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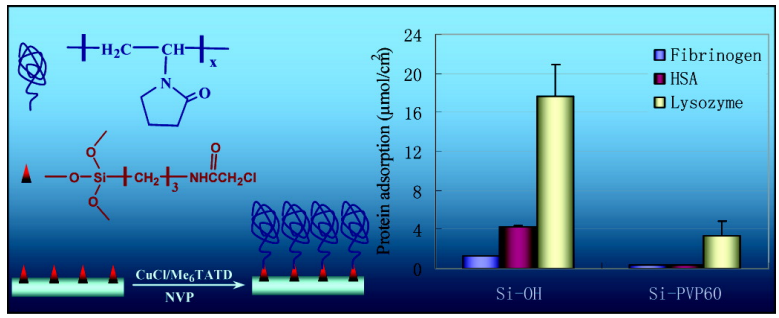

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Protein Adsorption on Poly(*N*-vinylpyrrolidone)-Modified Silicon Surfaces Prepared by Surface-Initiated Atom Transfer Radical Polymerization

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Well-controlled poly(*N*-vinylpyrrolidone) (PVP)-grafted silicon surfaces were prepared by surface-initiated atom transfer radical polymerization (SI-ATRP) with 1,4-dioxane/water mixtures as solvents and CuCl/5,7,7,12,14,14-hexamethyl-1,4,8,11-tetraazacyclotetradecane (Me_6TATD) as a catalyst. The thickness of the PVP layer on the surface increased with reaction time, suggesting that the ATRP grafting of *N*-vinylpyrrolidone (NVP) from the silicon surfaces was a well-controlled process. The water contact angle and X-ray photoelectron spectroscopy (XPS) were used to characterize the modified surfaces. The protein adsorption property of the PVP-grafted surfaces was evaluated using a radiolabeling method. Compared with unmodified silicon surfaces, a Si−PVP60 surface with a PVP thickness of 15.06 nm reduced the level of adsorption of fibrinogen, human serum albumin (HSA), and lysozyme by 75, 93, and 81%, respectively. Moreover, the level of fibrinogen adsorption decreases gradually with an increase in PVP thickness. However, no significant difference in fibrinogen adsorption was found when the PVP layer was thicker than the critical thickness of 13.45 nm.

Introduction

The uncontrolled, nonspecific adsorption of proteins onto synthetic surfaces causes a complex cascade of undesired events, including coagulation, complement activation, platelet adhesion and activation, immunological reactions, and other undesirable responses in the applications of biomaterials.[1−4] There is thus considerable interest in surfaces that might inhibit or prevent protein adsorption to improve the biocompatibility of biomaterials.[5] Over the past two decades, grafting poly(ethylene glycol) (PEG) onto a surface has been extensively investigated as the most common method of constructing nonfouling surfaces[6−10] because PEG has good biocompatibility, low toxicity, nonimmunogenicity, and high water solubility.[11] Although PEG-modified surfaces can effectively resist nonspecific protein adsorption, they have limited success in preventing long-term Biofilm formation.[12] It was reported that PEG can be oxidized enzymatically in vivo, allowing proteins and cells to attach.[13] The susceptibility of PEGs to oxidative damage in the presence of oxygen and transition metal ions reduces their utility for applications that require long-term material stability.[14]

Poly(*N*-vinylpyrrolidone) (PVP) has a long history as a synthetic polymer for biomedical applications since its discovery in Germany in 1930 and was widely used as a blood plasma substitute during World War II.[15] Research on PVP has attracted considerable interest due to its excellent water solubility, biocompatibility,[16,17] chemical stability, and biological inertness.[18] Furthermore, polymeric substrates blended with PVP can effectively resist nonspecific protein adsorption.[19−21] For example, the membranes made from blends of PVP have been found to reduce fouling by bovine serum albumin (BSA),[22] human serum albumin (HSA), and fibrinogen.[23] However, a significant disadvantage of polymeric substrates blended with PVP is the

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leaching of PVP from the blended polymeric substrates for a long-term application.

Compared with the blend method, covalent grafting of PVP onto substrates is an effective way to solve the PVP leaching problem.[24−26] Pieracci et al.[24] reported graft polymerization of *N*-vinylpyrrolidone (NVP) onto poly(ethersulfone) membranes by photolysis using ultraviolet light. The modified membranes showed a 49% decrease in fouling using a BSA solution. Higuchi et al.[25] reported graft polymerization of NVP onto polysulfone hollow fibers by free radical polymerization. The modified membranes gave a low level of protein adsorption from plasma solution and reduced the amount of adhered platelets. Rovira-Bru et al.[26] reported that the level of adsorption of lysozyme onto zirconia surfaces modified with terminally grafted PVP via free radical polymerization decreased by up to 76%. Although grafting PVP onto a surface via conventional free radical polymerization is achievable, this grafting strategy offers little control over graft density (number of chains per unit area) and chain length (number of monomer units per chain), which are believed to be two key parameters in determining protein-resistant behavior.[27,28] Therefore, the relationship between these parameters and protein-resistant behavior for PVP-grafted substrates is difficult to investigate.

