

Diagnostic Accuracy of Sentinel Lymph Node Biopsy with Methylene Blue for Detection of Sentinel Lymph Node Metastasis Taking Histopathology as Gold Standard

GUL RUKH KHAWAR¹, MUHAMMAD AZAM TAYYAB², SAAD ARSHAD², UZAIR AHMED QURESHI², ABDUR REHMAN ALVI³

¹Gul Rukh Khawar, Postgraduate Trainee, Department of Surgery, DHQ Teaching Hospital, Gujranwala

²Senior Registrar, Department of Surgery, DHQ Teaching Hospital, Gujranwala

³Chief Consultant Surgeon, DHQ Teaching Hospital, Gujranwala

Correspondence to: Saad Arshad, Email: saadihere@gmail.com, Cell: 0331-6453506

ABSTRACT

Objective: The objective of this study was to determine the diagnostic accuracy of sentinel lymph node biopsy using methylene blue for diagnosis of sentinel lymph node metastasis taking histopathology as a reference standard.

Material and Methods: The cross-sectional study was conducted at the Surgery Department, DHQ Hospital, Gujranwala. 126 patients accomplishing the inclusion criteria were included in this research. The procedure was carried out under general anesthesia. After draping, 5ml injection of 1% methylene blue was administered in the subareolar area, divided in 3 injections. Massage was done for 10–15 min. A valuable anatomical landmark was made to perform the incision 1cm beneath the axillary hairline. Dissection of skin and subcutaneous tissue was made followed by the dissection of the clavipectoral fascia to invade the axilla, and lymphatics having blue staining were recognized. Sensitivity, specificity, PPV, NPV, and diagnostic accuracy of SLNB was calculated using 2x2 table, considering histopathology as a reference frame.

Results: Mean age of the study participants was 54.02 ± 12.07 years with minimum age as 25 years and maximum age as 75 years. There were 59(46.8%) females who were diagnosed of sentinel lymph node with methylene blue and 58(46%) females were positive with Sentinel lymph with histopathology. The sensitivity, specificity, positive predictive value, negative predictive value and overall diagnostic accuracy of methylene blue was 89.66%, 89.71%, 88.14%, 91.04% and 89.68% respectively. Stratification of data was done for age groups, duration of disease, marital status, parity, and type of treatment and high sensitivity, specificity, PPV, NPV, and diagnostic accuracy was found in each strata with significant p-value <0.001.

Conclusion: It is concluded that on the basis of results of current study the sensitivity, specificity, PPV, NPV, and overall diagnostic accuracy of methylene blue was highly significant in assessing sentinel lymph nodes. Hence, we can implement the results of this study for our population and implement the utilization of SLNB method making use of methylene blue dye instead of going for the excision biopsy. It poses fewer complications as compared to other comparable procedures. Also, it is an accurate, easily accessible and cheap method to evaluate the status of metastasis of lymph nodes in the axilla.

Keywords: Axillary lymph node, methylene blue, Sentinel lymph node biopsy, Breast cancer.

INTRODUCTION

Globally, the most prevalent cancer (CA) among women is the breast cancer with approximately greater than 2 million newly detected cases in the year 2020¹ and is one of the principal causes of deaths related to cancer in females². In Pakistan, breast cancer accounts for more than 19% of the cancers being encountered in both sexes and around 36.77% of all cancers occurring in women³. Histopathology is regarded as the reference frame for the evaluation of CA Breast 4. Status of the lymph nodes in the axilla is of great importance in defining the prognosis of breast cancer as well as choosing the right treatment^{4,5}. The lymphatic route is the principal way of breast cancer to metastasize from the origin. Therefore, nodal assessment is a vital part of the CA breast management^{5,6}.

The most common ways to assess the axillary lymph nodes are either the dissection of the lymph nodes in the axilla or the sentinel lymph node biopsy (SLNB)⁷. Dissection of the axillary lymph node is associated with significant adverse events (15-20%), including nerve damage, numbness, lymphedema, and wound infection⁸. Therefore, SLNB is now a widely practiced and a comparatively safe procedure for staging of the lymph node in early CA breast⁹.

The first lymph node that gets lymphatic drainage from tumor is termed as the sentinel lymph node⁵. A lot of different dyes are being utilized to observe the sentinel lymph node. But, methylene blue dye is regarded to have a comparatively lower possibility of allergic reactions, be inexpensive and easily accessible^{10,11}. One study reported that the sensitivity and specificity of SLNB with methylene blue were 89% and 100% respectively in cases of breast carcinoma¹². One more research reported that specificity and sensitivity were 85.7% and 100% respectively of SLNB with methylene blue in cases of breast carcinoma⁷. Some other study showed 85% sensitivity while 100% specificity of SLNB with methylene blue in cases of CA breast¹³. Another study reported the sensitivity of SLNB with methylene blue in CA breast as 90.48%,

specificity as 85.71%, positive predictive value (PPV) as 90.48%, negative predictive value (NPV) as 85.71%, and accuracy as 88.57%⁹. One more study conducted in Pakistan reported that the positive axillary lymph nodes for CA were 71.7% and the sensitivity, specificity and accuracy of SLNB with 1% methylene blue dye were 96.8%, 86.3%, and 94.1% respectively⁴.

