Sectioning Protocol Determines Accuracy of Intraoperative Pathological Examination of Sentinel Lymph Node in Cervical Cancer: A Systematic Review and Meta-analysis

Hua Tu[1], Haifeng Gu[1], He Huang[1], Kaijiang Liu[2], Jingping Yun[1], Jieping Chen[1], Xinke Zhang[1], Hideaki Yahata[3], Kenzo Sonoda[3], Haoyang Zhang[4], and Jihong Liu[1]

[1]Sun Yat-sen University Cancer Center
[2]Affiliation not available
[3]Kyushu University
[4]Sun Yat-Sen University

April 28, 2020

Abstract

Background In cervical cancer, the benefits of sentinel lymph node biopsy (SLNB) have long been confined by the lack of precise intraoperative pathological examination. Objective To determine the diagnostic performance and optimal protocol of frozen section examination (FSE) in SLNB for cervical cancer. Search Strategy PubMed, EMBASE, Web of Science, Cochrane Library, Wanfang Data and China National Knowledge Infrastructure were searched from inception to July 30, 2019. Selection Criteria Studies reporting the data of SLNB combined with FSE in cervical cancer were included. Data Collection and Analysis Two independent reviewers extracted the data. Bivariate mixed-effects regression model was applied for analyses. Sensitivity of FSE in detecting SLN metastasis was the primary diagnostic indicator for evaluation. Main Results The pooled sensitivity of FSE among 31 eligible studies (1887 patients) was 0.77 (95% CI 0.66–0.85) with high heterogeneity (I^2=69.73%). Two representative FSE protocols were identified from 26 studies, described as equatorial (E-protocol, SLN was bisected) and latitudinal (L-protocol, SLN was cut at intervals). Meta-regression showed that FSE protocol was the only source of heterogeneity ($p<0.001$). The pooled sensitivity was 0.86 (95% CI 0.79–0.91, I^2=0%) and 0.59 (0.46–0.72, I^2=58.47%) for FSE using L- and E- protocol, respectively. The pooled sensitivity of FSE using L-protocol would reach 0.97 (0.89–0.99) if only marcometastases were considered. These findings were robust to sensitivity analyses. Conclusions With L-protocol, FSE can provide precise intraoperative pathology for SLNB, which enables immediate decision-making for individualized managements.
Keywords Cervical cancer, sentinel lymph node, metastasis, frozen section, sensitivity, meta-analysis.

Sectioning Protocol Determines Accuracy of Intraoperative Pathological Examination of Sentinel Lymph Node in Cervical Cancer: A Systematic Review and Meta-analysis

Hua Tu*, Hai-Feng Gu*, He Huang*, Kai-Jiang Liu, Jing-Ping Yun, Jie-Ping Chen, Xin-Ke Zhang, Hideaki Yahata, Kenzo Sonoda, Hao-Yang Zhang, Ji-Hong Liu

*Contributed equally as joint first authors

Running title: Frozen section of sentinel nodes in cervical cancer

Department of Gynecologic Oncology (H Tu MD, H-F Gu MD, H Huang MD, J-P Chen MD, Prof J-H Liu MD PhD), **Department of Pathology**(X-K Zhang MD, Prof J-P Yun MD), **Sun Yat-sen University Cancer Center, State Key Laboratory of Oncology in South China, Collaborative Innovation Center for Cancer Medicine, Guangzhou, China.**

Department of Gynecologic Oncology (Prof K-J Liu MD),

Renji Hospital, Medical College of Shanghai Jiaotong University, Shanghai, China.

Departments of Obstetrics and Gynecology (Prof H Yahata MD PhD, Prof K Sonoda MD PhD), **Graduate School of Medical Sciences, Kyushu University, Fukuoka, Japan.**

School of Data and Computer Science (H-Y Zhang MSc),**Sun Yat-sen University, Guangzhou, China.**

Correspondence to: Prof Ji-Hong Liu, Department of Gynecologic Oncology, Sun Yat-sen University Cancer Center, State Key Laboratory of Oncology in South China, Collaborative Innovation Center for Cancer Medicine, Guangzhou, 510060, China. Tel: +86-20-87343014.

liujih@mail.sysu.edu.cn.

Background

In cervical cancer, the benefits of sentinel lymph node biopsy (SLNB) have long been confined by the lack of precise intraoperative pathological examination.

Objective

To determine the diagnostic performance and optimal protocol of frozen section examination (FSE) in SLNB for cervical cancer.

Search Strategy

PubMed, EMBASE, Web of Science, Cochrane Library, Wanfang Data and China National Knowledge Infrastructure were searched from inception to July 30, 2019.

Selection Criteria

Studies reporting the data of SLNB combined with FSE in cervical cancer were included.

Data Collection and Analysis

Two independent reviewers extracted the data. Bivariate mixed-effects regression model was applied for analyses. Sensitivity of FSE in detecting SLN metastasis was the primary diagnostic indicator for evaluation.

Main Results

The pooled sensitivity of FSE among 31 eligible studies (1887 patients) was 0.77 (95% CI 0.66–0.85) with high heterogeneity (I^2=69.73%). Two representative FSE protocols were identified from 26 studies, described as equatorial (E-protocol, SLN was bisected) and latitudinal (L-protocol, SLN was cut at intervals). Meta-regression showed that FSE protocol was the only source of heterogeneity ($p<0.001$). The pooled sensitivity was 0.86 (95% CI 0.79–0.91,I^2=0%) and 0.59 (0.46–0.72,I^2=58.47%) for FSE using L- and E-protocol, respectively. The pooled sensitivity of FSE using L-protocol would reach 0.97 (0.89–0.99) if only marcometastases were considered. These findings were robust to sensitivity analyses.

Conclusions

With L-protocol, FSE can provide precise intraoperative pathology for SLNB, which enables immediate decision-making for individualized managements.

Keywords

Cervical cancer, sentinel lymph node, metastasis, frozen section, sensitivity, meta-analysis.

Funding

This work was supported by the Health and Medical Cooperation Innovation Special Program of Guangzhou Municipal Science and Technology (grant number: 158100075).

Tweetable abstract

In cervical cancer, sentinel nodal metastases can be precisely detected by frozen section examination using proper protocol.

