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超分子聚合物刷PVP-Chol表面图案化及其力诱导的可逆结构转变

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摘要: 傅里叶变换红外(FT-IR)光谱、表面压力–分子面积(π – A)等温线和原子力显微镜(AFM)结果表明, 聚乙烯吡咯烷酮(PVP)与胆固醇分子(Chol)在溶液中和气/液界面上可通过氢键作用形成刷状的超分子聚合物PVP-Chol。当表面压力低于 $2.5 \text{ mN} \cdot \text{m}^{-1}$ 时, 界面膜主要由富含胆固醇的微区与PVP-Chol纳米纤维构成的微区共存。在相对较低的表面压力下($< 2.5 \text{ mN} \cdot \text{m}^{-1}$), PVP-Chol微区形貌随界面膜压缩发生有序的变化: 从最初的无规结构逐渐变为月牙形、心形和圆形结构; 表面压超过 $2.5 \text{ mN} \cdot \text{m}^{-1}$ 后, 圆形的PVP-Chol微区最终消失并转变为少量的纤维聚集结构。值得注意的是, 在 $1.0 \text{ mN} \cdot \text{m}^{-1}$ 之前, PVP-Chol纳米纤维高度随AFM成像过程中压电陶瓷外加电压的变化在 1.8 到 4.3 nm 之间出现了可逆转变, 表明扫描探针针尖与样品之间的作用力可诱导超分子聚合物刷PVP-Chol发生从圆柱状到椭圆柱状的可逆结构转变。

关键词: 氢键; 气/液界面; 微区; 纳米纤维; 结构演变; 针尖-样品作用力

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Surface Patterning and Force-Induced Reversible Structural Transformation of a PVP-Chol Supramolecular Polymer Brush

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Abstract: A supramolecular polymer brush composed of polyvinylpyrrolidone-cholesterol (PVP-Chol) was fabricated based on intermolecular hydrogen bonding between the ketone groups of the PVP moieties and the hydroxyl groups of the cholesterol, both in solution and at the air/water interface. These confirmations were confirmed by Fourier transform infrared (FT-IR) spectroscopy, surface pressure *versus* molecular area (π – A) isotherms, and atomic force microscopy (AFM). At surface pressures up to $2.5 \text{ mN} \cdot \text{m}^{-1}$, the interfacial film was composed of cholesterol-enriched regions and PVP-Chol nanofibril domains. Interestingly, oversurface pressures, the structure of the PVP-Chol-enriched domains evolved from initial irregular shapes, to crescent and heart-shaped zones, and then to circular shapes. This transition indicated the controllability of such domains during compression of a complex monolayer at relatively low surface pressures. Above $2.5 \text{ mN} \cdot \text{m}^{-1}$, the circular PVP-Chol domains disappeared and fibrous aggregates were formed at the air/water interface. The height of the comb-like PVP-Chol nanofibrils could be reversibly varied between approximately 4.3 and 1.8 nm by adjusting the voltage applied to piezoceramics during the AFM scanning process. These results demonstrate that the reversible structural transformation of the PVP-Chol complex from cylindrical to elliptic cylindrical is induced by the applied force of the AFM tip up to a value of $1.0 \text{ mN} \cdot \text{m}^{-1}$. These data provide important and useful information that improves our understanding of the relationship between the structures and performances of supramolecular polymer brushes, and should extend the range of applications of cholesterol-functionalized

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polymers fabricated *via* a simple mixing strategy.

Key Words: Hydrogen bonding; Air/water interface; Domain; Nanofibril; Structural evolution; Tip-sample force

1 Introduction

The decoration of linear polymer backbones with pendant functional groups can significantly affect performances of polymers, e.g., leading to the formation of helical polymers or polymeric nanoparticles^{1–3}. Such a polymer is composed of a linear main chain and pendants as side chains or groups connected onto the backbone through covalent or noncovalent bonds, similar to a representative synthetic or supramolecular comb-like polymer or cylindrical polymer brush⁴. Regular comb-like polymers exhibit the architecture of cylindrical brushes if the side chains are densely grafted, namely that every monomer unit of the main chain bears a side chain and if the main chain is much longer than the side chains. To date, comb-like polymers have attracted broader interest due to their great potential as facile building blocks for the creation of self-organizing materials. In this aspect, architectures and performances of molecular brushes can be tuned *via* varying the inherent nature, grafting density and length of side chains, and the property of the backbone as well^{5–7}. Here, each of these variables can affect their properties, but it is the nature of side chains, such as the steric hindrance and the intra- or intermolecular interactions between pendants, that has a significant effect on conformations and properties of brush-like polymers^{1,2}. As a result, so far various types of functional side chains had been utilized to decorate polymeric backbones.

