



Dual-Incision Laparoscopic Spleen-Preserving Distal Pancreatectomy: Merits Compared to the Conventional Method

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Abstract

Background Herein, we assess the safety and feasibility of dual-incision laparoscopic spleen-preserving distal pancreatectomy (DILSPDP) through lateral approach with reduced trocars for benign and low-grade malignancy in pancreas tail. We compare DILSPDP with surgical outcomes of conventional laparoscopic spleen-preserving distal pancreatectomy (LSPDP).

Methods Patients with benign pancreas tail mass that had been scheduled for LSPDP were selected to undergo DILSPDP. These patients had spleen-preserving distal pancreatectomy with the dissection in lateral-to-medial fashion using a multichannel trocar in the right lateral decubitus position of patient. We compared the demographics and operative outcomes of DILSPDP with those of conventional LSPDP which was performed with dissection in medial-to-lateral fashion using four or five trocars in supine position.

Results Twenty two cases of DILSPDP and 26 cases of conventional LSPDP were reviewed. There was no difference in terms of demographic features including diagnosis or tumor size, although the location of the tumor was fundamentally different between the two groups. Significantly shorter operative times and reduced blood loss were observed in DILSPDP group ($p = 0.004$ and 0.011 , respectively) and the preservation of splenic vessels was more successful with DILSPDP than conventional surgery (95.5% vs. 65.4% , $p = 0.013$).

Conclusions DILSPDP appears to be a feasible method of spleen-preserving distal pancreatectomy for benign or low-malignancy of pancreas tail and is accompanied by advantages in terms of splenic vessel preservation and reduced parietal trauma.

Keywords Dual incision · Spleen-preserving · Distal pancreatectomy · Laparoscopy

Abbreviations

DILSPDP	Dual-incision laparoscopic spleen-preserving distal pancreatectomy
LSPDP	Laparoscopic spleen-preserving distal pancreatectomy
MIS	Minimal invasive surgery
LDP	Laparoscopic distal pancreatectomy

Introduction

Spleen-preserving distal pancreatectomy may be achieved with either complete preservation of, or with sectioning of splenic vessels called the Warshaw method.¹ Because the sacrifice of splenic vessels could result in insufficient blood supply to the spleen which can cause morbidities such as splenic abscess or infarction,^{2–4} the complete preservation of vessels is theoretically the ideal option and would avoid these disadvantages. However, its technical complexity and the difficulty of dissection compared to the Warshaw method has been an obstacle to its adoption as a widely used procedure. In fact, splenic vessel preservation is very difficult in cases where the mass is located on the far distal portion of pancreas because the splenic vein near the pancreas tail usually runs with embedded pancreatic parenchyma and it can easily create bleeding during dissection that results in the sacrifice of these vessels.

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Minimally invasive surgery (MIS) has recently seen improvements in terms of reducing the number of trocars, resulting in less parietal trauma during surgery, and the scope of its application has gradually increased as reported in various fields of surgery such as appendectomy,⁵ herniorrhaphy,⁶ or cholecystectomy.⁷ Although adoption of MIS for pancreas surgery has also increased, attempts to perform MIS with reduced numbers of trocars, are rare in cases of splenic vessel-preserving laparoscopic distal pancreatectomy (LDP).^{8–11}

In 2015, we were the first to design the dual-incision laparoscopic spleen-preserving distal pancreatectomy (DILSPDP) which is a novel technique for LSPDP through lateral approach with reduced trocars and has been applied to masses located on pancreas tail. Herein, we analyzed the surgical outcomes of DILSPDP, comparing it with the conventional method to assess feasibility and safety of technique.

Materials and Methods

From March 2015 to February 2018, all patients admitted at the Department of Surgery, Seoul St. Mary's Hospital and scheduled for LSPDP were enrolled in this retrospective study. All operations were performed by two specialized pancreas surgeons at our institution. The indications for surgery were cases with pancreas tumor in the pancreas body or tail which were presumed as benign or of low-grade malignancy based on preoperative radiologic study, or tissue pathology obtained from ultrasound-guided biopsy. Cases, in which the tumor exceeded the right lateral border of superior mesenteric vein toward the right side of body, were not applicable for spleen-preserving LDP. While the conventional LSPDP had been performed by two surgeons (You and Hong), surgeon Hong first introduced the DILSPDP method in March 2015 as previously reported.⁸ This method was consecutively applied for selected cases in which the tumors were located in the pancreas tail dominantly. That means the central portion of the mass exceeds the left lateral border of aorta toward the pancreas tail on preoperative imaging. In all cases, the preservation of splenic vessels and spleen was the original operative strategy. However, the Warshaw method or even LDP with splenectomy was conducted under specific conditions in which the dissection of vascular structures was too difficult, or it bled too seriously.

