Effect of Proline and its Derivatives on the Properties of Silk Fibroin Microneedles

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Abstract

With its good biocompatibility and excellent mechanical properties, silk fibroin microneedles can transport drugs to the body fluid circulation system and then act on the affected area, so as to replace intravenous injection and oral administration, and achieve the purpose of treating diseases. In the process of processing and use, silk fibroin microneedles are non-toxic, harmless, pollution-free and biodegradable to human body and environment. Therefore, the application prospect and application range of silk fibroin microneedles are very wide.

In this paper, the effects of proline and its derivatives prolinamide and hydroxyproline on the performance of silk fibroin microneedles were studied on the basis of the previous experiments of constructing microneedles to carry drugs. The composite silk fibroin microneedles were obtained by pouring the amino acid/silk fibroin mass ratio of 0/10, 1/10, 2/10, 3/10 and 4/10 into a polydimethylsiloxane mold, after vacuum defoaming and drying. The length of the microneedles was about 600 μm. The aggregation structure of amino acid/silk fibroin microneedles was measured by X-ray diffraction (XRD), Fourier transform infrared spectroscopy (FTIR) and Raman Scattering Spectroscopy. The mechanical properties of the microneedles were measured by texture analyzer. The results showed that: (1) The silk fibroin microneedles prepared by adding proline and its derivatives had predominately Silk I crystal structure; (2) When the mass ratio of proline and its derivatives to silk fibroin reached 2/10, it had a higher swelling degree and a lower dissolution rate; (3) The silk fibroin microneedles prepared by proline and its derivatives have good mechanical properties. The following conclusion was drawn: with the addition of proline and its derivatives, silk fibroin microneedles with higher swelling degree and lower dissolution rate can be obtained. The crystal structure of Silk I is formed inside the microneedles, which has good penetration and fracture properties. It is expected that the microneedles can be used as swelling microneedles for drug transdermal delivery.

Keywords: Silk Fibroin; Microneedle; Proline; Silk I Crystalline Structure

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1 Introduction

In the past few decades, transdermal delivery has been an attractive route for drug delivery [1-3]. Skin is the largest organ in the human body. It receives one-third of the blood supply of the entire body and was not used as a drug delivery route until the end of the 20th century. The mechanism of microneedle transdermal drug delivery is to use microneedles to penetrate the tightly arranged stratum corneum of the skin [4], and directly deliver the drug to the dermis. Microneedle penetration strengthens the drug delivery channel into the skin, and the microneedle will not penetrate too deep, which can protect the human skin. The administration process is almost painless, which can improve the compliance of patients. In addition, the dosage of drugs delivered by skin is usually lower than oral drugs, which can avoid the side effects caused by unstable absorption and metabolism of drugs in gastrointestinal tract [5]. In the case where oral drug delivery is difficult, transdermal drug delivery technology can be easily applied on the skin, and provide effective blood concentration level [6].

Different forms of microneedles have different ways of releasing drugs. There are five types of microneedles: solid microneedles, hollow microneedles, swelling microneedles, soluble microneedles, and coated microneedles [7]. The main advantage of solid microneedles is their firmness, which makes it easier to penetrate the skin and can be used to pretreat the skin. Henry et al [8]. First demonstrated the increase of transdermal flux of calcein after the silicon microneedles were prepared by ion etching. Dissolving microneedles are mainly made of polymers or polysaccharides, which release drugs through dissolution after piercing the skin [9]. Hollow microneedles have the same empty cavity as a traditional hypodermic needle to deliver drugs to the skin or through the skin into the blood, but the microneedles are shorter and the flow rate can be controlled by a micropump or syringe [10]. Coated microneedles are made from a substrate coated with drugs that can be delivered quickly to the skin and increase the long-term stability of the active drug, although the amount of drug can be uncontrolled. Swelling microneedles can absorb the interstitial fluid of the skin and provide channels for drug delivery in the microneedles. When used, the integrity can be retained without residual accumulation in the skin, and it is convenient for patients to use and improve patient dependence. Swelling microneedles have the function of rate control and prolong the time of drug administration by adjusting the swelling performance [11]. Now, the most widely used is the use of microneedles to carry drugs, after the microneedle enters the epidermis, the needle body swells and the drugs enter the human skin.

Silk fibroin protein is a kind of natural polymer material with good biocompatibility and natural degradation, and rarely has sensitization reaction [12]. Silk fibroin protein can also maintain the biological activity of the drugs it carries for a long time, so it is extremely suitable for the preparation of biological materials [13]. Based on previous studies by scholars, it can be known that pure silk fibroin protein microneedles have good mechanical properties and can successfully achieve the purpose of drug release by microneedles. However, the untreated pure silk fibroin is random coil structure, its molecular chain arrangement is disordered, and water molecules can enter smoothly, which makes the microneedles dissolve quickly when contacting body fluid. In this paper, we explored the effects of proline and its derivatives on the structure and properties of silk fibroin microneedles, and explored a more balanced addition strategy to prepare the swelling silk fibroin microneedles with low solubility, high swelling and good mechanical properties. It provides new methods and ideas for the improvement of microneedle transdermal drug delivery materials.