Our own interest in supramolecular comb-like polymers, derived from noncovalent bonding, has focused on the relationship between their structures and performances. In our previous work, we fabricated a bottlebrush-like supramolecular polymer *via* a noncovalent bond linking, i.e., hydrogen bond, between polyvinylpyrrolidone (PVP) and *p*-dodecylphenol (PDP), and proposed a formation process of hierarchical helices from primary to quaternary structures⁸. In that case, PDP carries a hydroxyl group and a linear alkyl chain with 12 carbon atoms. In order to understand the role of the pendant size in properties of side-chain-substituted polymers, on one hand, we utilized a combination of PVP-cholesterol and a PDP-PVP polymer analogue, in which cholesterol (with a larger molecular size compared with PDP) was chosen as a pendant. This is due to two factors as follows: (i) cholesterol is a major component of mammalian plasma membranes, and its functionalized polymers play an important role in liposomal formulation of drugs for controlled and targeted delivery^{9–13}; and (ii) cholesterol was commonly selected as a substituted side group for modifying polymeric backbones and preparing polymer capsules¹⁴. On the other hand, PVP was still used as the polymeric backbone, because that it has been known that PVP is a macromolecular compound with widespread use in cosmetic, surfactant, food, pharmaceutical industry and other related industrial fields for various purpose^{15–22}; extensive toxicological studies, including carcinogenicity and teratogenicity studies, had indicated that PVP was biologically inert, i.e., non-toxic and biocompatible^{23–26}; and the molecular structure of PVP could be used as a simple protein model from a biological point of view. In this work, we sought to investigate assembling structures with uniquely physicochemical properties of cholesterol-modified supramolecular polymer (PVP-Chol) through hydrogen bonding.

Herein, a supramolecular polymer PVP-Chol was successfully prepared by means of the formation of hydrogen bonds between PVP and cholesterol and its self-assembling behavior was subsequently investigated at the air/water interface. Results show the presence of domains with a facile control over their surface patterns at relatively lower surface pressures. In particular, a reversible transformation in the height of PVP-Chol nanofibrils, induced by varying the magnitude of the tip-sample force, was observed in the process of atomic force microscopy (AFM) imaging. These results may give an insight into the influence of side groups on behaviors and architectures of cholesteryl-substituted polymers, which is favorable for providing important and useful information on understanding the relationship between structures and performances of supramolecular comb-like polymers.

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2 Experimental

2.1 Materials

Polyvinylpyrrolidone (PVP, $M_n = 24000$, $\geq 99\%$) and cholesterol (Chol, $\geq 99.7\%$) were purchased from Aladdin Chemistry Co. Ltd., which were used without further purification. Chloroform (CHCl_3 , AR, $\geq 99\%$) was purchased from Shanghai Chemistry Reagent Co. Ultrapure water ($18.2 \text{ M}\Omega \cdot \text{cm}$) was purified by a Milli-Q plus water purification system.

2.2 Monolayer preparation

The surface pressure *versus* molecular area (π - A) isotherms at the air-water interface and Langmuir-Blodgett (LB) depositions onto a solid substrate (mica) were conducted using a LB trough (KSV NIMA medium trough, Helsinki, Finland). 60 μL of pure PVP, cholesterol and their mixed solution with the molar ratio 1 : 1 in CHCl_3 were uniformly deposited and distributed onto the water surface, respectively, and then left to evaporate a period of about 15 min. Herein, the concentration of all samples was $5.0 \times 10^{-4} \text{ mol} \cdot \text{L}^{-1}$. Simultaneously, the subphase temperature, which was detected by an attached temperature probe, was maintained at $(20.0 \pm 0.1)^\circ\text{C}$ *via* circulating water from a thermostat circulated through the base plate of the trough. π - A isotherms were recorded as compressing the barriers at a constant speed of $10 \text{ mm} \cdot \text{min}^{-1}$.

The LB films of PVP-Chol were deposited by a vertical dipping method at different surface pressures and the deposition rate was

10 mm·min⁻¹. One-layer LB films were transferred onto freshly cleaved atomically flat mica surfaces, then dried in a desiccator at room temperature for subsequent atomic force microscope (AFM) characterization. The transfer ratio of the upstroke is about 0.9–1.05.

2.3 Fourier transform infrared spectroscopy

Fourier transform infrared (FT-IR) spectra were carried out on a Nicolet iS10 infrared spectroscopy (US) using the liquid film method. The dilute solution of samples (5.0×10^{-4} mol·L⁻¹ in repeat unit for PVP and cholesterol) was spread onto the surface of CaF₂ plate and dried in the infrared drying oven (WS70-1, Shanghai Jin Ping) over 2 min. The spectra with a resolution of 4 cm⁻¹ were obtained at room temperature.