Introduction

Over the past decades, sentinel lymph node biopsy (SLNB) has become an attractive surgical procedure in many malignancies.[1-5]The benefits of SLNB mainly include a reduction in surgical morbidity and time cost, achieved by replacing lymphadenectomies in selected patients,[6,7] and revelation of aberrant drainage regions that are probably omitted during routine lymphadenectomies.[8] Furthermore, by pathological ultrastaging of sentinel lymph nodes (SLN), the micro-metastatic burden in lymphatic system can be evaluated conveniently, enabling more precise individualized treatments.[9-11] With these benefits and high accuracy.[12] SLNB is currently recommended as an alternative of pelvic lymph node dissection (PLND) in early-stage cervical cancer.[13,14]

However, in clinical practice, only a few authors have attempted to perform SLNB alone without a further PLND.[6,7,15] A recent international survey by the Gynecologic Cancer Intergroup showed great divergence regarding the SLNB strategy,[16]reflecting worldwide mistrust on intraoperative decisions made based on SLNB. Indeed, previous studies had shown that the accuracy of SLNB largely relied on postoperative ultrastaging,[17,18]which is time consuming and unavailable for intraoperative decision-making. As a result, many gynecologists choose to directly replace PLND with SLNB in radical surgery and wait for final pathology. However, nodal metastasis has been included as IIIC stage in the latest International Federation of Gynecology and Obstetrics (FIGO) staging system,[19] so it is becoming increasingly important to acquire the lymphatic status before deciding treatment modality. The European Society of Gynaecological Oncology/European Society for Radiotherapy and Oncology/European Society of Pathology guidelines (2018) therefore recommended submitting SLNs for intraoperative assessment to immediately triage patients towards radical surgery or definitive chemoradiotherapy.[14] Consequently, accurate intraoperative pathology of SLNs is urgently required.

Frozen section examination (FSE) is the most common method for intraoperative SLN assessment.[20] Compared with other methods, FSE has a natural superiority of almost 100% specificity.[21-23]However, the sensitivity of FSE varies considerably between the published studies.[24-35] Some authors had cautioned the high false-negative rate of FSE in SLNB,[24-27,29] whereas others provided satisfying results.[30,32-35] The reason for such a discrepancy remains unclear and may be associated with the heterogeneity among these studies, including the differences in methodologies, patients' characteristics, volume of metastases, as well as pathologists' experiences. However, few studies had concerned the impacts of these factors on the sensitivity of FSE.[29,33] It spontaneously interests us whether there exists an optimal protocol by which the FSE can yield the highest sensitivity for intraoperative decision-making.

To our knowledge, there are several ongoing international multicenter trials targeting the validation of SLNB in early-stage cervical cancer.[36,37] All these trials were designed with an intraoperative randomization or assignment depended on the results of FSE. Therefore, it is of great importance to validate the accuracy of FSE first. In this systematic review and meta-analysis, we analyzed the available data on this issue, in order to determine the diagnostic performance and optimal protocol of FSE in SLNB for cervical cancer and provide evidence for ongoing and future studies.

Methods

Searchstrategy and selection criteria

We performed and reported this systematic review and meta-analysis in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analysis of Diagnostic Test Accuracy (PRISMA-DTA) guideline. The study protocol was registered with PROSPERO, number CRD42019130044.

We systematically searched PubMed, EMBASE, Web of Science, Cochrane Library, Wanfang Data and

China National Knowledge Infrastructure (from inception to July 30, 2019) for studies on SLNB with FSE in cervical cancer. The full search strategy was provided in supplementary (p4). No publication date or language restrictions were applied. We manually searched the references of all resulting publications to identify any studies possible missed by the initial search. Besides, we contacted experts in this field with questionnaires for possible unpublished data (supplementary p4–6).

The population of interest was women with cervical cancer initially managed by surgery involving a SLNB procedure combined with FSE. Studies containing these individuals were potentially eligible for review. There were no restrictions for the study purpose or design, methods for SLN detection, or surgical approaches. For qualitative and quantitative synthesis, we included only studies reporting the results of FSE and definitive pathological examination (DPE). Besides, we excluded studies in which the sample size was less than ten, no metastatic case was found, or FSE were selectively performed. In addition, we checked the repetition between studies from the same institution. If two or more studies were found to had irremovable overlaps in studied population, only the most recent study reporting adequate information was included while others were excluded.

Data extraction and quality assessment

We extracted detailed information from the full texts of included studies (supplementary p7). Outcomes of interest was the diagnostic performance of FSE of SLN, including sensitivity, true positives (TP) and false negatives (FN). The reference standard was DPE based on the paraffin sections of SLN. Sensitivity was assumed as the primary indicator of the diagnostic performance and defined as the ratio of cases having both positive FSE and DPE within all DPE-positive patients. TP were defined as cases having both positive FSE and DPE, and FN as cases having negative FSE and positive DPE. Isolated tumor cells (ITC) were defined as tumor diameter $<$0.2 mm, micrometastasis (MIM) as between 0.2 and 2 mm, and macrometastasis (MAM) as $>$2 mm. We took these parameters directly from the original papers or calculated them with available information. We excluded cases who had no SLN detected or did not underwent FSE. Whenever a calculation was impossible, we contacted the authors for necessary information.

In previous publications the term FN usually referred to cases presenting positive non-SLN without a positive SLN. Of note, in this study we redefined these cases as true negative given that their SLNs were tumor-free. We assessed the qualities of the included studies using Quality Assessment of Studies of Diagnostic Accuracy-2 (QUADAS2) tool.

Statistical analysis

We examined the heterogeneity assumption using Cochran's Q test and quantified it using Higgins I^2 statistic, with$I^2 > 50\%$ considered statistically heterogeneous. We used bivariate mixed-effects regression model to calculate pooled sensitivity and 95% confidence intervals (CI). We performed meta-regression and subgroup analysis to investigate the potential impact of variates on sensitivity and the source of heterogeneity. Results were displayed graphically on forest plots. We conducted sensitivity analysis to evaluate the robustness of the results by removing studies with high weight or debatable methodology, modifying stratified criteria, or altering statistical model. We did all statistical analyses with STATA (version 15.0; Stata Corporation, College Station, TX, USA)

Funding

This work was funded by Health and Medical Cooperation Innovation Special Program of Guangzhou Municipal Science and Technology. The funders had no role in the design and conduct of the study; collection, management, analysis, and interpretation of the data; preparation, review, and approval of the manuscript; or decision to submit the manuscript for publication.

Results

We yielded a total of 1261 publications by database searches, of which 131 studies were identified after exclusion of reduplicative and irrelevant studies. After abstracts screening, 73 unique studies remained, and all full texts were obtained. After reviewing of the full texts, we excluded 44 studies. The reasons for each exclusion of these studies were listed in supplementary (p8–9). Besides, we obtained five reports of unpublished data and two of them were eligible (supplementary p15–16). Finally, 31 eligible studies (29 publications) were included. Figure 1 displays the PRISMA (2009) flow diagram for study selection.

The publication years of the 31 identified studies were between 2002 and 2019. Most of these studies were done in Europe, Asia, and the USA. A total of 1887 eligible patients were identified from the 31 studies. Each of them underwent SLNB for cervical cancer, with at least one SLN detected and examined by intraoperative FSE.

The characteristics of the 31 finally included studies were listed in supplementary (p17). Patients with early-stage disease accounted for 66.1–100% of whole populations, with 19 out of 31 (61.3%) studies having 100% patients at early stage. The most common histological types were squamous cell carcinoma, followed by adenocarcinoma. The rate of nodal metastasis varied between 5.4–36.4% among the included studies.