To obtain well-controlled PVP-grafted surfaces and ultimately achieve surfaces that can control protein adsorption behavior, one of the most effective methods is surface-initiated atom transfer radical polymerization (SI-ATRP). SI-ATRP is a "living" polymerization method that has been extensively investigated for grafting polymers onto surfaces in a controlled manner, including graft density, chain length, and chemical composition.[29] Recently, there are numerous reports referring to the grafting of polymers such as poly(2-methacryloyloxyethyl phosphorylcholine) (PMPC),[27,28] poly[oligo(ethylene glycol) methyl methacrylate] (POEGMA),[7] poly(sulfobetaine methacrylate) (PSBMA),[30] and poly(carboxybetaine methacrylate) (PCBMA)[31] by SI-ATRP and their anti-biofouling abilities. However, NVP is a nonconjugated monomer. The controlled/"living" radical polymerization of NVP is difficult because the propagating radicals are so reactive that they have a tendency to undergo various side reactions during the polymerization.[32] Only one publication which was concerned with the atom transfer radical polymerization (ATRP) of NVP[33] has been found so far. Moreover, in this work,[33] the polymerization was carried out in solution, rather than on a surface.

In this study, surfaces grafted with PVP were prepared with the well-controlled process, i.e., SI-ATRP on silicon wafers. The ability of these surfaces to suppress protein adsorption was assessed by investigating the adsorption of fibrinogen, HSA, and lysozyme on these surfaces. The three proteins have significantly different sizes and charges. The influence of the thickness of grafted PVP on fibrinogen adsorption was also examined.

Experimental Section

Materials. Silicon wafers [n-doped, (100)-oriented, 0.56 mm thick, and 100 mm in diameter] were purchased from the laboratory of Guangzhou Semiconductor Materials (Guangzhou, China). The as-received silicon wafers were polished on one side and cut into square chips of ∼0.5 cm × ∼0.5 cm using a diamond-tipped glass cutter. *N*-Vinylpyrrolidone (NVP, Acros, 98%) was purified by distillation under reduced pressure to remove the inhibitors before use. CuCl (Aldrich, 98%) was purified by being stirred in acetic acid, washed with methanol, and then dried in vacuum. 3-Aminopropyltriethoxysilane (APTES) and methyl 2-chloropropionate (MCP) were purchased from Aldrich and used as received. 1-Pyrenebutyric acid hydrazide was synthesized according to the published method.[34] *N*,*N*-Dimethylformamide (DMF), triethylamine (TEA), chloroacetyl chloride, and all other solvents were purchased from Shanghai Chemical Reagent Co. and used as received without further purification. Nitrogen gas was of high-purity grade. The cyclic ligand 5,7,7,12,14,14-hexamethyl-1,4,8,11-tetraazacyclotetradecane (Me_6TATD) was prepared according to the method described by Hay and Lawrance:[35] yield 73.5%; ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.12 (m, 6H, CH_3), 1.32 (m, 12H, CH_3), 1.53 (m, 4H, CH_2C), 2.32 (m, 2H, CH), 2.58 (b, 4H, NH disappeared with addition of D_2O), 2.69−2.92 (m, 8H, NCH_2).

Fibrinogen (plasminogen-free) was purchased from Calbiochem (La Jolla, CA). Lysozyme was obtained from Sigma Chemical Co. (St. Louis, MO) and was used as received. HSA was purchased from Sigma Chemical Co. Tris-buffered saline (TBS, pH 7.4) was prepared by mixing a 0.05 mol/L tris(hydroxymethyl)aminomethane aqueous solution with 1.0 mol/L HCl to give the required pH value.

Surface Treatments. The pretreatment of silicon wafers was carried out according to the published procedure.[36] To obtain a hydroxy-terminated Si(100) surface (Si−OH surface), the clean wafers were exposed to UV irradiation in a rotary photochemical reactor (model UER 20-172, manufactured by Ushio Electric) at 28 °C for 10 min. After UV irradiation, a uniform hydroxy-terminated silicon surface was obtained.[37]

Preparation of PVP-Grafted Silicon Wafers. *Synthesis of Amino-Functionalized Silicon Wafers.* The freshly prepared hydroxy-terminated silicon surfaces were placed in a solution of 0.4 mL of APTES (1.7 mmol) in 20 mL of dry toluene. After being treated at 80 °C for 24 h, the amino-functionalized silicon substrates were washed successively with toluene, dichloromethane, methanol, and water and then dried in a nitrogen stream.

