Novel Antifouling Surface with Improved Hemocompatibility by Immobilization of Polyzwitterions onto Silicon via Click Chemistry

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Abstract

A novel procedure a novel procedure is presented and demonstrated to develop an antifouling silicon surface with improved hemocompatibility by using a zwitterionic polymer, poly(sulfobetaine methacrylate) (polySBMA). Functionalization of the silicon surface with polySBMA involves the following three steps: (1) an alkyne terminated polySBMA was synthesized by RAFT polymerization; (2) a self-assembled monolayer with bromine end groups was constructed on the silicon surface, and then the bromine end groups were replaced by azide groups; and (3) the polySBMA was attached to the silicon surface by azide–alkyne cycloaddition click reaction. Membrane characterization confirmed a successful silicon surface modification with almost 100% coverage by polySBMA and an extremely hydrophilic surface after such modification. The polySBMA-modified silicon surface was found to have excellent anti-nonspecific adsorption properties for both bovine serum albumin (BSA) protein and model bacterial cells. Whole blood adsorption experiments showed that the polySBMA-modified silicon surface exhibited excellent hemocompatibility and effective anti-adhesion to blood cells. Silicon membranes with such antifouling and hemocompatible surfaces can be advantageously used to, for example, drastically extend the service life of artificial kidney devices.

Keywords: Zwitterionic polymer; Click chemistry; Antifouling; Hemocompatibility; Silicon membrane; Artificial kidney.

1. Introduction

Kidney not only serves as a biological filter of the human body to remove wastes from blood, but also helps regulate the body electrolyte balance, acid-base balance, blood pressure, and hormone secretion. However, about 26 million American adults suffer from chronic kidney diseases, many of which have developed to the end-stage renal disease (ESRD) and thus the function of kidney is completely lost.[1] Although dialysis is considered useful for treating ESRD, the survival rate at three years after the start of the therapy is only 50%.[2] In addition, a routine thrice-per-week dialysis treatment can be very costly and time-consuming, let alone seriously deteriorate the patients' life quality. Although kidney transplantation may achieve a much higher survival rate (91%) at three years and significantly improve the patients' life quality,[2, 3] such alternative treatment has been severely limited by the shortage of donor organs. Therefore, implantable artificial kidneys that use a manmade membrane filtration system to substitute the function of human kidney are highly promising for ESRD treatment.[4, 5]

In recent years, significant progress has been made in the development of implantable artificial kidneys.[4, 5] As the core component of an artificial kidney, membranes function as a filter to remove toxic compounds and wastes while retaining biomacromolecules in blood. In order to achieve such precise separation, membrane pore sizes must be fine-tuned to allow for only an extremely narrow distribution. Conventional polymeric membranes, which typically have an irregular structure and wide pore size distribution, are thus not suitable for this purpose. Nanostructured silicon membranes with a uniform geometry and precise pore sizes have been recently developed and shown excellent separation performance in an

implantable artificial kidney device.[2] However, silicon membranes are susceptible to nonspecific adsorption of biomacromolecules (e.g., platelets, proteins) from blood, thereby leading to full-scale platelet adhesion/activation and ultimately the formation of thrombosis/embolism that can be fatal to patients.[6] In addition, the adhesion of biomacromolecules can also clog membrane pores, reduce membrane permeability and selectivity, and hence severely shorten the lifetime of an artificial kidney device.

Surface functionalization is an extremely useful approach to prevent nonspecific adsorption and improve hemocompatibility of silicon membranes. Poly(ethylene glycol) (PEG) has been widely used and proven very efficient to reduce nonspecific adsorption on surfaces.[7-11] Unfortunately, the autoxidation of PEG in the presence of oxygen and transition metal ions alters the chemical structure of the polymer and eliminates its ability to repel nonspecific adsorption.[12, 13] For this reason, the application of PEG in blood-contacting environment where various metal ions, organic compounds, and oxygen exist, is very limited.[14]