The surgical approaches were affected by the FSE results in 18 studies, of which 13 performed additional para-aortic lymphadenectomy in positive cases. Only four studies omitted PLND in cases of negative FSE. Four studies applied negative FSE as a prerequisite of fertility preservation. Eight studies transferred radical surgeries to concurrent chemoradiotherapy in positive cases. Besides, one study performed simple hysterectomy in cases of negative FSE. Twelve studies reported follow-up results and the oncological outcomes were generally good (supplementary p22).

The sectioning protocols of FSE consisted of three different approaches as following:

1. SLN was bisected, one section was taken from the maximum surface of one half SLN (9 studies);
2. SLN was bisected, adjacent sections were taken from the maximum surface of each half SLN (4 studies);
3. SLN was cut into pieces at certain intervals (varied from 2 to 5 mm, perpendicularly to their long axis), and one or more sections were taken from each piece (13 studies).

For subgroup analysis, we defined the two "bisected" protocols together as equatorial (E), and the third one as latitudinal (L) (figure 2). The next steps were similar in which the sections were examined after staining with hematoxylin and eosin (H&E) and the rest tissue were embedded in paraffin. For DPE, additional ultrastaging were performed in 24 studies, while routine pathological examination was performed alone in seven. Among the 24 studies using ultrastaging, serial sectioning combined with immunohistochemistry (IHC), serial sectioning alone, and IHC alone were performed in 18, 2 and 4, respectively.

In published pathological studies at least four further step sections combined with IHC examination were recommended for SLN ultrastaging.[38] We utilized this criterion to evaluate the stringencies of ultrastaging techniques and their potential influences. DPEs meeting this criterion were judged as adequate, otherwise as inadequate. The descriptions and judgements of FSE and DPE protocol for each study were presented in supplementary (p20–21).

The process of data extraction for meta-analysis were detailed in supplementary p10–16. Overall, there were 363 patients having SLN metastases confirmed by DPE, of whom 115 were misdiagnosed by FSE. The sensitivities of FSE varied over a wide range of 0%–100% among the 31 included studies (table 1). Specificities were 100% in all studies except two reporting false positive. Both were due to misdiagnosis of endosalpingiosis in SLN.[6,30]Pooled analysis using mixed-effect model yielded an estimated rate of 0.77 (95%CI: 0.66–0.85, figure 2) for sensitivity. Heterogeneity test for sensitivities showed high heterogeneity among the included studies (Q=99.09,I^2=69.73%, p<0.001).

Among 26 studies with definable pathological protocols, 19 reported FN results. The metastatic types of FNs were available in 18 studies, including 24 MAM, 51 MIM, and 29 ITC in 101 patients (table 2). The metastatic types of TPs were available in nine studies, including 95 MAM, 14 MIM, and one ITC in 110 patients (table 3). The pooled sensitivity of FSE were 0.79 (95%CI: 0.70–0.86) if ITC were not considered, and 0.94 (95%CI: 0.85–0.98) if only MAM was considered. Notably, only four of the 24 (16.7%) FN-MAM were missed, whereas 13 of the 15 (86.7%) TP-MIM/ITC were detected under L-protocol.

We conducted meta-regressions to investigate the source of heterogeneity in sensitivities. Studies were categorized into subgroups by the study design (prospective and retrospective), SLNB strategy (whether the surgical approaches were affected by FSE results: yes and no), sample size (<60 and [?]60), overall metastatic rate (<20% and [?]20%), reference standard (DPE protocol: adequate and inadequate), and index test (FSE protocol: E and L). Five studies were excluded from meta-regression due to undefinable methodologies. Finally, FSE protocol was found to be the only source of heterogeneity (p<0.001, table 4).

Subgroup analysis showed decreased heterogeneities in both subgroups stratified by FSE protocol (Q=7.59, I^2=0%, p=0.82 for L-protocol; Q=28.90, I^2=58.47%, p<0.001 for E-protocol). The sensitivity pooled achieved 0.86 (95%CI: 0.79–0.91) in the L-protocol subgroup (13 studies, 650 patients), whereas it was 0.59 (0.46–0.72) in the E-protocol subgroup (13 studies, 1047 patients). The difference reached statistical significance (P<0.001). If ITC was not considered, the pooled sensitivities would be 0.88 (0.81–0.93) and 0.64 (0.52–0.75) for L and E-protocol subgroup, respectively (p<0.001). If only MAM was considered, the pooled sensitivities would be 0.97 (95%CI 0.89–0.99) and 0.86 (0.74–0.93) for L and E-protocol subgroup, respectively (p=0.01). In sensitivity analyses (supplementary p23–27), whatever the alterations made in study setting or statistical model, the observation that sectioning protocol determined the accuracy of FSE, remained unchanged.

Discussion

Main findings

In this study, we systematically investigated the diagnostic performance of FSE in SLNB for cervical cancer, based on the available data over nearly twenty years. To our knowledge, this is the first systematic review and meta-analysis on this topic. The sensitivity of FSE in SLNB, which had been surrounded by controversies, was found to be dissatisfactory in pooled analysis (0.77, 95%CI 0.66–0.85). This data is very close to that previously reported in breast cancer.[54]However, a more important finding of this study is that the sectioning protocol of FSE had great impact on diagnostic accuracy, which also generated a high heterogeneity (I^2=69.73%). Significantly improved sensitivities (pooled: 0.86, 95%CI: 0.79–0.91) and low heterogeneity (I^2=0) were observed among the studies using L-protocol, whereas an even lower sensitivity (0.59, 0.46–0.72) was pooled under E-protocol. This difference was so remarkable that we expected it would hardly be denied or reversed in future studies. Thus, our study provides strong evidence supporting L-protocol as the standard for intraoperative pathological examination of SLNs.

PLND shares the same incision with hysterectomy, lowering the priority of a two-step surgery strategy.[29] In addition, it is still inconclusive whether PLND can be completely replaced by SLNB in early-stage cervical cancer. For these reasons, intraoperative pathological diagnosis remains an important element in SLNB for cervical cancer. Despite wide recognition, the benefits of SLNB in cervical cancer have long been confined by the lack of precise intraoperative pathology.[18]Similar dilemmas are also encountered in other malignancies such as gastric cancer.[55] The major contribution of this meta-analysis is that, for the first time, we identified a simple method to achieve more precise intraoperative SLN assessment, which enables immediate decision-making for individualized treatments.[7]

Strengths and Limitations

This study has several strengths. First, a meticulous screening was carried out to ensure that the data for analyzing did not contain any reduplicative or ineligible individual. On the other hand, we had done our best to re-analyze the existing data, collect unpublished studies and contact the authors to obtain precise diagnostic information for each included individual (supplementary p10–16). Each step of data synthesis was detailed to make our results reproducible. Besides, most of the included studies were prospective, used multiple tracers for SLNB and ultrastaging for DPE, indicating low risk of bias in the results. Furthermore, the L and E-protocol can be clearly distinguished and were evenly used in previous studies, suggesting the generalizability for both. In addition, the studies in which FSE was selectively performed (usually for SLNs with suggestive appearance) were excluded because there were high risks of bias (supplementary). Finally, the sensitivity analyses demonstrated the robustness of the main findings (supplementary).

