Original Article
Intraoperative sentinel lymph node mapping guides laparoscopic-assisted distal gastrectomy for distal gastric cancer

Naiqing Liu[1,2*], Zhengchuan Niu[2*], Wei Niu[2*], Cheng Peng[2], Xueqing Zou[2], Shuxiang Sun[1], Obo Shinichi[2], Muhammad Shahbaz[2], Qinli Sun[1], Niu Jun[2]

[1]Department of General Surgery, Yishui Central Hospital, Linyi 276400, Shandong, P. R. China; [2]Department of Hepatobiliary Surgery, Qilu Hospital, Shandong University, Jinan 250012, Shandong, P. R. China. *Equal contributors.

Abstract: Aims: The aim of this retrospective study is to explore the effects of sentinel lymph node (SLN) mapping guided laparoscopic-assisted distal gastrectomy (LADG) for distal gastric cancer. Methods: Two hundred patients were enrolled in this study. One hundred and one patients undergoing SLN guided LADG were designated as the SLN group. Ninety-nine patients having conventional LADG with D1 or D2 lymph node dissection were designated as the control group. Intraoperative and postoperative indicators such as the number of lymph nodes dissected, intraoperative and postoperative conditions, flow cytometry analysis of T lymphocyte subsets and natural killer (NK) cells, survival rates, recurrence rates and postoperative complications were investigated between these two groups. Results: The number of lymph nodes dissected in the SLN group was significantly lesser than that in the control group. Furthermore, in the SLN group, the patients achieved better immunization status, improved intraoperative and postoperative conditions and decreased postoperative complications. There were no significant differences were found in the positive lymph nodes detected, the distance between proximal and distal cutting edge, postoperative survival or recurrence rates. Conclusions: SLN guided LADG for gastric cancer is a safe and effective method and could achieve an equal clinical effect as traditional laparoscopic D1 or D2 radical operation with less operation trauma and better recovery.

Keywords: Sentinel lymph node mapping, laparoscopic-assisted distal gastrectomy, gastric cancer

Introduction

Laparoscopic-assisted distal gastrectomy (LA-DG) for gastric cancer was first performed in 1994 [1]. In LADG, the extent of tumor resection and patients' prognosis are both determined by the status of lymph node metastasis. It is important to investigate safe and effective ways for lymph node dissection in laparoscopic gastrectomy.

Laparoscopic D1 or D2 resection in advanced gastric carcinoma was first conducted in 1997 [2]. It is safe and feasible and it has the same extent of lymph node dissection as the traditional open surgery [3]. Nodal involvement in gastric cancer occurs in 2-18% of T1 and in about 50% of T2 tumors (TNM staging) [4]. The preventive and extensive lymph node dissection in laparoscopic D2 resection for gastric carcinoma included might affect the immune function of the organism and increase the surgical trauma. The incidence of postoperative complications in laparoscopic D1 or D2 resection for gastric carcinoma is about 22%-54%, including lymphatic fistula, reduced stomach capacity, gastric emptying dysfunction, dumping syndrome, alkaline reflux gastritis, and so on, which severely affected the post-operative living quality of the patients [5]. It is urgent to investigate the way to selectively diminish the extent of lymph node dissection and reduce postoperative complications without weakening the effect of radical resection.

