High-density collagen patch prevents stricture after endoscopic circumferential submucosal dissection of the esophagus: a porcine model

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Background and Aims: Extensive excision of the esophageal mucosa by endoscopic submucosal dissection (ESD) frequently evokes a luminal stricture. This study aimed to determine the efficacy of a high-density collagen patch for the prevention of esophageal stricture in extensive ESD.

Methods: Six pigs underwent circumferential esophageal ESD under general anesthesia. In 3 pigs, artificial ulcers were covered by 2 collagen patches. The other 3 pigs underwent circumferential ESD only.

Results: The 2 collagen patches were settled onto the ulcer surface using a general endoscope and instruments. The collagen patch-treated group showed significantly better patency rates on both the oral and anal sides of the wound area compared with the control group at day 14. The mucosal re-epithelization ratio was significantly promoted, and the extent of mucosal inflammation and fibrosis was significantly decreased with the collagen patch treatment in the wound area. The frequency of cells positive α-smooth muscle actin was significantly reduced in the collagen patch-treated group compared with the control group.

Conclusions: We have established a high-density collagen device that can reduce the esophageal stricture associated with extensive ESD. This easy-to-handle device would be useful during superficial esophageal cancer treatment by ESD. (Gastrointest Endosc 2017;85:1076-85.)

Endoscopic submucosal dissection (ESD) was originally developed as a resection procedure for stomach neoplasms.[1] Recently, ESD has been used for esophageal squamous cell neoplasms.[2,3] Esophageal ESD is a revolutionary treatment technique because it is a much less invasive procedure than conventional surgical treatment. However, postoperative stricture frequently develops after extensive esophageal resection by ESD.[4] Although this stricture causes dysphagia and is life-threatening, no methods have yet been established to prevent the esophagus from constricting after ESD.[5]

Abbreviations: αSMA, α-smooth muscle actin; CVP, collagen vitrigel patch; CTGF, connective tissue growth factor; ESD, endoscopic submucosal dissection; HE, hematoxylin-eosin; MD, mucosal dissected tissue diameter; ND, non-epithelized tissue diameter; PSR, picrosirius red.

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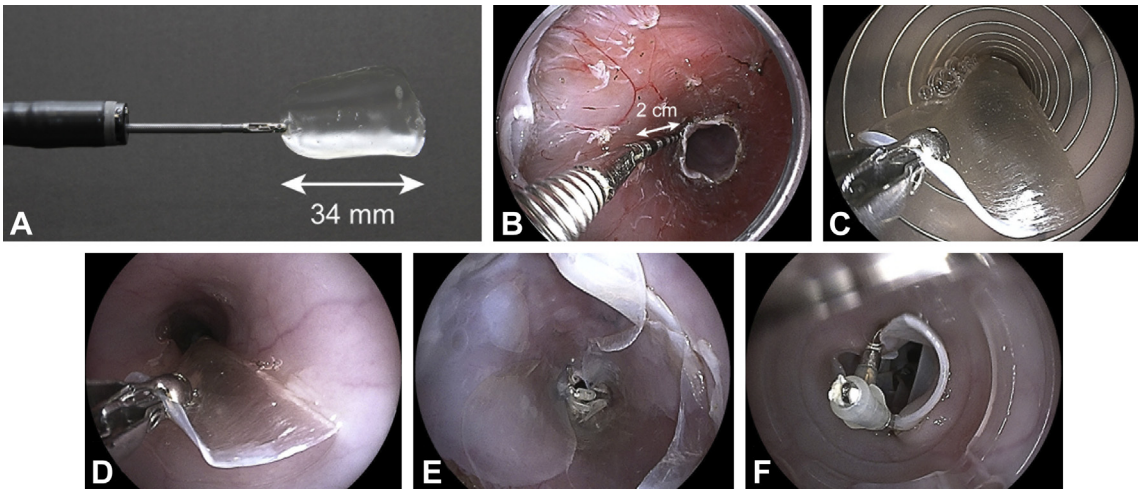

Figure 1. Collagen vitrigel patch (CVP) and esophageal endoscope. **A**, The appearance of the CVP grasped with biopsy forceps. **B**, Circumferential mucosal dissection. **C**, The grasped CVP is passed through the overtube. **D**, The grasped CVP is passed through the esophageal lumen. **E,** Artificial ulcer covered by 2 CVPs. The CVPs are clipped to the mucosa at the ulcer edge on the anal side. **F**, The CVPs are clipped to the coverage edge of the normal mucosa on the oral side.