2.4 Atomic force microscopy

The AFM topographic images were carried out on a Solver P47-PRO (NT-MDT Co., Moscow, Russia) microscope in tapping mode with a triangular microfabricated cantilever (Mikro Masch Co., Estonia) with a length of (130 ± 5) μm, a Si pyramidal tip with Au reflective side and a spring constant of ~ 5.1 N·m⁻¹. A resonance frequency in the range of 55–500 kHz was used and resonance peaks in the frequency response of the cantilever typically at 110–180 kHz were chosen for the tapping mode oscillation. The curvature radius of the tip was less than 10 nm. The experiments were operated under ambient conditions (temperature: ~ 23 °C, humidity: $\sim 45\%$). Prior to AFM measurements, LB films were dried in a desiccator at least for 24 h. It should be

noted that the tip-sample force is nonlinearly increased with the increment of the applied voltage of piezoceramics within limits.

2.5 Transmission electron microscopy

One-layer of PVP and cholesterol mixture was deposited onto pure carbon-covered Cu grids (400 meshes) by a horizontal dipping method (Langmuir-Schaefer (LS) method) at the surface pressure of 1.0 mN·m⁻¹. LS film was further stained by exposing them to the phosphotungstic acid solution (concentration (mass fraction) 1%, pH 4–6). Transmission electron microscopy (TEM) image was acquired on a JEOL JEM-2100 (Japan) instrument operating at 200 kV.

3 Results and discussion

3.1 Hydrogen bonding between PVP and cholesterol

FT-IR spectra were carried out to illustrate the formation of hydrogen bond between PVP and cholesterol in CHCl₃ with 1 : 1 molar ratio of pyrrolidone group to cholesterol, given in Fig.1A. In Fig.1A, pure PVP has a characteristic and relatively broad and strong infrared absorption peak at 1672 cm⁻¹, ascribed to the C=O stretching vibration, but as for the spectrum of cholesterol, there is only a weak absorption peak around 1669 cm⁻¹, attributed to an intra-annular C=C double bond in the basic ring of the sterol. Nevertheless, after PVP and cholesterol were mixed at 1 : 1 molar ratio, this strong peak assigned to C=O remarkably shifted to 1668 cm⁻¹. Such shift indicates that the weak interaction between the ketone group of PVP and cholesterol occurs indeed²⁷. On the

Fig.1 (A) FT-IR spectra of PVP, Chol, and PVP-Chol complex in CHCl₃ at 5.0×10^{-4} mol·L⁻¹; (B) π -A isotherms of PVP, Chol, and PVP-Chol complex at 5.0×10^{-4} mol·L⁻¹ at 20 °C; (C) schematic illustration of supramolecular assembling structure of PVP-Chol and the hydrogen bonding mode

In B, as for PVP-Chol, the molecular area is based on the average molecular weight of PVP repeating unit and cholesterol. The inset is the amplification of π -A isotherm of PVP-Chol complex at the lower surface pressures of (a) 0 mN·m⁻¹, (b) 0.2 mN·m⁻¹, (c) 0.5 mN·m⁻¹, (d) 1.0 mN·m⁻¹, (e) 2.0 mN·m⁻¹, and (f) 5.0 mN·m⁻¹.

other hand, the C—O stretching vibration of pure cholesterol appears at 1055 cm^{-1} and PVP has no any characteristic absorption peaks in the range of 1000 to 1100 cm^{-1} , whereas a pronounced blue shift of the C—O stretching vibration is observed at 1066 cm^{-1} when we detected a stoichiometric PVP-cholesterol mixture. This implies that the weak interaction is relevant to 3β -hydroxyl group of cholesterol. Taken together, both results indicate that the weak interaction between the ketone group of PVP and the hydroxyl group of cholesterol does take place, namely that a hydrogen bond is formed as schematically shown in Fig.1C, in which the proposed structure of PVP-Chol complex is a brush-like nanofibril when the hydroxyl group of cholesterol bonds to the ketone group of PVP. In general, the carbonyl is a proton acceptor, and easily bonds with the hydroxylated species. The formation of the hydrogen bond is in great agreement with those results reported in previous literature^{8,18,27–30}. For instance, Karavas *et al.*¹⁸ found that PVP could improve dissolution of drug, such as felodipine (a poorly soluble antihypertensive drug), *via* the intermolecular hydrogen bonding. In our previous work, a supramolecular polymer, PVP-PDP, was also fabricated through the formation of the intermolecular hydrogen bond between PVP and PDP⁸. Therefore, FT-IR spectra suggested that the supramolecular polymer PVP-Chol complex had been formed when PVP and cholesterol mixed in CHCl_3 , which was further verified by the Langmuir isotherms in Fig.1B.