The concept of sentinel lymph node (SLN) method is to predict the lymph node status by a sim-
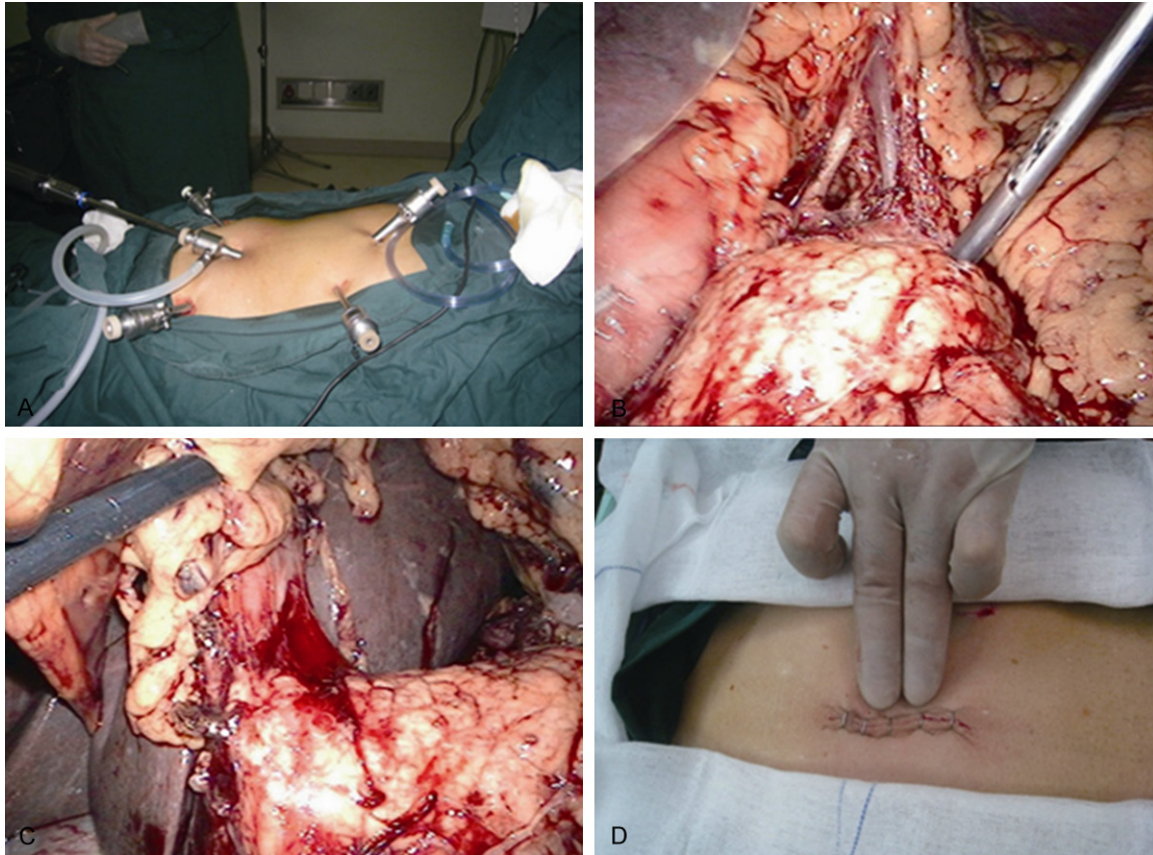
Figure 1. Procedure of LADG with D1 or D2 lymph node dissection for gastric cancer. A: The patient's position and Trocar port location were shown. B: Laparoscopic lymph node dissection. C: After freeing, the stomach was extracted outside of the peritoneal cavity for resection and anastomosis. D: The small, minimally-invasive incision. LADG, laparoscopic-assisted distal gastrectomy.

ple pick-up biopsy in patients with a high risk of metastases to lymph nodes. If the SLN indicates the patient is negative for metastasis, unnecessary lymph node dissection can be avoided [6]. The feasibility and effects of SLN biopsy in guiding the lymph node dissection have been confirmed in open radical resections for gastric cancer. Patent blue was first used in SLN biopsy in gastric cancer in 1999. Then Aikou presented the idea that there might be a potential significance of SLN biopsy in the lymph node dissection of early gastric carcinoma in 2001 [7]. Kitagawa reported a Laparoscopic pylorus-preserved partial gastrectomy based on SLN biopsy [8]. These studies suggest a potential role of SLN in guiding lymph node dissection during gastrectomy.

In this study, the feasibility and clinical effects of SLN mapping on guiding LADG for distal gastric cancer via sub-mucosal blue dye injection were investigated. The effects of SLN guided

LADG were compared with those of traditional laparoscopic D1 or D2 radical operation.

Materials and methods

Patients

This study was approved by the Ethics Committee of Shandong University. The patients were fully involved in the decision making process. Written informed consent was obtained from all the patients.