This study also has several limitations. First, only nine studies provided the sizes of TP and the question how many MIM/ITC were detected under different protocols has not been satisfactorily answered. Furthermore, due to the multidisciplinary nature of SLNB technique, the sample size of single study was usually small. We could not eliminate the impact of pathologists' experience although all involved institutions were highly specialized. This also poses a further question whether our results can be validated in hospitals without enough experience in SLNB. Besides, due to the length of time elapsed, some authors could not be contacted or provide requested information, including the diagnostic data, protocols of FSE and DPE, and the metastatic types.

Interpretation

In this study we found that many pathologists examined only one H&E-stained frozen section for each SLN in order to reserve more tissue for DPE.[25-29] This consideration may be more reasonable for assessing SLN of breast cancer, in which the axillary lymphadenectomy can be performed asynchronously and usually replaced by chemoradiotherapy.[56] Actually, the use of FSE in SLNB for breast cancer has significantly decreased during the past years.[57] In early-stage cervical cancer, however, recent viewpoints have begun to emphasize the avoidance of combination of surgery and radiotherapy, since there was a significantly increased morbidity.[14] So the management will be challenging once the FSE result was found to be false. Therefore, best efforts are required in intraoperative diagnosis and the FSE protocol should be given enough attention, especially in fertility-preserving surgeries.[58]

Generally, FN results can be caused by technical errors in sectioning processes or judgment errors in reviewing processes. Gortzak-Uzan and colleagues[49] reported a technical error on MAM with diameter of 4-mm, which was not observed in the frozen sections but hided in the remaining tissue. Only four MAM were missed among the 13 studies using L-protocol. However, such FN results seemed more common in the studies using E-protocol since there were 20 MAM omitted in total. In the study by Slama and colleagues[29] one-level section was examined for each node and nine of 48 MAM were missed by FSE. The median diameter of these FN-MAM was 3.94 mm and the largest one reached 8.4 mm, which could hardly be neglected in reviewing processes. So, it is reasonable to speculated that most of these FNs were technical errors and could have been avoided by taking sections at short intervals.

Some may doubt that the high FN rates were due to strict ultrastaging in which more occult metastases might be revealed. This explanation also seems reasonable. However, in this meta-analysis, most of the studies used both serial sectioning and IHC examination for ultrastaging. We classified these techniques using a recommended criterion by previous pathological studies.[38] Yet only the FSE protocol was found to be a source of heterogeneity in meta-regression, whereas the DPE protocol showed minimal impact on sensitivity. This observation was further confirmed by sensitivity analyses (supplementary). A more reasonable explanation is that, in E-protocol more lymph tissue was reserved for DPE, which inevitably carried higher opportunity to have metastasis within, regardless of the method for detection.

The clinical significance of MIM/ITC in SLN remains to be clarified.[59,60] Okamoto and colleagues found that non-SLN were seldom involved if SLN harbored merely MIM/ITC.[61] In the SENTICOL study, only one recurrence was observed among 16 patients having MIM/ITC in SLN.[10] Besides, three included studies showed favorable oncological outcomes despite that PLND were omitted in FN-FSE cases.[6,15,49] Taken together, these evidences suggested that MIM/ITC only represented the very beginning of lymphatic spread, and their impacts might be negligible provided that metastatic SLNs were removed. This inference was encouraged by the findings from a breast cancer study (IBCSG 23-01) supporting the exemption of axillary lymphadenectomy in patients presenting only MIM/ITC in SLN.[5] If MIM/ITC was not considered, the pooled sensitivity for L-protocol would reach 0.97 (95%CI 0.89–0.99), which is high enough for intraoperative decision-making.

In the E-protocol subgroup there remained moderate heterogeneity in sensitivities, which may due to the remaining methodological difference. Since our aim was to determine the optimal protocol, the heterogeneity in this subgroup was less important. The sectioning intervals were 2–5 mm in L-protocol and the pooled sensitivity further increased when we restricted the criterion (supplementary p18). However, shortened sectioning intervals may increase the pressure upon pathologists and result in loss of tissue for DPE.[18] Yamashita and colleagues examined 3 to 5 sections for each SLN and reported that the diagnosis usually finished within 30 minutes.[30] This may be a rational workload.

The survival data of patients whose PLND were exempted for negative FSE-SLN is still insufficient. Only three observational studies[6,15,49] and one randomized controlled trial (SENTICOL II=NCT01639820)[7] had addressed this issue and the outcomes were generally good. However, the sample sizes of these studies are relatively small. High-quality evidence should be expected from several ongoing multicenter trials (SENTIX=NCT02494063, CSEM010=NCT02642471, SENTICOL III=NCT03386734, supplementary p28) in which patients with FSE-negative SLN are exempted from further PLND,[37] or intraoperatively randomized into arms with or without PLND.[36] The suggestion by this meta-analysis is to adopt L-protocol to reduce the risk of inadequate treatment and ensure the applicability of future findings.

Conclusion

This systematic review and meta-analysis summarized the available data on the review question and identified an optimal protocol for FSE. With short sectioning intervals perpendicular to the long axis of SLN (L-protocol), FSE will yield high sensitivity in detecting metastases, which allows for a precise intraoperative pathological assessment of SLNs. This finding may redefine the role of FSE in SLNB and should be considered in future practice of this technique.

Acknowledgments

We thank Prof Shingo Fujii (Kyoto University, Japan), Prof Aikou Okamoto (School of Medicine, Jikei University, Japan), Prof László Ungár (Duna Medical Center, Budapest, Hungary), Prof Wei-Guo Lu (Women's Hospital, School of Medicine, Zhejiang University, China), and Dr Liang Chen (Shandong Cancer Hospital, Shandong University, China) for their assistances in contacting the authors. We thank Prof Tomohito Tanaka (Osaka Medical College, Japan), Prof Yoshito Terai (Kobe University Graduate School of Medicine, Japan), Prof Robert Póka (University of Debrecen, Hungary), and Prof Han-Mei Lou (Zhejiang Cancer Hospital, Zhejiang University, China) for their works and patience in re-examining the pathological data of their patients. We thank all the respondents in our questionnaire survey no matter whether they provided eligible data. This study was funded by Health and Medical Cooperation Innovation Special Program of Guangzhou Municipal Science and Technology (grant number: 158100075).