All patients underwent the same clinical protocol during hospitalization regardless of the type of surgery. The patients were given parenteral nutritional support on the first day after operation if their vital signs were stable, and then oral feeding was resumed routinely on postoperative day 3, unless there was any evidence of pancreas leakage. Unless the case had obvious evidence of postoperative pancreatic fistula (POPF) clinically, the octreotide was not used routinely. On postoperative day 5, patients were evaluated by abdominal CT scans to confirm the

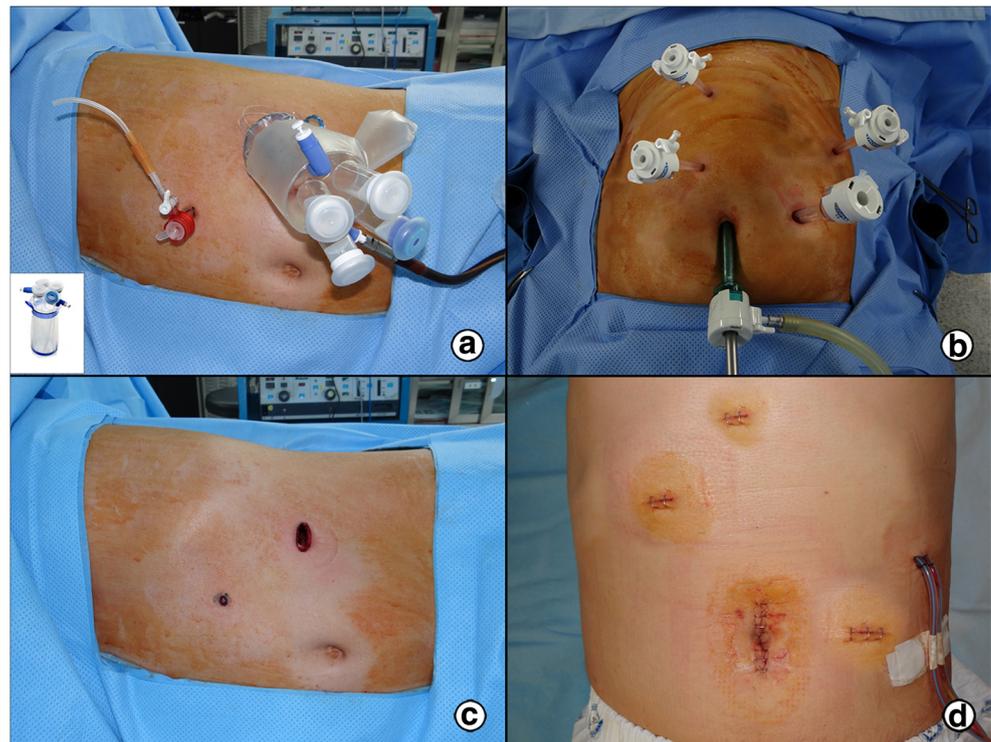
absence of pancreas leakage or any other intraabdominal complications. The drains were kept at least 5 days after surgery and removed if the drainage volume was less than 100 ml per day without evidence of bleeding or pancreas leakage. For analysis of postoperative pain, the visual analysis scale (VAS) was assessed from 0 (no pain) to 10 (worst pain imaginable) at first, third, and fifth postoperative day. Postoperative morbidities were reviewed and classified according to the Clavien-Dindo classification.¹² POPF was defined according to the revised definition of the International Study Group of Pancreatic Fistula,¹³ as the drain output of any measurable volume with an amylase level greater than three times the upper limit of normal serum amylase values, associated with a clinically relevant condition related directly to POPF which requires changes in the postoperative management.