A total of 200 cases of patients with preoperative histologically confirmed gastric cancer were enrolled in this study. Their gastric lesions were located in the lower or middle third of stomach. They all underwent LADG by the operation team of Pro. Jun Niu. The patients who had gastric cancer with a diameter greater than 5 cm, possible invasion beyond the muscularis propria, preoperative evidence of metastatic
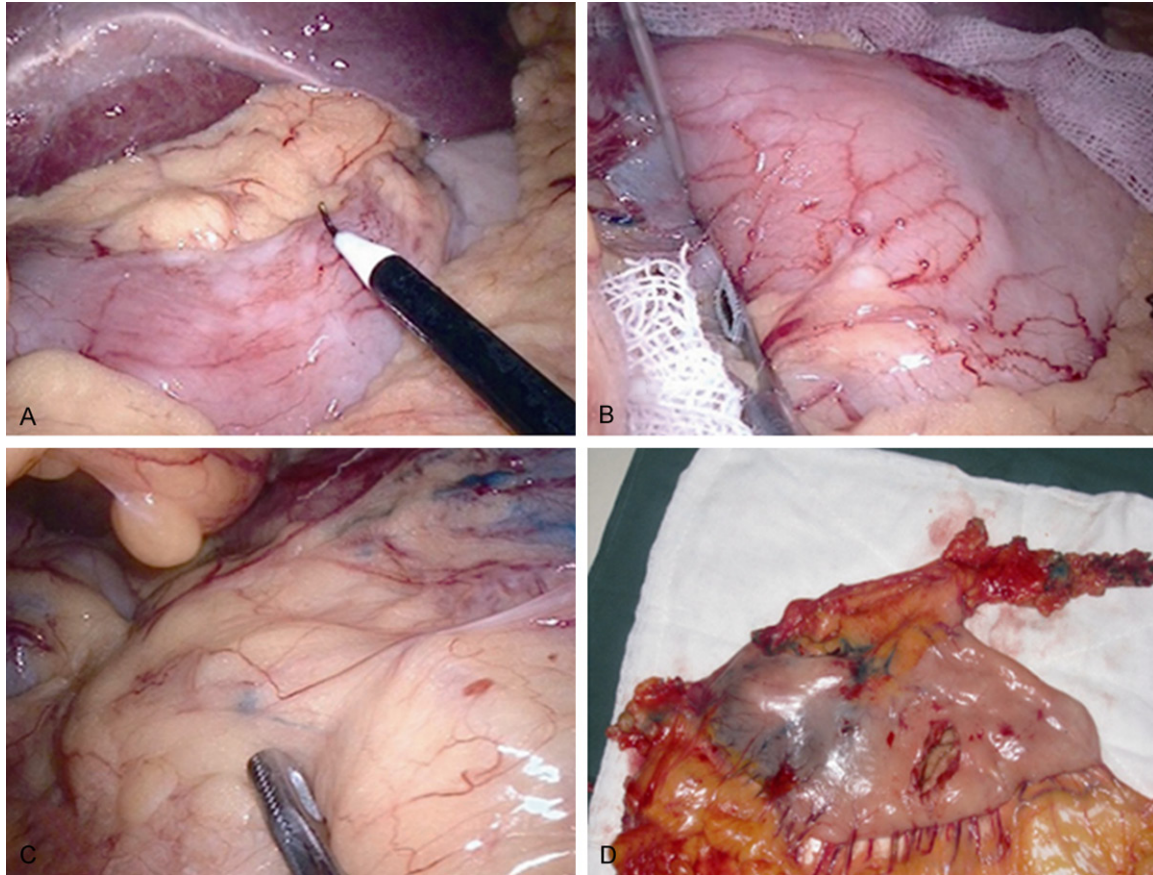
Figure 2. Procedure of SLN mapping guided LADG for gastric cancer. A: Laparoscopic exploration was performed to define the location of tumor lesions, indicated by a coagulation hook. B: Laparoscopic assisted blue dye injection around the tumor was carried out for SLN mapping. C: SLNs were shown by the blue dye. D. Selective gastric partial resection was performed based on SLN mapping. SLN, sentinel lymph node; LADG, laparoscopic-assisted distal gastrectomy.

disease, or reported intolerance to Patent blue were excluded from the study. Among these 200 patients, 101 patients (52 males and 49 females, mean 51.69 years) underwent SLN guided LADG and were designated as the SLN group. Ninety-nine patients (51 males and 48 females, mean 53.72 years) underwent conventional LADG with D1 (68 cases) or D2 (31 cases) lymph node dissection and were designated as the control group. All patients received infection prevention and total parenteral nutrition support postoperatively.

