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Original Article

A sutureless method for digestive tract reconstruction during pancreaticoduodenectomy in a dog model

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Abstract: Development of pancreatic fistulas as a result of anastomotic gaps is still a major complication after pancreaticoduodenectomy, and can cause post-operative death. Therefore, safer and more effective methods of anastomosis are needed to avoid leakage and decrease mortality. Materials and methods: Twenty domestic dogs with body weights ranging from 15 to 25 kg were used, regardless of gender. A model of common bile duct and pancreatic duct dilatation was surgically prepared in these dogs. Pancreaticobiliary stents combined with magnetic anastomoses (PB-MA), and controls were treated with fibrin glue were studied in terms of and efficacy by measurement of serum amylase, incidence of complications, and survival times. Results: The mean time required to create the fibrin glue pancreaticoenterostomy was 9 ± 2.05 min, while the mean time required to create the magnet cholangioenterostomy was 5 ± 0.9 min. The total operative time was 2.7 ± 0.6 h. Eighty percent of the dogs that underwent the operations were still alive for 15 days after the operations and none developed pancreatic fistulas. Examination by macroscopic observation, and hematoxylin and eosin staining of the pathological specimens showed that the anastomoses were completed healed. Conclusions: The use of a PB-MA in sutureless digestive tract reconstruction for pancreatoduodenectomy resulted in an elimination of pancreatic fistulas, and shortening of the stent removed time. In addition, the procedure is simple to perform, fast, and appears to be safe in this dog model.

Keywords: Pancreatic fistula, pancreaticoduodenectomy, magnetic sutureless anastomosis

Introduction

Although, pancreaticoduodenectomy has improved dramatically as a therapeutic operation for the treatment of carcinoma of the head of pancreas, ampulla and chronic pancreatitis, there are still many deficiencies. Pancreatic fistula (PF) continues to be a major complication, ranging from 3% to 26% [1-7], usually caused by inadequate anastomosis, long operation time, and prolonged requirement for internal stents. To solve these problems, surgeons have attempted to improve the operative technique using binding panreaticojejunostomies [8], external or internal stents [9, 10] and pancreaticogastrostomies [7, 11]. However, these techniques are not widely applicable. Moreover, they have not significantly decreased the incidence of complications.

Previous studies have showed that PF is caused by gaps in sutured anastomoses, due to increased pressure in the jejunal lumen, and digestion by pancreatin which is activated by bile and intestinal juice. Roux-en-Y choledochojejunostomy is a common operation for bypassing extrahepatic biliary obstructions and establishing biliary-enteric continuity after resections for benign or malignant biliary diseases [12, 13]. The traditional procedure is also time consuming which may also be directly related to the PF: 1. The potential gaps between the sutures of the anastomosis can enable pancreatic juice to leak out ; 2. Postoperatively, bile and pancreatic juice frequently accumulate in the jejunal lumen, leading to increases in anastomotic tension, which can cause a pancreatic fistula . 3. The location of the suture can result in poor blood supply and healing. 4. Bile mixed with pancreatic juice in the jejunal lumen after operation can activate trypsin resulting in digestion of the anastomosis. Although stapled anastomoses have been used in an attempt to