Collagen vitrigel is a collagen-based biomaterial consisting of high-density collagen fibrils equivalent to connective tissues in vivo that is created by a vitrification process.[6] Collagen vitrigel membranes possess excellent transparency, mechanical strength, and permeability to proteins with high molecular weight.[7] Collagen xerogel membranes, defined as dried collagen vitrigel membranes without free water, can be prepared by simply re-vitrifying collagen vitrigel membranes on a separable sheet.[8] We previously established an artificial skin based on collagen vitrigel and demonstrated that it can promote epithelization of regenerative skin and inhibit scar formation and inflammation.[9] The surface of the esophagus and skin is covered by a squamous cell layer, and fibroblasts are located in the respective subepithelial connective tissue, ie, the lamina propria and dermis.[10] Furthermore, myofibroblasts play a central role in pathologic scar formation and luminal stricture in these organs.[11,12] Considering the histologic similarity and biological commonality between the esophageal mucosa and skin, we developed a collagen vitrigel patch (CVP) as a device to prevent esophageal stricture after extensive ESD.

The purpose of the present study was to assess the possibility of using the CVP as a stricture-preventing device after extensive resection by esophageal ESD. We evaluated the efficacy of the device after circumferential mucosal dissection in a porcine model to determine whether the newly devised membrane is feasible for clinical use.

METHODS

All experimental procedures were approved by the Institutional Animal Ethics Committee of IVTec (Kobe, Japan) and Saga University. Six 24-week-old female pigs weighing 30.3 to 38.6 kg (LWD pigs; Aratoyama SPF Farm, Okayama, Japan) were used. The pigs were regarded as healthy based on daily visual inspections during an acclimatization period of 7 days. The pigs were fed a commercial semisolid swine feed (MP-A; Oriental Yeast, Tokyo, Japan) and housed in groups during the acclimatization period.

Collagen vitrigel and CVP

A CVP appropriate for esophageal mucosa regeneration was prepared by reference to related studies.[9,13] A raw material of 0.5% atelocollagen solution was prepared by uniformly mixing equal volumes of 1.0% acidic solution of porcine-derived atelocollagen for regenerative medicine (Nippon Meat Packers, Tokyo, Japan) and serum-free Dulbecco's modified Eagle's medium, supplemented with 20mM N-2-hydroxyethylpiperazine-N'-2-ethanesulfonic acid, 100 units/ml penicillin and 100 ug/ml streptomycin (Gibco, Rockville, Md). A collagen vitrigel membrane containing 11.0 mg collagen/1.0 cm^2 unit area was fabricated by gelation, vitrification, and rehydration with phosphate-buffered saline solution after pouring 20.0 mL of the porcine-derived atelocollagen solution into a separable container comprising a cylinder with an inner diameter of 34 mm and a plain-bottom plate wider than the cylinder's outer diameter. Subsequently, the membrane was converted to a collagen xerogel membrane by revitrification on a separable sheet. This round-shaped collagen xerogel membrane comprised a CVP for an artificial esophageal ulcer (Fig. 1A). The CVP had a diameter of 34 mm and thickness of 70 to 80 μm.

ESD procedure

After anesthetization, an overtube (JMDN70244000; TOP Corporation, Tokyo, Japan) was inserted into the esophagus. An artificial ulcer (2-cm-long dissection) in the lower esophagus was prepared by a standard ESD