# ACCURACY OF INTRAOPERATIVE RADIONUCLIDE SCINTIGRAPHY FOR DETECTION OF SENTINEL LYMPH NODE USING 99MTC LABELED NANOCOLLOIDS IN EARLY BREAST CARCINOMA

**Zaigham Salim Dar, Omer bin Abdul Aziz*, Tanveer Sajid**, Mujahid Khalid Ali**

Armed Forces Institute of Pathology/National University of Medical Sciences (NUMS) Rawalpindi Pakistan, *Combined Military Hospital Kohat/National University of Medical Sciences (NUMS) Pakistan, **Combined Military Hospital Quetta/National University of Medical Sciences (NUMS) Pakistan

## ABSTRACT

***Objective:*** To determine the accuracy of intraoperative radionuclide scintigraphy for detection of sentinel lymph node (SLN), using 99mTc-Nanocolloids, in patients with early breast carcinoma.
***Study Design:*** Cross sectional analytical study.
***Place and Duration of Study:*** Nuclear Medical Centre, Armed Forces Institute of Pathology, Rawalpindi and department of Surgery, Pak Emirates Military Hospital, Rawalpindi, from Jul 2012 to Jun 2013.
***Patients and Methods:*** After fulfilling the inclusion and exclusion criteria and taking written informed consent, 67 patients of carcinoma breast were enrolled in the study. Each patient was injected with 37 MBq of 99mTc-Nanocolloids, via sub areolar, peri-tumoural route, pre-operatively. SLNs were identified using handheld gamma probe followed by excision biopsy. Patients, then, underwent modified radical mastectomy with dissection of axillary lymph nodes (ALN) and the samples were sent for histopathology.
***Results:*** Out of 67 patients, 28 to 77 years of age (mean ± SD of 49.61 ± 11.572 years), 17.9% (12) patients had T1tumour while 82.1% (55) had T2 tumour (mean ± SD of 3.48 cm ± 1.050 cm). Histopathology revealed no metastasis in SLNs as well as ALN of 55.2% (37) patients while 40.3% (27) patients were positive for both. However, in 4.5% (3) patients SLNs were negative for metastasis with positive ALN. Diagnostic accuracy was calculated to be 95.5%, sensitivity 90%, false negative rate 4.5% and negative predictive value 92.5%.
***Conclusion:*** Intraoperative radionuclide scintigraphy, using 99mTc-Nanocolloids and handheld gamma probe is a reasonably accurate technique for localization of SLNs in early breast carcinoma.

**Keywords:** Carcinoma Breast, Sentinel lymph node, 99mTc-Nanocolloids.



## INTRODUCTION

Carcinoma breast is the most common non skin cancer and 5th most common cause of cancer death in females worldwide while it is the commonest cancer in female population of Pakistan[1]. Among all the factors that determine the prognosis of carcinoma breast, metastatic involvement of the axillary lymph nodes remains the most important[2]. This not only determines the prognosis but is also an important factor in deciding the treatment plan regarding surgery and adjuvant chemo-radiation therapy and predicting their outcome. Sentinel lymph node (SLN) biopsy has gained considerable acceptance among the clinicians. It is considered to be the standard diagnostic procedure for assessing metastatic status of axillary lymph node (ALN), which can craft the treatment plan[3]. However, recently conducted multi-institutional studies have reported false negative rates for SLN biopsy ranging from 5.5% to 16.7%[4-7] which are higher than the target of <5% set in the 2005 guidelines of American society of clinical oncology[2]. In order to localize SLN, either a combination of direct visualization technique, employing Isosulfan Blue dye and scintigraphy, is used[4-6] or scintigraphic technique alone is employed[7]. Carbon nanoparticles have been the latest addition to the dyes used in direct visualization technique, whereby 150 nm carbon particles are used to stain the SLN and drainage track black[8]. While doing scintigraphy, different radio pharma-