As a new antifouling material, zwitterions have received growing attention over recent years due to their excellent anti-adhesive property against protein adsorption and bio-attachment.[15-18] It has been reported that zwitterionic monomers or polymers are effective, stable materials that can prohibit protein fouling and have an excellent anti-biofouling property against bacterial attachment in a biological system. The anti-adhesive nature of polyzwitterions can be attributed to their high hydration capability and conformational structure. Zwitterion is a molecule containing both positively and negatively charged functional groups, which neutralize each other and thus maintain an

overall neutrality. Zwitterionic polymers are made by polymerization of zwitterions, thus containing localized charges within every repeating side chain while the overall charge neutrality is maintained. Compared with PEG, the zwitterionic polymer is able to strongly bind to water molecules with localized charges. Such electrostatically induced hydration can greatly inhibit the attachment of biomacromolecules to a polymer surface due to the physical swelling of the polymer. Overall, zwitterions are very hydrophilic, non-cytotoxic, and with an acceptable endotoxin level, making them an ideal antifouling material for surface modification of silicon membranes used in an artificial kidney.

Depending on different charged functional groups carried by the polymers, zwitterionic polymers used for generating antifouling surfaces are typically classified as sulfobetaine, carboxybetaine and phosphobetaine. Phosphobetaine, a mimic of the phospholipids of the outer surface of cell membranes, is the very first type of zwitterions ever studied.[13, 19] Recently, sulfobetaine and carboxybetaine have attracted an intense interest due to their superior antifouling properties compared with phosphobetaine, and they have been used to functionalize various surfaces such as polymeric membranes,[20-27] silicon/silica,[14, 28-30] glass,[29, 31, 32] gold,[33-35] and steel.[36, 37] Studies have shown that carboxybetaine may exhibit better anti-adhesion properties than sulfobetaine.[38] When fouling is a primary concern, however, sulfobetaine is more often used to prevent/mitigate nonspecific adsorption. This is because the charged functional groups (of a “strong-strong” type) of sulfobetaine maintain much more stable localized charge properties under different conditions. In a wide pH range (e.g., 3 to 10), a sulfobetaine polymer can still retain its zwitterionic property, while carboxybetaine with charges of a “strong-weak” type loses the localized negative charge from

carboxyl groups under acidic conditions.[39] As a result, the zwitterionic property of the nanobrush is impaired and the polymer exhibits overall positive charge instead. This positive charge may adsorb biomacromolecules with negative charges and thus lead to severe fouling of the surface.

In this study, we employed a zwitterionic polymer, poly(sulfobetaine methacrylate) (polySBMA), to improve the antifouling properties of silicon surfaces. The polySBMA was immobilized onto the silicon surface using a highly efficient azide-alkyne click reaction, following a three-step procedure: (1) synthesis of alkyne terminated polySBMA by a reversible addition-fragmentation chain-transfer (RAFT) mediated radical polymerization, (2) functionalization of silicon by self-assembled monolayer (SAM) with bromine end groups and conversion of bromine groups to azide groups by nucleophilic substitution, and (3) immobilization of alkyne terminated polySBMA to the silicon surface by azide-alkyne click reaction. Fourier transform infrared spectroscopy (FTIR), X-ray photoelectron spectroscopy (XPS) and water contact angle measurement were carried out to characterize and confirm the functionalization of silicon surfaces. The anti-nonspecific adsorption ability of the modified samples was evaluated by protein adsorption using quartz crystal microbalance with dissipation (QCM-D) and bacterial cell adhesion experiments. The hemocompatibility of the modified surface was evaluated by whole blood contact experiments.