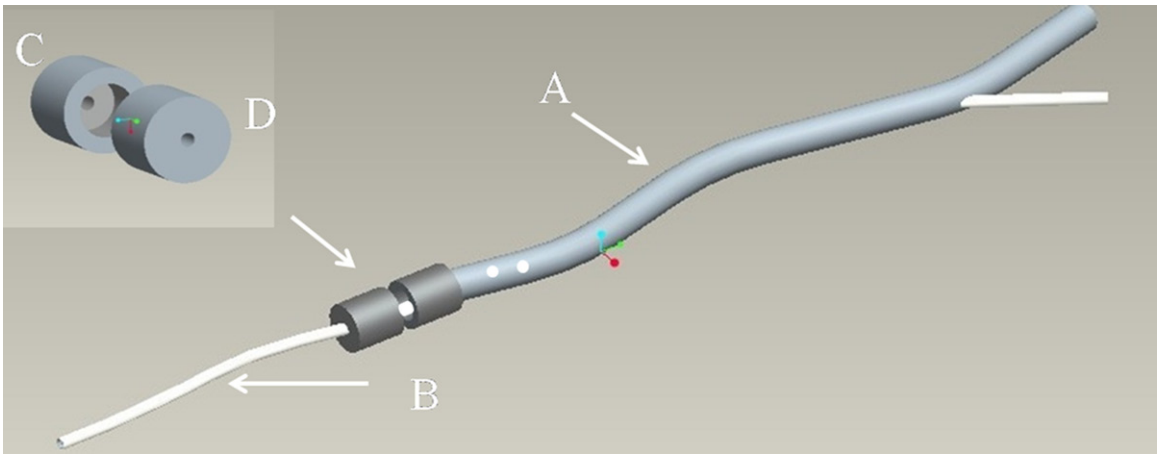

Figure 1. The structure of the PB-MA. A: Stent A (bile duct stent). B: Stent B (pancreatic duct stent). C: Magnet C with two central holes (a bigger central hole on one side, a smaller one on the other side). D: Magnet D with a central hole.

decrease the operation time, the diameter of the common bile duct cannot be sufficiently widened to accommodate the stapler [14].

Surgeons have attempted to utilize the advantages of magnetic compressive anastomoses for choledochojejunostomy to reduce operation time [15-19]. Magnetic compression anastomosis has proven to be a safe surgical technique that is equivalent or superior to anastomoses created by traditional sutures or stapling techniques [20].

The aim of current study was to design a PB-MA for sutureless digestive tract reconstruction for pancreaticoduodenectomy, shunt the pancreatic juice and bile completely, simplify the anastomosis technique, reduce operation time and external stent time.

Materials and methods

The pancreaticobiliary combined stent with magnet

PB-MA consisted of four components (**Figure 1**): stent A (bile duct stent), stent B (pancreatic duct stent), magnet C and magnet D. Stent A and B were made of polyvinylchloride. The length of stent A was 250.0 mm and the diameter was 6.0 mm. Stent B was 450.0 mm length and the diameter was 1.8 mm. Both stents had 4 side holes. Magnets C and D had the same height (5.0 mm), diameter (6.0 mm), and inner diameter of the central hole (2.2 mm). Magnet C had a larger central hole with the 4.0 mm inner diameter and a 2.5 mm depth at one side. Magnets C and D were made of neodymium-iron-boron, NdFeB (Northwest Institute For Non-ferrous Metal Research, China), and the stent was remade by ourselves using polyvinylchloride.

Animals

Twenty domestic dogs were used regardless of gender, with body weights ranging from 15 to 25 kg (where did you get these dogs). The study was performed at the Surgical Dream Works Laboratory in Medical College of Xi'an Jiaotong University, following approval by Institutional Research Committee, and the authority of Xi'an prefecture for experimental animal protocols. All dogs received humane care in compliance with the Guide for the Care and Use of Laboratory Animals published by the National Institutes of Health. The dogs were maintained on a standard light-dark cycle.

Operation technique

Following with 12 h fasting and 6 h water deprivation, twenty dogs were anaesthetized with pentobarbital, 0.03 mg/kg, i.p.. Before operation, all dogs were intubated, ventilated with supplemental oxygen and continuous O_2 monitoring. No urinary catheterization was used in this operation. The saphenous veins were used to establish intravenous access for intraoperative and post-operative IV hydration. Animals were given benzyl penicillin sodium before incision and at the end of the operation, to prevent

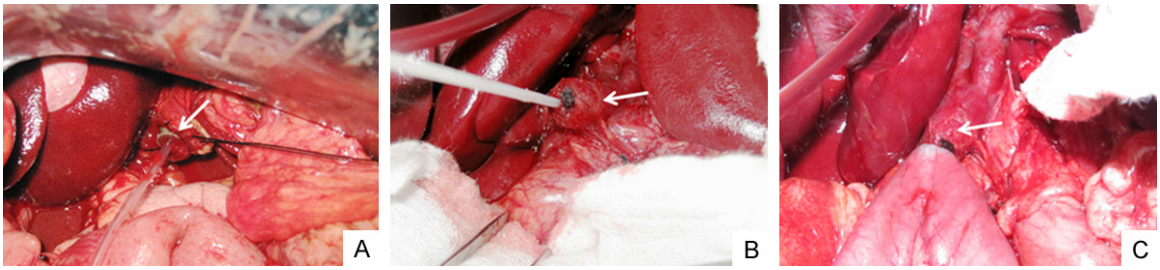

Figure 2. A: After the stent B was passed through the central hole of magnet C, the magnet C was fixed in the stump of the common bile duct. B: The magnet C was purse-string sutured with 3-0 Prolene in the stump of the common bile duct. C: The biliary enteric anastomosis was constructed with the magnet C and magnet D attracting.