2 Materials and Methods

2.1 Experimental Materials and Instruments

2.1.1 Experimental Materials

Silkworm cocoon shell (Suzhou siruibao Biotechnology Co., Ltd., China), sodium carbonate, sodium bicarbonate, lithium bromide (Tiancheng Chemical Co., Ltd., China), dialysis bag (MWCO: 8-12kd, Pierce), L-proline (Shanghai Yuanye Biotechnology Co., Ltd.), L-prolinamide (Shanghai Aladdin Biochemical Technology Co., Ltd.), L-hydroxyproline (Shanghai Aladdin Biochemical Technology Co., Ltd.) etc.

2.1.2 Experimental Instruments

FA2004 electronic balance, SHY-2 digital display water bath constant temperature oscillator, DT5-2 low speed table centrifuge, DHG-9246A electrothermal constant temperature blast drying oven, 84-1A magnetic stirrer, SmartSpec Plus spectrophotometer, X'PERT-PRO MPD X-ray diffractometer, Nicolet is 5 intelligent Fourier transform infrared spectrometer, TMS-PRO texture analyzer.

2.2 Experimental Part

2.2.1 Preparation of Silk Fibroin Solution

Weigh a certain proportion of sodium carbonate and sodium bicarbonate and dissolve in water at 100 °C. Add 80 g cocoon shell and cook for 30 min. Clean boiled cocoons with deionized water to remove sericin from the surface. The cleaned cocoon shell is boiled in sodium carbonate/sodium bicarbonate buffer solution again for 30 min and cleaned. Repeat three times. Put the degummed silk fibroin into 60 °C oven for drying. Weigh 15 g of dried degummed silk fibroin and dissolve it in LiBr solution (9.3 M) at 65 °C for 60 min. After cooling, the solution was put into a dialysis bag and placed in 4 °C deionized water for dialysis for 3-4 days. The obtained silk fibroin solution was stored in a 4 °C refrigerator for later use.

2.2.2 Preparation of Microneedles by Die Casting

Proline and its derivatives were prepared into an aqueous solution with a concentration of 150 mg/mL for use. Mix according to the mass ratio of amino acids to silk fibroin of 0/10, 1/10, 2/10, 3/10, 4/10, stir fully, and pour the mixed solution into the PDMS mold. After vacuuming for several times, the formed microneedles were dried for 24 h in a constant temperature and humidity room.

2.2.3 X-ray Diffraction

X-ray diffraction sample preparation: The silk fibroin fibers were cut into fine powder particles and various silk fibroin microneedles were cut into pieces with scissors and screened by 80 μm sieve. The particles that could pass the sieve were used for testing.

X-ray diffraction analysis: Using automatic X'PERT PRO MPD X-ray diffractometer. The diffraction intensity curve was recorded between 5° and 45° under the conditions of 10°/min scanning speed, 40 kV, and 30 mA.

2.2.4 Infrared Detection

The prepared silk fibroin film was tested on the Nicolet is 5 intelligent Fourier transform infrared spectrometer. The scanning range was 400∼4000 cm^{-1}, and the infrared absorption spectrum was obtained.

2.2.5 Raman Scattering Spectroscopy

Raman spectrum was measured using a Japanese HORIBA Raman Microscopy (HORIBA Ltd., Kyoto City, Japan). The excitation wavelength was 532 nm, slit width was 100 μm, and 1200 gr/mm grating was selected. The scanning time of the fixed sample was 20 s, and the Raman scattering spectrum recording step range was 200∼2000 cm^{-1}.

2.2.6 Moisture content of microneedles

Different amino acids/silk fibroin protein microneedles were weighed, denoted as M1, placed in an oven at 105 °C, dried for 2 hours, and weighed M2. The moisture content of the microneedle was calculated by the formula (1).

$$\text{Moisture content} = \frac{\text{M1} - \text{M2}}{\text{M2}} * 100\% \tag{1}$$

2.2.7 Detection of Dissolution and Swelling of Microneedles

The silk fibroin microneedle was balanced at the same room temperature for 24 hours, then the M1 was weighed and put into a centrifuge tube. PBS buffer (pH = 7.4) was added at a bath ratio of 1:100(W/V), and the silk fibroin microneedle was shaken in a constant temperature shaker at 37 ° water bath for 24 hours. Clean the solid silk fibroin microneedles in the solution with deionized water for 3 times, then absorb surface moisture with absorbent paper and weigh M2. After the solution in the centrifuge tube was centrifuged at 3000 r/min, the supernatant was taken and the absorbance of the solution at 278 nm was measured by ultraviolet spectrophotometer. The swelling rate Q(%) and silk fibroin dissolution rate C(%) of microneedles were calculated by formula (2) and (3) respectively.

$$\text{Q} = \frac{\text{M2} - \text{M1} \times (1 - F)}{\text{M1} \times (1 - F)} \times 100\% \tag{2}$$

$$\text{C} = \frac{KAV}{\text{M1} \times (1 - F)} \times 100 \tag{3}$$

Q — Silk fibroin swelling degree (%);

M1 — The initial weight of the sample (g);

M2 — Weight of sample after soaking (g);

F — Moisture content (%);

C — The dissolution rate of c-silk fibroin (%);

K — UV absorption constant of silk fibroin solution;

A — Absorbance;

V — Volume of PBS solution (mL).

2.2.8 Measurement of Compression Strength of Microneedles

Different amino acid/silk fibroin microneedles were evenly cut into 3× 3 array, 5 parallel samples in each group, the needle tip was placed upward under the TMS-PRO texture analyzer, and the breaking strength of the needle tip was detected. Test conditions: initiation force 0.02 N, maximum detection range 25 N, deformation 80%, compression speed 10 mm/min.