Surgery

In the control group, LADG procedure with D1 or D2 lymph node dissection was performed according to the reports of the intraoperative frozen sections [9, 10] (**Figure 1**). SLN guided LADG was performed in the SLN group as previ-

ously reported (**Figure 2**) [4]. The lesion location was determined by laparoscope. Patent blue dye was carefully injected through abdominal wall into gastric sub-mucosal layer at four quadrants around the tumor with a 25-gauge needle. Within 5 minutes [4], blue colored lymph nodes were selected for mapping and biopsy individually. The frozen pathology reports were confirmed by more than 3 experienced pathologists. Among the 101 patients in the SLN group, pylorus preserving was performed in 15 cases, gastric wedge resection was performed in 35 cases, and Birroth II procedure was conducted in 51 cases. And, all of them had individual lymph node dissection.

Observation indexes

The following indexes were observed and compared, including counting of total lymph nodes

Table 1. Comparison of effects of tumor radical resection (mean ± SD)

Group	N	Distance of proximal cutting edge (cm)	Distance of distal cutting edge (cm)	Number of total lymph nodes	Number of positive lymph nodes
SLN group	101	4.67 ± 1.25	4.90 ± 1.64	5.32 ± 2.58	4.18 ± 2.93
Control group	99	4.63 ± 1.12	4.58 ± 1.11	12.15 ± 7.47	4.15 ± 2.54
P		0.786	0.103	< 0.001	0.945

Table 2. Intraoperative and postoperative conditions

Group	N	Intraoperative conditions		Postoperative conditions		
		Operative time (min)	Intraoperative blood loss (ml)	Time to flatus (day)	Time to defecation (day)	Time to walking (day)
SLN Group	101	167.6 ± 29.31*	117.43 ± 55.32*	2.86 ± 1.31*	3.62 ± 1.85*	2.08 ± 0.38*
Control Group	99	208.5 ± 30.02	168.78 ± 63.46	3.98 ± 1.50	4.81 ± 1.62	3.12 ± 0.61

Note: The data represents means ± SD. *P < 0.05 versus control group.

and positive lymph nodes determined by routine pathology, intraoperative blood loss, postoperative recovery time, evaluation of radical effect, counting of T lymphocyte subsets and natural killer (NK) cells in peripheral blood (1 week after surgery), complications, postoperative quality of life, survival and relapse indicators, et al.

Statistical analysis

All data were processed using SPSS 11.5 statistical package. The data were presented as means ± standard deviation (SD). The methods of t-test, χ^2-test and Fisher exact probability method were used to compare values between two groups. Log-rank test was conducted to compare the difference in survival rate. P < 0.05 was considered to be statistical significant.

Results

Effects of SLN mapping on lymph node dissection and tumor radical resection

Firstly, the effects of SLN on lymph node dissection and tumor radical resection were analyzed. All the frozen section reports correlated with the final histopathology results of the lymph node status. As shown in **Table 1**, there was no significant difference in the number of positive lymph nodes dissected between the SLN group and the control group (4.18 ± 2.93 versus 4.15 ± 2.54, P > 0.05). However, the number of total lymph nodes in the SLN group was significantly lesser than that in the control group (5.32 ± 2.58 versus 12.15 ± 7.47, P <

0.001). There were no significant differences in the distances of both proximal cutting edge (4.67 ± 1.25 cm versus 4.63 ± 1.12 cm, P > 0.05) and distal cutting edge (4.90 ± 1.64 cm versus 4.58 ± 1.11 cm, P > 0.05) between the SLN group and the control group.

Effects of SLN mapping on intraoperative and postoperative conditions

Then the effects of SLN on intraoperative and postoperative conditions were investigated. As shown in **Table 2**, the average operative time in the SLN group was significantly shorter than that in the control group (167.6 ± 29.31 min versus 208.5 ± 30.02 min, P < 0.001). The average blood loss of the SLN group was significantly lesser than that of the control group (117.43 ± 55.32 ml versus 168.78 ± 63.46 ml, P < 0.001). Moreover, in the SLN group, the times to flatus, defecation, and walking were significantly shorter than those in the control group (**Table 2**, P < 0.001). These results indicate that the intraoperative and postoperative conditions of patients in the SLN group are significantly improved.

