



Clinical and Inflammatory Predictors of Sentinel Lymph Node Involvement in T1 Early-stage Breast Cancer with Unfavorable Histology

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Abstract

Aim: Despite advances in early detection and surgical de-escalation strategies in breast cancer, accurately predicting sentinel lymph node involvement (SLNI) among patients with T1 disease remains a clinical challenge, particularly in those with unfavorable histologic subtypes. In this context, this study aimed to examine clinical factors related to pathological SLNI among patients with T1 early-stage breast cancer (ESBC) and unfavorable histologic subtypes.

Methods: This retrospective analysis included 128 patients with clinically node-negative T1 ESBC and unfavorable histology who underwent surgery between January 2010 and December 2020. Clinicopathological and preoperative laboratory parameters were analyzed. To identify the independent risk factors associated with SLNI, logistic regression analyses were performed.

Results: Thirty-six (28.1%) patients were SLNI-positive, and 92 (71.9%) were SLNI-negative. In univariate analysis, the presence of lymph nodes with a thickened cortex on preoperative ultrasonography ($p=0.016$), lymphovascular invasion (LVI) ($p=0.002$), larger tumor size ($p=0.002$), and higher neutrophil levels ($p=0.046$) were significantly associated with SLNI positivity. SLNI-positive patients also demonstrated significantly lower serum albumin levels ($p=0.002$), while monocyte levels exhibited a tendency toward lower values ($p=0.064$). In multivariate analysis, serum albumin levels ($p=0.015$), neutrophil count ($p=0.031$), monocyte count ($p=0.035$), and LVI ($p=0.040$) remained independently associated with SLNI.

Conclusion: Selected clinicopathological and inflammatory parameters were independently associated with SLNI in patients with T1 ESBC and unfavorable histology and may help identify patients at higher risk of nodal involvement.

Keywords: Breast neoplasm, sentinel lymph node biopsy, inflammation, albumin, retrospective study

Introduction

In patients with clinically node-negative (cN0) early-stage breast cancer (ESBC), sentinel lymph node biopsy (SLNB) is currently used as the standard approach for axillary staging

(1,2). Minimally invasive SLNB is still a surgical procedure (3). In the era of treatment de-escalation, the routine use of axillary staging has increasingly been debated, particularly in patients with small primary tumors (4-6). The prevalence of sentinel lymph node involvement (SLNI)

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in patients with T1 tumors has been reported to range from approximately 18% to 36% (2,3). Similarly, Wang et al. (4) reported SLNI rates of 2.8% in T1mic, 4.5% in T1a, 9.3% in T1b, and 21.0% in T1c tumors. This indicates that a considerable proportion of patients undergo axillary surgery without nodal disease. To address this issue, several prediction models based on clinicopathological characteristics and advanced imaging techniques have been developed to estimate the preoperative risk of SLNI in ESBC. However, none of them have been universally adopted as a standardized tool in routine practice (7,8). Recent randomized trials, including SOUND and INSEMA, have further suggested that omitting SLNB in carefully selected patients may be non-inferior to performing SLNB. This supports the concept of a more selective approach to axillary surgery in T1 ESBC with respect to oncological outcomes (5,6).

Tumor progression and prognosis have been associated with the systemic inflammatory (SI) response (9). In particular, lymphocyte-to-monocyte ratio (LMR) and neutrophil-to-lymphocyte ratio (NLR) have been reported as prognostic biomarkers in breast cancer (BC) patients (9). Fuji et al. (10) also stated that low serum albumin levels have been associated with poorer overall survival (OS) and recurrence-free survival (RFS) in ESBC patients. Several studies have evaluated the association between preoperative inflammatory markers and SLNI in cN0 T1 BC (2,3). However, the clinical utility of these inflammatory markers for guiding axillary surgery decision-making in T1 ESBC remains unclear.

We hypothesized that in patients with T1 BC and unfavorable histologic types, certain clinical factors are associated with an increased risk of pathological SLNI. Accordingly, the aim of this study was to evaluate the clinical factors associated with pathological SLNI in ESBC patients with T1 tumors and unfavorable histologic types. Improved preoperative identification of patients at increased risk of SLNI may help refine patient selection for SLNB and provide additional information for individualized axillary management in ESBC.