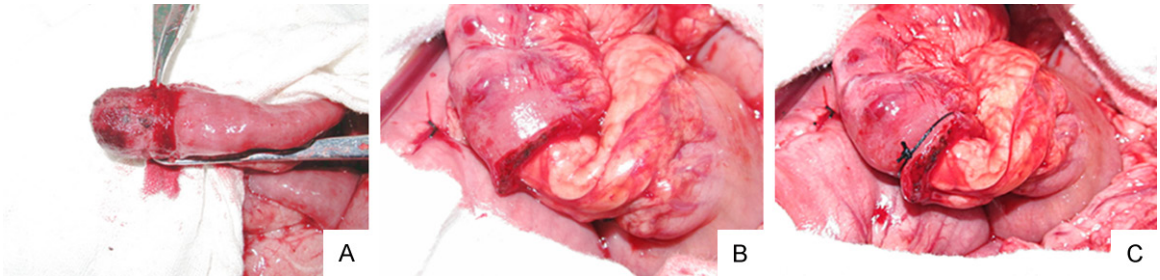

Figure 3. A: The distal cut end of the jejunum was everted and destroyed the exposed jejunal mucosa. B: The pancreatic stump was wraped after fibrin glue was applied. C: Looped around the entire circumference of the anastomosis with a suture 1 cm from the cut end of the jejunum.

peri-operative infection. Once an animal was infected, another appropriate antibiotic would be used, such as gentamicin, ornidazole, and cephalosporin.

All procedures were aseptic. After skin preparation, the abdomen was entered through the *linea alba*. The left lobe and up to 1 cm of the right lobe of the pancreas and the lower common bile duct were ligated to prepare the pancreatic duct and common bile duct dilatation model. After 8 d, reoperation was performed to resect the right lobe of pancreas and the lower common bile duct without resection of duodenum and gall bladder. The length of the free pancreatic stump was cleaned to expose approximately 3.0 cm. Hemostasis was obtained with fine nylon suture ligatures. The 1st loop of the jejunum was identified and transected. Then, stent A and B were passed into the liver parenchyma via the bile duct, and stent B was passed through the central hole of magnet C with the larger central hole facing the outside, and the magnet C was purse-string sutured with 3-0 Prolene® (Ethicon; Johnson & Johnson, Somerville, New Jersey, USA) at the stump of the common bile duct (**Figure 2A**, **2B**). The jejunum was then punched 10 cm distal to the raised loop in order to pass through stent B. Magnet D was precisely coupled to the magnet C guided by stent B protruding through the central hole of the magnet D. The biliary enteric anastomosis was constructed with the magnet anastomat (combination of magnet C and D) (**Figure 2C**). The sinus tract of liver was treated with fibrin glue and absorbable gelatin sponge.

The stump of the pancreatic remnant was exposed for a distance of 3 cm. The pancreatic duct was defined, and the stent B was inserted to a distance of 3-5 cm. Three centimeters of the distal cut end of the jejunum was everted. The exposed jejunal mucosa was removed either by electrical coagulation (**Figure 3A**). Contact between the everted jejunum and the pancreas was maintained by suturing each side of the anastomosis with one stitch, but without piercing the jejunal serosa. The exudates on the surface of the everted jejunum and the pancreatic were wiped clean. Fibrin glue was evenly applied to the pancreas and the everted jejunum. The everted jejunum was then returned to its normal position to wrap over the pancreatic stump (**Figure 3B**). A 1-0 Vicryl suture was

Table 1. The complications after operation

	Cases	Time of death	Cause of death
Bile leakage	2	on the 3rd day	The dog liver was too thin and too soft that could not retract after the PB-MA through the liver. The bile flowed into the abdominal cavity by the way of the PB-MA leading to acute bile peritonitis.
The pulling of the PB-MA	1	on the 4th day	The stent was dislodged leading to anastomotic dehiscence.
MODS*	2	on the 5th day	Electrolyte imbalance

*Multiple Organ Dysfunction Syndrome.

