Am J Transl Res 2023;15(10):6115-6121
www.ajtr.org /ISSN:1943-8141/AJTR0151239

Original Article

Endometrial cancer intraoperative sentinel lymph node identification can effectively guide treatment

Fanchen Zhou[1], Bangruo Qi[2]

[1]*Department of Gynecology, Dalian Central Hospital, Dalian, Liaoning, China;* [2]*Department of Obstetrics and Gynecology, Sanya Women and Children's Hospital, Sanya, Hainan, China*

Received May 16, 2023; Accepted September 15, 2023; Epub October 15, 2023; Published October 30, 2023

Abstract: Objective: To explore the significance of intraoperative sentinel lymph node (SLN) identification in endometrial cancer. Methods: We retrospectively analyzed the clinical data of 56 patients with intraoperative SLN recognition (group A) and 50 patients without intraoperative SLN recognition (group B). SLN and pelvic abdominal lymph node distribution, SLN recognition rate, SLN recognition effect, mortality, the incidence of adverse events, and cumulative survival rate were statistically analyzed. Results: SLN were identified and removed in 41 of the 56 patients, with a recognition rate of 82.14% (46/56). The sensitivity of SLN was 83.72%, the specificity was 84.62%, and the negative predictive value was 61.11%. There were 15 patients with no SLN metastasis found in the pathological examination during the operation, among which two patients with poorly differentiated adenocarcinoma and clinical stage II patients underwent immunohistochemical staining, and three patients showed SLN micro-metastasis but no cancer tissue metastasis in the lymph node dissection. There was no significant difference in the incidence of total adverse events between group A and group B (P>0.05). The cumulative survival rate of group A was higher than that of group B (P=0.018). Conclusion: Intraoperative SLE identification can avoid false negative results, is safe and feasible, and can prolong the survival time of patients with endometrial cancer.

Keywords: Endometrial cancer, sentinel lymph node, identification, pathological examination, accumulate survival rate

Introduction

Endometrial cancer (EC) is a malignant tumor type of the female reproductive system, and its incidence is second only to cervical cancer, with an increasing trend of occurrence [1]. At present, the primary treatment for EC is a surgery, which mainly includes hysterectomy and pelvic and abdominal lymph node dissection [2]. Study [3] has pointed out that lymph node metastasis of endometrial cancer is random, and the possibility of lymph node metastasis is only 10% in patients with clinical stage I to stage II. Another study [4] showed that 80% of endometrial cancers were diagnosed at the early stage, and there was only less than 4% developed lymphatic metastasis in early endometrial cancers without high-risk factors. Some clinical studies have shown that systematic lymph node dissection for patients with early-stage endometrial cancer cannot improve survival [5]. In addition, extensive blind lymph node dissection is prone to complications (such as postoperative lymphocytes and lower limb lymphedema) and does not improve the prognosis of patients. Therefore, how to evaluate the status of lymph node metastasis individually is a priority in the treatment of endometrial cancer. The sentinel lymph nodes (SLN) are the first lymph nodes through which tumor metastasis occurs, and it can reflect lymph nodes involved in all subsequent areas [6]. Intraoperative SLN identification and pathological examination can help determine whether to perform lymph node dissection and the scope of dissection, which has been successfully applied in cervical cancer, breast cancer, vulvar cancer and other cancers, providing guidance for lymph node dissection scope for malignant tumors [7-9]. After reviewing the literature, we found a few reports on the application of intraoperative SLN recognition in endometrial cancer, and whether SLN recognition is necessary for endometrial cancer treatment is still in