Operative Technique

Dual-Incision Laparoscopic Spleen-Preserving Distal Pancreatectomy

Under general anesthesia, the patient was placed in the right lateral decubitus position in a reverse Trendelenburg position and then bent at the waist to increase the space between the subcostal margin and the iliac crest. We made a 3-cm transverse incision at the mid-clavicular line in the left mid-abdominal quadrant, and this was extended down to the layer of the abdominal fascia. (Fig. 1a) We used a multichannel trocar (Glove port, Nelis, Seoul, Korea), which was composed of four trocars and two rings with gas insulation and exsufflation gates. Specific details about this specially designed trocar can be found in our previous report.⁸ This trocar provided access for a 10-mm 30-degree laparoscope and a 12-mm endovascular stapler (endoscopic linear stapler) and 5-mm laparoscopic instruments all together. Use of this specialized multichannel trocar enabled the operator to manipulate several instruments using his right hand, simultaneously with a laparoscopist. An additional 5-mm trocar was placed in the subxiphoid area and used as the access point for an additional 5-mm laparoscopic grasper or dissector; it served as the working channel for the operator's left hand. This method created a more ergonomic environment, enabling the operator to use instruments with both of his hands harmoniously. If the omentum attached to the stomach is too bulky to interfere the operative field, we pulled the omentum and attached it to the anterior abdominal wall using percutaneous suture in order to obtain the appropriate view without additional use of the trocar (Fig. 2a). The lateral incision was extended to the level of the short gastric vessels. Then, the avascular plane between spleen and pancreas tail was firstly dissected using the ultrasonic shears and the splenic vessels were usually identified at the upper border of the pancreas, and the splenic artery and vein were gently dissected and

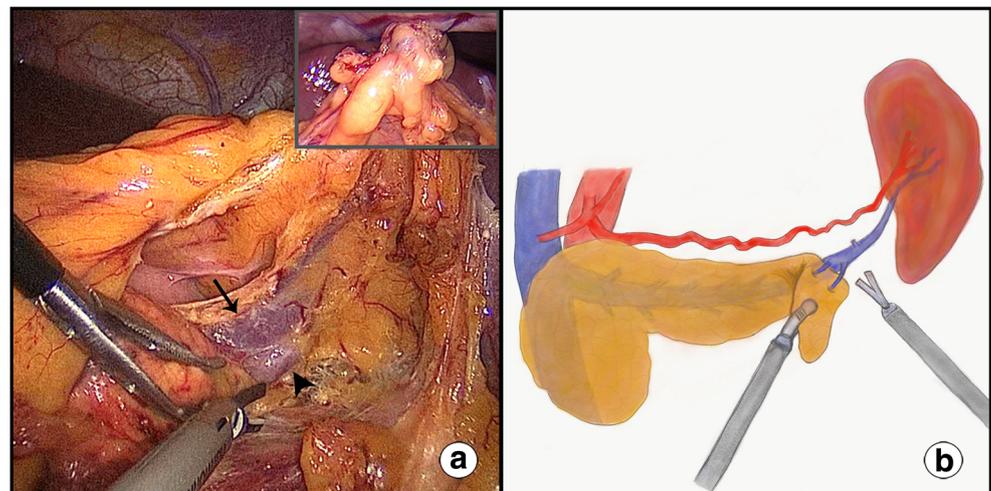
Fig. 1 Extracorporeal view of **a** the dual-incision laparoscopic spleen-preserving distal pancreatectomy using multichannel trocar (Glove port, Nelis, Seoul, Korea) and **b** the conventional laparoscopic spleen preserving distal pancreatectomy. Comparisons of the external wound after removing the trocars are presented in **c** the dual-incision laparoscopic spleen-preserving distal pancreatectomy and **d** the conventional laparoscopic spleen-preserving distal pancreatectomy



freed from the pancreas (Fig. 2a). Small branches of splenic vessels that penetrate the pancreas parenchyma were initially sacrificed using a vessel-sealing device (LigaSure V; Tyco Healthcare, Tokyo, Japan). After that, the pancreas tail was lifted anteriorly and the dissection was continued in lateral-to-medial fashion until the distal pancreas was released. Pancreas transection was performed using an endoscopic linear stapler (Echelon Flex™ 60 Endopath® Stapler with blue or gold cartilage, Ethicon Endo-surgery Inc., Cincinnati, OH, USA), allowing an adequate margin from the mass (about 1 cm). Reinforcement by laparoscopic suturing of the stapled

area was occasionally performed when the pancreas stump was crushed during the stapling and pancreas juice leakage was a concern. After transection, fibrin glue was applied to the stump. One closed suction drain was placed in the pancreas stump through the subxiphoid 5-mm port site and the specimen was bagged. It was retrieved through the 3-cm transverse incision after removing the multichannel trocar with or without extension of incision. If the tumor size was less than 3 cm, the incision was used as is without extending, but if necessary, due to the tumor size being greater than 3 cm, the incision was extended as little as possible.