Improved immunization status in the SLN group

To determine the effects of SLN mapping on postoperative immunization status of the patients, analysis of T cell subsets and NK cells was performed in the patients one week after the operation using flow cytometry. As shown in **Table 3**, in the SLN group, the percentages of CD3^{+} cells (T cells), CD4^{+} T cells (T helper cells)

Table 3. Peripheral blood T cell subsets and NK cells

Group	N	CD3$^+$ cells (%)	CD4$^+$ cells (%)	CD8$^+$ cells (%)	CD4$^+$/CD8$^+$ cells	NK cells (%)
SLN Group	101	78.14 ± 8.99*	45.99 ± 7.20*	32.57 ± 6.15*	1.55 ± 0.31*	14.49 ± 3.41*
Control Group	99	73.61 ± 8.38	36.44 ± 6.60	36.95 ± 6.70	0.95 ± 0.23	8.12 ± 1.42

Note: The data represents means ± SD. *P < 0.001 versus control group. NK cells, natural killer cells.
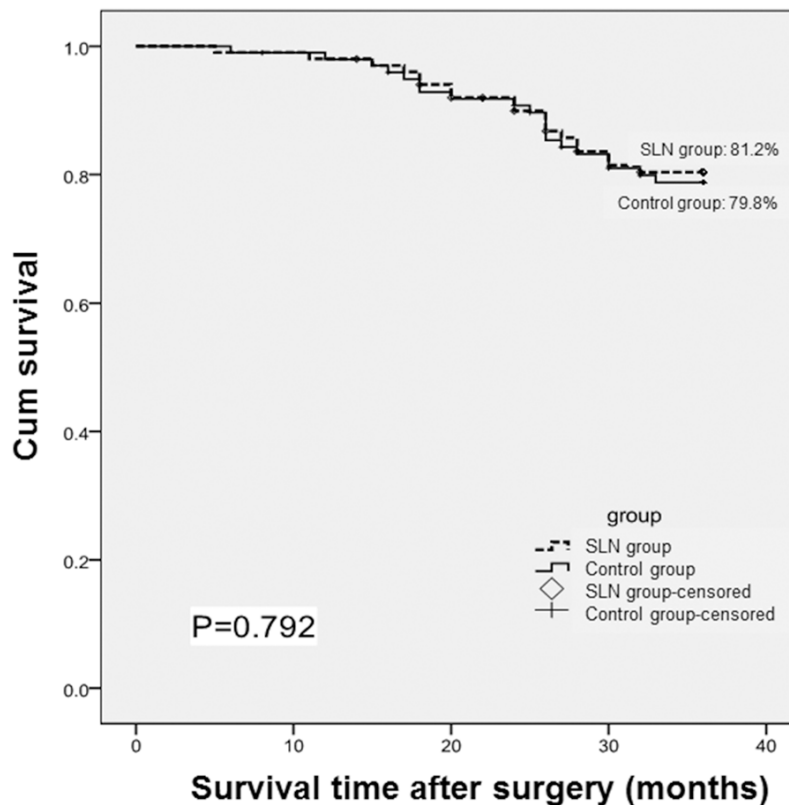

Figure 3. The 3-year survival rate after surgery. The 3-year survival rate after surgery of the patients in the SLN group and the control group were show. There was no significant difference in the 3-year survival rate between these two groups. The log-rank test, P = 0.792.

and NK cells in peripheral blood were 78.14 ± 8.99%, 45.99 ± 7.20% and 14.49 ± 3.41%, respectively, which were significantly higher than those in the control group (73.61 ± 8.38%, P < 0.001; 36.44 ± 6.60%, P < 0.001 and 8.12 ± 1.42%, P < 0.001, respectively). The level of CD8$^+$ T cells (cytotoxic T cells) in the SLN group was significantly lower than that in the control group (32.57 ± 6.15% versus 36.95 ± 6.70%, P < 0.001). Furthermore, in the SLN group, the CD4$^+$/CD8$^+$ T-cell ratio was significantly higher than that in the control group (1.55 ± 0.31 versus 0.95 ± 0.23, P < 0.001). These results indicate that postoperative immunization status of the patients in the SLN group is better than that in the control group.