Synthesis of Initiator-Functionalized Silicon Wafers. The amino-functionalized silicon wafers described above were immersed in a solution of 1.0 mL of TEA (7.0 mmol) in 20 mL of dichloromethane. After the sample had cooled to 0 °C, chloroacetyl chloride (0.5 mL, 6.6 mmol) was added dropwise into the mixture. The reaction was carried out at 0 °C for 0.5 h and then at room temperature for an additional 12 h. The obtained initiator-functionalized silicon wafers were purified following procedures similar to those described for the synthesis of amino-functionalized silicon substrates.

Surface-Initiated ATRP of NVP from Initiator-Functionalized Silicon Wafers. A typical procedure for grafting PVP onto the initiator-functionalized silicon wafers follows. CuCl (9.9 mg, 0.1 mmol), Me_6TATD (28.4 mg, 0.1 mmol), NVP (1.04 mL, 10 mmol), and 2.0 mL of the mixture of 1,4-dioxane (90 vol %) and water (10 vol %) were added separately to glass tubes that were capped with a rubber septum and equipped with a magnetic stir bar. After being stirred for 15 min under a nitrogen atmosphere, the mixture was degassed

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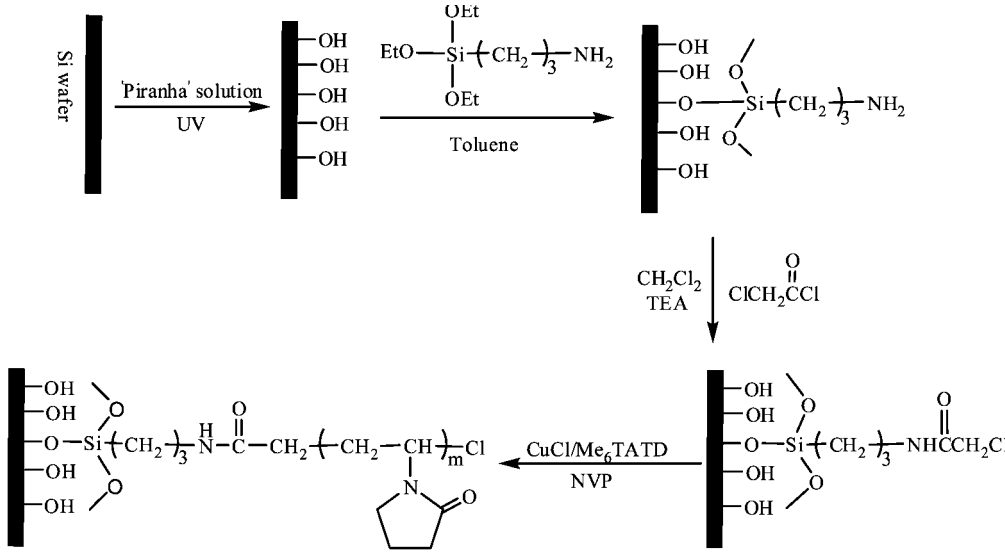
Scheme 1. Surface-Initiated ATRP of NVP



by three freeze−pump−thaw cycles. In the frozen state, initiator-functionalized silicon wafers were placed into these glass tubes. The tubes were then subjected to two additional freeze−pump−thaw cycles. After the contents had thawed, the tubes were kept at 30 °C. After desired periods for polymerization, the obtained PVP-grafted silicon wafers were removed from solution, rinsed with acetone, cleaned ultrasonically in acetone for 30 min, rinsed again with acetone and water, and then dried under a nitrogen flow.

Nucleophilic Substitution Reaction of PVP-Grafted Silicon Wafers. The Si−OH and PVP-grafted silicon wafers were placed in a solution of 1-pyrenebutyric acid hydrazide (0.15 g, 0.5 mmol) in 5 mL of DMF. After being treated at 110 °C for 12 h, the resulting silicon substrates were rinsed successively with DMF, dichloromethane, methanol, and water and then dried in a nitrogen stream.

Surface Characterization. The chemical composition of the modified silicon surfaces was determined with an ESCALAB MK II X-ray photoelectron spectrometer (XPS) (VG Scientific Ltd.). The static water contact angles of the unmodified and modified silicon surfaces were measured on a Ráme-Hart (Mountain Lakes, NJ) goniometer using the sessile drop method. The thickness of the PVP grafted on the silicon substrate was determined with an M-88 spectroscopic ellipsometer (J. A. Woollam Co., Inc.). The topology of the modified silicon surfaces was studied with a DI Nanoscope V atomic force microscope (AFM) (Vecco). The fluorescence of the silicon surfaces after the nucleophilic substitution reaction was detected with a Leica-SP2 confocal laser scanning microscope (CLSM) (Leica Ltd.).

