THE MECHANISM OF SILK FORMING FOR SPIDERS AND SILKWORMS¹⁾

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It is well known that the silk fibers secreted and spun out from the silk gland of spiders and silkworms hav high strength and high tenacity. The fine organization of —sheet and —helix structures in silk is commonly recognized as the primary contribution to the extraordinary mechanical properties. But how these factors such as concentration, shear rate, tension stress of fibroin solution and geometric shape of silk gland regu late the process from fibroin gel to solid-state silk fiber still remains unclear. The silks spun by different silkworm such as bombyx mori and Antheraea pernyi and by the different silk glands such as the major ampullate gland and the agelliform gland have a large variety of mechanical properties. The amino acid sequences of their fibroin also have their own motifs, e.g. [GAGAGX]_n in Bombyx mori or [A]_n in Antheraea pernyi and spider. So another arguable issue is whether the motifs of the amino acid sequences dominate the fold and interaction of molecules in fibroin gel.

The effect of concentration of fibroin solution on the secondary structures of molecules was studied in the present work. The regenerated fibroin solution was obtained by dissolving the degummed cocoon silk of *Bombyx mor* in salt solution. The final fibroin solution was diluted into a series of solutions with different concentrations. The concentrations of fibroin solutions were determined by the methods of BCA assay kit and OD280. Then the contents of secondary structures were computed from the circular dichroism spectra. Finally atomic force microscopy (AFM) was used to observe the molecules cast on the slides under the differen concentration. The results showed that the contents of secondary structures of -sheet, -helix, -turn and coil structures were about 40%, 10%, 20% and 30%, respectively and were almost independent of the concentration. AFM observation showed that the fibroin molecules seemed to fold into micelles and along with the fibroin solution being diluted, the micelles gradually dispersed in the solution. So it can be conducted that the regenerated fibroin solution might be mainly comprised of a large number of micelles and the above quantitative measurement of the contents of secondary structures will be helpful to establish the theoretical model of silk forming mechanism.

To probe the unfolding process of single fibroin molecule and the interactions of multiple molecules, AFM was used to measure the force spectrum curves of the fibroin of bombyx mori, Antheraea pernyi and an artificial synthetic spider's dragline polypeptide. Some typical force-displacement curves for the different fibroin molecules were found and the corresponding unfolding trajectories were explained by using the theoret ical model basing on the statistical mechanics.

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