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Electrospinning of Silk Biomaterials

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Abstract

Silk fibers have been processed and used to create fabrics and threads for centuries. However, the solubilizing of silk fibers, thereby turning it into a versatile pre-polymer solution is a much newer technology. Solubilized silk can be processed in many different ways to create a biocompatible material with controllable mechanical properties.

This video introduces the processing of silk from silk worm cocoons, and shows how the silk solution can be used to create a fiber mat via electrospinning. Several applications of this technique, such as its use as a structural material in engineered tissue scaffolds, are then introduced.

Transcript

Biologically derived materials possess key properties that are beyond the reach of synthetic materials, enabling the fabrication of complex structures with improved properties. Biomaterials are materials that are created or produced from living or once-living organisms. These materials, such as silk, typically undergo various processing steps to reach a usable and controllable state. The processed biomaterial can then be utilized to form specific structures such as fibrous mats, hydrogels, and films. This video will introduce the processing of silk from silkworm cocoons, followed by electrospinning of the processed material to form fibrous mats.

Silks are protein polymers that are spun into fibers by various insects, such as spiders. However, the silk used in bioengineering is most commonly derived from the larvae of the silk moth species Bombyx mori, commonly called the silkworm, and has been used commercially for medical sutures for decades. The B. mori silk material is extracted from the cocoon, which consists of the fibroin protein encased in a sericin coat. Sericin is a glue-like protein that holds the cocoon together. The fibroin protein is characterized by a highly repetitive amino acid sequence, with a predominance of alanine, glycine, and serine residues, which in turn leads to homogeneity in the protein's secondary structure. Silk fibroin exhibits repetitive beta-sheet structures, which are laterally connected strands forming a pleated sheet. These structures cause the polymer protein to be highly crystalline, which in combination with its hydrophobicity, provides incredibly strength and toughness. For example, silk's tensile strength, its capacity to withstand loads through elongation, can be up to four times that of bone. Silk is typically processed into a polymer solution prior to its applications in bioengineering. First, sericin is removed from the fibers, followed by solubilization of the fibroin. Silk solution has no secondary structure, and therefore has diminished mechanical properties. However, treatment with methanol can induce partial recovery of the beta sheet structures. The silk solution can be processed using a technique called electrospinning, where a high voltage is applied between a collection surface and a syringe needle. The syringe needle slowly dispenses the biomaterial solution. The electrostatic forces cause the biomaterial droplets to stretch into fibers. These fibers collect randomly on the collector, creating a nanofiber mat. Now that the basics of silk fibroin processing have been outlined, let's take a look at the silk processing procedure and see how it's used to create microfiber mats via electrospinning.

To prepare silk fibroin solution from Bombyx mori, the cocoons are first cut into small pieces, and the worm and other insect debris discarded. Next, the cocoon pieces are boiled in a sodium carbonate solution in order to remove the sericin. After boiling, the silk fibroin is removed from the solution and washed several times with clean water. Next, the silk fibers are dried overnight. The dried silk fibers are then dissolved in a lithium bromide solution at 60 degrees for four hours. Once the silk is solubilized, the solution is transferred to a dialysis cassette and dialyzed against water. The water must be changed frequently to ensure removal of the lithium bromide. After dialysis, the aqueous silk solution is transferred to tubes and centrifuged to remove any remaining silk fibroin particulates. The silk solution is stored at four degrees until needed.

To begin electrospinning, the biomaterial solution is loaded into a syringe and onto a syringe pump. The high-voltage source is connected to the syringe needle tip, called the spinneret. The collection source, often a strip of foil, is placed opposite the spinneret and grounded. The distance between the spinneret and source is adjusted to a pre-determined distance. If the distance is too small, fibers may not have time to solidify before deposition, forming inconsistent ribbons. However, if the distance is too high, fibers may become clumped or non-uniform. The syringe pump flow rate is set and the high-voltage source turned on. As the voltage increases, the electrostatic forces counteract the surface tension of the solution. The meniscus is drawn into a cone, called the tailor cone, and then eventually into a fluid jet with higher voltage. The fibers are then collected on the foil surface in a randomly arranged mat. A rotating collection surface can also be used in order to obtain aligned fiber mats.

There are many different types of biomaterials that are used in bioengineering applications, ranging from tissue implants to bioelectronics and biosensing. Electrospun mats of silk and other biomaterials are often used to create tissue scaffolds, which provide structure to an artificial tissue. Cells are seeded directly onto these structures, enabling the formation of three-dimensional tissue. Tissue scaffolds are designed to promote favorable cell interactions, thus mimicking the structure and function of real tissue. They are also engineered to possess the desired properties of a specific tissue. For example, scaffolds designed to behave like muscle must possess more aligned fibers than those designed to behave like skin. Although silk is commonly derived from the silkworm, spider silk is often used, as it provides higher mechanical strength. Since spiders can not be easily farmed, the material is produced synthetically using biotechnology. In this example, the recombinant silk protein was electrospun onto filter devices. The addition of this silk mesh to the filter greatly improved efficiency, as the pore sizes were significantly smaller than the manufactured filter, thereby enabling their retention of smaller particles. Finally, biomaterials are often prepared from man-made polymers such as polycaprolactone and polylactic acid. The materials are synthesized in the lab and used to mimic bio-derived materials as they are biodegradable and bio-compatible. The properties of the materials can easily be tailored using fabrication techniques to create bio-compatible surfaces for the assembly of bio-molecules or to create hydrophobic surfaces as shown in this example.

You have just watched JoVE's Introduction to Silk Biomaterials. You should now understand how silk is prepared and electrospun into mats and how other biomaterials are used in the bioengineering field. Thanks for watching.

