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SENTINEL LYMPH NODE DETECTION IN BREAST CANCER – FIRST EXPERIENCE

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Abstract

Introduction: Breast cancer accouns for 22.9% of all cancers in women and 13.7% of cancer deaths. Positive axillary lymphnodes (ALN) predict the development of distant metastases. The status of the sentinel lymphnode (SLN) is crutial for the treatment selection.

Aim: To determine the benefits of SLN detection in patients with breast cancer.

Material and methodology: 38 female patients (pts), age 44 ± 12 years, with T1-2 N0 M0 breast cancer, without enlarged ALN on ultrasound (US), were included. SLN detection was performed using gamma camera and gamma detection probe after periareolar subcutaneous and/or peritumoral injection of (99m-Technetium-SENTISCINT). Blue dye was administered 20 min before the operation. SLN was extirpated and ex tempore histopathology was performed.

Results: Ex tempore SLN evaluation was negative and the lymphatic pathways preserved in 28/38 (74%) pts. In 10/38 (26%) pts SLN was positive, followed by radical surgery. In 3/28 ex tempore negative patients, histopathological analysis showed metastatic involvement (false negative). In 3/10 ex tempore positive patients micro metastases 0,2–2 mm were detected. 12 pts had 2 SLN, 8/12 (66%) had negative and 4/12 (34%) had positive SLN. 3 pts had a rare double drainage to axilla and a. mammaria int.

Conclusion: Our results confirm that SLN detection technique is non-invasive, safe and reliable and should be incorporated into the guidelines for breast cancer pts (T1-2 N0 M0). The most reliable option for colloid application is the combined technique of periareolar and peritumoral injection. Patients with drainage to a. mammaria interna should be selected for adjuvant protocols.

Key words: breast cancer, sentinel lymph node, blue dye.

Introduction

Breast cancer accounts for 22.9% of all cancers in women and 13.7% of cancer deaths. Auxiliary lymph node (ALN) status is the most important prognostic factor in breast cancer patients. Sentinel lymph nodes (SLN) are the first nodes of the lymphatic drainage pathway in which cancerous cells could spread after they leave the primary tumor. SLN detection and intraoperative ex tempore pathohistological evaluation with high sensitivity and specificity provides valid information about the tumors lymphatic spread and enables mapping of the lymphatic drainage [1, 2]. It also determines the type and extensity of the surgical intervention – selective or radical mastectomy/lymphadenectomy. If SLN are free of metastases, selective lymphadenectomy (only the SLN) is performed, but if metastases are detected, tumor up staging (from N0 to N1) is obligatory, and radical lymphadenectomy is performed. The literature data indicate that this technique could

be considered as a successful alternative to the conventional radical axillary lymph node dissection [1, 2].

Lately, the status of intramammary lymph nodes increases its predictive role in breast carcinoma grading and modification of the postoperative therapeutic protocols. [3] If positive, they are always associated with worse disease outcome and therapeutic protocol redefinition-contribute to the selection of patients for adjuvant ontological protocols [4, 5].

Material and methods

38 female patients (pts), age 44 ± 12 years, (17 pts from the University Clinic for Thoracic vascular surgery, Skopje in the period 2012–2014, and 21 pts from the University Clinic Rebro, Zagreb in the year 2013) were included in the study. All pts were T1-2 N0 M0 staged breast cancer (confirmed with mammography and core biopsy), without enlarged ALN, neither clinically nor on ultrasound (US). Exclusion criteria were: pregnancy, lactation, previous breast or axillary surgery at the same site, multifocal cancer, T3-4, N1 stage and M1 stage tumors. After SLN detection and extirpation, ex tempore histopathology was performed, which determined the extent of the surgical intervention. The postoperative follow up of the patients included regular ontological checkups.

SLN detection included preoperative subcutaneous/peritumoral application (16 h prior tothe surgical procedure) of 4mCi (150MBq) ^{99m}Tc-radiolabeled colloids (SENTISCINT) subdivided in 4 separate doses (each dose of 1mCi/37MBq respectively) and injected into 4 separate periareolar and/or peritumoral locations [1]. Considering the fact that radiotracers were used, we followed the ALARA principles (best diagnostic presentation with the minimum radiation burden to the patient). The patients were familiarized with the procedure and written informed consent was obtained.