3 Experimental Results and Analysis

3.1 Silk Fibroin Protein Blend Membrane

In order to clearly observe whether proline and its derivatives are compatible with silk fibroin, silk fibroin blend membrane was prepared.

As shown in Fig. 1, proline and prolinamide are better fused with silk fibroin in any proportion. When the ratio of hydroxyproline to silk fibroin reached 3/10, white precipitates appeared on the film, which was the self-crystallization of small molecules. This indicates that when the proportion is high, the compatibility between hydroxyproline and silk fibroin becomes poor, and this kind of phase separation material is not suitable for microneedle material because of its unstable structure.

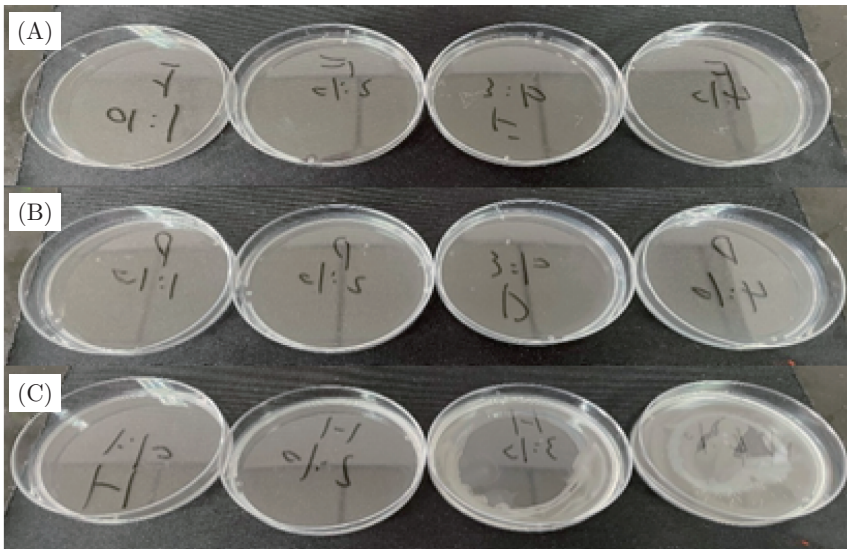

Fig. 1: Silk fibroin protein films of different amino acids: (A) proline/silk fibroin, (B) prolinamide/silk fibroin, (C) hydroxyproline/silk fibroin

3.2 Morphology of Microneedles

The microneedle mold determines the external shape of the microneedle. Here, we used the mold of the same specification to make the microneedle prepared by adding different amino acid silk protein solutions have the same needle shape. As shown in Fig. 2(A), the microneedle patch consists of 225 (15×15) microneedles evenly distributed. In Fig. 2(B), we can see the microneedles prepared by the mold were slender and tapered, with a length of about 600 μm.

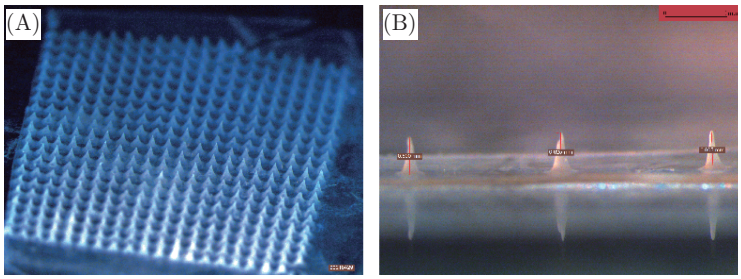

Fig. 2: Morphology of microneedles

3.3 X-ray Diffraction Analysis

In the X-ray diffraction pattern, the crystal peaks of Silk I appeared at 12.2°, 19.7°, 24.7°, 28.2°, 32.3°, 36.8° and 40.1°, while the crystal peaks of Silk II appeared at 9.1°, 18.9°, 20.7° and 24.3° [14].

As shown in Fig. 3(A), the X-ray diffraction curve of pure silk fibroin microneedles shows a steamed bread shape without obvious absorption peak. The results showed that the molecular