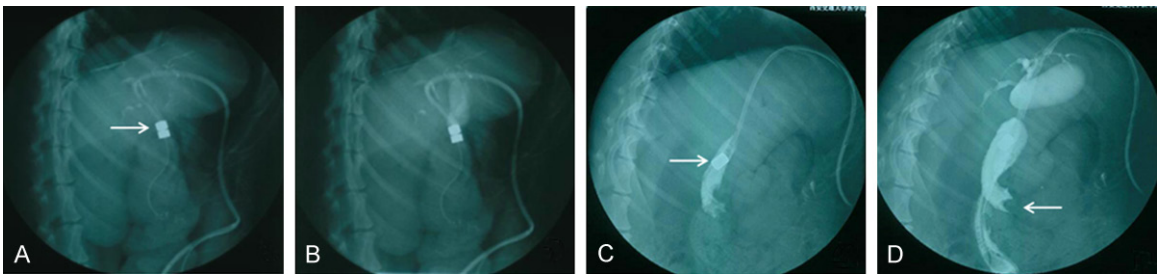

Figure 4. A, B: The 1st postoperative day radiography imaging; C, D: The 10th postoperative day radiography imaging. A: The PB-MA was introduced, '→' with the magnet anastomat. B: The common bile duct was introduced, but the contrast-medium did not flow out to jejunum. C: The biliary-enteric anastomosis was completed with the magnet ('→') in the jejunal lumen. D: The contrast-medium did not leak into the abdomen through any anastomosis. '←' indicates the pancreatojejunal anastomosis.

looped around the entire circumference of the anastomosis, 1 cm from the cut end of the jejunum. A bundle of vessels for maintaining the blood supply to the jejunal cut end distal to the binding ligature was preserved (**Figure 3C**).

The gut continuity was restored with an end to side jejunojejunal anastomosis in a Roux-en Y fashion, using a Stapler (SDH25; Johnson & Johnson, Somerville, New Jersey, USA). Finally, the abdomen was closed with a running 1-0 polyglycolic acid suture, and the abdominal skin was closed with a running 4-0 polyglycolic acid stitch.

Postoperatively, all animals received benzyl penicillin sodium for 3 days, and parenteral nutrition consisting of medium/long-chain fat emulsion infusion (250 ml of 20% Huarui Pharmaceutical Co., Ltd., force and energy c): 10% glucose infusion (provided by the First Affiliated Hospital Medical College of Xi'an Jiaotong University) in a 1:2 proportion to provide, 25 kcal/kg per day via the femoral vein. The dogs were not fed solid food for 2 days after the operation, but water was freely available. After 2 days, the food supply (pellet food; Jiruihe Pet Food Sales Co., Ltd., Tianjin, China) was gradually increased to twice per day (200-250 g at each feeding). Radiography imaging via PB-MA was carried out using cardiografin on 1st and 10th postoperative days using a C-arm X-ray machine PLX7000B (PERLOVE Nanjing, China). The PB-MA was removed on 12th postoperative day. On the 15th postoperative day, the dogs were anaesthetized with pentobarbital 0.03 mg/kg i.v, and re-explored to observe the anastomotic healing. Then, the dogs were sacrificed using 2 g of KCl i.v. The biliary-enteric and pancreatojejunal anastomoses were excised and inspected for evidence of defects or abscess. All samples were fixed in 10% formalin at pH 7.0 and processed according to standard histological procedures. Sections (4-6 μm) were stained with hematoxylin and eosin for light microscopy. The presence of pancreatic fistulas was diagnosed by radiologic examination and re-exploration.

Results

The baseline serum amylase concentration in normal dogs was 597.9 ± 96.6 IU/L, The serum amylase on the 8th day post pancreatic duct ligation was 1258.0 ± 55.5 IU/L. Inner diameter of dog left pancreatic duct was 1.0 ± 0.2 mm pre-ligation and increased to 2.0 ± 0.48 mm 8 days after ligation. Post-ligation, the pan-