2. Materials and Methods

2.1. Polyzwitterion synthesis

All chemicals are analytical grade or higher and were obtained from Sigma-Aldrich (St. Louis, MO). The deionized (DI) water was used in all experiments. PolySBMA was synthesized in our lab by a RAFT mediated radical polymerization.[27] Briefly, azodiisobutyronitrile (AIBN) was used as an initiator, and alkyne-terminated S-1-Dodecyl-S'-(R,R'-dimethyl-R''-acetic acid)trithiocarbonate (alkyne-SDDAT) as a chain transfer agent. First, 5 g SBMA ([2-(methacryloyloxy)ethyl]dimethyl-(3-sulfopropyl) ammonium hydroxide), 0.2 g alkyne-SDDAT, and 50 mg AIBN were mixed with 100 ml pure ethanol in a single-necked flask. Nitrogen was then bubbled through the solution for 30 min to remove oxygen from the flask. Next, RAFT polymerization was performed at 55 °C for 24 h to obtain alkyne-polySBMA, followed by cooling the reactor in ice water. The synthesized polymer was then collected by filtering the solution through a vacuum filter (Cole Parmer, Vernon Hills, IL), rinsing with ethanol, and subsequently drying overnight in oven at 65 °C. The collected alkyne-polySBMA was stored in desiccator prior to use in the grafting experiments.

2.2. Silicon surface functionalization

The overall modification procedure for silicon surface is illustrated in **Figure 1**. Before modification, a silicon wafer was first cleaned in UV/Ozone chamber for 30 min, rinsed by DI water and ethanol, and then dried with nitrogen gas. The silicon wafer was then brominated by immersing it in (3-bromopropyl)trichlorosilane (BPTS) (0.005% v/v) anhydrous toluene solution to form an SAM on the surface. After reacting in a sealed vessel placed in a desiccator for 24 h, the wafer was taken out, thoroughly rinsed with toluene, DI

water, and pure ethanol, and finally dried with nitrogen air. Next, the bromine groups were converted to azide groups by placing the wafer in 10 g/L sodium azide solution in a small vessel, and the reaction continued at room temperature for 24 h. Finally, the wafer was washed with DI water and subsequently dried with N₂ air.

PolySBMA was immobilized onto the silicon surface by click chemistry through azide-alkyne Huisgen cycloaddition. The polymer solution for the click reaction was prepared by dissolving 0.50 g alkyne-polySBMA and 0.011 g CuSO₄ in 50 mL DI water. The polymer solution was degased by nitrogen for 30 min and then azide-functionalized silicon wafer was immersed in the solution. Afterwards, 0.024 g sodium ascorbate was added into the solution to initiate the click reaction. The flask was placed on a shaking table for 24 h to complete the click reaction. The polySBMA immobilized silicon wafers were taken out and thoroughly washed with DI water for 24 h on a shaker by changing water frequently, followed by rinsing with ethanol and drying with nitrogen gas.

Figure 1

2.3. Surface characterization

FTIR measurement was carried out on a Thermo Nicolet Nexus 670 spectrometer with a Smart Golden Gate accessory and a diamond crystal window (Thermo Fisher, Madison, WI). Data collection and analysis was performed using the OMNIC software 6.2 provided with the instrument. XPS was used to characterize the chemical composition in each modification step. The measurement was taken on a Kratos AXIS 165 system (Shimadzu Corporation, Japan) equipped with monochromatic Al X-ray sources. Data analysis was carried out with the CasaXPS processing software. Water contact angle was analyzed using a sessile drop

method with a Kruss G10 goniometer (Matthews, NC). First, a water drop ($\sim 5 \mu\text{L}$) was injected from a syringe onto the membrane surface. After waiting 5 s to ensure the settlement of the water drop, a picture was taken to determine the contact angle.

2.4. Bacterial cell adhesion experiment

Escherichia coli (*E. Coli*) K12 MG1655 was used as a model bacterial cell. The *E. coli* cells were tagged with a plasmid coding for green fluorescent proteins to allow easy visualization via microscopy. A detailed cell culture procedure can be found in our previous study [40]. Briefly, *E. coli* was pre-cultured in 50 mL of LuriaBertani (LB) broth in a 250 mL flask. Then, 0.1 mL of the pre-cultured *E. coli* stock was transferred to another 250 mL culture flask with newly prepared 50 mL of LB broth. The second flask was then incubated at 37 °C in the shaking water bath until *E. coli* grew to the mid-exponential phase. The cells were harvested and washed three times by centrifugation at 3000 rpm for 15 min, and then re-suspended in 154 mM NaCl solution (*i.e.*, under cell isotonic condition). The cell suspension was then used in the adsorption experiments.