SENTISCINT is a MEDI-RADIOPHARMA LTD Hungary commercial kit, composed of human serum albumin nano-sized colloid particles with diameter of 100–600 nm in a form of sterile lyophilized powder.

Quality control was performed with ascendant paper chromatography. The injected volume at single site did not exceed 0.2–0.3 ml. In case of intravascular application, the SLN detection procedure was considered unsuccessful.

Postinjection, we performed dynamic acquisition (30 minutes; 60 seconds per frame, 256 × 256 matrix) followed by the static AP, AL and AO positions (300 seconds per position) at 30 min, 1 h, 2 h and 16 h post injection [1], using the dual headed gamma camera Mediso DHV Nucline Spirit. We used cobalt source Featherlite Co57 flood source MED 3709 for body contour drawing, and the SLN detection was performed with gamma detection probe EUROPROBE SYSTEM CE 0459. Preoperatively we used the gamma probe to detect the SLN location, quantify and mark the skin site above it in 2 positions AP and AL/AO in order to help the surgeon to determine the best operation incision approach for minimally invasive surgical procedure. Intraoperative, the gamma probe was used for radio guided surgery, activity quantification of the extirpated SLN and possible additional SLN or remnant detection.

The sensitivity and specificity of the method were increased with the use of sterile solution of metilen blue dye (indigo carmine) 0.5–1 ml, injected intradermally 15–20 min. before the operative procedure. The histopathological evaluation of the extirpated SLN was performed by routine hematoxylin and eosin (H&E) staining (ex tempore) and immunohistochemistysection analyses.

Results

Prior to using the preserving surgical protocols, during 2004, 2005 and 2006 a learning curve of 15 pts, using Re-S-colloid-Tc99m tracers with diameter 200–300 nm (in house produced) and blue dye, was performed which showed 93% sensitivity and 96% specificity of the method. The results were in favor of introducing the preserving surgical protocol and correlated with the scientific data worldwide (Fig 1).

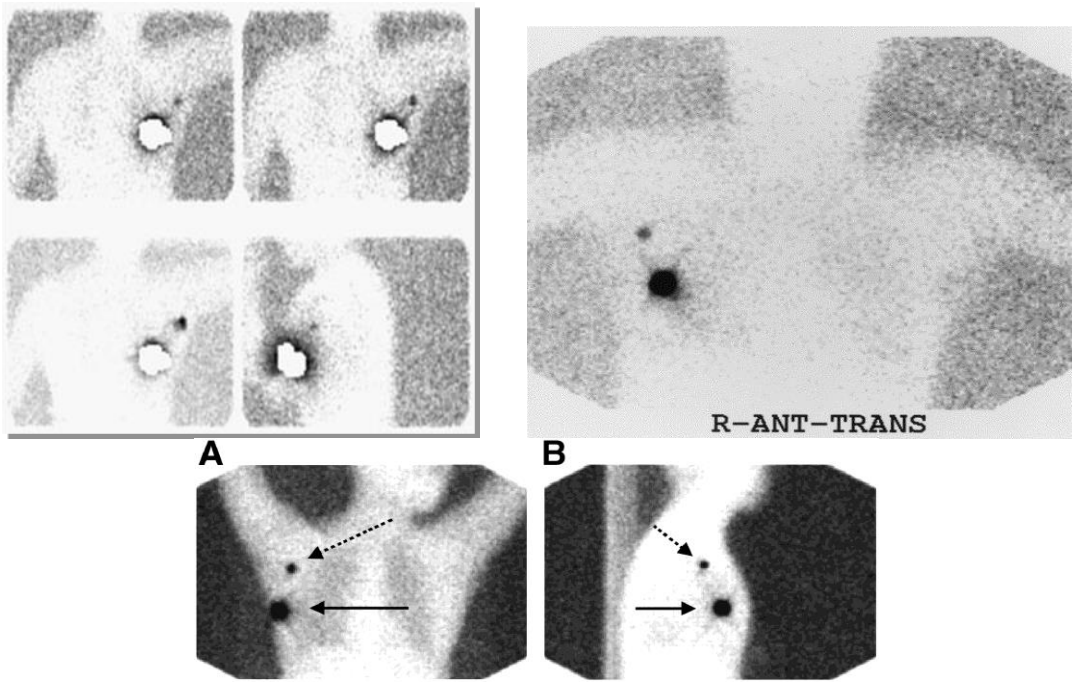

Figure 1 – Scintigraphic images obtained during the learning curve (visualization of the SLN). Bold arrows presenting the site of periareolar/peritomoral injection. Dotted arrows presenting radiotracer accumulation in SLN

SLN localized with radiocolloid were detected in 97.6% (periareolar injection) and 98% (combined periareolar and peritumoral injection) and blue dye in 77.7% of the cases. Ex tempore SLN evaluation was negative in 28/38 (74%) pts, which implicated the preserving surgical protocol and the lymph node pathways were preserved, while in 10/38 (26%) pts it was positive and radical surgical intervention was performed (Fig 2).