Author Contributors

HT and J-HL designed the study and wrote the manuscript. HT and H-FG wrote and registered the study protocol. H-FG and HH developed the search strategy and completed the literature search. HT and J-HL formulated the definitions for pathological protocols, designed the questionnaire survey, and contacted the authors. HT, H-FG, HH and J-HL reviewed the literature search results and extracted data. J-PC and H-YZ did the quality assessment. J-PC, X-KZ, and J-PY did the methodological judgement. H-FG, HH and H-YZ developed the statistical analysis methods. K-JL, HY, and KS provided original data. All authors reviewed the manuscript and provided contributions to the interpretation of the data.

Disclosure of interests

HT and J-HL report being investigators of an ongoing multicenter trial regarding SLNB in cervical cancer (CSEM 010, NCT02642471) funded by a government project (Health and Medical Cooperation Innovation Special Program of Guangzhou Municipal Science and Technology, grant number: 158100075). J-HL receives lecture fees from AstraZeneca and Roche, outside the submitted work. All other authors declare no competing interests.

Details of ethics approval

Not applicable.

References

1. O'Boyle JD, Coleman RL, Bernstein SG, Lifshitz S, Muller CY, Miller DS. Intraoperative lymphatic mapping in cervix cancer patients undergoing radical hysterectomy: A pilot study. *Gynecol Oncol*2000; **79** (2): 238-43.

2. Frumovitz M, Plante M, Lee PS, et al. 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. *The Lancet Oncology* 2018; **19** (10): 1394-403.

3. Leiter U, Stadler R, Mauch C, et al. Complete lymph node dissection versus no dissection in patients with sentinel lymph node biopsy positive melanoma (DeCOG-SLT): a multicentre, randomised, phase 3 trial. *The Lancet Oncology* 2016; **17** (6): 757-67.

4. Rossi EC, Kowalski LD, Scalici J, et al. A comparison of sentinel lymph node biopsy to lymphadenectomy for endometrial cancer staging (FIRES trial): a multicentre, prospective, cohort study. *The Lancet Oncology* 2017; **18** (3): 384-92.

5. Galimberti V, Cole BF, Viale G, et al. Axillary dissection versus no axillary dissection in patients with breast cancer and sentinel-node micrometastases (IBCSG 23-01): 10-year follow-up of a randomised, controlled phase 3 trial. *The Lancet Oncology* 2018;**19** (10): 1385-93.

6. Niikura H, Okamoto S, Otsuki T, et al. Prospective study of sentinel lymph node biopsy without further pelvic lymphadenectomy in patients with sentinel lymph node-negative cervical cancer. *Int J Gynecol Cancer* 2012; **22** (7): 1244-50.

7. Mathevet P, Lecuru F, Magaud L, Bouttitie F. Sentinel lymph node biopsy for early cervical cancer: Results of a randomized prospective, multicenter study (Senticol 2) comparing adding pelvic lymph node dissection vs sentinel node biopsy only. *Gynecol Oncol* 2017;**145** : 2-3.

8. Balaya V, Mathevet P, Magaud L, et al. Predictive factors of unexpected lymphatic drainage pathways in early-stage cervical cancer. *Gynecol Oncol* 2019; **154** (1): 102-9.

9. Sniadecki M, Wydra DG, Wojtylak S, et al. The impact of low volume lymph node metastases and stage migration after pathologic ultrastaging of non-sentinel lymph nodes in early-stage cervical cancer: a study of 54 patients with 4.2 years of follow up. *Ginekol Pol* 2019;**90** (1): 20-30.

10. Guani B, Dorez M, Magaud L, Buenerd A, Lecuru F, Mathevet P. Impact of micrometastasis or isolated tumor cells on recurrence and survival in patients with early cervical cancer: SENTICOL Trial. *Int J Gynecol Cancer* 2019; **29** (3): 447-52.

11. Barranger E, Cortez A, Commo F, et al. Histopathological validation of the sentinel node concept in cervical cancer. *Ann Oncol* 2004;**15** (6): 870-4.

12. Selman TJ, Mann C, Zamora J, Appleyard TL, Khan K. Diagnostic accuracy of tests for lymph node status in primary cervical cancer: a systematic review and meta-analysis. *CMAJ* 2008; **178** (7): 855-62.

13. NCCN-National Comprehensive Cancer Network. NCCN Clinical Practice Guidelines in Oncology: Cervical Cancer. Version 1.2019.

14. Dostalek L, Åvall-Lundqvist E, Creutzberg CL, et al. ESGO survey on current practice in the management of cervical cancer. *Int J Gynecol Cancer* 2018; **28** (6): 1226-31.

15. Yahata H, Kobayashi H, Sonoda K, et al. Prognostic outcome and complications of sentinel lymph node navigation surgery for early-stage cervical cancer. *Int J Clin Oncol* 2018; **23** (6): 1167-72.

16. Vercellino GF, Erdemoglu E, Lichtenberg P, et al. A GCIG international survey: clinical practice patterns of sentinel lymph node biopsies in cervical cancer. *Arch Gynecol Obstet* 2019;**300** (1): 191-9.

17. Tax C, Rovers MM, de Graaf C, Zusterzeel PL, Bekkers RL. The sentinel node procedure in early stage cervical cancer, taking the next step; a diagnostic review. *Gynecol Oncol* 2015; **139** (3): 559-67.

18. Cibula D, McCluggage WG. Sentinel lymph node (SLN) concept in cervical cancer: Current limitations and unanswered questions.*Gynecol Oncol* 2019; **152** (1): 202-7.

19. Bhatla N, Aoki D, Sharma D, Sankaranarayanan R. FIGO Cancer Report 2018. Cancer of the cervix uteri. *Int J Gynecol Obstet* 2018;**143** (143): 22-36.

20. Baker P, Oliva E. A practical approach to intraoperative consultation in gynecological pathology. *Int J Gynecol Pathol*2008; **27** (3): 353-65.

21. Okamoto S, Niikura H, Nakabayashi K, et al. Detection of sentinel lymph node metastases in cervical cancer: assessment of KRT19 mRNA in the one-step nucleic acid amplification (OSNA) method. *Gynecol Oncol* 2013; **130** (3): 530-6.

22. Lee YS, Rhim CC, Lee HN, Lee KH, Park JS, Namkoong SE. HPV status in sentinel nodes might be a prognostic factor in cervical cancer.*Gynecol Oncol* 2007; **105** (2): 351-7.

23. Kohler C, Le X, Dogan NU, et al. Molecular diagnosis for nodal metastasis in endoscopically managed cervical cancer: the accuracy of the APTIMA test to detect high-risk human papillomavirus messenger RNA in sentinel lymph nodes. *J Minim Invasive Gynecol* 2016;**23** (5): 748-52.

24. Marchiolè P, Buenerd A, Scoazec JY, Dargent D, Mathevet P. Sentinel lymph node biopsy is not accurate in predicting lymph node status for patients with cervical carcinoma. *Cancer* 2004; **100** (10): 2154-9.

