Optical Transparency and In Vitro Biocompatibility of Recombinant Silk-Elastinlike Protein Polymer

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Introduction:

Recombinant protein polymers have been evaluated extensively as biomaterials for applications in drug delivery and tissue engineering. In this study, we examine the optical transparency and in vitro biocompatibility of films composed of a genetically engineered silk-elastinlike protein polymer, SELP-47K, and explore its potential for ophthalmic applications.

Methods:

Mechanical characterization: The protein polymer SELP-47K was generously provided by Protein Polymer Technologies, Inc. (San Diego, CA). SELP-47K films were cast from aqueous solutions, treated using methanol (MeOH) vapor, and are denoted as MeOH-treated films. Some MeOH-treated films were further crosslinked with glutaraldehyde (GTA), and are denoted as MeOH-GTA-treated films. Samples were preconditioned for 10 cycles with 5 minutes off-load between cycles. The stress-strain relationship of preconditioned samples was examined in 1x PBS at 37 °C using a PerkinElmer dynamic mechanical analyzer under various cyclic deformations.

Optical Transparency: The transmittances of non-, MeOH-, and MeOH-GTA-treated films were measured using a Cary 5000 UV-Vis-NIR spectrometer.

Cell Viability and Proliferation: The proliferation of NIH/3T3 fibroblasts on tissue culture polystyrene (TCPS) wells (as a control), and on MeOH- and MeOH-GTA-treated films cultured in FBS-containing MEM medium was analyzed by the MTS assay on days 1, 3, 5, and 7.

Results: Compared to non-treated films, which are unstable in PBS, MeOH-treated films display enhanced mechanical strength. Fully hydrated, MeOH-treated films show nearly linear deformation response to external applied loads, with an estimated average Young's modulus of 1.66 ± 0.37 MPa. They possess strain at failure of 190 ± 60% and ultimate tensile strength of 2.5 ± 0.4 MPa. After GTA crosslinking, stress-strain analysis of SELP-47K films reveals increases in Young's modulus (3.34 ± 0.26 MPa), deformability (245 ± 58%) and ultimate tensile strength (5.4 ± 1.1 MPa).

UV-Vis spectroscopy reveals that SELP-47K thin films are optically transparent to visible light but opaque to ultraviolet (UV) light as shown in Fig. 2. All three types of thin films display comparable transparency greater than 90% at wavelengths of 450 to 800 nm.

As shown in Fig. 3, the SELP-47K films can support cell proliferation. During the culture period of 7 days, NIH/3T3 fibroblasts cultured onto SELP-47K films continuously grew. Both MeOH- and MeOH-GTA-treated SELP-47K film scaffolds demonstrated cell proliferation profiles comparable to the culture on TCPS.

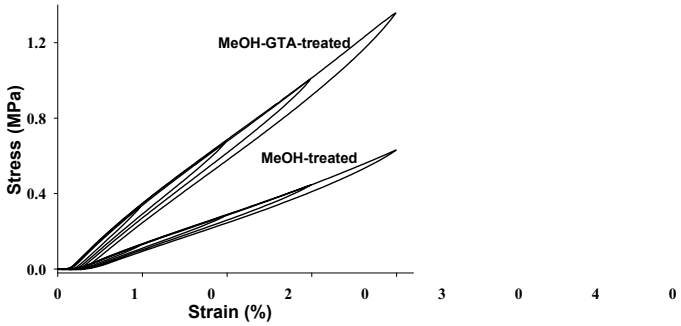

Figure 1. Stress-strain curve of preconditioned MeOH- and MeOH-GTA-treated SELP films under cyclic loading.

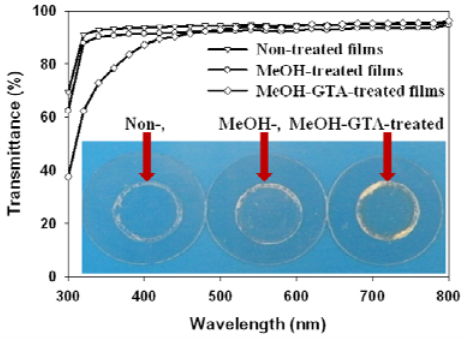

Fig. 2 Transmittance of non-, MeOH- , and MeOH-GTA-treated films of 30μm in thickness.

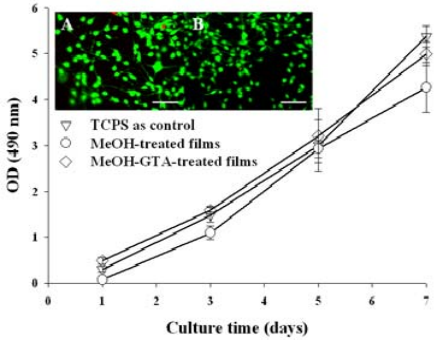

Fig. 3 Proliferation of NIH/3T3 fibroblasts grown on TCPS, MeOH-treated, and MeOH-GTA-treated films. The inset shows the fluorescent stained cells (live cells in green and dead cells in red) on MeOH- (A) and MeOH-GTA-treated (B) films. Scale bar: 100 μm.

Conclusions:

Genetically engineered SELP-47K films display excellent optical transparency to visible light. Together with the outstanding mechanical properties and in vitro biocompatibility, SELP-47K possesses the potential as a promising biomaterial for ophthalmic applications such as contact lenses, synthetic corneas, and ocular drug delivery matrices.