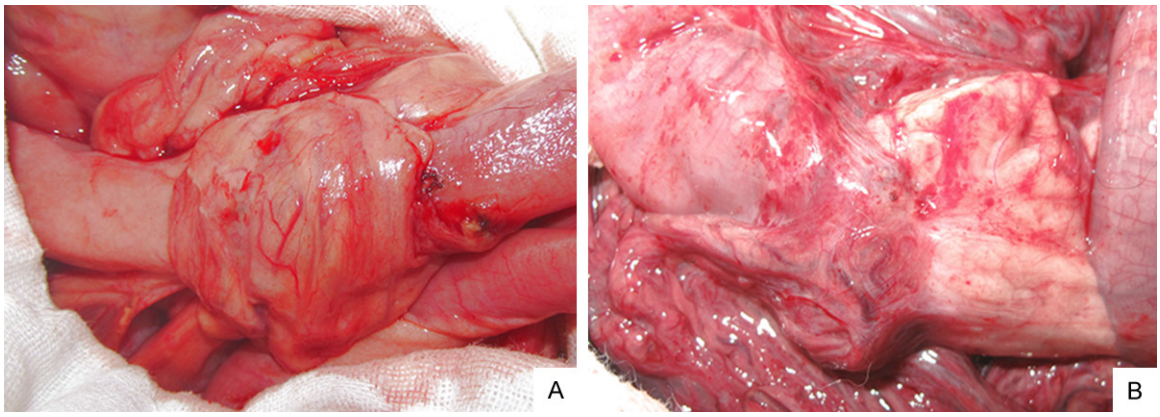

Figure 5. A, B: The intact pancreatojejunal anastomosis on the 15th postoperative day.

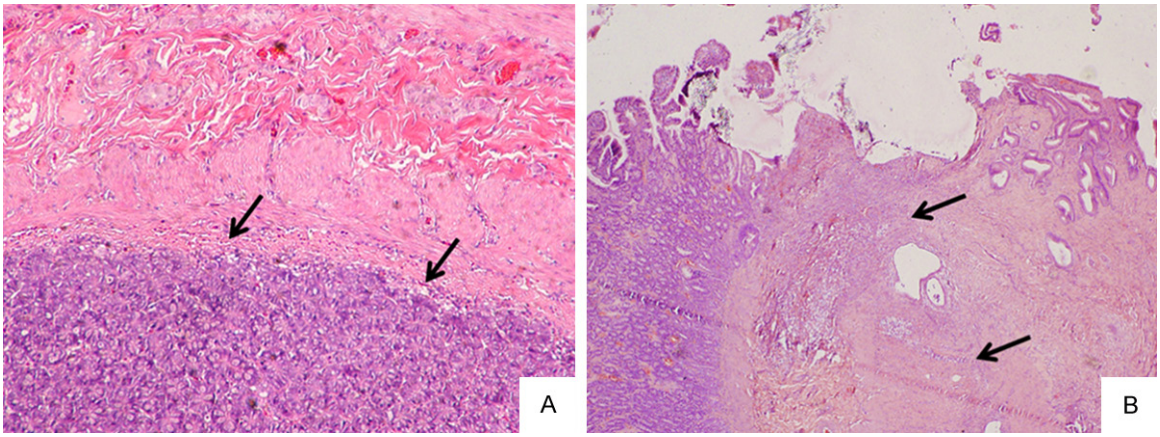

Figure 6. A: Cross section of the pancreatojejunal anastomosis on the 15th postoperative day (arrows indicate the junction of the pancreas and jejunum). B: Longitudinal section of the biliary-enteric anastomosis on the 15th postoperative day (arrows indicate junction of the bile duct and jejunum).

creas had mild edema, without gross hemorrhagic or exudative changes. The pancreatic texture was slightly hard, and the capsule intact.

There were no intraoperative deaths. The mean time required to perform fibrin glue pancreaticoenterostomy was 9 ± 2.05 min, while the mean time required to perform the magnet cholangioenterostomy was 5 ± 0.9 min with a total operative time of 2.7 ± 0.6 h. The dog serum amylase was 935.9 ± 76.4 IU/L on the 15th postoperative day. Sixteen dogs survived until the end of the experiment (15th postoperative day). Four animals died on 3rd to 5th postoperative day because of the complications in **Table 1**.

On the 1st postoperative day, stent A (bile duct stent) drained about 154.5 ± 43.2 ml bile per day while stent B (pancreatic duct stent) drained about 14.8 ± 4.9 ml pancreatic juice per day. From the 5th day on, stent A drainage reduced gradually, ranging from 20 to 45 ml, but there was no obvious change for stent B.