Rationale of the current study was to determine the diagnostic accuracy of SLNB with methylene blue for diagnosis of metastasis of sentinel lymph node taking histopathology as reference standard. Literature has shown that SLNB using methylene blue is highly accurate to detect metastasis or involvement of the lymph node in CA breast patients. But varied data has been retrieved from literature. So to confirm the evidence, this study was conducted. The present study including comparatively larger sample size than the already conducted researches in Pakistan^{4,13} will assist in the better selection of appropriate treatment of CA breast patients. So that in the future, the results of this study can be implemented in local hospital settings and the utilization of SLNB method using methylene blue dye can be implemented instead of going for the excision biopsy.

MATERIAL AND METHODS

It was a cross-sectional research carried out at the Surgery department, District Headquarter Hospital, Gujranwala from Nov 21, 2019 till May 21, 2020. Sample size (n=126) was calculated with 95% confidence interval, using 56% expected percentage of sentinel lymph node¹⁴ with sensitivity and specificity of SLNB with methylene blue, taking 12% margin of error, as 85.7%, and 71.4%, respectively⁷. Study included 126 consecutive female cases, aged between 25-75 years, presented with CA breast and planned to undergo assessment of sentinel lymph node through histopathology. Patients having positive history of systemic problems for example diabetes mellitus, liver disease, abnormal blood clotting profile and patients receiving neo-adjuvant treatment

or those having inflammatory CA breast were not included in the study.

Following the approval from the institutional ethical committee and an informed written consent from each participant willing to take part in the study, baseline information (age, marital status, parity, duration of CA, and type of treatment taking for CA) was recorded. Then all patients underwent sentinel lymph node biopsy. The biopsy was performed under general anesthesia. After draping, 5ml injection of 1% methylene blue was given in the subareolar area, divided in 3 injections. Massage was done for 10–15 min. A valuable anatomical landmark was made to perform the incision 1cm beneath the axillary hairline. Dissection of the skin and subcutaneous tissue was made followed by the dissection of the clavipectoral fascia to invade the axilla, and lymphatics having blue staining were recognized. These nodes were dissected and labeled positive or negative. It was labeled as positive if node adopted the blue color of methylene used in the biopsy and was labeled as negative if the node did not adopt blue color. The breast tissue or excised mass along with the remaining axillary lymph nodes were histopathologically examined separately from sentinel lymph nodes and patients were regarded as either negative or positive. It was labeled as positive if the hypothetical first lymph node drained cancer and was labeled as negative if the node did not drain cancer.

SPSS version 20 was utilized to enter and analyze all the collected data. Quantitative data such as age and duration of CA was exhibited by mean and standard deviation. Whereas, frequency and percentage was utilized to demonstrate qualitative data such as parity, marital status, type of treatment for CA and metastasis. Sensitivity, specificity, PPV, NPV, and diagnostic accuracy of SLNB were calculated using 2x2 table, considering histopathology as a reference frame. Chi square test was applied and p value less than 0.05 were considered significant.

RESULTS

Baseline parameters of the participants were demonstrated in Table 1. Mean age of the patients was 54.02 ± 12.07 years with minimum age as 25 years and maximum age as 75 years. There

were 52(41.3%) females who were 25-50 years old and 74(58.7%) females were 51-75 years old. The mean duration of disease was 5.56 ± 1.90 weeks with minimum and maximum duration as 2 and 8 week. There were 24(19%) females who had duration of disease as <4 weeks and 102(81%) females had duration as 4-8 weeks. There were 97(77%) married and 29(23%) un-married females. Females who had parity 1-2 were 46(36.5%) and 51(40.5%) females had parity > 2. Females who were treated with Chemotherapy were 71(56.3%) and 55(43.7%) females were treated with Radiotherapy

Table 1: Baseline parameters of study participants (N=126)

Age of patients* (years)		54.02 ± 12.07
Age groups of patients* (years)	25-50	52(41.3%)
	51-75	74(58.7%)
Duration of disease *(weeks)		5.56 ± 1.90
Duration of disease groups* (weeks)	<4	24(19.0%)
	4-8	102(81.0%)
Marital status*	Married	97(77.0%)
	Un-married	29(23.0%)
Parity*	Null	29(23.01%)
	1-2	46(36.5%)
	>2	51(40.5%)
Type of treatment*	Chemotherapy	71(56.3%)
	Radiotherapy	55(43.7%)

*: continuous data is given as mean ± standard deviation while discrete data as number and Percentage; n (%) = number (percentage).