In a typical cell adsorption experiment, silicon samples were immersed in the cell suspension and then kept at 37 °C for 30 min. For an incubation experiment, the samples were incubated with 0.1 mL pre-cultured *E. coli* stock in the presence of LB broth until the cell density reached 2.58×10^8 cell/mL. After that, the samples were taken out and rinsed with DI water. A fluorescence microscope equipped with a digital camera was then used to observe the sample surfaces. For each sample, pictures were taken from three different spots, and from each picture the bacterial cells were counted at three different locations.

The average of the nine counts was used as the final cell adsorption result.

2.5. Evaluation of nonspecific protein adsorption

QCM-D experiments were performed with a Q-sense E4 system (Q-sense, Sweden) to compare the nonspecific adsorption of proteins on unmodified and polySBMA immobilized silicon surfaces, respectively. To start a QCM-D experiment, a silicon sensor was assembled into the cell at a temperature of 20 °C. A baseline was generated with DI water, and then either electrolyte or protein solutions were injected at a constant flow rate of 100 $\mu\text{L}/\text{min}$. The changes in frequency and dissipation were observed, recorded, and processed by the Q-Tools software supplied by the manufacture (Q-sense, Sweden).

2.6. Evaluation of whole blood adsorption

To evaluate the effect of polySBMA on improving the hemocompatibility of silicon surfaces, a whole blood adsorption experiment was carried out with modified and unmodified silicon samples, respectively. The use of human blood in this research and the related protocol were approved by the Institutional Review Boards. Venous blood was drawn from a healthy adult blood donor into polypropylene tubes coated with anticoagulant heparin and was immediately put into a cooler box and transported to the lab. Each silicon sample was put in a small glass vial and 5 mL blood was poured in it. After being incubated at 37 °C for 1 h on a shaker at 70 RPM, the sample was taken out and rinsed with phosphate buffer solution (PBS) (100 mM, pH = 7.4). Then the sample was fixed with 2.5 wt%

glutaraldehyde in PBS buffer for 30 min and rinsed again with PBS buffer. After that, the sample was dehydrated with a series of ethanol/water mixtures (20, 40, 50, 60, 80, 100 vol.% ethanol; 30 min in each mixture) before being sputter-coated with gold and observed with SEM.

3. Results and Discussion

3.1. Immobilization of polySBMA on silicon surface by click chemistry

As shown in **Figure 1**, the immobilization of polySBMA on a silicon surface involved three major steps. The first step was to synthesize the clickable polySBMA by RAFT mediated polymerization. The chain transfer agent used in this study has an alkyne group, which subsequently remained at one end of the polymer chain. In the second step, an SAM layer with bromine end groups was first formed on the silicon surface and then bromine was converted to an azide group by reacting with sodium azide. The last step was to immobilize polySBMA onto the silicon surface by a click reaction between the alkyne groups on polySBMA and the azide groups on the silicon surface. The same procedure was applied for both silicon wafer and QCM-D sensor.

3.2. Characterization of polySBMA immobilization

FTIR was employed to qualitatively confirm the immobilization of polySBMA. As shown in **Figure 2**, there is no obvious peak on the spectrum of the unmodified silicon surface, while four new peaks can be observed on the spectrum of the polySBMA-immobilized sample surface. Peaks at 1725 cm^{-1} , 1165 cm^{-1} and 1034 cm^{-1} can

be assigned to the stretching vibration of C=O in ester bond, stretching vibration of the sulfonate groups, and stretching vibrations of C-O, respectively. All these three groups are from the structure of polySBMA (**Figure 1**). An additional broad peak between 3000 and 3600 cm^{-1} can be most likely ascribed to the H_2O adsorbed on the polySBMA-modified sample surface, since polySBMA is an extremely hydrophilic polymer.[41]

