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Clinical Study of Double Contrast-Enhanced Ultrasound Combined with Dye Method and Marker Placement to Identify and Locate Sentinel Lymph Nodes in Patients with Breast Cancer

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Abstract

Objective: To explore the value of percutaneous ultrasonography combined with transvenous ultrasonography for accurate localization of sentinel lymph nodes and diagnosis of metastatic lymph nodes in patients with breast cancer. **Methods:** 18 cases of patients with breast cancer attending the Hainan General Hospital from May 2022 to June 2024 who were proposed to undergo axillary lymph node dissection were selected, and the ultrasonographic agent was injected subcutaneously through the areola on the 1st day before the operation, and the marker localization of the manifestation of the Sentinel lymph nodes and draw the lymphatic vessel alignment for drainage on the body surface, and record the manifestation of SLN by conventional ultrasound and dual ultrasonography. At the time of surgery, intraoperative melphalan localization was used to identify the SLN, the difference between the number of ultrasound and melphalan localization was observed, and resection was performed for pathological examination to determine whether they were metastatic or not. **Results:** There were 8 metastatic lymph nodes and 18 non-metastatic lymph nodes among 31 SLN. A total of 62 SLN were localized by intraoperative melphalan, of which 31 were consistent with ultrasound localization and 31 were not identified by ultrasound. The diagnostic sensitivity of SLN metastasis diagnosed by transcutaneous ultrasonography was 62.50%, specificity was 91.30%, positive predictive value was 71.43%, negative predictive value 87.50%, accuracy was 83.87%, and the AUC was 0.769; the diagnostic sensitivity

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of transvenous ultrasonography diagnosed was 75.00%, specificity was 75.00%, and the accuracy was 83.87%, 75.00%, specificity 91.30%, positive predictive value 75.00%, negative predictive value 91.30%, accuracy 87.10%, AUC 0.832; dual ultrasonography diagnostic sensitivity 87.50%, specificity 91.30%, positive predictive value 77.78%, negative predictive value 95.45%, accuracy 90.32%. The AUC was 0.894. **Conclusion:** Transcutaneous ultrasonography combined with transvenous ultrasonography can accurately localize sentinel lymph nodes and improve the sensitivity and accuracy of the diagnosis of metastatic SLN.

Keywords

Sentinel Lymph Nodes, Breast Cancer, Ultrasound, Ultrasonography, Axillary Lymph Nodes

1. Introduction

Breast cancer is currently the most common malignant tumor in Chinese women, and localized metastasis in axillary lymph nodes is an important prognostic factor for overall survival in breast cancer. Sentinel lymph node biopsy (SLNB) results suggest whether the axillary lymph nodes are metastatic or not, which directly affects the clinical staging of breast cancer patients, and if metastatic SLN exists, patients need further chemotherapy or radiotherapy, and some patients may need to undergo additional axillary lymph node dissection [1]. Therefore, if the sentinel lymph nodes can be accurately localized and diagnosed with metastasis before surgery, the decision of whether to perform axillary lymph node dissection can be directly decided to avoid the damage of secondary surgery. Currently, nuclide or intraoperative melphalan are commonly used to locate the sentinel lymph nodes, both of which can locate the sentinel lymph nodes well, but neither of them can meet the requirement of preoperative diagnosis of metastatic sentinel lymph nodes well, and nuclide is radioactive, which is also harmful to patients and healthcare personnel. Percutaneous sentinel lymph node ultrasonography is a relatively new method to locate sentinel lymph nodes, and its enhancement mode can assist in diagnosing the nature of lymph nodes, and transvenous ultrasonography also has advantages in determining the nature of lymph nodes, and the combination of the two can accurately locate and diagnose metastatic sentinel lymph nodes [2], and the aim of this study is to explore the value of transvenous ultrasonography for the localization and diagnosis of metastatic sentinel lymph nodes for patients with breast cancer, in order to provide more optimized and precise treatment options for breast cancer patients.

2. Information and Methods

2.1. Objects of Study

Eighteen patients with breast cancer who attended the Hainan General Hospital from May 2022 to June 2024 for axillary lymph node dissection were selected.