Protein Adsorption Experiments. Fibrinogen, HSA, and lysozyme were radiolabeled with ^{125}I (Isotope Company of China, Beijing, PR China) using the iodine monochloride (ICl) method. Unbound radioactive iodide was removed by ion exchange chromatography on an AG-1-X4 resin column (Bio-Rad Laboratories, Hercules, CA). For studies of protein adsorption from buffer, labeled and unlabeled proteins were mixed (1/19, labeled/unlabeled) at a total concentration of 1 mg/mL. The clean, unmodified, and modified silicon wafers were first immersed in TBS-filled multiwell plates for 12 h to achieve complete hydration prior to the adsorption experiments.

The samples were placed in wells containing protein solutions, and adsorption was allowed to proceed for 3 h at room temperature. The silicon surfaces were then rinsed three times, 10 min each time, with TBS, wicked onto filter paper, and transferred to clean tubes for radioactivity determination using a Wizard 3″1480 Automatic Gamma Counter (Perkin-Elmer Life Sciences). Protein adsorption was expressed as moles or mass per unit surface area (micromoles per square centimeter or micrograms per square centimeter).

Table 1. Layer Thicknesses and Sessile Drop Water Contact Angles (advancing) of Unmodified and Modified Silicon Wafer Surfaces

surface	time (min)	layer thickness (nm)[a]	contact angle θ (deg)[b]
Si−OH	—	—	2
Si−APTES	—	3.05 ± 0.21	71
Si−initiator	—	3.25 ± 0.17	66
Si−PVP5	5	1.66 ± 0.14	47
Si−PVP15	15	5.62 ± 0.26	36
Si−PVP30	30	9.90 ± 0.37	30
Si−PVP45	45	13.45 ± 1.24	24
Si−PVP60	60	15.06 ± 0.99	24
Si−PVP120	120	33.56 ± 1.36	24

[a] Ellipsometry thickness measured in air-solid mode. Data are means ± the standard error ($n = 6$). [b] Water contact angle measured at room temperature.

Results and Discussion

Polymerization of PVP on Silicon Wafer Surfaces by SI-ATRP. Scheme 1 illustrates the procedure used for the synthesis of PVP-grafted silicon wafer surfaces. First, the initiator-functionalized silicon wafers were prepared by treatment with APTES, followed by amidation with chloroacetyl chloride using TEA as the catalyst. Then surface-initiated ATRP of NVP at 30 °C was carried out on the initiator-modified surfaces.

For the SI-ATRP of NVP on silicon wafers, we chose CuCl/Me_6TATD as the catalyst system because of its high activity for the atom transfer radical polymerization of NVP.[33] Because of the difficulty in obtaining the molecular weight and molecular weight distribution of the grafted polymer,[38] we used the development of PVP thickness as a function of polymerization time to monitor the surface polymerization process. The data on layer thickness of PVP at different reaction times are summarized in Table 1 and Figure 1. In Table 1, the layer thickness before and after the initiator conjugation was 3.05 ± 0.21 and 3.25 ± 0.17 nm, respectively. The other thickness values for Si−PVP surfaces are the thickness of the PVP layer only at a different reaction time; i.e., the thickness of the initiator layer was deducted from the whole thickness. The result shows that the thickness of the PVP layer increases with an increase in reaction time, and the greatest thickness of 33.56 nm is obtained when the reaction

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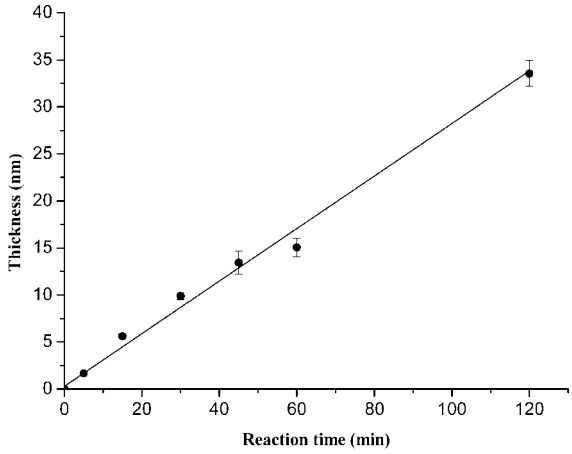


Figure 1. PVP thickness vs polymerization time at a 100/1/1 [NVP]/[CuCl]/[Me_6TATD] ratio.