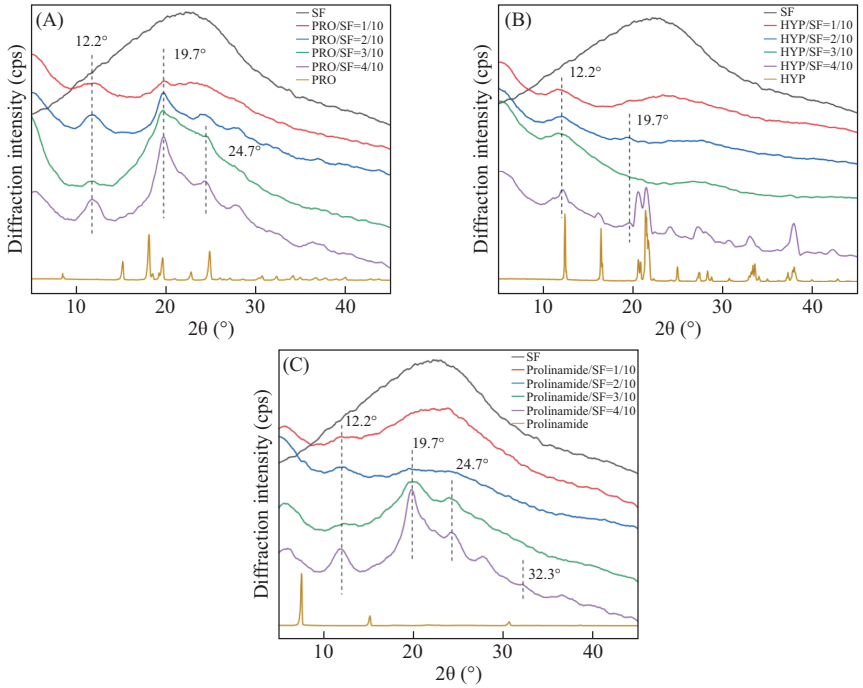

Fig. 3: X-ray diffraction curves of amino acid/silk fibroin microneedles: (A) proline/silk fibroin, (B) hydroxyproline/silk fibroin, (C) prolinamide/silk fibroin

chain of silk fibroin was in random coil state, and no crystal was formed in the microneedles. Therefore, pure silk fibroin microneedles are easily soluble in water. When the ratio of proline to silk fibroin is 1/10, 2/10, 3/10 and 4/10, the crystal peaks appear at 12.2° and 19.7°. With the increase of the proportion of proline, the peak pattern becomes more acute, indicating the formation of the crystal structure of Silk I. Proline mainly induces silk fibroin protein to form the crystal structure of Silk I. The addition of proline greatly reduced the dissolution rate of silk fibroin microneedles.

In Fig. 3(B), when the ratio of hydroxyproline to silk fibroin was 1/10, the curve showed no significant difference from that of pure silk fibroin, presenting a state of random coiling. At 2/10, 3/10 and 4/10, the crystal peaks of 12.2° and 19.7°. As the proportion of hydroxyproline increases, the peak shape becomes sharper. When the ratio of hydroxyproline to silk fibroin was 4/10, a large number of crystallization peaks of hydroxyproline appeared. The above results indicate that when hydroxyproline/silk fibroin is at 1/10, hydroxyproline does not induce silk fibroin to form Silk I crystallization. When the ratio reached 2/10, 3/10 and 4/10, the silk fibroin protein mainly formed the crystal structure of Silk I under the induction of hydroxyproline. When hydroxyproline/silk fibroin is 4/10, hydroxyproline begins to crystallize, which indicates that the compatibility between hydroxyproline and silk fibroin protein is poor at this time. This is the same as with the blending film.

In Fig. 3(C), 1/10 prolinamide/silk fibroin barely changed compared to pure silk fibroin. At 2/10, 3/10 and 4/10, the crystal peaks of Silk I appeared at 12.2° and 19.7°, and gradually became sharp with the increase of prolinamide dosage. When reaching 3/10, the crystal peaks of 24.7° and 32.3° appeared, indicating that a large number of crystal structures of Silk I were formed at this time. It can be seen from the figure that prolinamide has good compatibility with silk fibroin, and induces silk fibroin to mainly form the crystal structure of Silk I.

3.4 Infrared Detection

In order to further explore the aggregation structure of the modified silk fibroin microneedle, Fourier transform infrared spectrometer was used to test its secondary structure. In the infrared spectrum, the untreated silk fibroin microneedle had obvious absorption peaks at 1635 cm^{-1}, 1530 cm^{-1} and 1235 cm^{-1}, which were characteristic peaks of random coil.

In the infrared spectrum, it can be seen that the silk fibroin microneedles have no other obvious absorption peaks compared with pure silk fibroin microneedles after adding proline and its derivatives. Absorption peaks were observed at 1635 cm^{-1} (amide I), 1515 cm^{-1} (amide II) and 1235 cm^{-1} (amide III) [15]. With the increase of amino acids, the absorption peaks of silk fibroin in the amide I, the amide prism and the amide prism remain basically unchanged. In general, it is difficult to distinguish random coil from α-helix directly by FTIR. By combining FTIR spectra with XRD data, we can determine that the absorption peak should be the coexistence and superposition of α-helix, random coil and β-folding peak.

3.5 Raman Scattering Spectroscopy

To further illustrate the effect of proline and its derivatives on silk fibroin structure, we examined the changes of Raman spectra of silk fibroin material.