SLN mapping does not affect postoperative survival and recurrence rate

To study the effects of SLN mapping on postoperative survival and recurrence rate, the 3-year follow-up assessment was undertaken. Seventeen of the 200 patients were lost to follow-up (SLN group, 8 patients; control group, 9 patients). The survival time ranged from 5 months to 36 months. Thirty-nine of the 200 patients died during the 3-year follow-up period with the average 3 year survival rate of 80.5%. In the SLN group, 19 of the 101 patients died with the average 3 year survival rate of 81.2%. In the control group, 20 of the 99 patients died with the average 3 year survival rate of 79.8%. There was no significant difference in survival rate between these two groups (P = 0.792, **Figure 3**). In the SLN group, 18 of the 101 patients relapsed during the 3-year follow-up period with the recurrence rate of 17.8%. In the control group, 17 of the 99 patients relapsed with the recurrence rate of 17.2%. There was no significant difference in recurrence rate between these two groups (P = 1.000). These results indicate that SLN mapping does not affect postoperative survival and recurrence rate.

Decreased postoperative complications in the SLN group

The effects of SLN mapping on postoperative complications were also investigated. As shown in **Table 4**, there were no significant differences between the two groups in hepatic metastasis,

Table 4. Postoperative complications

Group	Postoperative bleeding	Obstruction	Hepatic metastasis	Anastomotic or stump fistula	Alkaline reflux	Gastroparesis
SLN Group	3*	2	5	4	9*	1*
Control Group	11	3	3	3	21	7

Note: Fisher exact probability method, *P < 0.05 versus control group.

obstruction, and anastomotic or stump fistula (P > 0.05). There were significant differences between the SLN group and the control group in the postoperative bleeding incidence (3 versus 11, P = 0.028), alkaline reflux incidence (9 versus 21, P = 0.018), and gastroparesis incidence (1 versus 7, P = 0.034). These results indicate a better postoperative life quality of the patients in the SLN group.

Discussion

Laparoscopic gastrectomy for gastric cancer is a promising minimally invasive surgery [11]. In this study, the effects of SLN mapping on guiding LADG for distal gastric cancer were investigated. It is safe and effective with less operation trauma and better recovery by achieving individual lymph node dissection.

In this study, we found that the extent of lymph node resection in SLN group was significantly less than the control group, however, there was no statistical difference in the number of positive lymph node between the two groups. And, there was no statistical difference in recent recurrence rate between the two groups, suggesting the security and feasibility of intraoperative SLN guided LADG for distal gastric carcinoma. In addition, patients in the SLN group had better intraoperative conditions, postoperative conditions and immune status. Moreover, compared with control group, there was no significant difference of SLN group in radical effect of tumor resection, postoperative survival time, and cancer recurrence rate. Our findings indicate that intraoperative SLN-guided LADG for distal gastric carcinoma is safer and feasible and can achieve the same therapeutic effects as LADG with D1 or D2 lymph node dissection.

Ichikura performed function-preserved gastrectomy in 35 early gastric cancer patients with negative SLN. The function-preserved gastrectomy achieved larger gastric capacity than that of standard gastrectomy. There was no death caused by surgery or operative complica-

tions. The hospital stay time was shorter and the recent recurrence rate was lower [12]. A multicenter and prospective study from Sep. 2004 to Mar. 2008 carried out by JSNNS (Japan society of sentinel node navigation surgery) found that the detection rate of SLN in early gastric cancer, the sensitivity and specificity in diagnosis of lymph node metastasis were 97.5%, 93%, and 99% respectively; compared with traditional method, patients with SLN guided surgery had better postoperative life quality [13]. Our results were consistent with these reports, indicating that SLN guided LADG could achieve the effects of the selective function-preserved gastrectomy and achieve individual LN dissection with less postoperative complication and better life quality simultaneously .

At present, SLN-guided LADG for distal gastric carcinoma is still an experimental technique and none of the tried approaches has obtained general acceptance [14]. Combined this present study and previous researches, we concluded its limitations as follows: 1) An obstructed lymphatic vessel due to cancer invasion may lead to a false negative results. 2) Lymphatic basin dissection is inadequate in the case of skip metastases, and the SLN omitted the first lymph node level in about 20% of cases [4]. 3) This method requires further improvement in technical issues, such as selection and usage of the tracers. Further studies are needed before this method can be introduced into routine procedure.

In conclusion, LADG with blue dye-guided lymph node dissection for gastric cancer is a safe and effective method for laparoscopic gastrectomy. It can reduce the operation trauma and achieve individual lymph node dissection with less postoperative complications. However, further studies are needed before this method can become a routine procedure.