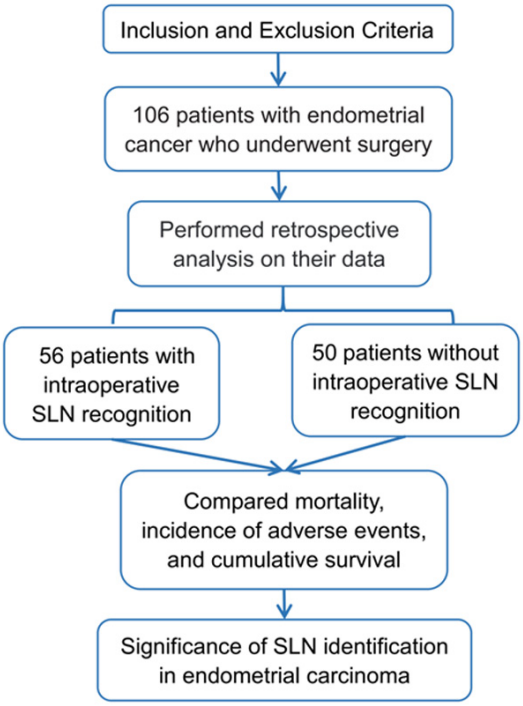

Figure 1. The technical steps of this study. To explore the significance of intraoperative sentinel lymph node (SLN) identification in endometrial cancer, clinical baseline data, SLN test data, and pathological examination results of 106 patients with endometrial carcinoma were analyzed retrospectively. SLN, sentinel lymph node.

debate. Therefore, this study analyzed the application value of intraoperative SLN identification and pathological examination in endometrial carcinoma.

Material and methods

Data source

The Medical Ethics Committee of Dalian Central Hospital approved the study. We performed a retrospective analysis of data from 106 patients with endometrial cancer who underwent surgery in our hospital from July 2020 to June 2021. Inclusion criteria: (1) Patients who met the clinical diagnostic criteria for endometrial cancer [10]; (2) Patients who were diagnosed and treated for the first time; (3) Patients who received surgical treatment under general intravenous anesthesia. Exclusion criteria: (1) Patients complicated with malignant tumors other than endometrial cancer; (2) Patients with incomplete data regarding clinical baseline data, SLN test data, and pathological examination results were missing. Patients with intraoperative SLN recognition were included in group A, and patients without intraoperative SLN recognition were included in group B. One percent methylene blue was the tracer used in patients undergoing intraoperative SLN identification, and these patients also underwent pathological examination. **Figure 1** is the flow chart of this study.

Clinical data collection

(1) Clinical baseline data were collected, including age, disease course, body mass index, clinical stage, histological type, degree of differentiation, muscular invasion, cervical interstitial, adnexal metastasis, vascular cancer thrombus, menopause, comorbidities, and tumor size. (2) SLN and pelvic abdominal lymph node distribution of the patients was collected. (3) Incidence of adverse events (including death, upper limb lymphedema, distant metastasis, and regional lymph node recurrence) and cumulative survival rate were also collected. The end time of follow-up was December 2022. Overall survival time was defined as the time from the discovery of EC to death or the end of follow-up.

Judging criteria

During the operation, doctors injected 1% methylene blue into the uterus and identified the first blue-stained lymph node. The SLN was removed and sent for frozen pathological examination. The positive expression of cytokeratin (CK) in SLN was observed by immunohistochemistry. CK positive expression is defined as cells with brown-yellow granules visible in the cytoplasm of lymph nodes. The patients with lymph node metastasis were performed with lymph node dissection.

SLN identification and inspection methods

(1) All patients underwent general intravenous anesthesia; (2) After laparotomy, doctors retained the peritoneal flushing solution for cytological examination; (3) We clamped (Supplier of forceps holder: Bochong Medical Technology (Shanghai) Co., LTD.) the roots of bilateral fallopian tubes, exposed the uterus, and protected the surrounding tissues and organs with gauze pads; (4) The dye tracer 1% methylene blue (Supplier: Shenzhen Zike Biotechnology Co., LTD.) was injected under the serous layer of the

Table 1. SLN and pelvic abdominal lymph node distribution in group A

Location	SLN (166 in total)		Pelvic and abdominal lymph nodes (813 in total)	
	Quantity	Proportion	Quantity	Proportion
External iliac region	63	37.95	325	39.98
Internal iliac	49	29.52	244	30.01
Common iliac	31	18.67	97	11.93
Obturator	13	7.83	81	9.96
Para-aortic	5	3.01	42	5.17
Near the palace	5	3.01	24	2.95

Note: SLN denotes sentinel lymph node.