Radiographic imaging showed that the PB-MA was clear and unobstructed. There was no shift in position or anastomotic leakage (**Figure 4**). The contrast-medium could enter the jejunal lumen on the 1st postoperative day (**Figure 4B**). Radiographic imaging on the 10th postoperative day showed that the contrast-medium flowed smoothly into jejunum confirming that the biliary-enteric anastomosis was patent, and not strictured (**Figure 4C**).

Autopsy of the surviving animals revealed multiple adhesions around the pancreatojejunal anastomosis without signs of pancreatic fistu-

las, peritoneal inflammation or any other intraabdominal pathologic changes (**Figure 5**).

Examination of sections of the anastomosis stained with hematoxylin and eosin confirmed that the anastomosis was intact. The pancreatic parenchyma was observed to be joined closely with the jejunal muscularis without any inflammatory reaction in the form of a thin band of granulation and fibrous tissue at the contact area between them (**Figure 6A**). The healing process of biliary-enteric anastomosis was almost complete with a high level of neutrophils infiltration n the lamina propria (**Figure 6B**).

Discussion

The concept of sutureless anastomosis of a hollow viscus with the aid of surgical sealants has been investigated in a number of experimental studies, most of which have used fibrin glue [21]. There are some advantages to this technique such as reduced operating time, simplicity of use, and lower leakage rate. Recently, fibrin glue has been used after pancreatic resections in various preclinical and clinical settings with favorable results [22-26]. Neodymium-iron-boron (NdFeB), a third-generation rare-earth permanent magnetic alloy is relatively inexpensive. Recently, Chao et al [27] reported that using the NdFeB in a dog choledochojejunostomy model was time-saving, safe and efficient.

In this study, we also showed that PB-MAthe PB-MA can shunt pancreatic juice and bile completely avoiding bile activation of pancreatic enzymes. Postoperatively, the PB-MA could be removed earlier than current clinical stent. At present, clinical pancreatic duct stents are mainly made from silica gel which require a long time for a sinus tract to fully form. Therefore, the stent should not be removed sooner than 1 month post-operation. The requirement leads to an increase in time with stents in place, and a poor postoperative quality of life. We used the PB-MA to pass through the liver parenchyma because the liver parenchyma is elastic. After replacement or removal of the PB-MA, the sinus tract can be closed by natural retraction of the liver to avoid hemorrhage and leakage of pancreatic, intestinal juice or bile along the sinus. Five days after operation, as a result of the completion of biliary-enteric anastomosis, most of the bile flowed into the intestinal lumen. In line with the preliminary experimental results, the anastomat resulted in biliary-enteric anastomosis within 3-5 days in the experimental group [27]. Therefore, it was possible to safely remove the stent on the 12th postoperative day.

Pathological examination of the biliary-enteric and pancreatic anastomoses revealed that healing had occurred and the anastomoses were closely connected. The fibrin glue was completely absorbed which did not affect the normal healing of the anastomosis. A negative result was observed in 5 dogs which died early in the postoperative period. The reasons were: 1. The dog liver was too thin and too soft so that it could not be retracted after the PB-MA passed through the liver [28]. Bile flowed into the abdominal cavity. However, human liver is thicker, and has a certain toughness and elasticity, so it may be suitable for clinical use in percutaneous transhepatic biliary drainage. 2. The PB-MA was dislodged by the dog leading to anastomosis dehiscence. 3. Poor tolerance to surgery, leading worse postoperative recovery.

This surgical technique is simple, less demanding than suturing, easy to complete, and may be suitable for clinical applications. The shortened exposure time of the abdominal viscera, potentially minimizes postoperative risk of infection may be conducive to patient recovery, and a shortened hospital stay.

In conclusion, the use of PB-MA for sutureless digestive tract reconstruction for pancreatoduodenectomy resulted in elimination of pancreatic fistulas, and a short stent removed time. In addition, the procedure is simple to perform, is not time-intensive, and appears to be safe in a dog model in terms of 0% biliary-enteric anastomosis leakage and pancreatic anastomosis leakage. If this procedure is performed on the clinic, it may shorten the operation time, improve postoperative recovery, and shorten the hospital stay. In addition, the procedure may be performed without requiring extensive training or special skills.

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Disclosure of conflict of interest

None.

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