Table 2. Atomic Concentrations of Unmodified and Modified Silicon Wafers from XPS Using 90° Takeoff Angles

surface	C (%)	O (%)	Si (%)	N (%)	Cl (%)
Si−OH	31.03	45.10	23.86	—	—
Si−APTES	55.22	25.81	10.78	8.20	—
Si−initiator	51.74	25.34	11.37	7.51	4.04
Si−PVP5	58.71	25.34	8.32	6.67	0.96

lasts ∼2 h. As the ellipsometry method provides averaged information about the polymer layer thickness, the combination of AFM and vacuum ultraviolet (VUV) radiation techniques was employed to characterize the localized thickness (Supporting Information). An average thickness of ∼33.14 nm for the Si−PVP120 surface was recorded with the AFM, which is in good agreement with the ellipsometry measurement (33.56 ± 1.36 nm). These results suggested that the ATRP grafting of NVP from the silicon wafer surfaces was a well-controlled process.

Charaterization of Silicon and Modified Silicon Surfaces. After the pretreatment with "piranha" solution and UV irradiation, the silicon substrates became strongly hydrophilic and exhibited extremely low water contact angles (−2°) (Table 1). After treatment of the hydroxy-terminated silicon surfaces with APTES and then chloroacetyl chloride, the water contact angles were 71° and 66°, respectively, indicating that the silicon substrates became hydrophobic. For the Si−PVP5 surface, the contact angle was 47°, representing a significant reduction in hydrophobicity compared with that of the Si−initiator surface. The contact angles decreased with an increase in PVP thickness, reaching a lower plateau of ∼24° for a thickness greater than 13.45 ± 1.24 nm (Si−PVP45). The higher water contact angles for Si−PVP5 and Si−PVP15 may be partially due to incomplete coverage of the silicon surface by PVP chains,[28] and the hydrophobic chloro end groups may also play an important role in this regard while the PVP chains are short. The decrease in water contact angle with an increase in PVP thickness suggested that coverage of the silicon surface increased with PVP thickness. In this case, the hydrophilic PVP dominates the surface property and eliminates the influence of silicon substrate and hydrophobic chloro end groups.

The chemical composition of the silicon wafer surfaces at various stages of surface modification was determined by XPS (see Table 2). Figure 2a shows a typical XPS survey spectrum for the Si−OH surface; the characteristic signals for silicon (Si2p at 103 eV and Si2s at 155 eV) and oxygen (O1s at 533 eV) are clearly detected. An additional carbon signal (C1s at 285 eV) is also detected. The latter feature can be attributed to the interference of unavoidable contamination of silicon wafer during analysis.[38] After treatment of the Si−OH surfaces with APTES, the

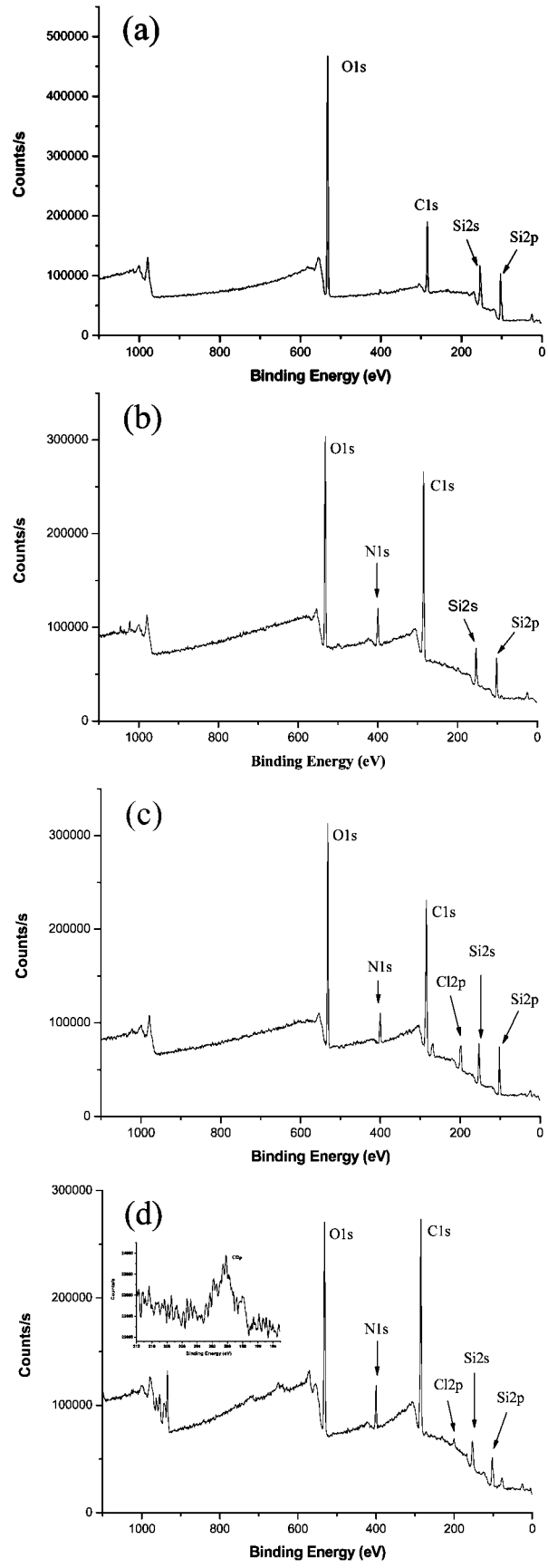
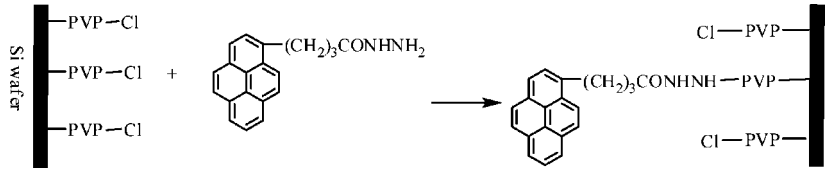