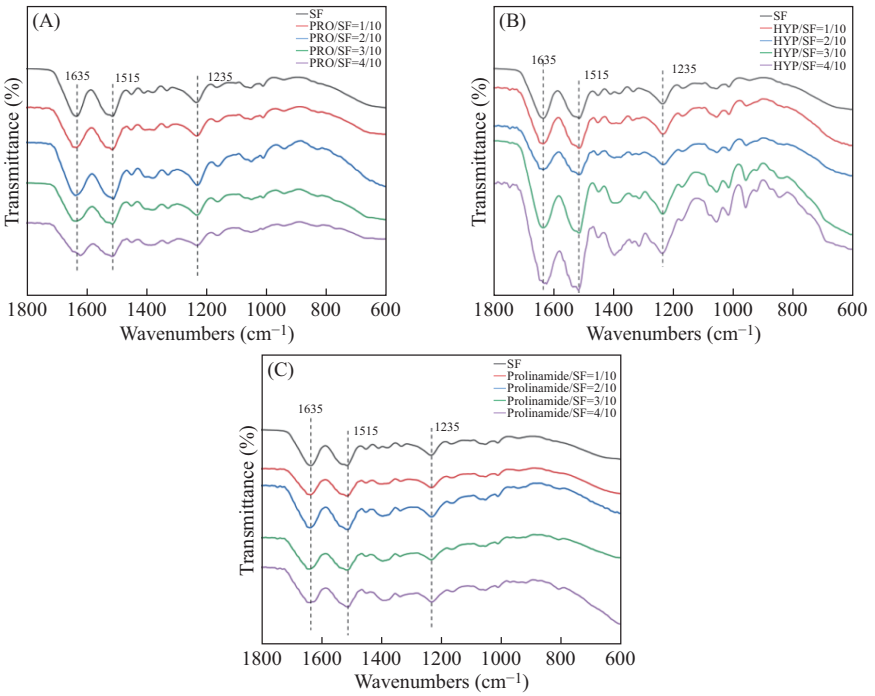

Fig. 4: FTIR of proline and its derivatives/silk fibroin microneedles: (A) proline/silk fibroin, (B) hydroxyproline/silk fibroin, (C) prolinamide/silk fibroin

In Raman spectroscopy (Fig. 5), it can be seen that proline and its derivatives have scattering peaks at 1667 cm^{-1} (amide I), 1245 cm^{-1} (amide III) and 1106 cm^{-1} [16]. With the increasing proportion of amino acids, the absorption peaks of silk fibroin at amide I and amide III were

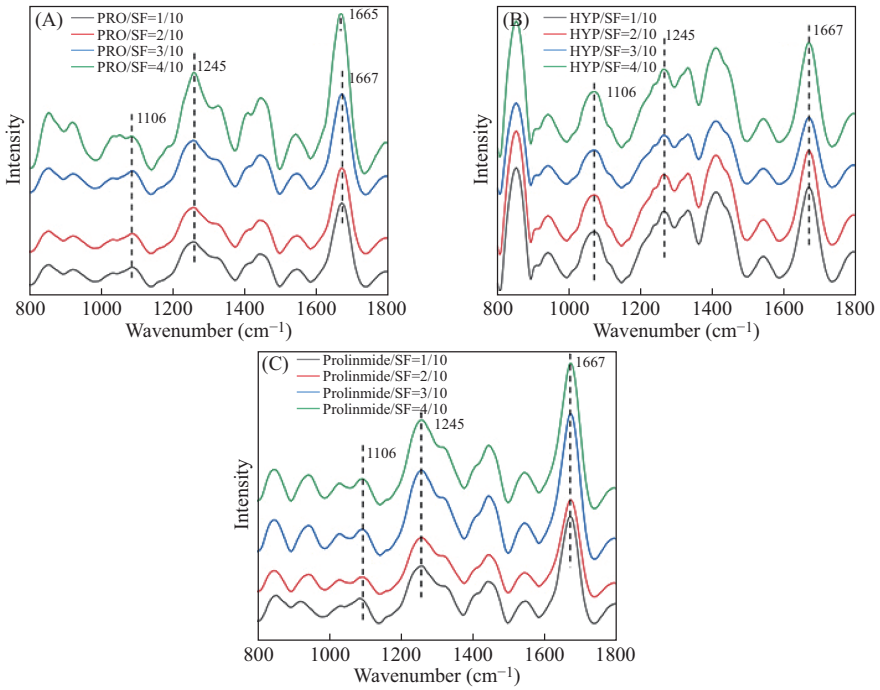

Fig. 5: Raman spectroscopy of proline and its derivatives/silk fibroin microneedles: (A) proline/silk fibroin, (B) hydroxyproline/silk fibroin, (C) prolinamide/silk fibroin

basically unchanged, which was a typical random curly conformation. Combining XRD data and FIRT spectra, we can further confirm that the structure of silk fibroin protein consists of a large amount of α-helix, random curl and a small amount of β-fold after the addition of proline and its derivatives.

3.6 Moisture content of microneedles

Tukey test was used for moisture content of different proportions of amino acids. As shown in Fig. 6(A), it can be found that the moisture content of different proportions of proline is about 9%. As shown in Fig. 6(B), the water content of hydroxyproline varies greatly with different proportions, and when the ratio of amino acids to silk fibroin reaches 3/10, the water content is 6.5%, which is the minimum. When reaching 4/10, the moisture content is the largest, which can reach 10.6%. In Fig. 6(C), it can be seen that there are significant differences between prolinamide. At 2/10, the moisture content is 14.7%, while at 1/10, the moisture content is only 6%.