Inclusion criteria: 1) patients with preoperative puncture biopsy confirmed breast cancer; 2) patients who did not receive any adjuvant treatments such as radiotherapy and chemotherapy; 3) patients with no previous history of breast mass cutting surgery; 4) patients with normal lymphatic drainage pathways of the affected side were not disrupted. Exclusion criteria: 1) pregnancy or breast-feeding; 2) patients without sentinel lymph node biopsy.

2.2. Research Methodology

Siemens Acuson Sequoia diagnostic ultrasound machine with 10L4 line array probe at low frequency was used. The study patients were subjected to ultrasonography, informed and consented to the application of the contrast agent prior to the examination and signed the informed consent for ultrasonography and informed consent for the use of the ultrasound instructions. SonoVue (SonoVue manufactured in Italy) lyophilized powder was prepared into a microbubble suspension of sulfur hexafluoride with 5 ml of 0 .9% saline according to the instructions, and was prepared by oscillating for 30 s.

Ultrasound scanning: the patient adopts the supine position, the arms of both sides are lifted up and abducted as much as possible, fully exposing the breast and axilla, the probe is swept radially with the nipple as the center, and after scanning the lesion, the lesion is observed from multiple views; in the areola area , the subcutaneous (about 2 mm) injections of the contrast agent SonoVuo 0.5 ml in the direction of the 3, 6, 9, and 12 o'clock positions are performed in order of total amount of 2 ml, and after the injection the local massage is carried out for 10 s, and the injection of the contrast agent is started at the same time. At the same time when injecting the contrast agent, turn on the imaging system and automatic timer in the instrument; real-time dynamic scanning from the injection site of the breast mass, tracking to the first lymph node enhanced in the axilla along the lymphatic vessels of enhancement, recording the time of the start of the enhancement of the contrast agent in the lymph node, the time of the enhancement reaching the peak, the time of the complete fading, the site of the lymph node, the number of the lymph nodes, and the characteristics of the contrast agent enhancement, tracking to the first or the first group of enhanced lymph vessels along the enhanced lymph vessels emanating from the injection site after injecting the ultrasound contrast agent. The first lymph node or the first group of lymph nodes with enhancement was identified as SLN, and the SLN and lymphatic drainage route were marked on the body surface with a marker pen, and the distance between the skin surface and the SLN was measured; then the SLN was imaged by peripheral venous ultrasonography, and the contrast agent perfusion pattern, enhancement characteristics, and the time of the beginning of enhancement, the time of the peak of enhancement, and the time of the complete regression were recorded and observed; and then the affected side of the upper limb adopted the position of the body at the time of surgery. The affected upper limb is then positioned in the surgical position. The SLN was positioned within the cortex of the SLN shown in the contrast under

ultrasound guidance, and a marker(or positioning hook silk) was placed under ultrasound guidance to locate the identified SLN (Figure 1 and Figure 2).

Methylene blue or melphalan was injected at least 5 min before surgery at four points inside and outside and above and below the skin in the areola area. After anesthesia took effect, surgery was performed. The blue-stained lymph nodes were found in the axilla, and after the SLN was cut, the rest of the axilla was probed again to make sure that there was no residue, and all the SLNs were cleared and sent to the Department of Pathology for frozen pathological section examination, and at the same time, the breast cancer was subjected to total mastectomy or breast-conserving surgery.

2.3. Type of Percutaneous Sentinel Lymph Node Ultrasonography

Transcutaneous subcutaneous ultrasonography classifies SLN into the following five types: type I, homogeneous hyperenhancement; type II, circumferential hyperenhancement; type III, diffuse inhomogeneous enhancement; type IV, localized perfusion deficit; and type V, no contrast perfusion (Figure 3).

2.4. Type of Transvenous Sentinel Lymph Node Ultrasonography

Transvenous ultrasonography: 2.0 ml of contrast microbubble suspension was injected through the elbow vein after complete contouring of percutaneous ultrasonographic contrast agent for ultrasonography of the SLN. The perfusion pattern was categorized as direction of perfusion: centripetal enhancement, centrifugal enhancement, diffuse enhancement and enhancement pattern: homogeneous, inhomogeneous (Figure 4).