Figure 2

XPS was used to more quantitatively confirm the immobilization of polySBMA. **Figure 3** shows the XPS spectra of the brominated and polySBMA-modified silicon surfaces, respectively. For the brominated silicon surface, significant signals from carbon (1s, 287.4 eV), oxygen (1s, 402.8 eV), and bromine (3d, 70.9 eV; 3p, 183.5 eV; 3s, 255.0 eV) can be found, proving the formation of bromine-ended SAM on the silicon surface. However, strong silicon signal (2p, 103.0 eV; 2s, 150.5 eV) can still be seen with a content of ~19% (**Table 1**). The observation of silicon underneath the SAM layer is most likely because the thickness of the SAM layer was less than the typical XPS detection depth (~10 nm). After the immobilization of polySBMA (**Figure 3(b)**), the carbon signal became the strongest with an almost doubled content compared with that of the SAM-coated surface, most likely due to the high content of carbon in the polymer backbone. Additionally, new peaks from nitrogen (1s, 402.8 eV) and sulfur (2s, 228.2 eV; 2p, 163.6 eV) further proved the presence of polySBMA. More importantly, the silicon peak nearly disappeared (only 0.89%) after polySBMA immobilization, indicating that the silicon surface was almost fully covered by polySBMA.

Figure 3, Table 1

Water contact angle measurement is an efficient way to evaluate the change in surface hydrophilicity due to surface modification. **Figure 4** shows the water contact angles and the water drop images on surfaces before and after each modification step. After the ozone/UV treatment, the silicon wafer surface was covered with hydroxyl groups, which led to very low water contact angle (15.5 degrees). The subsequent formation of bromine-terminated SAM layer, however, significantly increased the contact angle to 95.2 degrees due to the hydrophobic nature of the bromine groups. The water contact angle then decreased to 48 degrees by converting bromine groups to more hydrophilic azide groups. In the end, the attachment of polySBMA resulted in an extremely hydrophilic surface with a contact angle of only about 10 degrees. These data all agree with the surface chemistry changes in every step and give clear evidence for a successful surface modification.

Figure 4

3.3. QCM-D evaluation of nonspecific protein adsorption

We have performed the protein adsorption experiments using a QCM-D system to compare the nonspecific adsorption of proteins on the unmodified and polySBMA-immobilized silicon surfaces, respectively. As shown in **Figure 5(a)**, the unmodified silicon sensor suffered from obvious protein adsorption, and DI water washing did not regenerate the baseline, indicating the existence of irreversible protein adsorption on the surface. In contrast, the polySBMA-modified silicon sensor (**Figure 5(b)**) experienced no irreversible protein adsorption, as both f and D resumed the DI baseline after protein adsorption. These results proved that the grafted polySBMA exhibits an excellent resistance to nonspecific protein adsorption.

The mechanisms of the anti-adhesion properties of polySBMA can also be elucidated from the QCM-D experiments. As shown in **Figure 5(a-b)**, the unmodified and modified surfaces exhibited different responses to 20 mM NaCl solution. The decreased frequency and increased dissipation in NaCl solution are most likely due to water uptake by the zwitterionic layer on the surface, increasing the thickness and weight of the zwitterionic layer. As schematically illustrated in **Figure 5(c)**, the inter- and intra-chain electrostatic attractions between the positive and negative charges in a low salinity environment lead to a collapsed structure of polySBMA chains. In a high salinity environment, however, the presence of ions (Na^+ and Cl^- in our case) can shield charges on the zwitterionic polymer, thereby causing significant hydration to the polymer and making the flexible chains possess a more stretched morphology. [42] Therefore, it is likely that polySBMA adopts a swollen structure in a high salinity environment and further improves its antifouling properties.