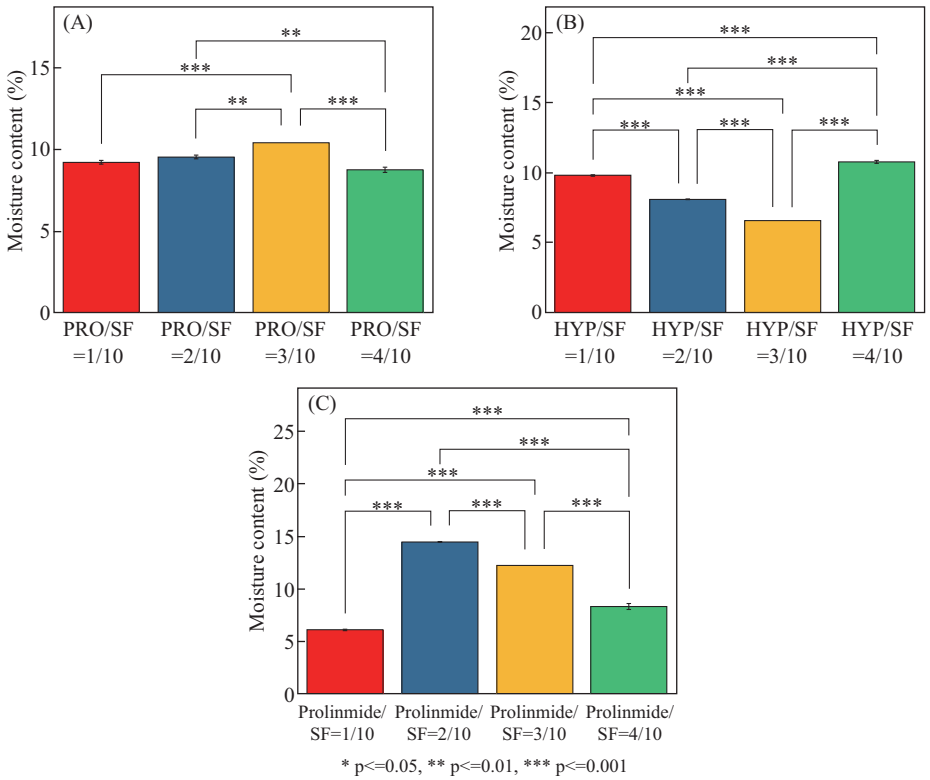

Fig. 6: Moisture content of microneedles: (A) proline/silk fibroin, (B) hydroxyproline/silk fibroin, (C) prolinamide/silk fibroin

3.7 Swelling and Dissolution Properties of Microneedles

Proline and its derivatives were added and mixed with silk fibroin protein to obtain silk fibroin microneedle with high swelling degree. By controlling the ratio of drugs and silk fibroin, different microneedles were prepared to compare their swelling and dissolution properties. In order to simulate the internal environment of human body, we soaked microneedles in PBS solution (pH = 7.4). Fig. 7 and Fig. 8 use Tukey test for significant difference analysis.

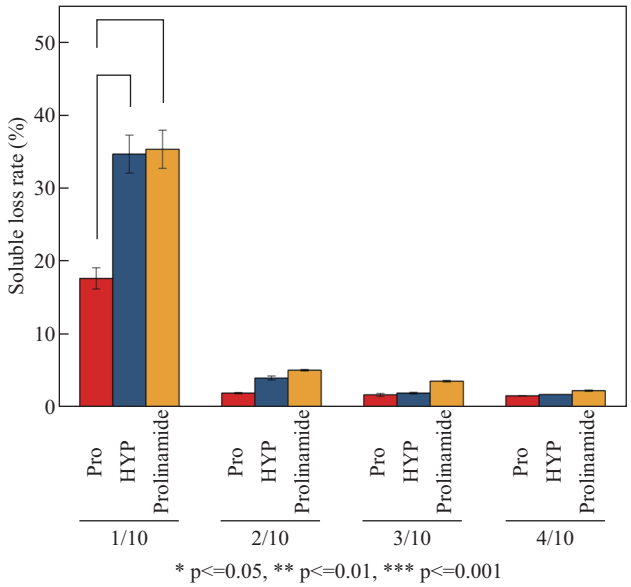

Fig. 7: Dissolution property of silk fibroin microneedles

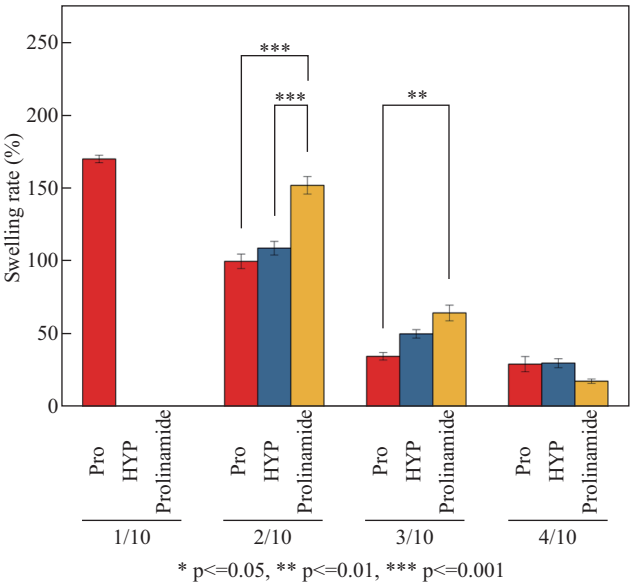

Fig. 8: Swelling properties of silk fibroin microneedles

It can be seen from Fig. 7 that when the ratio of proline and its derivatives to silk fibroin is 2/10, 3/10 and 4/10, the dissolution loss of the three microneedles is very small, less than 6%, and there is no significant difference. But at 1/10, hydroxyproline and prolinamide were significantly different from proline. At this time, the dissolution rate of hydroxyproline and prolinamide is as high as 35%, while that of proline is only 18%. This is because when hydroxyproline/silk fibroin and prolinamide/silk fibroin are 1/10, their structures are basically random coil, they are easily soluble in water, and the loss of protein is large, so the dissolution rate is relatively large.