2.5. Statistical Analysis

Statistical analysis using SPSS25.0, the measurement information was expressed as the mean earth standard deviation ($\overline{x} \pm s$), and the comparison between groups was made by the independent samples t-test, and the count information was expressed by frequency and rate, when $P < 0.05$ indicated statistical significance.

3. Findings

3.1. Analysis of General Information

In 18 patients, 18 breast masses were removed, including 12 cases of invasive carcinoma, 2 cases of intraductal papillary carcinoma, 2 cases of mucinous carcinoma, 1 case of intraductal carcinoma, and 1 case of invasive carcinoma with intraductal papillary carcinoma. After the results of postoperative lymph node pathology, there were 8 cases in the sentinel lymph node metastasis group and 23 cases in the non-metastasis group among the ultrasonographically localized SLN. There were 31 sentinel lymph nodes labeled by dual ultrasonography detection and 62 sentinel lymph nodes identified by intraoperative injection of methylene blue or melphalan (Table 1).

3.2. Double Contrast-Enhanced Ultrasound Characterization Statistics

Sentinel lymph node dual contrast characterization Of the 31 SLNs, percutaneous ultrasonography showed 13 type I SLNs, 11 type II SLNs, 3 type III SLNs, and 4 type IV SLNs. On transvenous ultrasonography, the arterial phase showed centripetal enhancement in 4, centrifugal enhancement in 14, diffuse enhancement in 13, homogeneous enhancement in 25, and inhomogeneous enhancement in 6.

3.3. Diagnostic Efficacy of Double Contrast-Enhanced Ultrasound

The diagnostic sensitivity of percutaneous ultrasonography was 62.50%, specificity 91.30%, positive predictive value 71.43%, negative predictive value 87.50%, accuracy 83.87%, AUC 0.769; transvenous ultrasonography was 75.00%, specificity 91.30%, positive predictive value 75.00%, negative predictive value 87.50%, AUC 0.769; transvenous ultrasonography was 75.00%, specificity 91.30%, positive predictive value 75.00%, negative predictive value 83.87%, AUC 0.769. The diagnostic sensitivity was 75.00%, specificity 91.30%, positive predictive value 75.00%, negative predictive value 91.30%, accuracy 87.10%, AUC 0.832. Dual ultrasound was positive when either one of the two was positive, and the diagnosis of metastatic lymph nodes was positive, with a sensitivity of 87.50%, specificity 87.50%, and accuracy of 0.769; transvenous ultrasonography was positive when either one of the two was positive. The sensitivity was 87.50%, specificity was 91.30%, positive predictive value was 77.78%, negative predictive value was 95.45%, accuracy was 90.32%, and AUC was 0.894 (Table 2, Figure 5).

Table 1. General information about the study population.

Characteristic	Quantities
Patients (n.)	18
Age (years)	45.89 ± 11.37
Outer Upper Quadrant (n.)	
Outer Upper Quadrant (n.)	8
Outer Upper Quadrant (n)	5
Outer Upper Quadrant (n.)	5
Sentinellymph nodes localization	
Dual ultrasonography localization (n.)	31
Intraoperative dye localization (n.)	62
Pathological type	
Invasive carcino (n.)	12
Intraductal papillary carcinoma (n.)	2
Mucinous carcinoma (n.)	2
Intraductal carcinoma (n.)	1
Invasive carcinoma with intraductal papillary carcinoma (n.)	1

Continued

Number of sentinel lymph nodes	
Transfers (n.)	8
Non-transferred (n.)	23

Table 2. Dual imaging characteristics of the sentinel lymph nodes.

Groups	Percutaneous sonography		Transvenous ultrasonography		Dual ultrasonography	
	Transfers	Non-transfer	Transfers	Non-transfer	Transfers	Non-transfer
Transfers (n = 8)	5	3	6	2	7	1
Non-transfer (n = 23)	2	21	2	21	2	21
Sensitivity (%)	62.50		75.00		87.50	
Specificity (%)	91.30		91.30		91.30	
Positive predictive value (%)	71.43		75.00		77.78	
Negative predictive value (%)	87.50		91.30		95.45	
Accuracy (%)	83.87		87.10		90.32	
AUC	0.769		0.832		0.894	
P-value	0.025		0.006		0.001	

$P < 0.05$ indicates statistical significance.