Figure 5

3.4. Cell adhesion experiment

To further confirm the anti-nonspecific adsorption property of the polySBMA-modified silicon surface, cell adhesion experiments were carried out. **Figure 6** shows the fluorescence microscope images taken for the unmodified, azide-functionalized, and polySBMA-modified surfaces, respectively. As shown in **Figure 6(a-b)**, fluorescent spots that came from the protein on *E. Coli* cell membranes can be observed on both unmodified and azide-functionalized surfaces, indicating severer cell adhesion on the surfaces. However, almost no fluorescence emission can be seen on the polySBMA-modified surface, demonstrating an excellent resistance to cell adhesion. To further validate the cell adhesion

resistance enabled by the polySBMA layer, we also performed the adsorption experiments under cultural conditions, in which bacteria grow, proliferate, and behave more similar to how they are in nature. As shown in **Table 2**, the number of bacterial cells adhered on the unmodified silicon surface is more than 38 times that on the polySBMA-modified surface. Besides, the azide-functionalized surface absorbed fewer bacterial cells than those on the unmodified sample, but still more than ten times those on the polySBMA-modified surface.

Figure 6, Table 2

3.5. Effects of hydrophilicity on nonspecific adsorption

The adsorption results also demonstrate that hydrophilicity is not the only contributor to the antifouling property of polySBMA surfaces. Normally the anti-nonspecific adsorption property of a surface is often attributed to the repulsion effect of the hydration layer brought by hydrophilic surfaces. In this study, however, the hydrophilic silicon surface (**Figure 4(a)**) did not suppress the nonspecific adsorption of either protein (**Figure 5(a)**) or bacterial cells (**Figure 6(a)**). We hypothesize that the high-density hydroxyl groups on silicon surface are typical hydrogen-bond donors and thus cause significant nonspecific adsorption. This hypothesis is consistent with the results from the literature,[12, 43-45] which theorized that besides hydrophilicity, three other criteria are needed for an effective non-fouling surface — no hydrogen-bond acceptors, no hydrogen-bond donors, and an overall charge neutrality.

Compared with silicon, polySBMA not only is highly hydrophilic but also possesses all the other criteria for a non-fouling surface. It is thus theorized that the combined effects of high hydrophilicity, no hydrogen-bond acceptors/donors, and overall charge neutrality are

responsible for the excellent anti-nonspecific adsorption property of polySBMA. Moreover, simulation studies from the literature have found that water molecules stay longer in the hydration layer of zwitterions than in PEG, indicating that the zwitterion surface binds water molecules more tightly than the PEG surface.[46] Therefore, the highly stable hydration layer of polySBMA is also believed to greatly contribute to its excellent anti-nonspecific adsorption ability.

3.6. Hemocompatibility

As the core part of an artificial kidney device, the silicon-based membrane directly contacts with the blood stream. Therefore, the hemocompatibility of the silicon surface is critical to the proper functioning and the lifetime of the artificial kidney. In general, plasma proteins adsorb onto the surface of blood-contacting materials and trigger subsequent platelet adhesion, which then activates a coagulation pathway and finally results in thrombus formation.[6, 47] So far, we have demonstrated that grafting polySBMA onto the silicon surface is highly efficient in resisting nonspecific protein/cell adsorption. In order to further confirm such resistance under the conditions resembling the actual environment, we employed the whole blood adsorption experiments to study the hemocompatibility of polySBMA.

The hemocompatibility results from the blood adsorption experiments are provided in **Figure 7**. After 1 h incubation in whole blood, obvious differences between the samples can be observed even with naked eyes. As shown in **Figure 7(a) and (b)**, the unmodified silicon and Si-N₃ samples exhibit cloudy surfaces, indicating significant adsorption of blood

materials. In contrast, the Si-polySBMA sample (**Figure 7(c)**) exhibits a very clear surface, indicating no or little adsorption.