Table 2 showed there were 59(46.8%) females who were diagnosed of sentinel lymph node with methylene blue and 58(46%) females were positive with Sentinel lymph with histopathology. There were 52 females who were diagnosed of Sentinel lymph node on both methylene blue and on histopathology and 61 females were not diagnosed of Sentinel lymph node on both methylene blue and on histopathology. There were 7 false positive and 6 false negative. Chi-square test was performed and it turned out to be 79.17 with a highly significant P-value = <0.001. The sensitivity, specificity, PPV, NPV, and overall diagnostic accuracy of methylene blue was 89.66%, 89.71%, 88.14%, 91.04% and 89.68%, respectively.

Table 2: Diagnostic Accuracy of SLNB with methylene blue taking histopathology as a reference standard

Sentinel lymph node		With Histopathology			p-value*	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	Diagnostic accuracy (%)
		+	-	Total						
With Methylene blue	+	52	7	59	<0.001	89.66	89.71	88.14	91.04	89.68
	-	6	61	67						
	Total	58	68	126						

SLNB = Sentinel lymph node biopsy; PPV =Positive predictive value; NPV = Negative predictive value; % = percentage; * = Chi square test was utilized to compute p value and less than 0.05 p value was regarded as significant.

DISCUSSION

The advantages of SLNB in CA breast patients have now been well established¹⁵. However the most beneficial technique to distinguish SLN and its biopsy has consistently been under deate. Accurate recognition of SLN always proved to enhance the experience of a surgeon. In current study the sensitivity, specificity, PPV, NPV, and overall diagnostic accuracy of methylene blue was 89.66%, 89.71%, 88.14%, 91.04% and 89.68% respectively.

One study reported that the sensitivity and specificity of SLNB with methylene blue were 89% and 100% respectively in cases of breast carcinoma¹². In another research, it was reported that the sensitivity and specificity of SLNB with methylene blue in CA breast cases were 100% and 85.7%, respectively⁷. One more study showed 85.7% sensitivity while 71.4% specificity of SLNB with methylene blue in cases of CA breast¹³.

Our results showed a higher diagnostic accuracy of 89.68%. A meta-analysis, including 18 researches, showed that the combined rate of identification was 91%, and the false negative rate was 13%. The collective sensitivity was shown to be 87%, NPV be 91%, and accuracy rate was 94%. So, it concluded that

acceptable rate of identification was obtained by plotting locations of SLN with methylene blue dye alone¹⁶.

Likewise, another local quasi experimental study was conducted to FIND OUT the diagnostic accuracy of methylene blue dye FOR THE DETECTION OF METASTASIS OF AXILLARY LYMPH NODES. The main findings revealed that 71.1% CASES HAD axillary lymph nodes positive for CA. The sensitivity was 96.8%, specificity 86.36%, and accuracy was 94.1%. Thus, technique using Methylene blue dye is an authentic and secure diagnostic technique for SLN identification in CA breast cases owing to its favorably elevated accuracy⁴. Another study revealed that sensitivity of SLNB with methylene blue in CA breast was 90.48%, specificity was 85.71%, PPV was 90.48%, NPV was 85.71%, and accuracy was 88.57%⁹.

The major strength of the current study is that the sample size was 126 which was comparatively larger than the already conducted researches in Pakistan. For instance, Bakhtiar et al.⁴ took in 85 patients, the study carried out by Sohail et al. comprised of only 30 patients¹³, whereas Vohra et al.¹⁷ took in only 30 cases. Also, the current study included the data stratification for age groups, duration of disease, marital status, parity, and type of

treatment. High sensitivity, specificity, PPV, NPV, and diagnostic accuracy was found in each strata with significant p-value <0.001, not discussed in other studies.

There is already an intense monetary burden on the health management system of Pakistan owing to its day by day growing population comprising of mainly females (>50%) who have an elevated risk of developing CA breast (1 in every 9 women)¹⁸. Hence it is very crucial to find out the cost effective ways to cater this serious issue. The outcomes of the present study have advocated the benefits of utilizing methylene blue dye in less privileged settings owing to its favorable accuracy rates without making use of complex and high-cost instruments.

CONCLUSION

It is concluded that on the basis of results of current study the sensitivity, specificity, PPV, NPV, and overall diagnostic accuracy of methylene blue was highly significant in assessing sentinel lymph nodes. Hence, we can implement the results of this study for our population and implement the utilization of SLNB method making use of methylene blue dye instead of going for the excision biopsy. It poses less complications as compared to other comparable procedures. Also, it is an accurate, easily accessible and cheap method to evaluate the status of metastasis of lymph nodes in the axilla.

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