For the purpose of giving an in-depth understanding of the weak interaction between PVP and cholesterol, Langmuir monolayers and LB films of PVP, cholesterol and their mixture were separately examined. This is due to three factors as following: (i) it was difficult to directly observe the subtle structure of the PVP-Chol complex on the ultrathin film through the general method, such as spin-coating films (Fig.S1 (Supporting Information)); (ii) interfacial assembly through Langmuir monolayer, which can provide an ideal two-dimensional plane with a confined environment at the air/water interface, as well as LB technique, affords excellent opportunities to construct the exquisite structure, and easily to exert molecular level control over the structure of organic thin films; (iii) the complexation of PVP with cholesterol would improve the hydrophobicity of PVP-Chol, endowing such complex an amenability to being constrained at the air/water interface, and hence it is feasible to obtain the structural information of PVP-Chol with the aid of LB method.

We monitored the surface pressure *versus* molecular area (π – A) isotherms of pure PVP, cholesterol and PVP-Chol complex over ultrapure water, respectively, given in Fig.1B. As a general method, the “additivity rule” of such mean area was usually utilized to address a nonideal interaction, either attraction or repulsion, between two components in mixed monolayers³¹. Taken the solubility of PVP into consideration, only the “liftoff” areas among three systems, i.e., PVP, cholesterol, and PVP-Chol, were used to empirically illustrate the intermolecular interaction between PVP and cholesterol. In Fig.1B, the “liftoff” area of PVP-Chol complex is ca 0.51 nm^2 , while that of the individual PVP and

cholesterol is 0.83 and 0.54 nm^2 , respectively. The former value is much smaller than half the sum of the last two values. This suggests that an intermolecular attractive association does take place between PVP and cholesterol.

It should be noted that the primary fraction of PVP-Chol curve resembles the cholesterol curve, especially when the surface pressure is up to $3.0\text{ mN}\cdot\text{m}^{-1}$, and the collapse pressure of PVP-Chol is comparable to that of cholesterol. The inset of Fig.1B is the amplification of π – A isotherm of PVP-Chol complex at the lower surface pressures. The isotherm of PVP-Chol is slowly upward at the lower surface pressures and has a sharper slope after $3.0\text{ mN}\cdot\text{m}^{-1}$. In particular, there appears an inflection point around $2.5\text{ mN}\cdot\text{m}^{-1}$ followed by a relatively horizontal plateau in the isotherm, indicative of the transition in the structure of PVP-Chol interfacial films³². Here, several points ($0, 0.2, 0.5, 1.0, 2.0, 5.0\text{ mN}\cdot\text{m}^{-1}$) were chosen to detect the changes in the structure and properties of the interfacial film. Meanwhile, given that the mean molecular area of PVP-Chol complex is calculated on the molecular weight of cholesterol (Fig.S2 (Supporting Information)), there is only a slight difference between PVP-Chol and cholesterol curves. Both results imply that the interfacial nature of PVP-Chol is to some extent identical to that of cholesterol, which will be discussed in the following section.

3.2 Evolution in surface structure of domains and fibril aggregates

AFM technique, combined with the LB method, was employed to visualize the structural transition of PVP-Chol interfacial films according to the inset of Fig.1B. The representatively morphological change of one-layer PVP-Chol LB films with the increasing surface pressures, corresponding to a – f (the inset of Fig.1B), are shown in Fig.2(A–F), while other typical examples in Fig.S3 (Supporting Information). As control experiments, the change in the morphology of PVP and cholesterol LB films with the increasing surface pressure was given in Figs.S4 and S5 (Supporting Information), respectively.

In Fig.2A, bright and irregular patches or domains distributed randomly were observed at the onset of the compressing process ($0\text{ mN}\cdot\text{m}^{-1}$). These domains as well as the darker region suggested the coexistence of two different structures in the complex film. With the surface pressure gradually increasing up to $1.0\text{ mN}\cdot\text{m}^{-1}$, the brighter domains become more and more ordered, with a structural change from the initial irregular shape at $0\text{ mN}\cdot\text{m}^{-1}$ (Fig.2A), to crescent at $0.2\text{ mN}\cdot\text{m}^{-1}$ (Fig.2B), and then to heart-shaped and circular shapes at 0.5 (Fig.2C) and $1.0\text{ mN}\cdot\text{m}^{-1}$ (Fig.2D), respectively. The morphological change in Fig.2 and Fig. S3 indicates that such surface patterning is tightly dependent on the surface pressure in the process of the compression. By contrast, as for PVP-PDP system with the ability to form a comb-like analogue, except for the fibrils with hierarchical helices, we did not observe such change in the structures with surface pressures under the same conditions⁸. This means that cholesterol with a larger molecular size compared with PDP plays a critical role in the formation of such domains.

Fig.2 AFM images of PVP-Chol LB films at the lower surface pressures of (A) 0, (B) 0.2, (C) 0.5, (D) 1.0, (E) 2.0, and (F) 5.0 $\text{mN} \cdot \text{m}^{-1}$

F is height image, while the others are phase images. The sizes of A, B, C, and D are $10 \mu\text{m} \times 10 \mu\text{m}$, while those of E and F are $30 \mu\text{m} \times 30 \mu\text{m}$ and $3 \mu\text{m} \times 3 \mu\text{m}$, respectively. Here, the average height of fibrous aggregates in F is $\sim 2 \text{ nm}$.