Fig. 2 **a** The intracorporeal view of the operation and **b** schematic diagram. The omentum attached to the stomach was pulled and attached to the anterior abdominal wall using percutaneous suture in order to obtain the appropriate view (inlet). The arrow and arrow head point to splenic vein and artery respectively



Conventional Laparoscopic Spleen-Preserving Distal Pancreatectomy

Conventional LSPDP was performed similarly as described in previous report of Kim et al.¹⁴ The patient was placed with legs apart in the supine position and tilted to the right side in the reverse Trendelenburg position and five trocars were usually used. (Fig. 1b) The direction of dissection is in a medial-to-lateral fashion and that is in direct contrast to the direction of dissection in DILSPDP. Nevertheless, the range of dissection was similar between the two types of surgery.² Briefly, after the adhesion between the posterior gastric wall and surface of the pancreas was released, dissection was performed along the inferior border of the pancreas within an avascular line, and then the posterior aspect of the pancreas was mobilized. After the splenic vessel was isolated and separated from the pancreas, the pancreas parenchyma was transected at a distance of at least 1 cm from the tumor by a linear stapler and then the separation of pancreatic gland from the splenic vessels was conducted toward the lateral end of the pancreas. Finally, the specimen was retrieved through the umbilicus trocar site with extensions of the incision, if necessary, according to the specimen length.

Statistical Analysis

All statistical analyses were conducted using SPSS statistical package software (version 22.0 for Windows; SPSS, Inc., Chicago, IL). Continuous data are presented as the mean \pm standard deviation (SD). For continuous data, overall differences were tested by Student's *t* test. The categorical variables were calculated using the Fisher's exact test or chi-square test. The descriptive statistics are described as mean \pm standard deviation and differences were regarded as statistically significant when $p < 0.05$.

Results

During the study period, a total of 74 patients underwent LDP for benign or borderline malignancy of distal pancreas. Among them, four patients who were diagnosed with pancreas cancer on the permanent pathologic report were excluded from analysis. Also, ten patients who planned splenectomy from the beginning due to severe inflammation around spleen, or the mass invaded the splenic hilum, were excluded. A total of 12 patients who underwent concurrent operation of other organs during the surgery were also excluded from the analysis. Finally, a total of 48 patients were analyzed in this study composed of 22 DILSPDP and 26 conventional LSPDP. Baseline characteristics and demographics of enrolled patients are presented in Table 1. There was no significant difference in terms of age, sex, diagnosis of disease between the two

groups, and the most common diagnosis was intraductal papillary mucinous neoplasm (18 cases, 37.5%) followed by neuroendocrine tumor (9 patients, 18.8%), mucinous cystadenoma (8 patients, 16.7%), serous cystadenoma (7 patients, 14.6%), and solid pseudopapillary epithelial neoplasm (6 patients, 12.5%). There was no difference of tumor size between two groups (2.7 ± 1.8 cm in DILSPDP vs. 3.6 ± 2.1 cm in conventional LSPDP group, $p = 0.138$) even if the total length of the retrieved specimen was significantly longer in conventional LSPDP than DILSPDP (6.5 ± 2.3 cm vs. 9.5 ± 2.7 cm respectively, $p < 0.001$). In terms of tumor location, 18 of 26 patients (69.2%) in conventional LSPDP group had pancreas tail tumors whereas all patients (100%) in DILSPDP group had the tumor located on pancreas tail ($p = 0.084$).