25. Fader AN, Edwards RP, Cost M, et al. Sentinel lymph node biopsy in early-stage cervical cancer: utility of intraoperative versus postoperative assessment. *Gynecol Oncol* 2008; **111** (1): 13-7.

26. Bats AS, Buenerd A, Querleu D, et al. Diagnostic value of intraoperative examination of sentinel lymph node in early cervical cancer: a prospective, multicenter study. *Gynecol Oncol* 2011;**123** (2): 230-5.

27. Diaz JP, Gemignani ML, Pandit-Taskar N, et al. Sentinel lymph node biopsy in the management of early-stage cervical carcinoma.*Gynecol Oncol* 2011; **120** (3): 347-52.

28. Roy M, Bouchard-Fortier G, Popa I, et al. Value of sentinel node mapping in cancer of the cervix. *Gynecol Oncol* 2011;**122** (2): 269-74.

29. Slama J, Dundr P, Dusek L, Cibula D. High false negative rate of frozen section examination of sentinel lymph nodes in patients with cervical cancer. *Gynecol Oncol* 2013; **129** (2): 384-8.

30. Yamashita T, Katayama H, Kato Y, et al. Management of pelvic lymph nodes by sentinel node navigation surgery in the treatment of invasive cervical cancer. *Int J Gynecol Cancer* 2009; **19** (6): 1113-8.

31. Klapdor R, Mucke J, Schneider M, et al. Value and advantages of preoperative sentinel lymph node imaging with SPECT/CT in cervical cancer. *Int J Gynecol Cancer* 2014; **24** (2): 295-302.

32. Deng XY, Zhang Y, Li DP, et al. Abdominal radical trachelectomy guided by sentinel lymph node biopsy for stage IB1 cervical cancer with tumors >2 cm. *Oncotarget* 2017; **8** (2): 3422-9.

33. Sonoda K, Yahata H, Okugawa K, et al. Value of intraoperative cytological and pathological sentinel lymph node diagnosis in fertility-sparing trachelectomy for early-stage cervical cancer.*Oncology* 2018; **94** (2): 92-8.

34. Papadia A, Morosi C, Wang J, et al. SLN mapping in early-stage cervical cancer as a minimal-invasive triaging tool for multimodal treatment. *Eur J Surg Oncol* 2019; **45** (4): 679-83.

35. Darlin L, Persson J, Bossmar T, et al. The sentinel node concept in early cervical cancer performs well in tumors smaller than 2 cm.*Gynecol Oncol* 2010; **117** (2): 266-9.

36. Lecuru FR, McCormack M, Hillemanns P, et al. SENTICOL III: an international validation study of sentinel node biopsy in early cervical cancer. A GINECO, ENGOT, GCIG and multicenter study. *Int J Gynecol Cancer* 2019; **29** (4): 829-34.

37. Cibula D, Dusek J, Jarkovsky J, et al. A prospective multicenter trial on sentinel lymph node biopsy in patients with early-stage cervical cancer (SENTIX). *Int J Gynecol Cancer* 2019;**29** (1): 212-5.

38. van Diest PJ. Histopathological workup of sentinel lymph nodes: how much is enough? *J Clin Pathol* 1999; **52** : 871–3.

39. Rhim CC, Jong-Sup P, Seog-Nyeon B, Sung-Eun NJJoKMS. Sentinel node biopsy as an indicator for pelvic nodes dissection in early stage cervical cancer. *J Korean Med Sci* 2002; **17** (4): 507-11.

40. Lambaudie E, Collinet P, Narducci F, et al. Laparoscopic identification of sentinel lymph nodes in early stage cervical cancer.*Gynecol Oncol* 2003; **89** (1): 84-7.

41. Sheng XG, Li DP, Liu NF, et al. Clinical significance of sentinel lymph nodes detection in patients with early stage cervical cancer.*Chin J Obstet Gynecol* 2004; **36** (1): 10-3.

42. Pijpers R, Buist MR, van Lingen A, et al. The sentinel node in cervical cancer: scintigraphy and laparoscopic gamma probe-guided biopsy. *Eur J Nucl Med Mol Imaging* 2004; **31** (11): 1479-86.

43. Schwendinger V, Müller-Holzner E, Zeimet AG, Marth C. Sentinel node detection with the blue dye technique in early cervical cancer.*Eur J Gynaecol Oncol* 2006; **27** (4): 359-62.

44. Altgassen C, Paseka A, Urbanczyk H, et al. Dilution of dye improves parametrial SLN detection in patients with cervical cancer.*Gynecol Oncol* 2007; **105** (2): 329-34.

45. Kushner DM, Connor JP, Wilson MA, et al. Laparoscopic sentinel lymph node mapping for cervix cancer–a detailed evaluation and time analysis. *Gynecol Oncol* 2007; **106** (3): 507-12.

46. Lou HM, Lv WG, Xie X, et al. Sentinel lymph node detection and intraoperational diagnosis in patients with early stage cervical cancer.*Natl Med J Chin* 2007; **87** (47): 3353-6.

47. Seong SJ, Park H, Yang KM, et al. Detection of sentinel lymph nodes in patients with early stage cervical cancer. *J Korean Med Sci*2007; **22** (1): 105-9.

48. Pluta M, Rob L, Charvat M, et al. Less radical surgery than radical hysterectomy in early stage cervical cancer: a pilot study.*Gynecol Oncol* 2009; **113** (2): 181-4.

49. Gortzak-Uzan L, Jimenez W, Nofech-Mozes S, et al. Sentinel lymph node biopsy vs. pelvic lymphadenectomy in early stage cervical cancer: is it time to change the gold standard? *Gynecol Oncol* 2010;**116** (1): 28-32.

50. Du XL, Sheng XG, Jiang T, et al. Sentinel lymph node biopsy as guidance for radical trachelectomy in young patients with early stage cervical cancer. *BMC Cancer* 2011; **11** : 157.

51. Póka R, Molnar S, Daragó P, et al. Intention-to-treat analysis of radical trachelectomy for early-stage cervical cancer with special reference to oncologic failures: single-institutional experience in Hungary. *Int J Gynecol Cancer* 2017; **27** (7): 1438-45.

52. Tanaka T, Terai Y, Ashihara K, et al. The detection of sentinel lymph nodes in laparoscopic surgery for uterine cervical cancer using 99m-technetium-tin colloid, indocyanine green, and blue dye. *J Gynecol Oncol* 2017; **28** (2): e13.

53. García JC, de la Riva Pérez PA, Jiménez IR, et al. Selective biopsy of the sentinel node in cancer of cervix: Experience in validation phase. *Rev Esp Med Nucl Imagen Mol* 2018; **37** (6): 359-65.

54. Poling JS, Tsangaris TN, Argani P, Cimino-Mathews A. Frozen section evaluation of breast carcinoma sentinel lymph nodes: a retrospective review of 1,940 cases. *Breast Cancer Res Treat* 2014;**148** (2): 355-61.