In Fig.1B, a striking feature is the presence of the inflection point around $2.5 \text{ mN} \cdot \text{m}^{-1}$ followed by a relatively horizontal plateau. In general, plateau regions in isotherms are usually associated with the formation of a new phase for conventional amphiphiles, such as lipids and surfactants. Many researchers had so far investigated the properties of such amphiphiles at the air/water interface and suggested that their monolayers exhibit a liquid expanded to a liquid condensed phase transition in the monolayer^{33–35}. Here, an inflection point followed by a relative plateau region might thus mean the presence of the structural transition analogous to that of conventional amphiphiles. Therefore, two points, before ($2.0 \text{ mN} \cdot \text{m}^{-1}$, Fig.2E) and after ($5.0 \text{ mN} \cdot \text{m}^{-1}$, Fig.2F and Fig.S6 (Supporting Information)) the plateau, were selected in this work to monitor the change in film structures during the compression. As expected, a transition from circular shape (before the plateau) to fiber structure (after the plateau) was observed as the compression proceeds. Besides, the morphology of PVP-Chol at $2.5 \text{ mN} \cdot \text{m}^{-1}$ (the inflection point) was also given in Fig.S7 (Supporting Information). The larger circular domains became loose and nanofibers were apparent. Nevertheless, we found the quantity of such fibrous aggregates did not accumulate with the increasing surface pressures, on the basis of the evaluation of more than 20 independent AFM images with a scanning size of $30 \mu\text{m} \times 30 \mu\text{m}$ at different surface pressures ($5.0, 10.0, 15.0$, and $25.0 \text{ mN} \cdot \text{m}^{-1}$). On the contrary, this value (the number of fibrous aggregates) had a tendency to reduce with the surface pressures. A representative example at $10.0 \text{ mN} \cdot \text{m}^{-1}$ is given in Fig.S8 (Supporting Information), a careful analysis indicates that

a substantial fraction of area, i.e., approximate 90%, is occupied by the featureless structure. Particularly, such featureless structure is identical to that of cholesterol at high surface pressures (Fig.S5), implying that the fibrous aggregates are expressed from the interfacial film as the surface pressure rises, meanwhile cholesterol is the primary component in this complex monolayer at the air/water interface.

3.3 Microstructure of the domains

To better explore the subtle structure of the brighter domains as well as the darker regions at the relatively low surface pressures, Fig.3 shows the locally amplified images of high-resolution AFM and TEM. Here, the AFM images were obtained *via* a larger tip-sample force.

At the beginning of the compression, the coexistence of the brighter domains and darker regions in Fig.3A indicates that there are two different structures. Remarkably, the darker region is heterogeneous, as shown in Fig.3B, which is analogous to that of cholesterol at lower surface pressure. This indicates that the dominant component of such region is cholesterol. Interestingly, the close inspection illustrates that such brighter domains are not homogeneous, and instead are made up of a large number of nanofibrils with an average height of ca 2 nm and a width of more than 10 nm, respectively, as shown in Fig.3C. Such fibrous structure is significantly different from that of individual components, i.e., PVP and cholesterol, at the air/water interface (Figs. S4 and S5), indicative of the formation of PVP-Chol complex. Generally, pyrrolidone groups can stochastically point out around the PVP backbone, so that the hydrogen bond between PVP and

Fig.3 AFM height images with the coexistence of two different structures in PVP-cholesterol mixture at 0 mN·m⁻¹ (A, B, C); TEM image with fibrous structure at 1.0 mN·m⁻¹ (D); illustration of different associating density of cholesterol (1: high-density; 2: low-density) at the air/water interface (E)

(A) the light domains and the adjacent and darker regions (10 $\mu\text{m} \times 10 \mu\text{m}$); (B) the amplified image of the darker region of A (1.5 $\mu\text{m} \times 1.5 \mu\text{m}$, the height scale: 2 nm); (C) the amplified image of the light domain with the structure of nanofibrils (2 $\mu\text{m} \times 2 \mu\text{m}$, the height scale: 3 nm); (D) the structure of nanofibrils acquired via LS method (the scale bar: 100 nm)

cholesterol may afford a comb-like structure of this PVP-Chol complex. Nevertheless, taken the molecular length of cholesterol (ca 1.8 nm, given a fully extended configuration, including a rigid moiety of the steroid and a branched hydrocarbon tail in the cholesterol) and ca 1 nm geometry of PVP into consideration, the inherent diameter of such complex should be 4.0–5.0 nm, which is inconsistent with the mean height of these nanofibrils. This is a likely consequence of the different associating density of cholesterol: (i) high-density but with a deformation in structures of PVP-Chol cylindrical polymer brush induced by the relatively larger tip-sample force during AFM scanning (Fig.3E-1) and (ii) low-density with or without this deformation (Fig.3E-2). Secondly, the width of these nanofibrils is more than 10 nm in the light of tip broadening correction calculation, suggesting that lateral aggregation occurs among them. The similar nanofibrils can also be

observed at other lower surface pressures by the high-resolution AFM (Fig.S9 (Supporting Information)) or TEM. Fig.3D gives a TEM result, where the average width of nanofibrils is ca 10 nm yet, comparable to the value obtained by AFM.