Disclosure of conflict of interest

None.

Address correspondence to: Qinli Sun, Department of General Surgery, Yishui Central Hospital, No. 17, Jiankang Road of Yishui, Linyi 276400, Shandong Province, P. R. China. Tel: +86-13854952789; Fax: +86-539-2251647; E-mail: sunqinli0808@163.com; Niu Jun, Department of Hepatobiliary Surgery, Qilu Hospital, Shandong University, No. 44, Wenhua Xi Road, Jinan 250012, Shandong Province, P. R. China. Tel: +86-531-82169203; Fax: +86-531-82169243; E-mail: niusdu@163.com

References

[1] Kitano S, Iso Y, Moriyama M and Sugimachi K. Laparoscopy-assisted Billroth I gastrectomy. Surg Laparosc Endosc 1994; 4: 146-148.

[2] Goh PM, Khan AZ, So JB, Lomanto D, Cheah WK, Muthiah R and Gandhi A. Early experience with laparoscopic radical gastrectomy for advanced gastric cancer. Surg Laparosc Endosc Percutan Tech 2001; 11: 83-87.

[3] Palaia R, Cremona F, Delrio P, Izzo F, Ruffolo F and Parisi V. Sentinel node biopsy in gastric cancer. J Chemother 1999; 11: 230-231.

[4] Cozzaglio L, Bottura R, Di Rocco M, Gennari L and Doci R. Sentinel lymph node biopsy in gastric cancer: possible applications and limits. Eur J Surg Oncol 2011; 37: 55-59.

[5] Ozmen MM, Ozmen F and Zulfikaroglu B. Lymph nodes in gastric cancer. J Surg Oncol 2008; 98: 476-481.

[6] Ichikura T, Morita D, Uchida T, Okura E, Majima T, Ogawa T and Mochizuki H. Sentinel node concept in gastric carcinoma. World J Surg 2002; 26: 318-322.

[7] Aikou T, Higashi H, Natsugoe S, Hokita S, Baba M and Tako S. Can sentinel node navigation surgery reduce the extent of lymph node dissection in gastric canscer? Ann Surg Oncol 2001; 8: 90S-93S.

[8] Kitagawa Y, Saikawa Y, Takeuchi H, Mukai M, Nakahara T, Kubo A and Kitajima M. Sentinel node navigation in early stage gastric cancer-updated data and current status. Scand J Surg 2006; 95: 256-259.

[9] Noshiro H, Nagai E, Shimizu S, Uchiyama A and Tanaka M. Laparoscopically assisted distal gastrectomy with standard radical lymph node dissection for gastric cancer. Surg Endosc 2005; 19: 1592-1596.

[10] DU XH, Li R, Chen L, Shen D, Li SY and Guo Q. Laparoscopy-assisted D2 radical distal gastrectomy for advanced gastric cancer: initial experience. Chin Med J (Engl) 2009; 122: 1404-1407.

[11] Lee HJ and Yang HK. Laparoscopic gastrectomy for gastric cancer. Dig Surg 2013; 30: 132-141.

[12] Ichikura T, Sugasawa H, Sakamoto N, Yaguchi Y, Tsujimoto H and Ono S. Limited gastrectomy with dissection of sentinel node stations for early gastric cancer with negative sentinel node biopsy. Ann Surg 2009; 249: 942-947.

[13] Kitagawa Y, Takeuchi H, Takagi Y, Natsugoe S, Terashima M, Murakami N, Fujimura T, Tsujimoto H, Hayashi H, Yoshimizu N, Takagane A, Mohri Y, Nabeshima K, Uenosono Y, Kinami S, Sakamoto J, Morita S, Aikou T, Miwa K and Kitajima M. Sentinel node mapping for gastric cancer: a prospective multicenter trial in Japan. J Clin Oncol 2013; 31: 3704-3710.

[14] Märkl B, Moldovan AI, Jähnig H, Cacchi C, Spatz H, Anthuber M, Oruzio DV, Kretsinger H and Arnholdt HM. Combination of ex vivo sentinel lymph node mapping and methylene blue-assisted lymph node dissection in gastric cancer: a prospective and randomized study. Ann Surg Oncol 2011; 18: 1860-1868.