55. Park JY, Kook MC, Eom BW, et al. Practical intraoperative pathologic evaluation of sentinel lymph nodes during sentinel node navigation surgery in gastric cancer patients - Proposal of the pathologic protocol for the upcoming SENORITA trial. *Surg Oncol* 2016; **25** (3): 139-46.

56. Donker M, van Tienhoven G, Straver ME, et al. Radiotherapy or surgery of the axilla after a positive sentinel node in breast cancer (EORTC 10981-22023 AMAROS): a randomised, multicentre, open-label, phase 3 non-inferiority trial. *The Lancet Oncology* 2014;**15** (12): 1303-10.

57. Bishop JA, Sun J, Ajkay N, Sanders MA. Decline in frozen section diagnosis for axillary sentinel lymph nodes as a result of the American College of Surgeons Oncology Group Z0011 Trial. *Arch Pathol Lab Med* 2016; **140** (8): 830-5.

58. Abu-Rustum NR, Neubauer N, Sonoda Y, et al. Surgical and pathologic outcomes of fertility-sparing radical abdominal trachelectomy for FIGO stage IB1 cervical cancer. *Gynecol Oncol* 2008; **111** (2): 261-4.

59. Cibula D, Abu-Rustum NR, Dusek L, et al. Prognostic significance of low volume sentinel lymph node disease in early-stage cervical cancer. *Gynecol Oncol* 2012; **124** (3): 496-501.

60. Fregnani J, Latorre M, Novik P, Lopes A, Soares F. Assessment of pelvic lymph node micrometastatic disease in stages IB and IIA of carcinoma of the uterine cervix. *Int J Gynecol Cancer* 2006;**16** : 1188-94.

61. Okamoto S, Niikura H, Yoshinaga K, et al. Detection of micrometastases in cervical cancer with a system that evaluates both sentinel and nonsentinel lymph nodes. *Int J Gynecol Cancer* 2009;**19** (4): 708-11.

Legends for figures

Figure 1: Study selection *gathered by questionnaire survey. FSE=frozen section examination. SLNB=sentinel lymph node biopsy. DPE=definitive pathological examination.

Figure 2: Two representative protocols for frozen section examination

Figure 3: Forest plots of sensitivity for all studies

Figure 4: Forest plots of sensitivity for subgroups (A) latitudinal; (B) equatorial

Hosted file

`META-TAB1.docx` available at https://authorea.com/users/311903/articles/442548-sectioning-protocol-determines-accuracy-of-intraoperative-pathological-examination-of-sentinel-lymph-node-in-cervical-cancer-a-systematic-review-and-meta-analysis

Hosted file

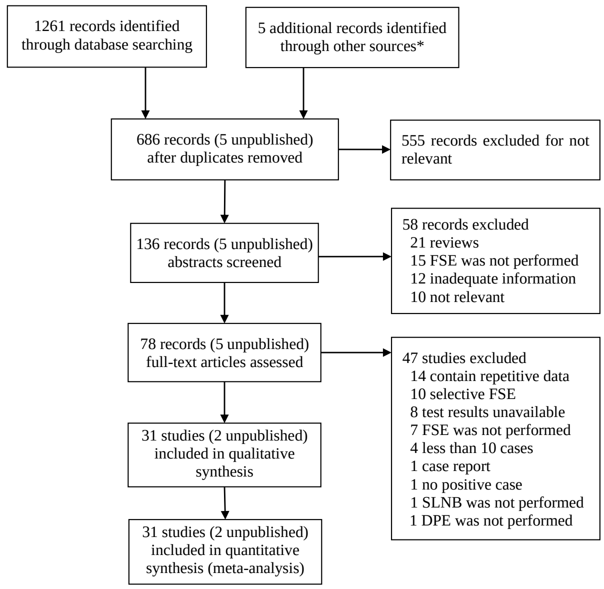
`META-TAB2.docx` available at https://authorea.com/users/311903/articles/442548-sectioning-protocol-determines-accuracy-of-intraoperative-pathological-examination-of-sentinel-lymph-node-in-cervical-cancer-a-systematic-review-and-meta-analysis

Hosted file

`META-TAB3.docx` available at https://authorea.com/users/311903/articles/442548-sectioning-protocol-determines-accuracy-of-intraoperative-pathological-examination-of-sentinel-lymph-node-in-cervical-cancer-a-systematic-review-and-meta-analysis