It should be noted that such brighter domains are abundant and can almost be seen everywhere at the lower surface pressures, suggesting that the quantity of nanofibrils of PVP-Chol complex is considerable. Although PVP is a soluble polymer, the association of itself with cholesterol may endow PVP-Chol complexes a more hydrophobic nature, favoring the formation of the interfacial film. However, after the inflection point, as described above, this value of fibrous aggregates becomes fewer and fewer with the increasing surface pressures. This implies that a large fraction of PVP in PVP-Chol complexes should dissolve into the subphase, which is a likely result of the competitive effect between the af-

finity of PVP with water and the hydrogen bond with cholesterol. As for cholesterol, the polar 3β -hydroxyl group provides a weakly amphiphilic nature, so that it can “stand” at the air/water interface when compressed up to a relatively high surface pressure. In contrast, PVP-Chol complex was also confined on the surface, PVP is liable to dissolve into the subphase due to its affinity with water and the reduction in the space as the compression proceeds.

3.4 Force-induced reversible structure transformation of PVP-Chol

In our previous work, AFM images were obtained in tapping mode (TM) with a relative soft tip^{8,36}. This operation mode enables one to minimize lateral forces and to investigate soft materials, especially for the soft elastomers without plastic deformation³⁷. In TM AFM imaging, the cantilever is oscillated at (or close to) its resonance frequency of several tens of kilohertz with an amplitude. Such amplitude represents the applied voltage of alternating current on the piezoceramics at the end of the cantilever beam, which is proportional to the oscillation amplitude of the cantilever. The larger oscillation amplitude provides a higher free magnitude,

meaning that the scanning tip has a higher energy. Thus, on the premise of definitely imaging, if other regulation parameters (gain, feedback, setpoint, etc.) are constant, the higher free magnitude, the greater force between the AFM scanning tip and the specimen surface within a certain range. It should be noted that the relationship of the free magnitude and the tip-sample force is non-linear. In other words, the tip-sample force presents nonlinear increase with the increment of the applied voltage of amplitude^{38–40}.

Fig.4 gives the variation in the height of domains (i.e., PVP-Chol nanofibrils) with the applied voltages of piezoceramics at the surface pressure of $0.5 \text{ mN} \cdot \text{m}^{-1}$. In Fig.4A, the inset is the locally amplified phase image, indicated by the blue circle, suggesting that such brighter domains consist of the fibers. When we tuned the applied voltage, an interesting and striking phenomenon was observed, namely that one can find a reversible variation in the height of these domains with the external force of AFM tips. If the applied voltage was set at 0.23 V, as shown in Fig.4A, the average height of PVP-Chol nanofibrils was ca 1.8 nm. This value almost remained the same when the voltage was decreased down to 0.18

Fig.4 Variation in the height of PVP-Chol nanofibrils at $0.5 \text{ mN} \cdot \text{m}^{-1}$ with different applied voltages of piezoceramics (A) 0.23, (B) 0.18, (C) 0.11, (D) 0.04, (E) 0.03, and (F) 0.18 V

The profile image is corresponding to the line of AFM height image. The inset of A is the locally amplified phase image ($2 \mu\text{m} \times 2 \mu\text{m}$).

(G) the illustration of reversible structure transformation with the applied force of AFM tips

V (Fig.4B), while it revealed an apparent increase from ca 2.3 nm at 0.11 V (Fig.4C) to ca 4.3 nm at 0.04 (Fig.4D) or 0.03 V (Fig.4E). Here, if we continued to reduce the applied voltage, we could not obtain any clear AFM images. Intriguingly, given that the applied voltage was reset at 0.18 or 0.23 V, the height can return to the initial value, ca 1.8 nm (Fig.4F). After more than 10 recycling measurements, this value almost kept constant, meaning that this process is reproducible. As a control system, a highly oriented pyrolytic graphite (HOPG) sample was utilized to verify that this process is convincing under the same experimental conditions, as shown in Fig.S10 (Supporting Information). As expected, there is not any variation in the height of the HOPG steps with the tip-sample force. The difference between HOPG and PVP-Chol complex may be attributed to that HOPG belongs to hard material relative to such supramolecular polymer.