**Correspondence: Dr Zaigham Salim Dar,** Nuclear Medical Center AFIP Rawalpindi Pakistan (*Email: xaighamaster@gmail.com)*
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ceuticals are used. These include Nanocolloids[4,7] (human albumin colloids), Sulfur colloids[5] and Antimony-sulfur colloids[6], all labeled with 99mTc. Likewise, different injection techniques are used. Majority injects radio-pharmaceutical peri-tumouraly[4-6], while some inject the radio-pharmaceutical sub dermally[7]. In Pakistan, intra-operative localization of SLN has largely been restricted to direct visualization technique using injection of blue dye, majorly Isosulfan blue, and the use of radio-pharmaceuticals and hand-held gamma probe has been restricted to a few centres[9,10]. The objective of this study was to find out the accuracy of SLN localization in our setup using 99mTc labeled nanocolloids and handheld gamma probe and employing it for treatment of early carcinoma breast. Study was planned as a cross sectional analytical study with non-probability consecutive sampling.

## PATIENTS AND METHODS

This study was carried out at Nuclear Medical Centre, AFIP Rawalpindi and department of surgery Pake Emirates Military Hospital Rawalpindi from 1st Jul 2012 to 30th Jun 2013. Sample size (n=67) was calculated by using WHO sample size calculator with confidence level 95%, Level of significance 5%, Power of test 90%, Anti-cipation of the population proportion (P0)=0.933 (sensitivity of SLN biopsy was 93.3%)[5], with required absolute precision 6%. Sixty seven patients were included, through non probability consecutive sampling, who fulfilled inclusion and exclusion criteria, were included. Female patients who were diagnosed with stage 1 (T1 N0 M0) or stage 2 (T2 N0 M0) carcinoma breast, either on FNAC or Trucut biopsy, and planned for modified radical mastectomy were enrolled in the study. Written informed consent was taken from all patients and approval of Hospital ethical committee was obtained. Patients with recurrent breast carcinoma or carcinoma beyond stage-II (T3N0 M0 or beyond) on post op assessment were excluded. Moreover, patients with recurrent tumours, history of previous breast surgery, chemo or radiation therapy, neo-adjuvant therapy or allergy to radio-colloids were also excluded from the study. All patients were given sub areolar, peritumoural injections of 37 MBq (1 mCi) 99mTc labeled nanocolloids (Nanocoll® GE Healthcare Inc.) pre-operatively, on surgery table. Total volume of injection was kept around 4 ml for all patients and it was instilled at 4 different spots around the tumour. Injection site was massaged for a few seconds to ensure absorption and uptake of radiopharmaceutical. After 10 minutes, an incision was given in the axilla and hand held gamma probe (Capintec® Inc.) was inserted. SLN was identified, by nuclear medicine physician, as a lymph node or a group of lymph nodes exhibiting max number of counts per second. SLN biopsy was then taken by the surgical specialist and sent for histopathology in a formaldehyde container. Later, all patients underwent modified radical mastectomy with axillary lymph node dissection and the samples of ALNs were sent for histopathology. The data was compiled comprising of demographic data of the patient, tumour size, pre- and post- operative stage of disease and the histopathological findings of Sentinel and rest of the Axillary lymph nodes. The data was analyzed using SPSS® version 12. Mean and standard deviation for the quantitative variable i.e. age of the patient, laterality of tumour, size of tumour, number of sentinel lymph nodes removed, were calculated. Moreover, percentages were calculated for all the categorical variables including true positive, false positive and true negative SLNs. Based on the

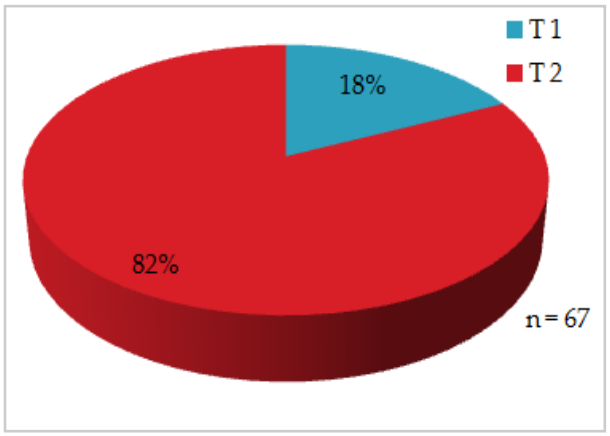


**Figure-1. Stage of tumour.**

data, accuracy, sensitivity and negative predictive value were computed.