Figure 2. XPS survey spectra of (a) Si−OH, (b) Si−APTES, (c) Si−initiator, and (d) Si−PVP45 surfaces.

Scheme 2. Nucleophilic Substitution Reaction of the PVP-Grafted Surface with 1-Pyrenebutyric Acid Hydrazide



characteristic signals attributed to silicon, oxygen, carbon, and additional signals assigned to N1s at 400 eV indicate that APTES has been tethered covalently to the Si−OH surfaces (Figure 2b). However, the carbon signal is noticeably stronger compared with the Si−OH surface, due to the additional carbon in the APTES.

For the initiator-functionalized silicon wafers (Figure 2c), the characteristic signals attributed to silicon, carbon, oxygen, and nitrogen are again detected. Moreover, the appearance of weak signals assigned to chlorine (Cl2p at 200 eV) indicates that the α-chloroacetyl initiator was formed on the silicon wafer surface with a thickness of 3.25 ± 0.17 nm measured by ellipsometry (see Table 1). Turning to the PVP-grafted silicon wafers, we still observed the characteristic signals attributed to silicon and chlorine, but with lower intensity, together with more intense carbon C1s signals (see Figure 2d), compared to the initiator-functionalized surface. The results show that the polymer layer had covered the surface homogeneously. The high-resolution spectrum of Cl2p (inset, Figure 2d) of the Si−PVP surface consists of a spin−orbit-split doublet ($Cl2p_{3/2}$ and $Cl2p_{1/2}$, with the binding energy of the peak components at ∼200.2 and ∼201.8 eV, respectively), suggesting that the PVP-grafted silicon wafer surfaces have a chloro end group.[39]

Inherent in the mechanism of ATRP is the incorporation of the halogen at the chain ends. These alkyl halide end groups can be transformed into other functionalities by means of standard organic procedures such as nucleophilic displacement reactions.[40] To further confirm the presence of the alkyl halide groups on the end of grafted PVP brushes, we chose nucleophilic substitution by reacting PVP-grafted silicon wafer surfaces with 1-pyrenebutyric acid hydrazide in DMF. Scheme 2 illustrates the nucleophilic substitution reaction. After reaction, the appearance of fluorescence attributed to pyrene groups on the PVP-grafted silicon substrate clearly demonstrates that the nucleophilic substitution reaction took place between the chloro end group of PVP and 1-pyrenebutyric acid hydrazide (see Figure 3). In contrast, no fluorescence was detected on the control surfaces (Si−OH surfaces; see the inset of Figure 3). All these results of XPS and nucleophilic substitution reactions indicated that the PVP-grafted silicon wafer surfaces with a high functionality of chloro end group were obtained, and this observation supported the idea that the growth of PVP from the surface proceeded in a "living" manner.

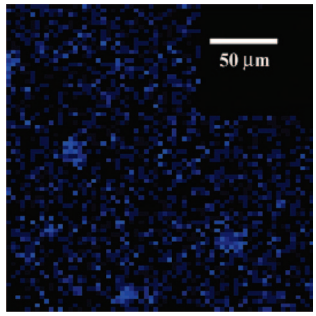


Figure 3. Fluorescent image of a Si−PVP30 surface after the nucleophilic substitution reaction obtained by CLSM (inset is the image of the control surface).