The variation in the height of PVP-Chol nanofibrils with the applied voltages suggests that the inherent height of these nanofibrils was not only 2 nm, as shown in Figs.3 and 4A or 4B, and instead ca 4.0 nm, which is basically consistent with the molecular diameter of PVP-Chol complex (4.0–5.0 nm, according to the molecular length of cholesterol and the geometry of PVP). Such height variation of comb-like polymers has not been reported in literature to our knowledge. Generally, the height of a soft film with a relatively higher thickness varies with the penetration depth of the AFM tip into the film⁴¹. Apparently, our results are different from this case because that the interfacial film in our study consists of one-layer fibrils lain horizontally. Here, the height variation of one-layer PVP-Chol LB film should be a likely result of the structure transformation of PVP-Chol supramolecular polymer brush induced by the change in the magnitude of tip-sample force. However, it must be pointed out that for these fibrous structures, the force-induced reversible height variation only appears at lower surface pressures. By contrast, at relatively higher surface pressures (exceeding $1.0 \text{ mN} \cdot \text{m}^{-1}$), the height of the nanofibrils remains the same, ca 2.0 nm (Fig.2(E, F)), no matter whether the applied voltage is high or low. The latter case may result from that the bonding content of cholesterol molecules onto PVP-Chol polymer brush decreases with the increasing surface pressures. In this case, the sparse association of cholesterol molecules onto PVP main chains could not form an actual and tight comb-like structure, leading to a ca 2.0 nm in the geometry of PVP and cholesterol complex. This is consistent with the morphological change with the surface pressure in Fig.2.

Taken together, these results suggest that PVP and cholesterol can form a comb-like PVP-Chol supramolecular polymer brush at the air/water interface in a high-density associating mode at lower surface pressures or in chloroform solution. Apparently, such brushes result in the formation of domains with different morphologies. However, compressing leads to the dissociation of cholesterol from PVP main chains due to the reduction in space and the competition between the hydrogen bond of PVP with cholesterol and the affinity of PVP with water. Therefore, the high-density association can result in a fibril with a diameter of ca 4.0

nm, an actual comb-like structure, while the low-density association is a complex with only 2.0 nm in the height of fibril. In this work, we propose a model about the reversible structure transformation of PVP-Chol complex at the lower surface pressures, as shown in Fig.4G. Interestingly, a cylindrical supramolecular polymer brush (Fig.4G-1) can be compressed into having an elliptic cylinder structure (Fig.4G-2) by a relatively larger tip-sample force at the vertical direction, and recover to its initial state when the applied tip-sample force is decreased.

4 Conclusions

The supramolecular polymer brush PVP-Chol was fabricated through the formation of the intermolecular hydrogen bond between PVP and cholesterol at the air/water interface or in chloroform solution, which was systematically investigated by FT-IR spectra, π -A isotherm and AFM images. The domains derived from PVP-Chol nanofibrils and darker regions coexist at the air/water interface. Interestingly, such domains undergo a structural evolution from initially irregular to crescent, heart-shaped and circular shapes in the process of the compression at relatively lower surface pressures, and finally these circular domains disappear and a small number of fibrous aggregates are formed after $2.5 \text{ mN} \cdot \text{m}^{-1}$. AFM results suggest that cylindrical PVP-Chol polymer brush with a diameter of $\sim 4.0 \text{ nm}$ can gradually transform into those fibrils with the height of only $\sim 2.0 \text{ nm}$ during the compression due to the dissolving of PVP into the subphase and the dissociation of cholesterol from PVP main chains. Especially, the reversible height variation of comb-like PVP-Chol nanofibrils between ~ 1.8 and $\sim 4.3 \text{ nm}$ with varying applied voltages of amplitude in the AFM scanning process indicates that the reversible structure transformation of PVP-Chol complex was induced by the applied force of AFM tip at relatively lower surface pressures. Such surface patterning with interfacial pressures and force-induced variation in the height of supramolecular polymer may help us understand the relationship between structures and performances and tune behaviors of cholesterol-functionalized polymers.

Supporting Information: available free of charge via the internet at <http://www.whxb.pku.edu.cn>.

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超分子聚合物刷 PVP-Chol 表面图案化及其力诱导的可逆结构转变

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Surface Patterning and Force-Induced Reversible Structural Transformation of a PVP-Chol Supramolecular Polymer Brush

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Fig.S1 AFM height images of the spin-coating film of PVP-Chol complex at 5.0×10^{-4} mol L⁻¹ in CHCl₃

(A) random coil structure; (B) spherical particle. The sizes of A and B are $10 \mu\text{m} \times 10 \mu\text{m}$.

In Fig.S1, the morphology of stoichiometric PVP-Chol complex in CHCl₃ presented two structures: one is the random coil structure, the number of such aggregates is ~30% on the basis of an evaluation of more than 30 independent AFM images with a scanning size of $10 \mu\text{m} \times 10 \mu\text{m}$; and the other is the spherical particle, the number is ~70%. The original structure of supramolecular polymer brush PVP-Chol can not obtain by spin-coating method.