Table 2. Clinical baseline data of Group A and B

Clinical baseline data	Group A (n=56)	Group B (n=50)	t/χ^2	*P*
Age ($\bar{x}\pm s$, years)	53.33±5.43	52.76±5.05	0.558	0.578
Course of disease ($\bar{x}\pm s$, months)	3.12±1.02	3.26±0.93	0.735	0.464
Body mass index ($\bar{x}\pm s$, kg/m^2)	21.91±3.22	22.03±3.14	0.194	0.847
Clinical stage (cases)			0.063	0.802
Stage I	30 (53.57)	28 (56.00)		
Stage II	26 (46.43)	22 (44.00)		
Histological types (cases)			0.020	0.990
Endometrioid adenocarcinoma	49 (87.50)	44 (88.00)		
Adenosquamous carcinoma	6 (10.71)	5 (10.00)		
Serous carcinoma	1 (1.79)	1 (2.00)		
Differentiated degree (cases)			0.366	0.833
High differentiation	26 (46.43)	24 (48.00)		
Moderately differentiated	21 (37.50)	20 (40.00)		
Poorly differentiated	9 (16.07)	6 (12.00)		
Muscle layer infiltration				
<1/2	48 (85.71)	43 (86.00)	0.002	0.966
>1/2	8 (14.29)	7 (14.00)		
Cervical stroma			>0.999	0.317
No involvement	52 (92.86)	47 (94.00)		
With involvement	4 (7.14)	3 (6.00)		
Annex transfer			1.514	0.219
No	56 (100.00)	49 (98.00)		
Yes	0 (0.00)	1 (2.00)		
Vascular tumor thrombus			2.548	0.110
No	54 (96.43)	49 (98.00)		
Yes	2 (3.57)	1 (2.00)		
Menopause			0.284	0.594
No	40 (71.43)	38 (76.00)		
Yes	16 (28.57)	12 (24.00)		
Complications				
Hypertension	9 (16.07)	5 (10.00)	0.849	0.357
Diabetes	10 (17.86)	7 (14.00)	0.292	0.589
Tumor size			0.242	0.623
<2 cm	34 (60.71)	28 (56.00)		
≥2 cm	22 (39.29)	22 (44.00)		

Figure 2. Sentinel lymph node stained with methylene blue of group A. In all group A patients, vascular blueness in the serous surface of the uterus and the pelvic funnel ligament was observed.

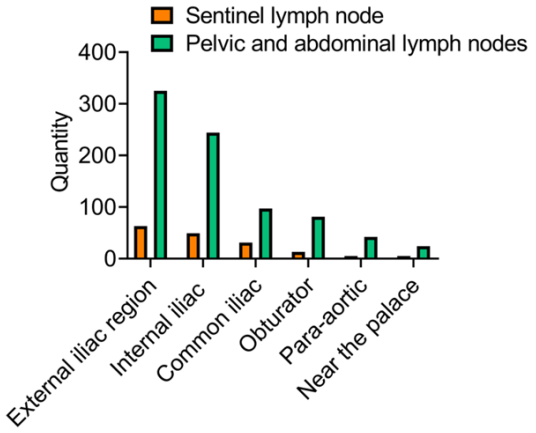

Figure 3. Distribution of sentinel lymph node and pelvic and abdominal lymph nodes in group A patients.

uterine body with a 5 mL syringe (Supplier: Anhui Kangtai Medical Equipment Co., LTD.). The injection sites were the midpoint of the bottom of the uterus, anterior wall midpoint, and posterior wall midpoint, with a total of 2 mL of 1% methylene blue injected; (5) They injected the two sides of the cervical isthmus with 2 mL 1% methylene blue; (6) Local compression and electrocoagulation were applied on injection point to prevent dye leakage; (7) The peritoneum was opened after injection of the dye tracer to expose the lymphatic drainage area; (8) After dissection along the direction of the blued lymphatic vessels, the first blued lymph nodes (the SLN) were observed; (9) The dyed SLNs were cut and frozen for pathological examination; (10) They dissected the uterus, bilateral adnexa, pelvic lymph nodes, and para-aortic lymph nodes.