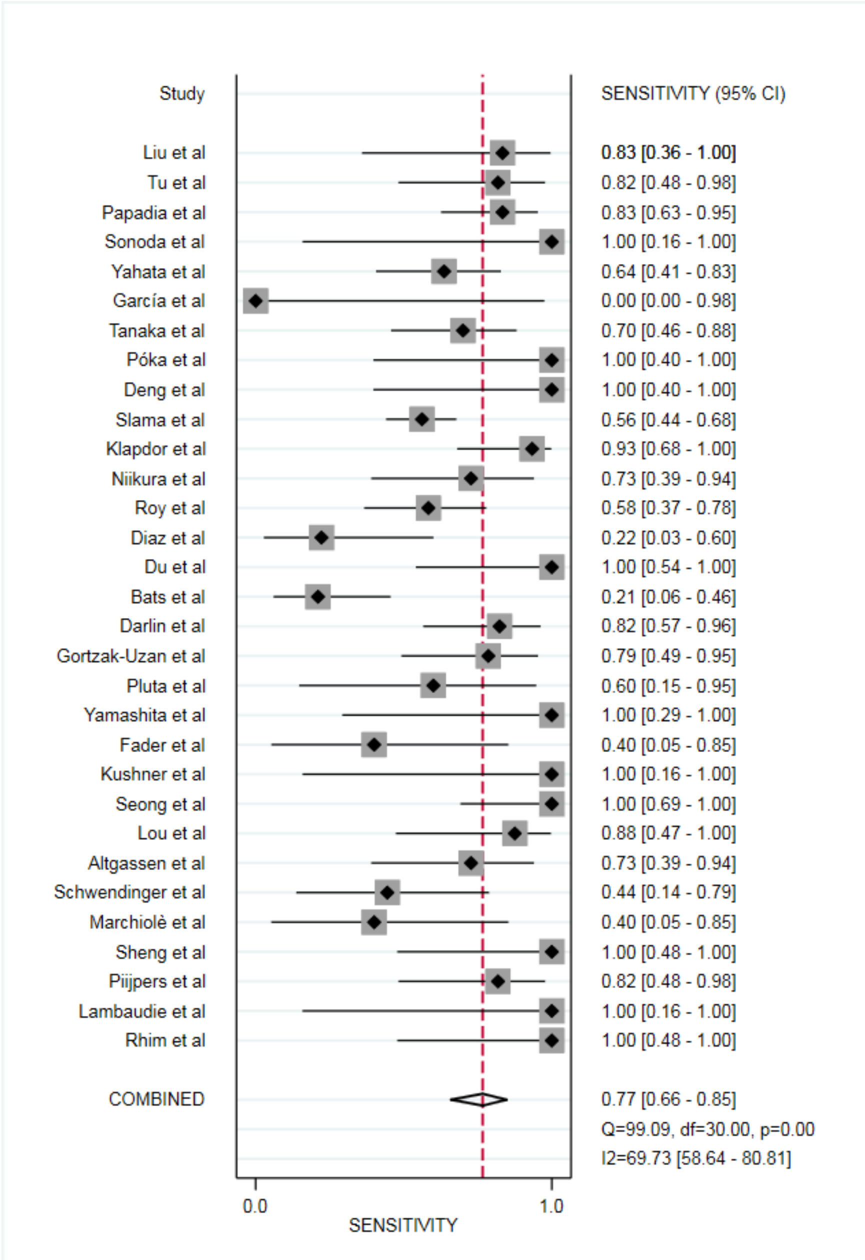
Hosted file

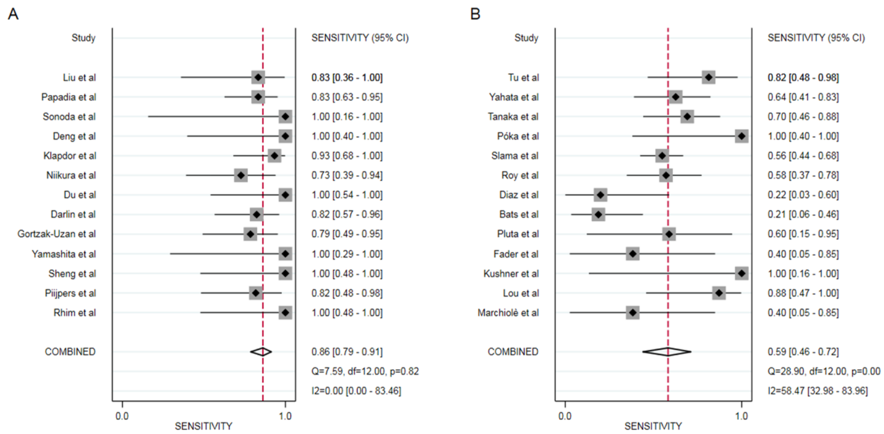
`META-TAB4.docx` available at https://authorea.com/users/311903/articles/442548-sectioning-protocol-determines-accuracy-of-intraoperative-pathological-examination-of-sentinel-lymph-node-in-cervical-cancer-a-systematic-review-and-meta-analysis



A. Equatorial protocol: SLN was bisected along the equator. One or more adjacent sections were taken from the maximum surface.

B. Latitudinal protocol: SLN was cut into pieces at short intervals, perpendicularly to the long axis. One or more adjacent sections were taken from each cut surface.


Study
SENSITIVITY (95% CI)
Liu et al 0.83 [0.36 - 1.00]
Tu et al 0.82 [0.48 - 0.98]
Papadia et al 0.83 [0.63 - 0.95]
Sonoda et al 1.00 [0.16 - 1.00]
Yahata et al 0.64 [0.41 - 0.83]
García et al 0.00 [0.00 - 0.98]
Tanaka et al 0.70 [0.46 - 0.88]
Póka et al 1.00 [0.40 - 1.00]
Deng et al 1.00 [0.40 - 1.00]
Slama et al 0.56 [0.44 - 0.68]
Klapdor et al 0.93 [0.68 - 1.00]
Niikura et al 0.73 [0.39 - 0.94]
Roy et al 0.58 [0.37 - 0.78]
Diaz et al 0.22 [0.03 - 0.60]
Du et al 1.00 [0.54 - 1.00]
Bats et al 0.21 [0.06 - 0.46]
Darlin et al 0.82 [0.57 - 0.96]
Gortzak-Uzan et al 0.79 [0.49 - 0.95]
Pluta et al 0.60 [0.15 - 0.95]
Yamashita et al 1.00 [0.29 - 1.00]
Fader et al 0.40 [0.05 - 0.85]
Kushner et al 1.00 [0.16 - 1.00]
Seong et al 1.00 [0.69 - 1.00]
Lou et al 0.88 [0.47 - 1.00]
Altgassen et al 0.73 [0.39 - 0.94]
Schwendinger et al 0.44 [0.14 - 0.79]
Marchiolè et al 0.40 [0.05 - 0.85]
Sheng et al 1.00 [0.48 - 1.00]
Piijpers et al 0.82 [0.48 - 0.98]
Lambaudie et al 1.00 [0.16 - 1.00]
Rhim et al 1.00 [0.48 - 1.00]
COMBINED 0.77 [0.66 - 0.85]
Q=99.09, df=30.00, p=0.00
I2=69.73 [58.64 - 80.81]
0.0
1.0
SENSITIVITY


A
Study
SENSITIVITY (95% CI)
Liu et al 0.83 [0.36 - 1.00]
Papadia et al 0.83 [0.63 - 0.95]
Sonoda et al 1.00 [0.16 - 1.00]
Deng et al 1.00 [0.40 - 1.00]
Klapdor et al 0.93 [0.68 - 1.00]
Niikura et al 0.73 [0.39 - 0.94]
Du et al 1.00 [0.54 - 1.00]
Darlin et al 0.82 [0.57 - 0.96]
Gortzak-Uzan et al 0.79 [0.49 - 0.95]
Yamashita et al 1.00 [0.29 - 1.00]
Sheng et al 1.00 [0.48 - 1.00]
Piijpers et al 0.82 [0.48 - 0.98]
Rhim et al 1.00 [0.48 - 1.00]
COMBINED 0.86 [0.79 - 0.91]
Q=7.59, df=12.00, p=0.82
I2=0.00 [0.00 - 83.46]
0.0
1.0
SENSITIVITY
B
Study
SENSITIVITY (95% CI)
Tu et al 0.82 [0.48 - 0.98]
Yahata et al 0.64 [0.41 - 0.83]
Tanaka et al 0.70 [0.46 - 0.88]
Póka et al 1.00 [0.40 - 1.00]
Slama et al 0.56 [0.44 - 0.68]
Roy et al 0.58 [0.37 - 0.78]
Diaz et al 0.22 [0.03 - 0.60]
Bats et al 0.21 [0.06 - 0.46]
Pluta et al 0.60 [0.15 - 0.95]
Fader et al 0.40 [0.05 - 0.85]
Kushner et al 1.00 [0.16 - 1.00]
Lou et al 0.88 [0.47 - 1.00]
Marchiolè et al 0.40 [0.05 - 0.85]
COMBINED 0.59 [0.46 - 0.72]
Q=28.90, df=12.00, p=0.00
I2=58.47 [32.98 - 83.96]
0.0
1.0
SENSITIVITY