Fig.S2 π -A isotherms of PVP, Chol, and PVP-Chol complex at 5.0×10^{-4} mol L⁻¹ with the subphase temperature of 20 °C

The molecular area of PVP-Chol is based only on the molecular weight of cholesterol.

In Fig.S2, the limiting area of PVP-Chol, by extrapolating the steepest portion of the π -A curves to $\pi = 0$, is $\sim 0.41 \text{ nm}^2$, close to the area of pure cholesterol (0.44 nm^2), illustrating that the majority of PVP macromolecules enters into the subphase during

increasing the operating pressure, and only a small quantity of PVP-Chol reserves and maybe continue to assemble with cholesterol molecules at the higher surface pressures.

Fig.S3 Morphology of phase separation of PVP-Chol complex at the lower surface pressures of (A) 0, (B) 0.2, (C) 0.5, (D) 0.7, (E) 1 and (F) 2 mN m^{-1}

A, C and E are height images, while the others are phase images. The sizes of all images are $30 \mu\text{m} \times 30 \mu\text{m}$.

In Fig.S3, with the surface pressure increasing, the brighter domains become more ordered, from the initially irregular shape at 0 mN m^{-1} (Fig.S3A) to crescent shape at 0.2 mN m^{-1} (panel B), then heart-shaped (panel C, 0.5 mN m^{-1} ; panel D, 0.7 mN m^{-1}) and circular (panel E, 1.0 mN m^{-1}) configuration. Subsequently, the circular domains become larger at 2.0 mN m^{-1} (panel F). This indicates that such surface patterning is tightly connected with the surface pressure at the air/water interface.

Fig.S4 AFM images of pure PVP at low surface pressures of (A) 0, (B) 2.0 mN m^{-1}

The AFM height images of PVP monolayer at low surface pressures show dense and random fibrous structure.

Fig.S5 AFM height images of pure cholesterol at different surface pressures of (A) 0, (B) 1.0 , (C) 5.0 and (D) 10.0 mN m^{-1}

The sizes of all images are $3 \mu\text{m} \times 3 \mu\text{m}$.

The surface topographies of cholesterol monolayer show a negligible change after the 1.0 mN m^{-1} with the dynamic compression of barriers.

Fig.S6 AFM height images of PVP-Chol at 5.0 mN m⁻¹
The sizes of A and B are 20 μm $\times$ 20 μm and 3 μm $\times$ 3 μm , respectively.

Fig.S7 AFM height images of PVP-Chol at 2.5 mN m⁻¹
The sizes of A and B are 10 μm $\times$ 10 μm and 5 μm $\times$ 5 μm , respectively.

Fig.S8 AFM height images of PVP-Chol complex at 10.0 mN m⁻¹
(A), the representative and amorphous structure of pure cholesterol in most areas of LB films (ca. 90%). Panel 2 (3 μm $\times$ 3 μm) is the zoomed in image of panel 1 (30 μm $\times$ 30 μm). (B), the coexistence of two different structures: one is the dominant structure of cholesterol (panel 2, 3 μm $\times$ 3 μm), the other is fibrous aggregates of PVP-Chol (panel 3, 3 μm $\times$ 3 μm) which accounts for ~10% of the whole area of LB films. Panels 2 and 3 are the amplified images of panel 1 (30 μm $\times$

30 μm).

In Fig.S8, the morphology of PVP-Chol complex at 10.0 mN m^{-1} shows the coexistence of two different structures. One is the featureless structure accounts for a substantial part of region, i.e., approximate 90%, according to the evaluation of more than 20 independent AFM images with a scanning size of $30 \mu\text{m} \times 30 \mu\text{m}$. Here, the whole topography of the three AFM images is this structure in four images (scanning size: $30 \mu\text{m} \times 30 \mu\text{m}$), as shown in Fig.S8A. Such featureless structure is identical to that of cholesterol at high surface pressures (Fig.S4). The other is fibrous aggregates of PVP-Chol which accounts for $\sim 10\%$ of the whole area of LB films in more than 20 independent AFM images (panel 3 of Fig.S8B).

Fig.S8 Fibrous structure of the brighter domains (A and B) 0 mN m^{-1} ; (C and D) 0.2 mN m^{-1} ; (E and F) 1.0 mN m^{-1}

B, D and F are the phase image of A, C and E, respectively. The sizes of all images are $3 \mu\text{m} \times 3 \mu\text{m}$.

In Fig.S9, the fibrous structure of the brighter domains can also be acquired. From

the morphology of panel C, one can see the structure of cholesterol around the crescent domain.

Fig.S10 Unchangeable height of highly oriented pyrolytic graphite (HOPG) with different set voltages

(A) 0.18, (B) 0.11, (C) 0.04, (D) 0.03 V