Electrospinning and hemostatic properties of gelatin/zein fibrous membranes

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Introduction

Advanced topical hemostatic agents are increasingly utilized to control traumatic hemorrhage. Numerous hemostatic agents including gelatin, collagen, chitosan and oxidized cellulose in various configurations such as powdered, granular, and flexible dressing formats are currently available. Microporous structure is designed to act as a sieve to dehydrate the blood and thus serve to accelerate the natural clotting process. Recently, a self-assembling peptide has been introduced to stop bleeding, which establishes a nanofiber barrier and incorporates it into the surrounding tissue to form an extracellular matrix (ECM). (Song H. Macromol. Biosci. 2010; 10: 33–39.) Electrospinning is a unique process for producing fibers with nanoscale diameters through the action of a high electric field. Nanofibrous scaffolds composed of electrospun fibers may promote platelet aggregation and coagulation by providing a three-dimensional meshwork for clotting to take place. Usually, electrospun scaffolds of biopolymer nanofibers require chemical crosslinking to maintain structural integrity in aqueous media. In the present study, electrospun gelatin/zein fibrous membranes were produced to fabricate nanostructured bicomponent scaffolds for hemostatic application. The fibrous membranes with a combination of hydrophilic and hydrophobic proteins without crosslinking showed structural integrity in contact with blood. Hemostatic properties and preliminary animal experiments of gelatin/zein composites were carried out to assess their potential applications as hemostatic dressings.

Methods

Gelatin (type B, from bovine skin) was purchased from Sigma Chemical Co. (St. Louis, MO, USA). Purified zein was purchased from Acros Organics (New Jersey, USA). Blend solutions at a final concentration of 24% (w/v) with various gelatin/zein weight ratios were electrospun to form fibrous membranes. Hemostatic properties of gelatin/zein fibrous membranes including fluid adsorption, whole blood clotting, platelet adhesion and thrombin generation were evaluated. For evaluating the hemostatic efficiency of gelatin/zein fibrous membranes, surgical procedures were performed on adult rabbits (white New Zealand rabbits, weighing 2.5-3.0 kg) in different models. Hemostasis was defined as the lack of active bleeding after pressure was released or within 1 minute (delayed bleeding). Bleeding time was noted after treatment with either gelatin sponge (control) or gelatin/zein fibrous membranes. Total blood loss was calculated by the weights of the sponge/membrane before and after removal from the wound site.

Results

The fibrous membranes exhibited uniform and smooth fibers morphology with various gelatin/zein weight ratios. After placed in phosphate buffer solution (PBS), the fibrous membranes, unlike other membranes made of hydrophilic biopolymers, did not disintegrate, even in the absence of crosslinking. After immersion for 2h in blood, the gelatin/zein fibrous membranes electrospun from blend solution with gelatin/zein weight ratio of 3/1 absorbed about 3 times its own weight (Fig. 1). There was no significant difference in fluid uptake between the fibrous membranes electrospun from blend solutions with gelatin/zein weight ratios of 1/2 and 1/1. In all animals with surgical applications, the fibrous membranes were adherent to the bleeding sites immediately. As shown in Fig. 2, placing the gelatin/zein fibrous membranes with gelatin/zein weight ratio of 3/1 to the bleeding site on the auricular vein of the rabbits, the bleeding time was 71.5±2.1s. On application of the fibrous membranes above trauma injury on auricular artery and liver, the arrest of bleeding took 75.5±4.9s and 87.5±2.1s, respectively.

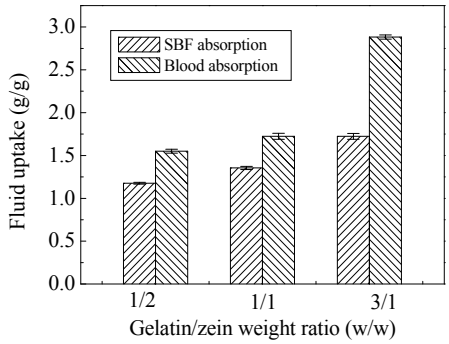

Figure 1. Blood and simulated body fluid (SBF) absorption capacities of gelatin/zein fibrous membranes

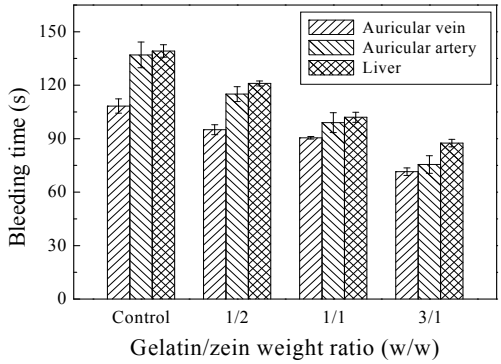

Figure 2. Hemostasis efficacy of gelatin/zein fibrous membranes in different models

Conclusions

Gelatin/zein fibrous membranes were electrospun and showed structural integrity in contact with fluids. The results suggested that the gelatin/zein fibrous membranes electrospun from blend solution with gelatin/zein weight ratio of 3/1 provided effective hemostasis of different models in preliminary animal experiments.