Nanomechanical Characterization and Evaluation of Silk Fibroin Electrospun Scaffolds for Regenerative-Engineered Orthopedic Products

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Introduction/Background

- Growing interest in biomimicking 3D living tissue and organs for regenerative medicine, leveraging superior mimicry of human physiology, supported by research evidencing improved replication of tissue architecture, with emphasis on mimicking the ECM through nano-scaled biopolymer fibers in 3D scaffold environments.
- Silk fibroin (SF), a natural bioopolymer found in silkworm cocoons, possesses excellent mechanical properties due to its embedded nano-fibrillar structure, contributing to a balance of strength, modulus, toughness, extensibility, lightweight, and flexibility.
- Research Objective:
  - Performing nanomechanical tests on electrospun-silk fibroin nanofibers via Atomic Force Microscopy is proposed to accurately quantify load-strain behavior and understand their mechanical interaction with anchorage-dependent cells on electrospun substrates.

Methods

Atomic Force Microscopy

- Atomic Force Microscopy (AFM) is a powerful tool used to image surfaces at the atomic level, providing high-resolution topographical information.
- Force-distance curves using atomic force microscopy (AFM) with a sharp tip possesses a very close to the sample surface, enabling the detection of changes in the mechanical properties of the sample.

Nanoindentation

- Nanoindentation measures the mechanical properties of a sample surface, enabling the determination of properties like hardness and modulus by analyzing the force-depth curve. This is performed via tapping mode (externally oscillating the cantilever at its fundamental resonant frequency while scanning the surface.)
- Young's modulus can be determined through analytical models such as the JKR model or Hertzian contact theory, which relate the contact mechanics of the indenter with the sample's properties.

Results

Youth's Modulus (E) - Degummed SF: 6.5 GPa
Youth's Modulus (E) - Electrospun SF: 5.15 GPa
Youth's Modulus (E) - Electrospun SF (Literature): 19.3 ± 2.8 GPa

Conclusion and Discussion

The objective of this project was to conduct nanomechanical tests on electrospun silk fibroin (SF) nanofibers using Atomic Force Microscopy (AFM) to accurately quantify load-strain behavior and understand their mechanical interaction with anchorage-dependent cells on electrospun substrates. This research is driven by the interest in advanced biomaterials for regenerative orthopedic products, aiming to enable the testing of nano stiffness with various forces to tailor cell differentiation. Nanoin dentation was employed via tapping mode of AFM to achieve this objective. The nanoin dentation measurements were obtained using a DNP probe with a radius of 60 nm. Young's modulus data were sourced from literature and supplemented with experimental results generated by members of the BioCAS laboratory. Post-processing of the bright image involved a first-order phase filter, while observation of tilt was made via the 3D image, which may be affected by scanner bias, potentially introducing errors.

Preliminary data obtained from nanoin dentation provided insights into the system's behavior, particularly through TM deflection sensitivity. To further advance this study, the next steps involve reprocessing results by measuring a single electrospun SF scaffold using a custom nanomechanical instrument. Additionally, conducting more AFM testing is essential to enhance the statistical validity of the findings. These endeavors are pivotal in advancing our understanding of nanomechanical properties in the context of electrospun SF nanofibers and their potential applications in regenerative orthopedics.