Pure silk fibroin microneedles dissolve rapidly in PBS solution, so it is impossible to measure its swelling degree. When the ratio of hydroxyproline/silk fibroin and prolinamide/silk fibroin was 1/10, the silk fibroin microneedles dissolved rapidly in solution, so the swelling degree could not

be measured. It can be seen from Fig. 8 that when proline and its derivatives/silk fibroin is 4/10, the swelling of the three microneedles is about 25%, and there is no significant difference. In the case of no significant difference in dissolution rate, there was a significant difference in swelling rate when amino acid/silk fibroin was 2/10. When the amino acid/silk fibroin ratio was 2/10, the swelling degree of silk fibroin microneedle was up to 150% when prolinamide was added, while the swelling degree of other microneedles were only about 100%. It can be considered to use the additive amount of 2/10 to get the higher swelling rate of silk fibroin microneedles. Prolinamide microneedles at 2/10 were significantly different from other microneedles. In addition, when the ratio of amino acid to silk fibroin was 3/10, the swelling degree of microneedles with prolinamide was 60%, while that of proline was only 34%.

With the increase of amino acid content, the swelling and dissolution of micro needle decreased gradually. This is because the small molecules of proline, hydroxyproline and prolinamide promote the formation of a small amount of silk fibroin crystallization, thus reducing the loss rate of silk fibroin, so it has certain swelling and insoluble. When PBS solution is added, the non-crystalline region expands, and a small amount of crystal region suppresses its infinite expansion. With the increase of the small amino acid molecules, silk fibroin was induced to produce more crystallized regions. The results showed that the dissolution rate of microneedle decreased with the increase of amino acid small molecule. With the increase of addition dose, hydroxyl, peptide bond and amino group in hydroxyproline, proline and prolinamide molecules can interact with polar groups such as amide bond, Unbound hydroxyl group or carboxyl group in silk fibroin molecular chain to form hydrogen bond. The formation of these hydrogen bonds further increases the interaction of silk fibroin proteins, so the swelling properties of microneedles also tend to decrease.

3.8 Mechanical Properties of Microneedles

Microneedles must be strong enough to penetrate the cuticle of the skin for their therapeutic purpose. The needle must not break in order to deliver the drug to the dermis, where it can enter the body's fluid circulation for transdermal release. Fig. 9 shows the shape changes of the microneedle before and after the strength test. When testing the mechanical properties of the microneedle with texture instrument, the sensor will sense two forces in the process from the contact of the sensor to the complete bending of the microneedle: the first is the bending force of the microneedle tip, and the second is the force of the whole microneedle body being broken. Here, the texture instrument records the first force, namely the breaking strength of the tip, to investigate the penetration performance of the microneedle, as can be seen in Fig. 10.

As shown in Fig. 9(A), the 3*3 microneedle array is not compressed. The microneedles in Fig. 9(A) have full tips and similar lengths. Fig. 9(B) shows the compressed 3*3 microneedle array, in which the microneedle tips are broken.

In Fig. 10(A), it can be seen that when the proline/silk fibroin ratio is 1/10, the displacement load of the microneedle is the largest, which is 10.1 N; when the proline/silk fibroin ratio is 4/10, the displacement load of the microneedle is the smallest, which is only 4.7 N.

In Fig. 10(B), when prolinamide/silk fibroin is 1/10, the maximum displacement load of the microneedle is 11 N, and when 4/10, the minimum displacement load of the microneedle is 7 N.

In Fig. 10(C), when hydroxyproline/silk fibroin is 2/10, the maximum displacement load of the microneedle is 13.3 N, and when it is 1/10, the minimum displacement load of the microneedle is 7.8 N.