Pathological examination

All the surgically removed lymph nodes and specimens were sent for pathological examination. The SLNs were embedded in paraffin. They dissected HE staining negative lymph nodes with a 400 um interval. A thickness of 4 um and immunohistochemical techniques were applied to detect the expression of CK. Micrometastasis is defined as the range of positive expression cell masses in successive sections exceeding 0.2-2.0 mm. CK monoclonal antibody (detected with immunohistochemical two-step method) was purchased from Wuhan Jinkairui Bioengineering Co., LTD.

Statistical methods

SPPS 23.0 was applied to process the data, and the mean ± standard deviation was applied to describe the measurement data such as age, disease course, and body mass index, and a t-test was carried out for analysis. The counting data were expressed as the number or percentage, and analyzed by chi-square test or Fisher's exact test. Log Rank (Mantel-Cox) test was used to analyze cumulative survival. The difference was statistically significant at $P<0.05$.

Results

Clinical baseline data of patients in group A and B

There were 56 cases in the group A and 50 cases in the group B. There was no statistical significance in the clinical baseline data between groups A and B (all P<0.05, **Table 2**), indicating the two groups were comparable.

Distribution of SLN and pelvic and abdominal lymph nodes in group A patients

Figure 2 shows SLN stained with methylene blue. The uterine serous surface and the vessels of the pelvic funnel ligament were blue in group A patients. There were 813 lymph nodes detected and an average of (30.36±5.27) lymph nodes removed. Forty-one patients with

Table 3. Incidence of adverse events in group A and B

Group	Number of cases	Deaths	Upper limb lymphedema	Distant metastasis	Regional lymph node recurrence	Total adverse events
Group A (n=56)	3 (5.36)	4 (7.14)	2 (3.57)	1 (1.79)	10 (17.86)	3 (5.36)
Group B (n=50)	2 (4.00)	2 (4.00)	2 (4.00)	0 (0.00)	8 (16.00)	2 (4.00)
χ^2						0.065
P						0.799

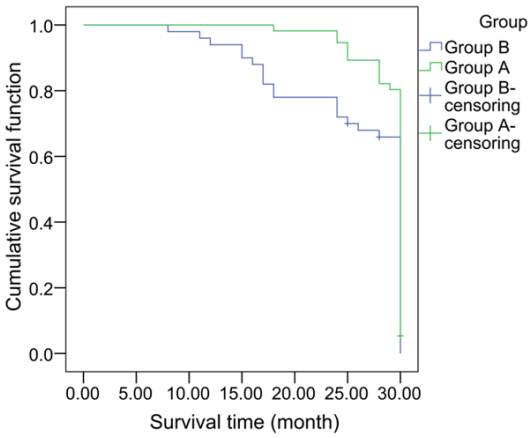

Figure 4. Kaplan-Meier survival curve of group A and group B.

SLN metastasis were successfully identified and removed during the operation, with a recognition rate of 82.14% (46/56). There were 166 (20.42%) SLNS removed. **Table 1** and **Figure 3** show the distributions of SLN and pelvic and abdominal lymph nodes.

There were 15 patients with no SLN metastasis found in the intraoperative pathological examination, including two patients with poorly differentiated adenocarcinoma and clinical stage II patients. Three patients showed CK-positive expression cell clusters through immunohistochemical staining, which was determined to be SLN micro-metastasis but no cancer tissue metastasis in the lymph node dissection.