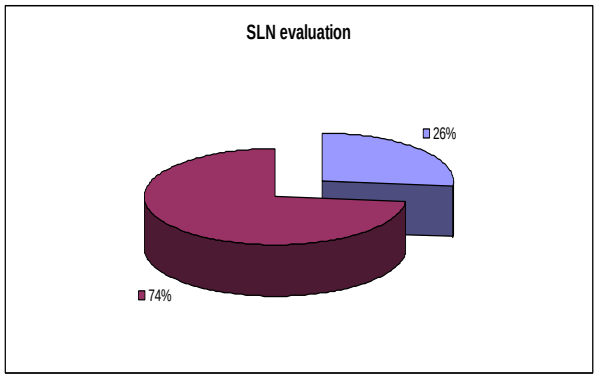

Figure 2 – Ex tempore SLN evaluation: 74% of pts – negative SNL for metastasis, 24% of pts – positive SLN for metastasis

In 3/28 ex tempore negative SLN, the postoperative histopathological analysis showed metastatic/micrometastatic involvement (false negative) and second phase radical surgical procedure was performed. In 3/10 ex tempore positive SLN micrometastases (0.2–2 mm) were detected, but nevertheless, radical surgery was performed.

Radiocolloid uptake was higher in SLN accumulating blue dye vs the non-accumulating (1643 ± 3216 counts/10 sec vs 526 ± 1284 counts/10 sec, $p < 0.001$). SLN with higher colloid uptake were free of metastases $p < 0.05$ vs SLN with lower colloid uptake $p < 0.01$, most probably due to lymph node destruction from metastatic tissue.

12 pts had more than one SLN (i.e. 2 SLN), 8/12 (66%) pts had negative and 4/12 (34%) pts had positive SLN. 3 pts had a rare double drainage to axilla and a. mammaria int. and were selected for adjuvant oncological protocols (Fig. 3).

Figure 3 – Rare double drainage to axilla and a. mammaria int. Solid arrow presenting radiotracer accumulation in intramammary SLN. Dotted arrow presenting radiotracer accumulation in axillary SLN

According to our data, there was a strong correlation between the size of the tumor and the metastatic involvement of SLN ($p < 0.001$). (Fig. 4) The number of pts with positive SLN increased with the increasing of the tumor size ($p < 0.001$), while thosewith negative SLN decreased with the increasing of the tumor size ($p < 0.001$)

Correlation between size of tumor and metastatic involvement of SLN		
Size of tumor and type of SLN	**Number of patients**	**Percentage**
Positive SLN; Tu ≤ 1cm.	**1**	**2,6%**
Positive SLN; Tu 1-2 cm.	**3**	**7,9%**
Positive SLN; Tu > 2 cm.	**6**	**15,8%**
Negative SLN; Tu ≤ 1 cm.	**24**	**63,2%**
Negative SLN; Tu 1-2 cm.	**3**	**7,9%**
Negative SLN; Tu > 2 cm.	**1**	**2,6%**
All	**38**	**100%**

Figure 4 – Correlation between size of tumor and metastatic involvement of SLN

Discussion

SLNB (sentinel lymph node biopsy) is currently accepted as the standard method in the evaluation of axillary status of stage I and II breast cancer. Almost 75% of early staged breast cancer patients benefit from the technique in terms of axillary dissection-related morbidity. Despite the cumulative experience on the subject, the ideal SLN localization technique has yet to be defined. A combined method using blue dye and radiocolloid has been proposed as the gold standard [1, 23].