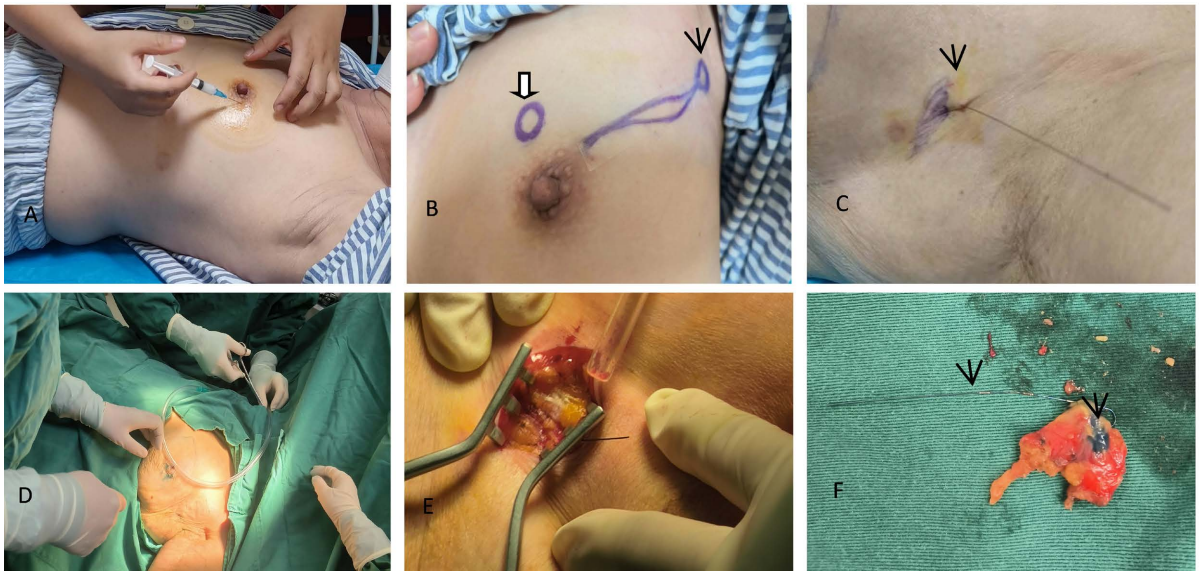

Figure 1. Sentinel lymph node ultrasonography and intraoperative resection to remove the sentinel lymph node. (A) Percutaneous injection of ultrasound contrast agent; (B) Sentinel lymph nodes were found along the lymphatic ducts were visualized and projected on the body surface at the location of the marked lymphatic ducts and lymph nodes (large arrows are breast lumps, small arrows are sentinel lymph nodes); (C) Ultrasound-guided placement of positioning hook silk in the sentinel lymph node; (D) Patient was injected with intraoperative melphalan to locate the sentinel lymph node; (E) Intraoperative finding of the sentinel lymph node marked with positioning hook silk; (F) Removal of the marked sentinel lymph node, with the positioning hook silk and blue color in the lymph node visible.

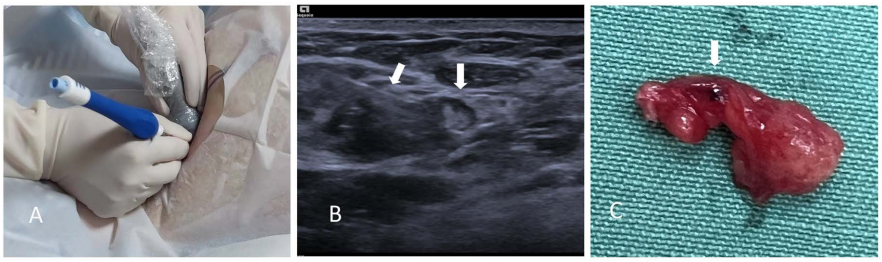

Figure 2. (A) Ultrasound-guided puncture, placed marker in the sentinel lymph node with suspected metastasis. (B) Needbes and sentinel lymph nodes seen on ultrasound (arrow); (C) Surgical removal of sentinel lymph node with internal marker (arrow).