Incidence of adverse events in group A and group B patients

There was no significant difference in the incidence of total adverse events between groups A and B ($P>0.05$). See **Table 3**.

Cumulative survival analysis

The cumulative survival rate of group A patients was significantly higher than that of group B patients ($P=0.018$, **Figure 4**).

Discussion

Abdominal or pelvic lymph node metastasis is a basis for the operation-pathological staging of endometrial cancer and a key factor affecting the prognosis and recurrence of the disease. Most studies believe that micrometastases of endometrial cancer significantly increase the possibility of recurrence [11, 12]. SLN identification and pathological biopsy can improve the detection rate of micro-metastases, thus improving surgical staging accuracy and the accuracy of prognosis and guiding the judgment of subsequent adjuvant therapy. Therefore, it is necessary to investigate the clinical significance of SLN identification and pathological biopsy for endometrial carcinoma.

SLN is the primary site of lymphatic drainage in malignant tumors, and the earliest metastasis of malignant tumors occur in SLN [13, 14]. Therefore, the intraoperative identification and pathological examination of SLN can accurately determine lymphatic drainage direction and make a preliminary determination of whether lymph nodes in the subsequent lymphatic drainage area have tumor tissue metastasis. For SLN patients without metastasis, blind and extensive lymph node dissection can be avoided, thus reducing trauma and complication occurrence [15]. Good tracer selection and appropriate injection sites are key factors to ensure successful SLN detection [16]. In this study, the tracer selected by the patients in group A in the process of SLN recognition was the commonly used 1% methylene blue. In line with relevant statistics, the recognition rate of this tracer dye was 70%-90% [17]. Methylene blue is a non-toxic and non-radioactive dye, requiring no special equipment for color development, and has the advantages of simple operation, non-toxic, safe, intuitive, low-cost, and easy to promote [18]. The injection sites were the patient midpoint of the uterus, the anterior and posterior wall midpoint, and two sides of the cervical isthmus. Cervical injection

is the most widely used method at present. Since uterine fibroids and endometrial lesions do not cause cervical deformation, the injection site has good accessibility and can increase the recognition rate [19]. According to the clinical data, 46 patients of group A were successfully identified and removed SLN, with a recognition rate of 82.14%. The reason for identifying SLE failure may be related to the unskilled and inexperienced medical personnel in technical operations [20]. Some studies show that the principal way of lymphatic drainage of the uterine body was through the internal iliac, obturator foramen, and common iliac lymph nodes [21]. Our study concluded that the top 3 SLN distribution proportions were external, internal, and total iliac lymph nodes in group A patients, confirming the uncertainty of uterine body lymphatic drainage again. Therefore, intraoperative SLE identification is indeed necessary. We found 15 cases of patients with no SLN metastasis in the intraoperative pathological examination, among which 2 cases had poorly differentiated adenocarcinoma with clinical stage II stained by immunohistochemistry in group A patients. Three patients had SLN micrometastases but had no metastasis in lymph node dissection. Therefore, we believe that SLN-negative patients, especially those with low tissue differentiation and high clinical stage, using immunohistochemical staining can avoid false negatives to conduct supplementary treatment for patients after surgery [22]. The results of this study showed that there was no significant difference in the incidence of total adverse events between patients who received SLN recognition and those who did not receive SLN recognition, indicating that SLN recognition had no significant damage to patients with endometrial cancer and did not significantly cause adverse events, indicating that this method was safe. In addition, the cumulative survival time of patients with SLN recognition was longer than that of patients without SLN recognition, which indicates that SLN recognition could prolong the survival of patients.

In conclusion, intraoperative SLE identification can avoid false negative results, is safe and feasible, and can prolong the survival time of patients with endometrial cancer. However, this is a retrospective single-center study with selectivity bias and needs further exploring on large sample size and prospective studies.

Disclosure of conflict of interest

None.