SLN detection rate varies according to the type of colloid and injection modality. In a recent international multicenter study, an overall SLN detection rate of 97% was reported. When different injection modalities were compared, periareolar intradermal injection was found superior for lymphatic mapping [21, 23]. In our study, we demonstrated 97.6% and 98% SLN identification rates respectively using the periareolar intradermal and the combined periareolar and peritumoral injection technique. We consider both techniques to be a reliable option for radiocolloid application. However, our results failed to recommend the use of blue dye as a single agent in SLN identification, since it was successful only in 77.7% patients. SLN with blue dye uptake had significantly higher count rates ($p < 0.001$). Additionally, count rates were influenced by the size of the colloid. Larger colloids yielded a significantly higher count rate probably due to slower clearance from the SLN. SLN with higher colloid uptake were free of metastases $p < 0.05$ vs SLN with lower colloid uptake $p < 0.01$, most probably due to lymph node destruction from metastatic tissue. We also found a strong correlation between the size of the tumor and the metastatic involvement of SLN ($p < 0.001$).

SLN number is closely related to the size of the radiocolloid. In accordance with the relevant literature, the number of SLNs in our study was higher in pts treated with small radiocolloids (Re-S-colloid-Tc99m) [20, 21]. Injection modality and size of the colloid have been described as major factors that influence visualization of parasternal nodes [18]. By using deep injections and small colloids, the rate of parasternal node visualization increases.

Removal of palpable lymph nodes with no radiocolloid or blue dye uptake is of great importance for reducing the false-negative rate in SLNB.

SLN detection technique was developed for precise diagnosis of metastases in the axillary lymph nodes in breast cancer patients with clinically and ultrasonographically negative lymph nodes staged as T1-2 N0 M0 [1, 6]. Krag DN et al in their randomized NSABP B-32 study compare the SLN resection with the conventional axillary-lymph-node dissection in clinically node-negative patients [20]. They point out that ifaxillary SLN are free of metastases on ex tempore evaluation, selective lymphadenectomy (only the SLN) should be performed, but if metastases are detected, a radical lymphadenectomy is to be performed. They confirm that the technique for SLN detection is a minimally invasive procedure but at the same time a very precise, safe and reliable evaluation method for axillary lymph node metastatic involvement and that this technique

could be considered as a successful alternative to the conventional radical axillary lymph node dissection. In this manner, the complications such as lymph edemas, seromas, infections, numbness and impaired movements could be avoided. This is a strong argument in favor of the preserving surgical interventions vs the radical surgery.

Subsequently Giuliano AE et al., from the American College of Surgeons Oncology Group, in their recent study compared axillary dissecttion vs no axillary dissection in women with invasive breast cancer and concluded that SLNB alone is safe and does not affect the survival of women who underwent this procedure, and avoided the radical axillary dissection [21].

According to the American society of clinical oncology guidelines (2014), women without SLN metastases should not receive axillary lymph node dissection (ALND). Women with one to two metastatic SLNs planning to undergo breast-conserving surgery with whole-breast radiotherapy should not undergo ALND (in most cases), while those with SLN metastases who will undergo mastectomy should be offered ALND. These three recommendations are based on RCTs. Women with operable breast cancer and multicentric tumors, with ductal carcinoma in situ (DCIS) who will undergo mastectomy, who previously underwent breast and/or axillary surgery, or who received preoperative/neoadjuvant systemic therapy may be offered SNB. Women who have large or locally advanced invasive breast cancer (tumor size T3/T4), inflammatory breast cancer, or DCIS (when breast-conserving surgery is planned) or are pregnant should not undergo SNB.

In the last years, the status of the intramammary lymph nodes increases its predictive role in N (nodal) breast carcinoma grading and postoperative therapeutic protocols modification. The interest for this lymphatic pathway dates many years before the introduction of lymphoscintigraphy and SLN detection methods, when during the radical Halsted mastectomy the intramammary lymph nodes had been extirpated and histopathologically analyzed [7, 8]. This radical type of mastectomy enabled the first insight in the intramammary lymphatic pathways and those first results also indicated that if positive for metastases, this lymph nodes correlate with worse disease outcome, more aggressive carcinoma variants and lower survival rate [9–11].

Shen et al. in their study point out the fact that metastatically positive intramammary lymph nodes are an independent factor for the decrease in survival rate [5]. Guth et al. and Hogan et al. in two independent research studies conclude that patients with positive intramammary lymph nodes have more aggressive tumors, with higher rate of lymphovascular invasion and axillary metastases [4, 12].

Patients diagnosed with positive intramammary lymph nodes and negative axillary lymph nodes, have similar 10 years survival rate, approximately 60%, as patients with positive axillary lymph nodes [6, 11].