## RESULTS

Total 67, patients were included in the study. None of the patients was dropped out or lost at any point in the study. The age distribution ranged from 28-77 years in the study. Mean age was 49.61 ± SD of 11.57 years. Majority, 28.3% (19/67), patients belonged to age group of 50-60 years followed by 26.9% (18/67) 40-50 years group, while least number of patients, 1.5% (1/67), belonged to the age group 20-30 years. 62.7% (42/67) patients had carcinoma in the right breast while remaining 37.3% (25/67) had carcinoma in the left breast with Right to Left ratio of 1.68:1. No Bilateral tumour was seen. Size of Tumour ranged from 1cm to 5cm with a mean size ± SD of 3.48 ± 1.05 cm. No patient with tumour size less than 1 cm was seen. Only 17.9% (12/67) patients had T1tumour (1-2 cm) while the rest 82.1% (55/67) had T2tumour (2-5 cm) as shown in fig-1. Varying number of Lymph nodes was removed while performing biopsy of SLN. In 20.9% (14/67) patients only 2 lymph nodes were removed. 50.7% (34/67) SLN biopsies had 3 lymph nodes removed and 28.3% (19/67) had 4 lymph nodes in the sample. Histopathology revealed that 59.7% (40/67) samples of SLN did not show any tumour metastasis while the rest of 40.3% (27/67) were positive for metastasis. However, histopathology of ALNs dissected from these patients showed metastasis in 44.8% (30/67) samples and metastasis free lymph nodes in 55.2% (37/67) patients. These results are elaborated in table-I & II. Based on the histopathology, 55.2% (37/67) cases were true negative and 40.3% (27/67) were found to be true positive (40.3%). However, 4.5% (3/67) cases were found to have false negative results. If SLN is positive for metastasis and ALN is negative, it is still considered as true positive thus accounting for zero false positive results. These results are shown in table-III & fig-2. Accuracy of sentinel lymph node localization using 99mTc labeled nanocolloids was calculated to be 95.5%,

**Table-I: Sentinel Lymph Node Histopathology.**

| | Frequency | Percentage (%) |
|---|---|---|
| Negative | 40 | 59.7 |
| Positive | 27 | 40.3 |
| Total (n) | 67 | 100 |

**Table-II: Axillary Lymph Node Histopathology.**

| | Frequency | Percentage (%) |
|---|---|---|
| Negative | 37 | 55.2 |
| Positive | 30 | 44.8 |
| Total (n) | 67 | 100 |

**Table-III: Distribution of true positives, True negative and False negative.**

| n=67 | ALN POS (30) | ALN NEG (37) |
|---|---|---|
| SLN POS (27) | 27 (TP) | - |
| SLN NEG (40) | 3 (FN) | 37 (TN) |

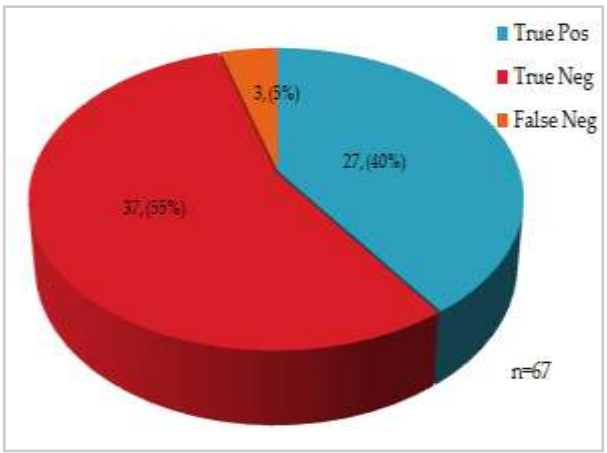


**Figure-2: Percentage of True Negative, True Positive and False Negative cases.**

Sensitivity 90%, Negative predictive value 92.5% and false negative rate 10%.

