Liquid Crystal Solution and Mechanically Strong Scaffolds of Human Recombinant Type-I Collagen, Produced in Transgenic Tobacco

Introduction

Collagen deposition by fibroblasts is an essential stage during wound healing process. Type-I Collagen scaffolds dominate the medical device market, providing an optimal environment for cell proliferation and tissue repair. To date, the collagen used is of animal origin, entailing several risks including pathogens and allergic reactions. In addition, collagen extracted from tissues is a mixture of monomers and higher molecular weight collagen species which hamper its ability to assemble highly ordered structures with desired mechanical properties essential in scaffolds for regenerative medicine. Our goal was to produce a "virgin" human recombinant Type-I collagen in transgenic plants that will allow us to fabricate mechanically strong collagen scaffolds from liquid crystal solutions.

Materials and Methods

We have expressed five human genes in tobacco plants to produce fully functional triple helical recombinant human type-I collagen, identical to native human collagen. A wound dressing sheet was fabricated by fibrillogenesis, freeze drying and cross linking by thermal dehydration. Strong membranes were fabricated from liquid crystal solution of human recombinant Type-I collagen, by using a film coater followed by glutaraldehyde cross linking.

Results

A highly porous cohesive wound dressing sheet was obtained, capable of absorbing large quantities of fluids, up to 40 times its own weight in a few seconds. The wound dressing sheets demonstrated denaturation temperature higher than 60^{o}C and pore size of average 100µm, comparable to commercial products. In human cell culture studies with key cells that participate in wound healing; dermal fibroblasts and endothelial cells, proliferation of cells on the human recombinant collagen scaffold was superior to commercially available products. This biofunctionality is expected to jump start tissue repair and wound healing *in vivo*.

Membranes fabricated from liquid crystal solution of human recombinant Type I collagen were compared to membranes derived from pure bovine collagen. HNMR analysis clearly indicated that liquid crystal solution of human recombinant collagen is significantly more homogenous than a similar preparation of bovine collagen. Furthermore, the ultimate tensile strength of these membranes was twice higher than that of bovine.

Discussion and Conclusions

In this work, we have demonstrated superior attributes of collagen sheets fabricated from recombinant human collagen over commercial products made of animal-origin collagen. We utilized collagens unique liquid crystalline and self assembly properties to create ordered scaffolds from recombinant human Type I collagen. Overall, we present a novel, safe and functional biomaterial that outperforms both biologically and mechanically existing devices in the arena of regenerative medicine.