The surface morphologies of unmodified and modified silicon wafers at the dry state were studied by tapping mode AFM. Figure 4 shows the changes in topography of the silicon surfaces after surface grafting modification by SI-ATRP. The freshly prepared Si−OH surface has a rather uniform and smooth surface, with a root-mean-square surface roughness value (R_a) of only ∼0.18 nm (Figure 4a). The initiator-functionalized silicon surface remains molecularly uniform with an R_a value of ∼0.27 nm (Figure 4b). After surface graft polymerization of NVP via ATRP for 15 min, a PVP layer 5.62 nm in thickness was formed on the silicon surface. The R_a value increased to ∼0.68 nm (Figure 4c). For a longer polymerization time of 60 min, the ATRP-mediated graft polymerization gave rise to a denser coverage of PVP on the silicon surfaces and an increased roughness of 1.40 nm was observed (Figure 4d). The formation of the nanosized islands probably resulted from the nanoscale phase aggregation of the grafted polymer chain after drying.[41] The results presented above suggest that the ATRP graft polymerization of NVP proceeded uniformly on the silicon surface.

Protein Adsorption. The protein adsorption property of the grafted PVP surfaces was tested by measuring the level of adsorption of fibrinogen, HSA, and lysozyme which have significantly different size and charge characteristics. Figure 5 shows the adsorption of fibrinogen, HSA, and lysozyme from TBS buffer for both PVP-modified (Si−PVP60) and control (unmodified silicon, Si−OH) silicon wafers. It was found that the control surface adsorbed more of all three proteins than the Si−PVP60 surface. Levels of fibrinogen, HSA, and lysozyme adsorption on control surface were 1.215, 4.305, and 17.640 μmol/cm^2, respectively. For the Si−PVP60 surface, the levels of adsorption of fibrinogen, HSA, and lysozyme were 0.300, 0.302, and 3.403 μmol/cm^2, respectively; i.e., reduction in the level of protein adsorption, 75% for fibrinogen, 93% for HSA, and 81% for lysozyme, was achieved, compared to the unmodified silicon. These results show that a dense and thick PVP layer on a surface can efficiently reduce the quantity of adsorbed protein. This PVP-modified silicon surface via a SI-ATRP procedure adsorbed a much smaller amount of protein than previous PVP-grafted surfaces that were prepared by conventional free radical polymerization,[24,26] but the protein adsorption values are still quite higher than 0.3 ng/cm^2 which was obtained from several kinds of surfaces that resist fibrinogen adsorption.[42] It was found that platelet still adhere to the surfaces even though the level of protein adsorption was lower than 0.3 ng/cm^2. Additionally, other researchers found that hydrophilic surfaces with −50 ng/cm^2 can support significant cell adhesion.[43] Together, these studies suggest that a more in-depth evaluation of our surfaces is required to assess biocompatibility.

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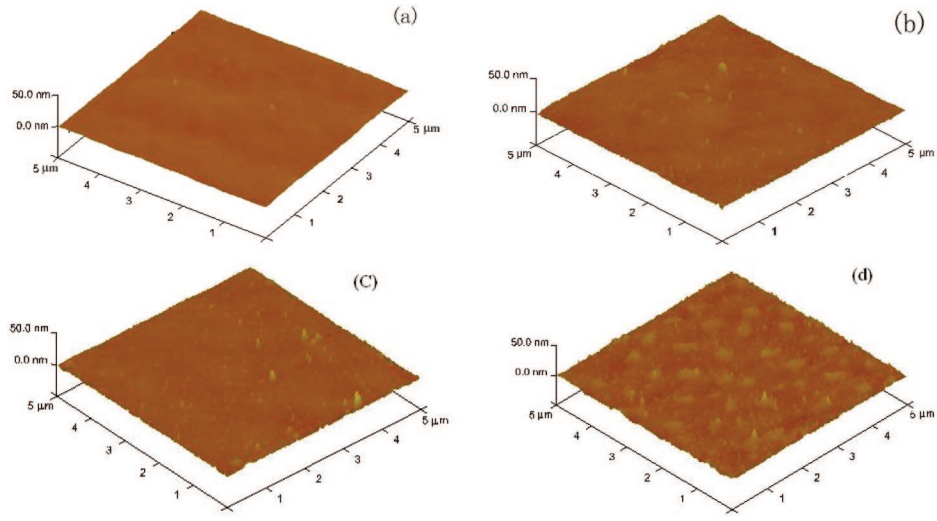

Figure 4. Tapping mode AFM images of (a) Si−OH, (b) Si−initiator, (c) Si−PVP15, and (d) Si−PVP60 surfaces.