Address correspondence to: Bangruo Qi, Department of Obstetrics and Gynecology, Sanya Women and Children's Hospital, No. 339 Yingbin Road, Jiyang District, Sanya, Hainan, China. Tel: +86-13262755666; E-mail: bangruoqi@126.com

References

[1] Oaknin A, Tinker AV, Gilbert L, Samouëlian V, Mathews C, Brown J, Barretina-Ginesta MP, Moreno V, Gravina A, Abdeddaim C, Banerjee S, Guo W, Danaee H, Im E and Sabatier R. Clinical activity and safety of the anti-programmed death 1 monoclonal antibody dostarlimab for patients with recurrent or advanced mismatch repair-deficient endometrial cancer: a nonrandomized phase 1 clinical trial. JAMA Oncol 2020; 6: 1766-1772.

[2] Nagar H, Wietek N, Goodall RJ, Hughes W, Schmidt-Hansen M and Morrison J. Sentinel node biopsy for diagnosis of lymph node involvement in endometrial cancer. Cochrane Database Syst Rev 2021; 6: CD013021.

[3] Frumovitz M, Plante M, Lee PS, Sandadi S, Lilja JF, Escobar PF, Gien LT, Urbauer DL and Abu-Rustum NR. Near-infrared fluorescence for detection of sentinel lymph nodes in women with cervical and uterine cancers (FILM): a randomised, phase 3, multicentre, non-inferiority trial. Lancet Oncol 2018; 19: 1394-1403.

[4] Dowdy SC. Improving oncologic outcomes for women with endometrial cancer: realigning our sights. Gynecol Oncol 2014; 133: 370-374.

[5] Park JY, Kim JH, Baek MH, Park E and Kim SW. Randomized comparison between sentinel lymph node mapping using indocyanine green plus a fluorescent camera versus lymph node dissection in clinical stage I-II endometrial cancer: a Korean Gynecologic Oncology Group trial (KGOG2029/SELYE). J Gynecol Oncol 2022; 33: e73.

[6] Gu Y, Cheng H, Zong L, Kong Y and Xiang Y. Operative and oncological outcomes comparing sentinel node mapping and systematic lymphadenectomy in endometrial cancer staging: meta-analysis with trial sequential analysis. Front Oncol 2021; 10: 580128.

[7] Balaya V, Guani B, Benoit L, Magaud L, Bonsang-Kitzis H, Ngô C, Le Frère-Belda MA, Mathevet P and Lécuru F. Diagnostic value of frozen section examination of sentinel lymph nodes in early-stage cervical cancer at the time of ultrastaging. Gynecol Oncol 2020; 158: 576-583.

[8] Vermersch C, Raia-Barjat T, Chapelle C, Lima S and Chauleur C. Randomized comparison between indocyanine green fluorescence plus 99mtechnetium and 99mtechnetium alone methods for sentinel lymph node biopsy in breast cancer. Sci Rep 2019; 9: 6943.

[9] Coleman RL, Ali S, Levenback CF, Gold MA, Fowler JM, Judson PL, Bell MC, De Geest K, Spirtos NM, Potkul RK, Leitao MM Jr, Bakkum-Gamez JN, Rossi EC, Lentz SS, Burke JJ 2nd, Van Le L and Trimble CL. Is bilateral lymphadenectomy for midline squamous carcinoma of the vulva always necessary? An analysis from Gynecologic Oncology Group (GOG) 173. Gynecol Oncol 2013; 128: 155-159.

[10] Professional Committee of Gynecological Oncology and Chinese Anti-Cancer Association. Guidelines for the diagnosis and treatment of endometrial cancer (2021 edition). Chin J Oncol 2021; 31: 501-512.