Such differences in blood adsorption can be further proved by the SEM images of the samples. As shown in **Figure 7 (a-1) and (a-2)**, the unmodified silicon surface was covered by a layer of thrombus and platelets with a tendency of aggregation. Moreover, it is noticed that all the platelets underwent shape change and pseudopodia emission, indicating an activated status of the attached platelets and a poor hemocompatibility of the silicon surface.[48] The Si-N₃ surface (**Figure 7 (b-1) and (b-2)**) showed much less platelet adhesion while the activation-related aggregation and size enlargement still existed, implying an unfavorable blood compatibility of this surface. Obviously distinguished from the afore-mentioned two samples, the polySBMA-modified surface exhibited no detectable thrombus or platelet adhesion (**Figure 7 (c-1) and (c-2)**). These results clearly demonstrate that polySBMA grafting can bring excellent antiadhesive and antithrombotic effects to the silicon surface and greatly improve the surface hemocompatibility. Therefore, silicon surface modification by polySBMA could potentially enhance the performance and prolong the service life of the silicon membranes in an artificial kidney device.

Figure 7

4. Conclusions

In this study we have established a click-based method to effectively immobilize polySBMA onto a silicon surface to improve its antiadhesive property and hemocompatibility. In this method, an alkyne-ended polySBMA was first synthesized via RAFT polymerization and the silicon surface was functionalized with SAM bearing azide groups. Subsequently,

an azide–alkyne cycloaddition click reaction was carried out to attach the polySBMA to the silicon surface. FTIR, XPS and contact angle measurements have all confirmed the successful surface modification with almost 100% surface coverage by polySBMA and an extremely hydrophilic surface after the modification. The polySBMA-modified silicon surface exhibited excellent anti-nonspecific adsorption of both BSA protein and model bacterial cells. The whole blood adsorption experiments clearly showed that the polySBMA-modified silicon surface had an excellent antiadhesive property to blood cells and outstanding surface hemocompatibility. Therefore, this method can be effectively used to improve the antifouling properties of silicon membranes in an artificial kidney device. By coating such a polySBMA layer on the silicon membrane, its blood-contacting surfaces will be highly resistant to the nonspecific adsorption of proteins, platelets, and other biomacromolecules in the plasma, thereby significantly prolong the lifetime of the artificial kidney device.

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Figure 1. Schematic illustration of the surface modification procedure.

Figure 2. FTIR spectra of the unmodified and polySBMA-modified silicon surfaces.
transmittance

Figure 3. XPS spectra of (a) brominated-modified and (b) polySBMA-modified silicon surfaces, respectively.

Figure 4. Water contact angles of (a) unmodified, (b) brominated-modified, (c) azide-functionalized, and (d) polySBMA-modified silicon surfaces, respectively.

Figure 5. Protein adsorption on (a) silicon-modified silicon surface and (b) polySBMA-modified silicon surface. (c) Mechanisms for the responses of polySBMA in different electrolyte solutions.

Figure 6. Fluorescence images of bacterial cells adsorbed on (a) unmodified, (b) azide-functionalized, and (c) polySBMA-modified silicon surfaces, respectively.

Figure 7. Whole blood adsorption on (a) unmodified, (b) azide-functionalized, and (c) polySBMA-modified silicon surfaces, respectively. SEM images for each sample were taken at 1,000 $\times$ (denoted by -1) and 10,000 $\times$ (denoted by -2) magnifications, respectively.

Table 1. Elemental composition of brominated and polySBMA modified silicon surfaces, respectively, from XPS characterization.

Sample	Percentage content (%)					
	C	N	O	Si	S	Br
Si-Br	37.75	-	38.70	19.06	-	4.49
Si-PolySBMA	75.48	2.99	17.37	0.89	3.27	-

Table 2. Number of cells adhered on different surfaces after culture with bacterial stock.

Sample	Cell density in suspension (cells/ml)	Adhesion on surface ($\times 10^4$ cells/mm ²)
Unmodified Si	2.58×10^8	7.24 ± 1.01
Si-N ₃		2.36 ± 0.65
Si-PolySBMA		0.19 ± 0.03

Graphical Abstract