Materials and Methods

Compliance with Ethical Standards

This study was conducted in accordance with the principles of the Declaration of Helsinki. Ankara Bilkent City Hospital Clinical Research Ethics Committee No. 1 approved the study (approval no.: E1/2236/2020, date: 15.12.2021) and waived the requirement for informed consent due to its retrospective nature.

Study Rationale

The literature indicates that SLNI plays a more critical role in guiding adjuvant and neoadjuvant treatment

decisions for T1 BC than for T2 tumors. Due to differences in neoadjuvant and adjuvant chemotherapy strategies between T1 and T2 disease, patients with T2 tumors were excluded from the present study. In addition, previous studies have indicated that the necessity of SLNB is controversial and that the incidence of SLNI is relatively low in small tumors with favorable histologic types. Therefore, these tumors were also excluded from the analysis.

Exclusion Criteria

Patients with isolated tumor cells or micrometastases in the sentinel lymph node (SLN), hematologic disorders, SI diseases, a history of other malignancies, active infections, or steroid use within the past month were excluded. Patients who received neoadjuvant therapy, had incomplete clinical or laboratory data, or demonstrated lymphocytic infiltration consistent with breast inflammation on histopathological examination were also excluded from the study.

Study Cohort

Patients with ESBC who underwent SLNB at Ankara Bilkent City Hospital between January 2010 and December 2020 were analyzed retrospectively. A total of 547 individuals who underwent SLNB for clinically ESBC were evaluated. Patients with favorable histological tumor types and those with tumors larger than pT1 were excluded. The study cohort consisted of 128 patients with pT1 and unfavorable tumors (Figure 1).

Patients were then grouped according to SLNI into Group 1 (SLNI negative) and Group 2 (SLNI positive). The association between clinical characteristics, preoperative inflammatory biochemical markers, and SLNI was then analyzed.

Preoperative Breast Imaging and Diagnostic Procedures

Bilateral breast ultrasonography (US) was performed preoperatively in all patients, while those aged ≥ 40 years also underwent bilateral mammography for diagnostic evaluation and the planning of BC treatment. Breast imaging was performed by radiologists specializing in breast imaging. Following imaging of a suspicious breast lesion, the diagnosis of BC was established using a core needle biopsy, stereotactic biopsy, or excisional biopsy, as appropriate.

Preoperative Axillary Evaluation

During preoperative axillary US, lymph nodes (LNs) that demonstrated a preserved hilum and a normal or mildly thickened cortex (< 3 mm) were classified as cN0. Lymph nodes showing cortical thickening without loss of the hilum underwent fine-needle aspiration biopsy, and cases with negative cytology were also considered cN0. SLNB was performed in patients classified as cN0. Axillary

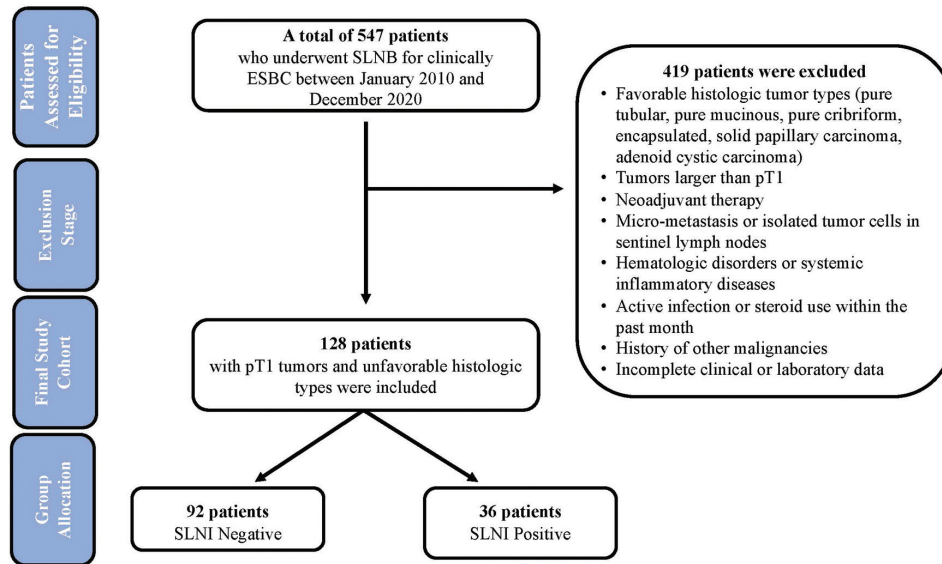


Figure 1. Study flow diagram

SLNB: Sentinel lymph node biopsy, ESBC: Early-stage breast cancer, SLNI: Sentinel lymph node involvement

LN with cortical thickness greater than 3 mm and an absent hilum were classified as clinically node-positive and excluded from the study.

