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The effect of ionized collagen for preventing postoperative adhesion

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ABSTRACT

Background: Collagen exhibits ideal multifactorial action for preventing tissue adhesions. This study examined the efficacy of ionized collagen in preventing tissue adhesion after surgical procedures.

Materials and method: Ionized collagen was prepared using the esterification technique of natural collagen. Three forms of collagen materials (membrane, film, and gel) were compared with three commercialized materials (oxidized regenerated cellulose membrane [OC membrane], hyaluronic acid and carboxymethylcellulose film, and gel [HC film and HC gel]) in a rat cecum abrasion model. Antiadhesive activity and histologic findings were assessed.

Result: The incidence of adhesion was reduced significantly in all test groups compared to the sham-operated control group (100% in control, 14.3% in collagen membrane, 63.6% in collagen film, 25.0% in collagen gel, 55.6% in OC membrane, 75% in HC film, and 83.3% in HC gel). All collagen materials of the three forms exhibited a significant reduction in adhesion grade compared with the sham operation, whereas no significant difference was found among these three different forms. The collagen membrane showed significantly less adhesion grade, less inflammation and more regenerative features compared to widely used conventional materials.

Conclusions: This preclinical investigation indicated that ionized collagen materials readily formed clinically suitable shapes for easy handling without the need for any complex processing and effectively reduced postoperative tissue adhesion profiles compared to conventional antiadhesive agents.

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Introduction

Postoperative adhesion is an inevitable issue in surgical patients that can cause intestinal obstruction,[1] chronic abdominal pain,[2] and infertility.[3,4] Multiple strategies to reduce the adhesions have been proposed including fibrinolytic agents, anti-inflammatory drugs, and antiadhesive barriers.[5,6] One of the promising results in clinical applications is a mechanical barrier between the damaged organ surface and abdominal walls. The barrier materials cover the damaged surface during the mesothelial regeneration. The chemical and physical properties of the materials should be considered for their application as antiadhesive agents. The materials should be biocompatible and should not induce inflammation, fibrosis, or encapsulation. Several types of antiadhesive materials have been studied and used in clinical settings. However, postoperative adhesions are still an important cause of surgical morbidity.

Collagen, which is the main component of the extracellular matrix, is one of the most suitable candidates for an implant material because of its excellent biocompatibility and physical properties.[7,8] Cross-linked collagen has been used in many medical applications.[9,10] Collagen, however, has some drawbacks in terms of the complexity of the fabrication process and limitations of clinical application in various surgical situations. In previous report, we developed the ionized collagen using esterification method. Owing to its water solubility, ionized collagen can be fabricated in various forms such as films, membranes, and gels. Also, esterified collagen has a net positive charge that can enhance cell attachment and proliferation.[11,12] Despite the excellent biocompatibility and biodegradability of collagen, it has not yet been fully developed as an adhesion prevention product.

The purpose of this study was to examine the efficacy of ionized collagen materials compared with commercially used antiadhesive materials in a rat cecum abrasion model for implementation of the novel antiadhesive agents in the clinic.

Materials and methods

Experimental animals and design

Male Sprague–Dawley rats, weighing 180-200 g, were used for the experiments. All animal experiments were performed according to the guidelines of the Institute of Laboratory Animal Resources, Asan Institute for Life Sciences, Asan Medical Center (Seoul, Republic of Korea; No. 2011-13-193). Adhesion lesions at the cecum and abdominal wall were performed on anesthetized rats according to the established protocols.[13-15] Three forms of antiadhesive esterified collagen (porous membrane, film, and gel) were compared with the control sham-operated group and commercially available antiadhesive agents including oxidized regenerated cellulose membrane (Interceed, Johnson &Johnson Medical Inc., Arlington, Texas), hyaluronic acid with carboxymethylcellulose film (Seprafilm, enzyme, Cambridge, Massachusetts), and hyaluronic acid and carboxymethylcellulose gel (Guardix gel, Hanmi medicare, Korea). Adhesion index, proportion and area were measured, and adhesion severity and strength were graded, 1 wk after surgery as shown in Table.[16-18] A total of 70 male rats were randomly divided into seven groups (10 rats in each group), including six treatment groups and a sham group.

Fabrication of ionized collagen-based antiadhesive materials

Three forms of antiadhesive collagen materials, including a porous membrane, film, and gel, were fabricated (Fig. 1A). For preparing the esterified collagen, lyophilized type I atelocollagen (DalimTissen Co. Ltd., Seoul, Korea) was immersed and agitated in ethanol, and then, the collagen suspension was neutralized with 0.5-M NaOH and centrifuged.[19] For fabricating the collagen films, collagen solution (w/v, pH 7.4) was poured into a petri dish and air-dried. Porous membranes were prepared by lyophilization of collagen solution. The film and membrane were cross-linked with 1-ethyl-3-(3-dimethylaminopropyl) carbodiimide (EDC) in ethanol for 24 h. Residual EDC and ethanol were washed out with autoclaved distilled water five times. Additionally, the 1% (w/v, pH 7.4) collagen gel was prepared by solubilizing in physiological sterilized saline.

Physical properties of collagen

To confirm the solubility of esterification of the natural collagen, the viscosity of natural collagen and esterified collagen solution (0.8 w/v%) were assessed by a digital viscometer (Model DV-II + Pro, Brookfield). Water uptake ability of the esterified collagen was determined by measuring the contact angle of the collagen film. The contact angle was measured using the sessile drop image. All contact angle values were obtained from five measurements.

For the assessment of weight loss of each material, each specimen was measured for initial weight and was then placed in a tube containing 5 mL of pH 7.4 PBS buffer. The samples were transferred from the vials and replenished with fresh medium twice a week for up to 8 wk; samples were then freeze-dried, and their weight loss was measured. The collagen compound was compared with commercialized cellulose film (SurgiWrap, Mast Biosurgery, California).

Table – Classification of severity and strength grades of the peritoneal adhesion.

Adhesion severity	Description
0	No adhesion
1	Thin filmy adhesion
2	More than one thin adhesion
3	Thick adhesion with focal point
4	Thick adhesion with planar attachment
5	Very thick vascularized adhesions or more than one planar