Up to date clinical research trials consider the postoperative systemic adjuvant chemo and radiotherapy as an option in this subgroup of patients [13–14]. No case of negative ISLN and positive ASLN has been reported in the literature. This fact raises the dilemma of possible primary breast carcinoma drainage towards the intramammary lymph nodes and remains as a scientific challenge in future. In this context, Vijan et al. in their scientific research conclude that patients with positive ISLN have high incidence of axillary metastatic involvement and they raise a hypothesis about intramammary lymph nodes being "sentinel nodes" for the axillary lymph nodes [18].

According to the scientific data, if intramammary lymph nodes are positive and axillary nodes are negative, radical axillary dissection could be avoided. The extent of the surgical procedure concerning the axilla should be based only on the status of the axillary lymph nodes [15–17, 19].

Conclusion

Our results confirm that the SLN detection technique is non invasive, safe and reliable and should be incorporated into the surgical guidelines in our country concerning the subset of breast cancer pts (T1-2 N0 M0), in order to avoid the unnecessary radical surgical procedures and postoperative complications. The most

reliable option for the radiolabeled colloid application is the combined technique of periareolar and peritumoral injection. Although the use of blue dye facilitated the procedure by adding a visual component, it was inefficient as a single agent. SLNs possessing higher radiocolloid count rates tended to accumulate blue dye more frequently. SLN with higher colloid uptake more often were free of metastases, while occurrence of metastatic SLN correlates positively with tumor size. Patients with drainage to a mammaria interna should be selected for adjuvant oncological protocols.

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Р е з и м е

ДЕТЕКЦИЈА НА СЕНТИНЕЛНИ ЛИМФНИ ЈАЗЛИ КАЈ КАРЦИНОМ НА ДОЈКА – ПРВИ ИСКУСТВА

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Вовед: Карциномот на дојка опфаќа 22,9% од сите карциноми кај женската популација и е одговорен за 13,7% од смртноста кај нив. Позитивните аксиларни лимфни јазли се предиктори за развој на дистантни метастази. Статусот на сентинелните лимфни јазли е круцијален за изборот на адекватна тераписка процедура.

Цел: Да се процени бенефитот од детекцијата на сентинелни лимфни јазли кај пациентите со карцином на дојка.

Материјал и методи: 38 женски пациенти, на возраст 44 ± 12 години, со T1-2 H0 M0 градуиран карцином на дојка, без зголемени аксиларни лимфни јазли на ултразвук беа вклучени во студијата. Детекцијата на сентинелните лимфни јазли беше изведена со гама-камера и гама детекциона сонда по периареоларно поткожно и/или перитуморално инјектирање на (99m-Technetium-SENTISCINT). Метиленско плаво беше аплицирано 20 мин. пред операцијата. Сентинелните лимфни јазли беа екстирпирани и подложени на ex tempore хистопатолошка евалуација.

Резултати: Сентинелните лимфни јазли беа негативни на ex tempore евалуација и лимфните патишта беа зачувани кај 28/38 (74%) пациентки. Кај 10/38 (26%) пациентки сентинелните лимфни јазли без позитивни, поради што следуваше радикална хируршка интервенција. Кај 3/28 ex tempore негативни пациентки, хистопатолошката анализа покажа метастатско зафаќање (лажно негативни). Кај 3/10 ex tempore позитивни пациентки беа откриени микрометастази 0,2–2 мм. 12 пациентки имаа 2 сентинелни лимфни јазли, од коишто 8/12 (66%) беа негативни, а 4/12 (34%) беа со позитивни лимфни јазли. Три пациентки имаа ретка двојна дренажа кон аксила и кон a. mammaria int.

Заклучок: Нашите резултати потврдуваат дека техниката за детекција на сентинелни лимфни јазли е неинвазивна, сигурна и прецизна и би требало да биде инкорпорирана во гајдлајнсите за пациентите со карцином на дојка во стадиум (T1-2 H0 M0). Најпрецизен начин за апликација на колоидниот трасер е комбинираната техника на периареоларно и перитуморско инјектирање. Пациентите со дренажа кон a. mammaria interna би требало да бидат селектирани за адјувантни онколошки тераписки протоколи.

Клучни зборови: карцином на дојка, сентинелни лимфни јазли, метиленско плаво.