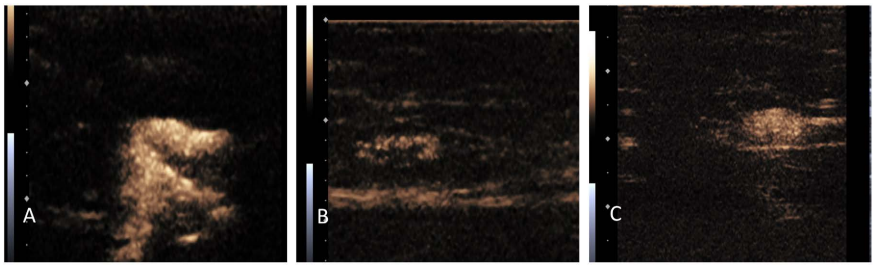

Figure 3. Transcutaneous injection of ultrasound contrast agent to locate anterior sentinel lymph nodes along the lymphatic vessels. (A) A central filling defect is seen (pathology with metastasis); (B) Homogeneous circumferential hyperenhancement (pathology without metastasis); (C) Homogeneous hyperenhancement is seen pathology without metastasis).

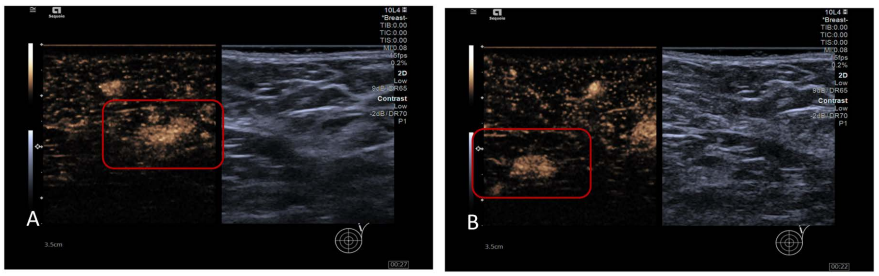

Figure 4. Transvenous ultrasonography. (A) shows inhomogeneous hyperenhancement (pathology with metastasis) and cortical filling defects; (B) homogeneous yperenhancement (pathology without metastasis).

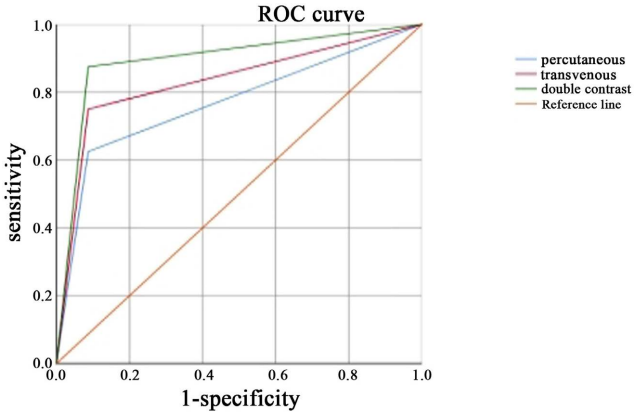

Figure 5. The ROC curve.

4. Discussion

The incidence of breast cancer is the highest among female malignant tumors in the world. According to the recent estimation of the International Agency for Research on Cancer (IARC) of the World Health Organization (WHO), there are 1.38 million new cases of breast cancer in the world each year, accounting for 23% of the incidence of all female malignant tumors, and 410,000 deaths, accounting for 14% of the deaths of all female malignant tumors. Lymph node metastasis of breast cancer directly affects the staging of breast cancer, the choice of treatment options and the prognosis of patients. Therefore, studying the mechanism of breast cancer lymph node metastasis and obtaining information about lymph node metastasis are key aspects of early prevention and treatment of breast cancer [3]. Traditional breast cancer surgery involves total excision of the patient's breast and sweeping of the axillary lymph nodes, and patients are prone to postoperative complications such as upper extremity edema: shoulder joint stiffness, pain, and numbness. Sentinel lymphatic node (SLN) is the first station in the lymphatic drainage of the tumor, which can be the first lymph node or the first group of lymph nodes, and the clinical application of breast-conserving mastectomy + sentinel lymph node biopsy has been a landmark advancement in breast surgery in recent years [4]. Among the currently used clinical methods to locate the sentinel lymph nodes, the radionuclide imaging method has some radiation, the dye method requires surgical dissection to search for colored lymphatic vessels and lymph nodes after injection, which is traumatic and increases the operation time, it is easy to dye the adjacent adipose tissue, and the dispersion is too fast and may flow to the secondary lymph nodes and result in false positives, and it is prone to anaphylactic reactions [5].