Comparisons of surgical outcomes according to the type of surgery are shown in Table 2. In DILSPDP group, the operation time and blood loss were both significantly reduced compared to those in conventional LSPDP group. (119.3 ± 50.4 min and 96.8 ± 165.3 cc in DILSPDP vs. 190.9 ± 43.7 min and 282.7 ± 305.3 cc in conventional LSPDP, $p = 0.004$ and 0.011 , respectively) Preservation of splenic vessels was achieved in 95.5% of DILSPDP (21 of 22 patients) and 65.4% of conventional LSPDP (17 of 26 patients) which was a significant difference ($p = 0.013$). There were four cases of splenectomy in this study and they were all in the conventional group. (15.4%, $p = 0.114$) Among them, three cases were converted to laparotomy due to severe intraoperative bleeding and then splenectomy performed. The other case resulted in splenectomy due to splenic infarction after converting to the Warshaw method. In terms of postoperative pain using VAS, lower pain score was observed in DILSPDP group at postoperative day 5, whereas there were no differences between groups at postoperative day 1 or 3. There was no difference in the length of hospital stay, postoperative morbidities between two groups, and no postoperative mortality was observed in the study.

Discussion

Division of splenic vessels has potential risks in terms of causing inadequate blood supply to the spleen, creating potential for splenic infarction or gastric variceal changes, due mainly to the overweight inflow of short gastric and left gastroepiploic vessels toward spleen. Symptomatic splenic infarction could require re-laparotomy for splenectomy and increases the risk of postoperative morbidities.^{15, 16} Despite these potential shortcomings of the Warshaw technique however, it has been considered preferable to splenic vessel-preservation LDP because of its technical ease.^{17, 18} Complete preservation of splenic vessels is quite difficult and technically challenging, especially in the case of tumors which are located close to the splenic hilum, because the

Table 1 Comparative analysis of baseline characteristics between two groups according to the surgical methods; DILSPDP versus Conventional LSPDP

Characteristics	DILSPDP (<i>n</i> = 22)	Conventional LSPDP (<i>n</i> = 26)	<i>p</i> value
Age (year)	51.5 ± 17 (18–74)	55.3 ± 15.1 (20–77)	0.416
Sex (M/F)	5 / 17	8/18	0.746
BMI	25.2 ± 4.5 (18.4–35.8)	24.7 ± 2.6 (19.8–29)	0.668
Diagnosis (%)			0.973
IPMN	8 (36.4)	10 (38.5)	
Serous cystadenoma	4 (18.2)	3 (11.5)	
Mucinous cystadenoma	3 (13.6)	5 (19.2)	
NET	4 (18.2)	5 (19.2)	
SPEN	3 (13.6)	3 (11.5)	
Tumor size (cm)	2.7 ± 1.8 (1–6.5)	3.6 ± 2.1 (1.4–10)	0.138
Specimen length (cm)	6.5 ± 2.3 (3.8–12.5)	9.5 ± 2.7 (3.5–16.2)	< 0.001
Tumor location ^a (%)			0.084
Body	0	8 (30.8)	
Tail	22 (100)	18 (69.2)	

DILSPDP dual-incision laparoscopic spleen-preserving distal pancreatectomy, *LSPDP* laparoscopic spleen-preserving distal pancreatectomy, *BMI* body mass index, *IPMN* intraductal papillary mucinous neoplasm, *NET* neuroendocrine tumor, *SPEN* solid pseudopapillary epithelial neoplasm

^a We classified the location of tumor per each case into the body or tail; if the boundary of mass exceeded over the left lateral border of aorta, it was regarded as the mass at tail of pancreas whereas the mass at body of pancreas if the mass was located within the left lateral border of aorta

vessels near the distal pancreas are usually embedded in the pancreas parenchyma.^{19–21} To overcome these vulnerable points, Nakamura et al.² firstly proposed the lateral approach for LSPDP which is a method quite different from the conventional method of medial approach for vessel preservation. The conventional approach performs the separation of pancreas parenchyma from the splenic vessels after the completion

of pancreas transection, from the medial side of pancreas toward the lateral end of pancreas. On the other hand, the lateral approach has separated the pancreas parenchyma from the vessels toward the medial side of pancreas from the tip of pancreas tail and then transects the pancreas parenchyma. The lateral approach has advantages for the preservation of splenic vein which is the most crucial stage for LSPDP on the

Table 2 Comparative analysis of surgical outcomes between two groups according to the surgical methods; DILSPDP versus conventional LSPDP