## DISCUSSION

Incidence of carcinoma breast in Pakistan is higher than the neighboring countries, almost 2.5 times the incidence in Iran and India, and is second to only Jewish population in the world. This incidence is rising with presentation at much younger age than the west[1]. Recent past has seen multiple, large scale, multi-institutional studies revealing SLN biopsy's false-negative rates ranging from 5.5% to 16.7%[4-7], higher than the target (<5%) set by the 2005 guidelines of the American society of clinical oncology[2]. In our study, the accuracy of the SLN detection with radionuclide technique was 95.5% comparable to that of ALMANAC validation study (97.6%) and NSABP B-32 trial (97.3%)[4,5]. The overall sensitivity of 90% in our study, was less than 93.3% as found out in the axillary lymphatic mapping against nodal axillary clearance (ALMANAC) validation study and 90.1% in national surgical adjuvant breast and bowel project (NSABP) B-32 trial[4,5]. The false negative rate in our study was 10% (3/30) which was not within the target (<5%) set by 2005 guidelines of American society of clinical oncology[2]. The false negative rate in our study was 8.6% (2/23) for T2 tumours and 14.3% (2/23) for T1tumours. In the ALMANAC validation study, overall false negative rate of 6.7% was observed, 7.7% in patients with T2 disease and 5.2% for T1tumours, with an accuracy of 97.6%[4]. Another study, the NSABP B-32 phase-III trial, observed false negative rate of 9.8%, with 8.9% for T2 tumours and 10.3% for T1 tumours, and an accuracy of 97.3%[5]. In the most recent randomized controlled surgical trial carried out by sentinel node versus axillary clearance (SNAC) trial group of the royal Australasian college of surgeons (RACS) and NHMRC clinical trials centre, the false negative rate was 5.5%. However this study was done with 99mTc labeled antimony sulfide colloids[6]. Our choice for radio pharmaceutical was nanocolloids (Human albumin colloids) because of smaller particle size than other commonly available agent i.e., sulfur colloids. However sulfur colloid particles passed through 0.22 µm filter gives equally good results[11]. Antimony sulfur colloid particles are smaller than Nanocolloids but are not available in Pakistan. In this study we preferred peri-tumoural injection as it is more widely used though some researchers have advocated sub dermal injection technique as well. However, certain studies that compared peri-tumour and subdermal injection and no significant difference were found in SNL localization[12]. In addition researches have worked on another debate about the number of injections that need to be administered. Some institutions have advocated 2 injections around the tumour while others have recommended 4 injection however a recent study has revealed no statistically significant difference in rate of identification of SLN with any of the technique[13]. Injection volume in our study was kept standardized at 4ml injection volume is one variable that has not been standardized for SNL localization. Researchers have used as low as 0.4 ml and as high as 8 ml to achieve results, however, success rate using larger volumes has remained high and it is hypothesized that larger volume produce high interstitial pressure leading to better lymphatic absorption[14]. SLN localization in our setup using 99mTc labeled Nanocolloids and handheld gamma probe and employing it for treatment of early carcinoma breast can save patients from significant morbidity associated with extensive axillary surgery especially in young patients who can undergo breast conservation surgery. Our study has some limitations, of which most important is being a small sample size which cannot be considered as representative of entire population. Further more, Lympho-scintigraphy was not performed prior to on table SNL localization which documents lymphatic drainage to nodes other than axillary group like internal mammary or supraclavicular basins and hence easily identifies the failed radiopharma-ceutical migration[11]. Moreover, cost effectiveness of the procedure was not calculated and no long term follow up was done.

## CONCLUSION

Intraoperative radionuclide scintigraphy, using 99mTc labeled Nanocolloids and handheld gamma probe, for sentinel lymph node metastasis detection can be employed as a reasonably accurate diagnostic technique in early breast carcinoma patients and can be used as a diagnostic procedure for assessment of axillary lymph node status for metastasis which may help tailor the treatment plan.

## CONFLICT OF INTEREST

This study has no confict of interest to be declared by any author.

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