Ultrasonography localization and detection of SLN has the unique advantage of 1) no radioactive damage; 2) marking the drainage lymphatic channel and the location of SLN on the body surface, which is convenient for clinicians to make a reasonable design of surgical incision and less traumatic; 3) showing the possibility of metastatic foci in the lymph nodes, and providing new differential diagnostic imaging features for the diagnosis of metastatic and reactive hyperplastic lymph nodes [6].

In this study, SLNs were identified by dual ultrasonography, the direction of lymphatic drainage and the location of SLNs were marked on the body surface, the lymph node enhancement pattern was observed at the same time, and ultrasound-guided puncture biopsy was performed to characterize the suspected metastasis of SLNs, and then a marker was placed to pinpoint SLNs under ultrasonography. The results showed that 31 sentinel lymph nodes were localized by dual ultrasonography and were bluish-stained in intraoperative operation, which is consistent with that of the ones identified by the US blue, but the number of those identified by US blue in intraoperative operation was 62, which was much different than that identified by the ultrasonography. However, there were 62 sentinel lymph nodes identified by Meilan intraoperatively, which differed

greatly from the number identified by ultrasound, and in the final pathology, those not identified by ultrasound were all non-metastatic, and the reason for this was analyzed that these bluish-stained lymph nodes were not true sentinel lymph nodes, but rather false-positive sentinel lymph nodes that appeared; there was one sentinel lymph node in the study, one of the three in the axilla of a patient, which was small and non-metastatic/no change in cortical echogenicity. One sentinel lymph node in the study, which was one of three in the axilla of a patient, was small in size and had no metastasis/altered cortical echogenicity and was in close proximity to another sentinel lymph node, which made ultrasound identification confusing, and therefore was not recognized by ultrasound, an error that needs to be addressed in order to improve the recognition rate. It was found that all the metastatic sentinel lymph nodes could be recognized by ultrasound, and those not recognized by ultrasound were not metastatic; therefore, dual ultrasound recognition of sentinel lymph nodes is clinically trustworthy.

In this study, dual ultrasonography not only efficiently and accurately identifies and localizes sentinel lymph nodes, but also has high efficacy in diagnosing metastatic sentinel lymph nodes.

Dual ultrasonography was more diagnostic than percutaneous ultrasonography and transvenous ultrasonography alone for metastatic anterior sentinel lymph nodes, and the specificity and sensitivity were higher than that of a single imaging modality. Therefore, dual ultrasonography can identify the sentinel lymph nodes and provide information about metastasis before surgery, which can provide effective and reliable information for precise and individualized treatment of patients.

The shortcoming of this study is that the number of cases is relatively small, so in the future, we will continue to expand the sample size and accurately assess the diagnostic value of dual ultrasonography on metastatic anterior lymph nodes so that it can be applied in the clinic early to bring benefits to patients.

5. Conclusion

This study shows that dual ultrasonography can precisely localize sentinel lymph nodes and improve the sensitivity and accuracy of metastatic SLN diagnosis. Ultrasonography can accurately identify and localize SLN in breast cancer patients, and determine whether SLN is metastatic before surgery, which further helps clinicians to determine the clinical staging to provide new guidance. Dual ultrasonography can dynamically observe the lymphatic drainage situation of breast cancer in real time, and observe the enhancement characteristics to determine the suspected metastatic site, and timely perform ultrasound-guided puncture biopsy on it, which is highly accurate and does not have radiation, which is more convenient and worth promoting in the clinic. It has high accuracy and is not radioactive and more convenient, which is worthy of clinical popularization.

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Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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