Characteristics	DILSPDP (<i>n</i> = 22)	Conventional LSPDP (<i>n</i> = 26)	<i>p</i> value
Operation time (min)	119.3 ± 50.4 (50–225)	190.9 ± 43.7 (94–260)	0.004
Blood loss (cc)	96.8 ± 165.3 (20–280)	282.7 ± 305.3 (50–500)	0.011
Preservation of splenic vessels (%)	21 (95.5)	17 (65.4)	0.013
Splenectomy (%)	0	4 (15.4)	0.114
Conversion to laparotomy (%)	0	4 (15.4)	0.114
Number of used trocars	2.7 ± 0.7 (2–4) ^a	4.9 ± 0.3 (4–5)	< 0.001
Length of hospital stay (day)	7.7 ± 1.3 (5–10)	8.0 ± 1.4 (6–13)	0.065
Postoperative pain (VAS)			
POD 1	3.1 ± 1.1 (1–5)	3.2 ± 1.4 (1–6)	0.962
POD 3	2.8 ± 1.3 (1–5)	3.5 ± 1.8 (1–7)	0.121
POD 5	1.8 ± 0.9 (1–3)	2.6 ± 1.3 (1–6)	0.026
Postoperative morbidities (%)	0	1 ^b (3.8)	1.000
Postoperative mortality (%)	0	0	

DILSPDP dual-incision laparoscopic spleen-preserving distal pancreatectomy, *LSPDP* laparoscopic spleen-preserving distal pancreatectomy, *VAS* visual analogue scale, *POD* postoperative day

^a The number of used trocars in DILSPDP group included the one multi-channel trocar

^b This one case had the wound infection at the site of umbilicus trocar which was grade I morbidities graded by Clavien-Dindo classification

strength of anatomical characteristics of pancreas, and the major benefit of DILSPDP is accruing from the lateral-to-medial dissection of pancreatic tail tumors. Although the splenic vein embedded in pancreas sulcus acts as an obstacle in separating it from the pancreas parenchyme especially in the distal portion of pancreas, it is characteristically fully exposed at the tip of the pancreas tail and the gap between spleen and the pancreas tail, as described in Fig. 2b. When performing DILSPDP, we firstly dissected the avascular plane of inferior border of pancreas, and the dissection proceeded to the gap between pancreas tail and spleen. Then, the several perforator vessels that are small splenic branches penetrating pancreas parenchyme along the upper border of pancreas could be obviously identified and sacrificed. Because of this approach, it is relatively easy to identify and expose the travel route of these small blood vessels and this allows a quick judgment of which vessel to scarify and the easy ligation of these vessels.

The reverse Trendelenburg position in pancreas surgery has generally been used in order to pull the pancreas and spleen toward the umbilicus by gravity to remove the omental tissue from view which helps with visibility. In DILSPDP, the right lateral decubitus position is added and it allows that the pancreas tail and spleen be raised to the left flank of the patient whereas the omental tissue and colon were moved downwards to the right side of the patient's body. This characteristic position of DILSPDP could be helpful to expose the operative field on pancreas tail effectively and enable easier approach to the pancreas tail. We successfully performed DILSPDP even in patients with a past history of upper abdominal surgery with midline incision, such as partial resection of transverse colon and primary closure of duodenal perforation accompanied with severe intraabdominal adhesion. Also, this maximizing use of gravity is very useful in cases of obese patients having huge fat tissual pad with omentum which usually covers the anterior aspect of pancreas. Indeed, two extremely obese

patients in our study having BMI over 35 underwent DILSPDP successfully. Of further note, the use of gravity reduces the need for an additional trocar to gain a surgical field, making the reduced port laparoscopic surgery possible.

Given these various advantages, we propose that DILSPDP could be very favorable for achieving spleen-preserving LDP with vessel preservation for benign pancreas tail mass. When considering the surgical results of LSPDP from previous literature (Table 3),^{2, 4, 22–26} a superior rate of successful preservation of splenic vessels was observed with DILSPDP in accordance with less intraoperative blood loss or shortened operation time, compared to other reports, even if an accurate comparison might be difficult since the status of the tumor in each study was not considered.