[11] de Boer SM, Powell ME, Mileshkin L, Katsaros D, Bessette P, Haie-Meder C, Ottevanger PB, Ledermann JA, Khaw P, D'Amico R, Fyles A, Baron MH, Jürgenliemk-Schulz IM, Kitchener HC, Nijman HW, Wilson G, Brooks S, Gribaudo S, Provencher D, Hanzen C, Kruitwagen RF, Smit VTHBM, Singh N, Do V, Lissoni A, Nout RA, Feeney A, Verhoeven-Adema KW, Putter H and Creutzberg CL; PORTEC Study Group. Adjuvant chemoradiotherapy versus radiotherapy alone in women with high-risk endometrial cancer (PORTEC-3): patterns of recurrence and post-hoc survival analysis of a randomised phase 3 trial. Lancet Oncol 2019; 20: 1273-1285.

[12] Vale CL, Tierney J, Bull SJ and Symonds PR. Chemotherapy for advanced, recurrent or metastatic endometrial carcinoma. Cochrane Database Syst Rev 2012; 2012: CD003915.

[13] Soderini A, Depietri V, Crespe M, Rodriguez Y and Aragona A. The role of sentinel lymph node mapping in endometrial carcinoma. Minerva Ginecol 2020; 72: 367-383.

[14] Matei D, Filiaci V, Randall ME, Mutch D, Steinhoff MM, DiSilvestro PA, Moxley KM, Kim YM, Powell MA, O'Malley DM, Spirtos NM, Small W Jr, Tewari KS, Richards WE, Nakayama J, Matulonis UA, Huang HQ and Miller DS. Adjuvant chemotherapy plus radiation for locally advanced endometrial cancer. N Engl J Med 2019; 380: 2317-2326.

[15] Frich L, Hermann R, Berentzen Å and Ryder T. Randomized study of wound drainage on early complications after lymph node dissection for melanoma. J Surg Res 2021; 267: 467-476.

[16] Persson J, Salehi S, Bollino M, Lönnerfors C, Falconer H and Geppert B. Pelvic sentinel lymph node detection in high-risk endometrial cancer (SHREC-trial)-the final step towards a paradigm shift in surgical staging. Eur J Cancer 2019; 116: 77-85.

[17] Paulinelli RR, Freitas-Junior R, Rahal RM, Oliveira LF, Vilela MH, Moreira MA, Alves KL, Peleja MB and Resende TC. A prospective randomized trial comparing patent blue and methylene blue for the detection of the sentinel lymph node in breast cancer patients. Rev Assoc Med Bras (1992) 2017; 63: 118-123.

[18] Thongvitokomarn S and Polchai N. Indocyanine green fluorescence versus blue dye or radioisotope regarding detection rate of sentinel lymph node biopsy and nodes removed in breast cancer: a systematic review and meta-analysis. Asian Pac J Cancer Prev 2020; 21: 1187-1195.

[19] Mueller JJ, Dauer LT, Murali R, Iasonos A, Pandit-Taskar N, Abu-Rustum NR and Grimm J. Positron lymphography via intracervical 18F-FDG injection for presurgical lymphatic mapping in cervical and endometrial malignancies. J Nucl Med 2020; 61: 1123-1130.

[20] Deken MM, van Doorn HC, Verver D, Boogerd LSF, de Valk KS, Rietbergen DDD, van Poelgeest MIE, de Kroon CD, Beltman JJ, van Leeuwen FWB, Putter H, Braak JPBM, de Geus-Oei LF, van de Velde CJH, Burggraaf J, Vahrmeijer AL and Gaarenstroom KN. Near-infrared fluorescence imaging compared to standard sentinel lymph node detection with blue dye in patients with vulvar cancer - a randomized controlled trial. Gynecol Oncol 2020; 159: 672-680.

[21] Tu H, Wan T, Zhang X, Gu H, Feng Y, Huang H and Liu J. Potential risks in sentinel lymph node biopsy for cervical cancer: a single-institution pilot study. World J Surg Oncol 2020; 18: 133.

[22] Bilchik AJ and Trocha SD. Lymphatic mapping and sentinel node analysis to optimize laparoscopic resection and staging of colorectal cancer: an update. Cancer Control 2003; 10: 219-223.