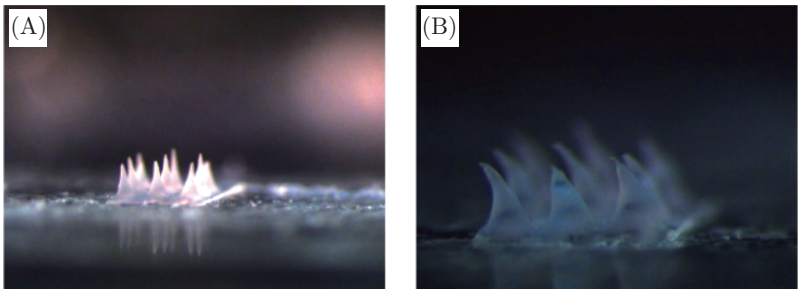

Fig. 9: Morphology of microneedle before and after mechanical test

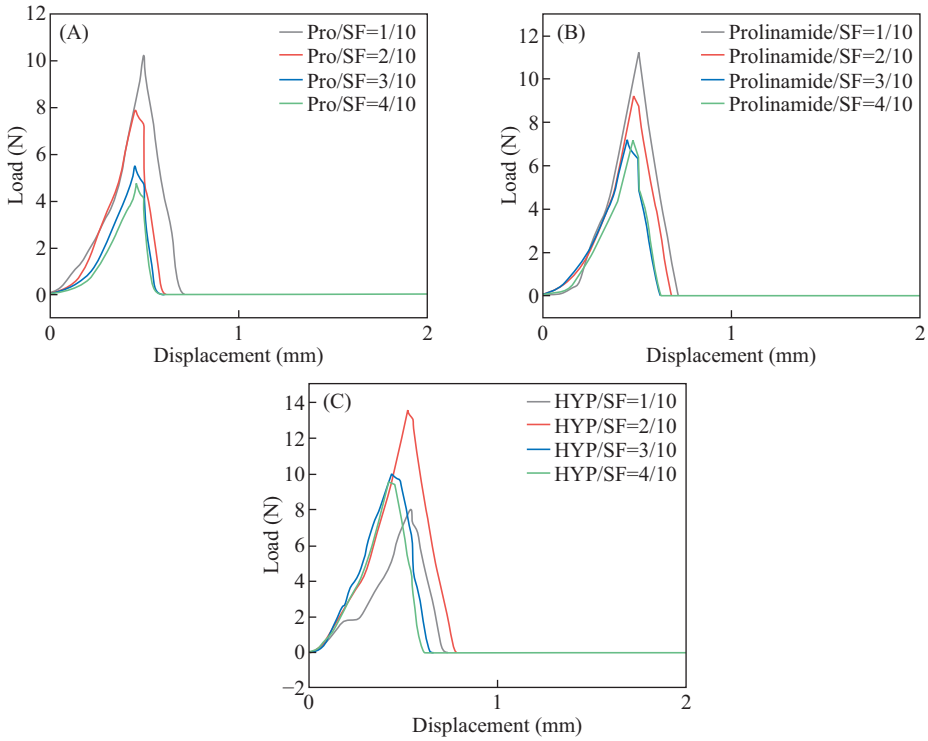

Fig. 10: Displacement load curve of proline and its derivative 3*3 microneedles: (A) proline/silk fibroin, (B) prolinamide/silk fibroin, (C) hydroxyproline/silk fibroin

Fig. 11 Significant difference analysis was performed using Tukey test. As can be seen from Fig. 11, there are significant differences among the three microneedles in any proportion. At 2/10, the maximum compression strength of hydroxyproline/silk fibroin single needle reached 1.4 N. According to the observation, except for 1/10, the compressive strength of single needle was hydroxyproline > prolinamide > proline. Therefore, the mechanical properties of hydroxyproline were the best, followed by prolinamide, and proline was the worst. In general, the breaking strength of microneedles decreased with the increase of amino acids, which was due to the fact that more small molecules were inserted between the molecular chains of silk fibroin to play the role of lubricant. When the external force acts on the microneedles, the relative slip of silk fibroin molecular chain is more likely to occur due to the action of small molecules, resulting in the decrease of its breaking strength. It has previously been reported that a single needle can break human skin with a force of 0.08 N. When the insertion force is 0.1-3 N, it is enough to penetrate the skin [17]. It can be seen from the figure, the strength of proline/silk fibroin microneedles,

hydroxyproline/silk fibroin microneedles and prolinamide/silk fibroin microneedles are very high, which is far greater than the force required to puncture the skin. Therefore, the microneedle has a good penetration performance, which can easily penetrate the cuticle of the skin without breaking, so as to achieve the role of drug delivery.

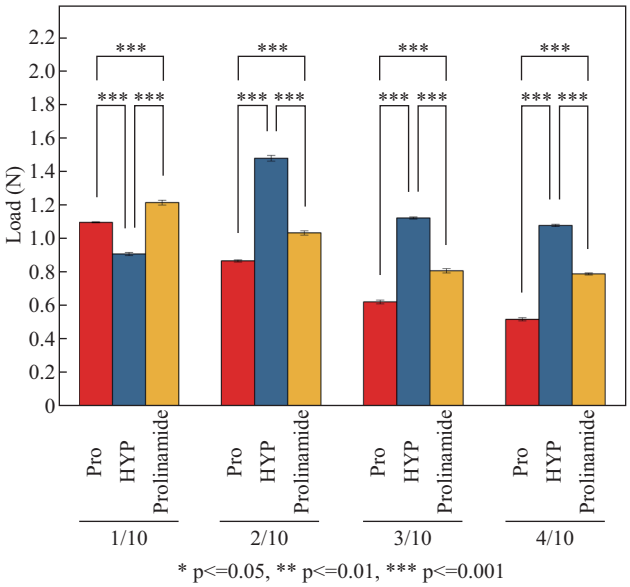

Fig. 11: Fracture strength of proline and its derivative/silk fibroin single microneedle

4 Conclusion

In this paper, silk fibroin was modified by proline and its derivatives to prepare swelling microneedles. In the above experiments, it can be found that high proportion of hydroxyproline and silk fibroin will produce white crystals after blending, which is not suitable for micro needle materials. In the XRD and FTIR images, it can be found that silk fibroin blends with proline and its derivatives mainly show the Silk I crystal structure, so water molecules can enter the internal structure of the microneedle to produce swelling. In the swelling and dissolution diagram, when the proline/silk fibroin is 1/10, the swelling degree can reach 165%, but the dissolution rate is as high as 18%, so it is not suitable for swelling microneedles. When the mass ratio was 2/10, proline and its derivatives showed the same trend, with higher swelling degree and lower dissolution rate. In order to prepare high swelling microneedles, materials with low dissolution rate, high swelling degree and good mechanical properties need to be selected. When prolinamide /silk fibroin is 2/10, the swelling degree is 150%, the dissolution rate is 5%, and the breaking strength of a single needle can reach 1 N. It can be used as a swelling microneedle for transdermal drug delivery.

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