The minimally invasive approach using the dual incision with multichannel trocar is also one of the characteristic features of DILSPDP which is different from other methods. The reduce-port laparoscopic surgery could be expected to have benefits such as reduced incisional pain and parietal tissue trauma, accompanying improved cosmetic outcomes.²⁷ In case of pancreatectomy, the pancreas specimen usually has bulky size and solid texture and this inevitably requires a certain length of incision to retrieve the specimen from the patient's body. Thus, it is a characteristic of DILSPDP that the transverse incision of such a length is made in advance and the multichannel trocar is used here to reduce the total parietal trauma and the number of trocars used. Moreover, an additional subxiphoid 5-mm trocar was used as the working channel for the left hand of the operator, separately. These arrangements of trocars named dual-incision could provide a comfortable environment to surgeons similar to that in conventional LSPDP using four or five trocars. It enables the operator to manipulate the laparoscopic instruments in both hands making the fine and complex movements easily parallel to the existing method. As demonstrated in Table 3, the surgical results of DILSPDP have favorable outcomes compared to

Table 3 Outcomes of our study (DILSPDP¹) and previously reported conventional spleen preserving laparoscopic distal pancreatectomy trials

Reference, year	No. of cases	No. of vessel-preserving LDP ²	Operative time ^a (min)	Intraoperative blood loss ^a (ml)	Length of hospital stay ^a (day)	Pancreas fistula ^a (%)	Morbidity ^a (%)
Nakamura et al. ² , 2011	23	14	NA ³	NA	–	0	–
Butturini et al. ²² , 2012	43	36	180	NA	8	29	58
Choi et al. ²³ , 2012	40	29	303	340	7.1	12.5	25
Adam et al. ^{4a} , 2013	140	55	214	342	8.2	3.6	27.3
Sato et al. ^{24a} , 2000	24	7	319	60	18	NA	0
Nakamura et al. ^{25a} , 2016	17	11	280	60	11	12.5	12.5
Nagakawa et al. ^{26a} , 2017	51	32	213	43	13.4	12.5	18.6
Current study (DILSPDP) ^a	22	21	119	97	7.7	0	0

DILSPDP dual-incision laparoscopic spleen-preserving distal pancreatectomy, LDP laparoscopic distal pancreatectomy, NA not available

^a Results based on an intention-to-treat splenic conservation

the results of previous reports using conventional LSPDP accompanied by cosmetic improvements on the strength of reduced trocars. (Fig. 1).

Despite these interesting outcomes, the result of the current study should be interpreted carefully, because the technical usefulness of DILSPDP would vary according to the location of the pancreas mass and this study presupposes the locational difference of tumors. Actually, the lateral approach in DILSPDP would not be suitable for situations where the mass is located at the body of pancreas, which means a mass exceeding the left lateral border of aorta inwardly. This is because in the lateral position, the medial position of pancreas, i.e., the body of pancreas is usually pressed making it difficult to obtain an operative field of view and it is unlikely that the benefit of DILSPDP would be found in pancreas body tumors as well. The authors presume that it would be inappropriate to apply DILSPDP to pancreas body tumors. Nevertheless, when comparing the surgical results only for pancreas tail masses between the two groups, three of 18 cases (16.7%) having pancreas tail mass in the conventional group had to be converted to laparotomy due to intraoperative bleeding, whereas no cases in the DILSPDP group had the conversion. Also, half of cases having pancreas tail mass in conventional group failed to preserve the splenic vessel during the operation and eventually converted to Warshaw method, while only one case in DILSPDP group failed the vessel-preserving LDP. The authors believe that DILSPDP is preferable to conventional spleen-preserving LDP in cases of pancreas tail mass, in contrast to the case of tumors located at pancreas body, and a tailored treatment strategy should be considered according to the tumor location. In our forthcoming study, we need to strictly observe the inclusion criteria in the prospective design of the study, so that the effect of tumor location could be completely excluded and the feasibility of procedure for the tail mass would be confirmed. We believe that the prospective large-scale study should be conducted to substantiate the results of the current study in near future.

In conclusion, the authors propose that DILSPDP would be a feasible and effective method of vessel-preserving LDP for benign or low-malignancy of pancreas tail which could overcome some disadvantages of the conventional method and also acquire the benefits of reduced parietal trauma, concurrently.

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Author Contribution Eun Young Kim and Tae Ho Hong contributed to all aspects of this study and article. Eun Young Kim, Young Kyoung You, Dong Goo Kim, and Tae Ho Hong contributed to study conception and design, experiments, collection of data, and critical revision of the article. All authors approved the final draft of the article.

Compliance with Ethical Standards

Conflicts of Interest No potential conflict of interest relevant to this article was reported.

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