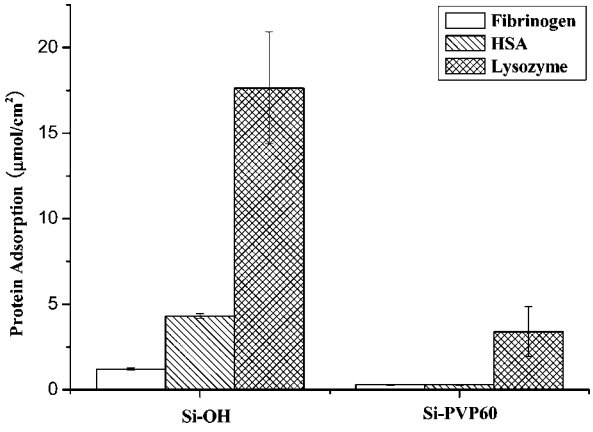

Figure 5. Fibrinogen, HSA, and lysozyme adsorption on Si−OH and Si−PVP60 surfaces in TBS for 3 h at room temperature. The protein concentration is 1.0 mg/mL. Data are means ± the standard error (n = 3).

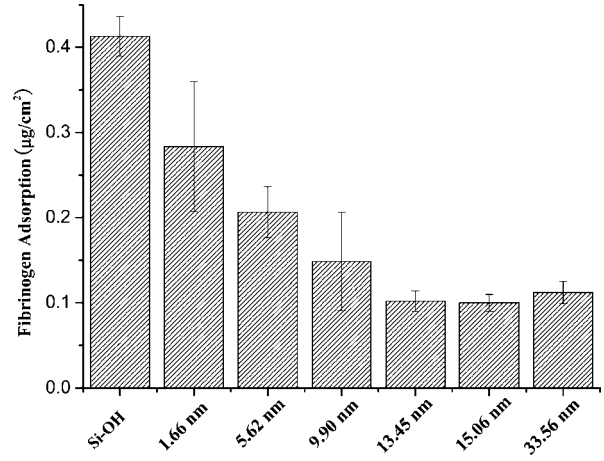

Figure 6. Adsorption of fibrinogen on PVP-grafted silicon surfaces with varying thicknesses in TBS buffer for 3 h at room temperature. The fibrinogen concentration was 1.0 mg/mL. Data are means ± the standard error (n = 3).

Both the graft density and the polymer chain length (film thickness) should be important factors for controlling the adsorption of proteins. Longer chains are needed to resist protein adsorption at a low graft density,[27] while shorter chains (more than one repeat unit) with a high graft density will be enough to resist proteins.[6] Since it is difficult to characterize the graft density in the SI-ATRP system directly, the graft density of PVP chains on the silicon surface was estimated using the thickness of the dry polymer layer and the molecular weight of PVP obtained in solution.[28] We calculated the graft density of PVP to be 0.26 chain/nm^2 (Supporting Information). It must be pointed out, however, that this graft density value is not very accurate as the chain length obtained on the surface might be different from that in solution. Fortunately, the thickness of PVP on the silicon surface can be easily obtained, so that the thickness of PVP layer was adopted as one major factor for examining its influence on protein adsorption. As shown in Figure 6, with a smaller PVP thickness (from 1.66 to 13.45 nm), the amount of fibrinogen adsorption decreased gradually with an increase in PVP thickness. The reductions in the level of adsorption versus the unmodified silicon ranged from 31 to 75%. The amount of adsorption reached a lower limit (~0.10 μg/cm^2) when the thickness of the PVP layer reached 13.45 nm. There were no significant differences among the surfaces when the layer thickness was >13.45 nm. These results show that the protein adsorption on PVP-grafted silicon surfaces is strongly dependent on the grafted PVP thickness. Feng et al.[27] also found that the level of protein adsorption decreased significantly when the poly(MPC) chain was short, while this change became slower when the chain length increased. This conclusion was reached at a higher graft density than 0.26 chains/nm^2, and no critical thickness was reported in their system. The layer thickness of 13.45 nm obtained in this work can be assumed as the critical thickness for fibrinogen adsorption. The influence of the density and thickness of grafted PVP on adsorption of different proteins is being examined.

Conclusions

In summary, well-controlled PVP-grafted silicon surfaces were prepared for the first time using SI-ATRP at 30 °C. The thickness of the grafted PVP layer was controlled by the reaction time. A set of surface samples with various thicknesses from 1.66 to 33.56 nm were prepared. The protein adsorption property of these PVP-grafted surfaces was evaluated by fibrinogen, HSA, and lysozyme adsorption experiments. The surfaces with large thicknesses showed a dramatic reduction in the level of adsorption of all the proteins. Moreover, the level of fibrinogen adsorption decreases gradually with an increase in PVP thickness. However, no significant difference in fibrinogen adsorption was found when

the PVP layer was thicker than the critical thickness of 13.45 nm. Considering the excellent water solubility and biocompatibility of PVP, grafted surfaces such as those reported here offer a new method for preparing surfaces with properties such as a reduced amount of protein adsorption.

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Supporting Information Available: Characterization of the localized thickness and calculation of PVP grafting density. This material is available free of charge via the Internet at http://pubs.acs.org.

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