PDF/530360/530360_21400BZZ00061A02_A_01_02.pdf. Accessed April 20, 2017.
18. Sartori M, Day S, De Rosa S, et al. Pharmacokinetic analysis of antibiotic adsorption (vancomycin and teicoplanin) by the Lixelle extracorporeal unit. Int J Artif Organs. 2015;38(1):8-12.
19. Reitamo S, Lalla M, Huipero A. Serum lysozyme: evaluation of a nephelometric assay. Scand J Clin Lab Invest. 1981;41(4):329-332.
20. Tanaka M, Mochizuki A. Clarification of the blood compatibility mechanism by controlling the water structure at the blood-poly(meth)acrylate interface. J Biomater Sci Polym Ed. 2010;21(14):1849-1863.
21. Ward RA. Protein-leaking membranes for hemodialysis: a new class of membranes in search of an application? J Am Soc Nephrol. 2005;16(8):2421-2430.
22. Hasegawa T, Iwasaki Y, Ishihara K. Preparation and performance of protein-adsorption-resistant asymmetric porous membrane composed of polysulfone/phospholipid polymer blend. Biomaterials. 2001;22(3):243-251.
23. Ward RA, Brinkley KA. A proteomic analysis of proteins removed by ultrafiltration during extracorporeal renal replacement therapy. Contrib Nephrol. 2004;141:280-291.
24. Iwasaki Y, Sawada S, Ishihara K, Khang G, Lee HB. Reduction of surface-induced inflammatory reaction on PLGA/MPC polymer blend. Biomaterials. 2002;23(18):3897-3903.
25. Kocakulak M, Ozgürtaş T, Ayhan H. Effect of poly (2-methoxyethyl acrylate)-coated oxygenators on haemolysis. J Biomater Sci Polym Ed. 2006;17(4):449-460.
26. Zheng Z, Ren L, Zhai Z, Wang Y, Hang F. Surface modification on polyethylene terephthalate films with 2-methacryloyloxyethyl phosphorylcholine. Mater Sci Eng C Mater Biol Appl. 2013;33(5):3041-3046.
27. Himmelfarb J, Stenvinkel P, Ikizler TA, Hakim RM. The elephant in uremia: oxidant stress as a unifying concept of cardiovascular disease in uremia. Kidney Int. 2002;62(5):1524-1538.
28. Kaysen GA. The microinflammatory state in uremia: causes and potential consequences. J Am Soc Nephrol. 2001;12(7):1549-1557.
29. Del Vecchio L, Locatelli F, Carini M. What we know about oxidative stress in patients with chronic kidney disease on dialysisclinical effects, potential treatment, and prevention. Semin Dial. 2011;24(1):56-64.
30. Daugirdas JT, Bernardo AA. Hemodialysis effect on platelet count and function and hemodialysis-associated thrombocytopenia. Kidney Int. 2012;82(2):147-157.