SLNB Procedure

After induction of general anesthesia, the procedure continued with the injection of 10 mL of blue dye into the subareolar region. The blue dyes used were patent blue, isosulfan blue, and methylene blue. A 12-minute massage was performed to facilitate lymphatic drainage. All blue-stained LNs were designated as sentinel lymph nodes (SLN) and were excised. Intraoperative pathological evaluation was performed using frozen section analysis, with hematoxylin and eosin staining of SLNs sectioned at 2-mm intervals.

Because a single-dye technique was used, patients with two or fewer identified SLNs underwent axillary dissection if at least one SLN was positive on frozen section analysis. Patients in whom no SLNs were identified also underwent axillary dissection. In patients with three or more identified SLNs, axillary dissection was performed only when more than two SLNs were found to be positive on frozen section analysis.

Data Collection and Reference Ranges

Patient and tumor parameters, including histologic type and grade, clinical and pathological tumor size, molecular subtype, presence of lymphovascular invasion (LVI), and Ki-67 score, were collected. Ultrasonography characteristics of axillary LN, SLNI, the total number of excised SLNBs, and preoperative levels of white blood cell count (WBC), lymphocytes, monocytes, neutrophils, red cell distribution

width (RDW), albumin, and platelets were recorded. Reference ranges used in our biochemistry laboratory were as follows: WBC $3.9-10.2 \times 10^9/L$, lymphocytes $1.1-4.5 \times 10^9/L$, monocytes $0.1-0.9 \times 10^9/L$, neutrophils $1.5-7.7 \times 10^9/L$, RDW 11.5-16%, platelets $150-400 \times 10^9/L$, and albumin 3.2-4.8 g/dL. The LMR represents the ratio of total lymphocytes to total monocytes; the platelet-to-lymphocyte ratio (PLR) represents the ratio of total platelet count to total lymphocyte count; and the NLR represents the ratio of total neutrophils to total lymphocytes.

Study Outcomes

The primary outcome of this study was the identification of clinicopathological and preoperative SI parameters that were independently associated with SLNI in patients with T1 ESBC and unfavorable histologic types.

The secondary outcome was the identification of these parameters in order to provide additional information to support clinical decision-making in axillary management.

Statistical Analysis

All statistical analyses were conducted utilizing SPSS version 22.0 software (IBM Inc., Armonk, NY, USA). Data normality was examined with the Kolmogorov-Smirnov test, together with histogram and Q-Q plot inspection. Continuous data were summarized as mean \pm standard deviation, the Independent Samples t-test was employed to evaluate differences between these variables for normally distributed data, while the Mann-Whitney U test was used for non-normally distributed variables. For categorical data, the chi-square test or Fisher's exact test was applied, as appropriate.

Statistical significance is defined as a p-value of less than 0.05. Variables that were statistically significant in univariate analysis were included in multivariate analyses. In logistic regression, a p-value of ≤ 0.05 was designated as the threshold for model inclusion, while a p-value of ≥ 0.10 was established for model exclusion. The results of the multivariable analysis were reported as odds ratios (OR) together with the corresponding 95% confidence intervals (CI).

Results

This study included 128 female patients with a mean age of 51.73 ± 10.55 years (range: 35-84). The histological distribution of tumors was as follows: invasive ductal carcinoma (115 patients, 89.8%), invasive lobular carcinoma (4 patients, 3.1%), mixed-type carcinoma (1 patient, 0.8%), micropapillary carcinoma (1 patient, 0.8%), apocrine carcinoma (3 patients, 2.3%), and medullary-pattern ductal carcinoma (4 patients, 3.1%).

Patients were classified into Group 1 (SLNI negative), comprising 92 patients (71.9%), and Group 2 (SLNI positive), comprising 36 patients (28.1%). A mean of 3.07 ± 1.59 SLNs (range 1-7) was excised overall. When analyzed according to SLNI status, the mean number of excised LNs was 2.98 ± 1.55 in Group 1 and 3.27 ± 1.71 in Group 2, with no statistically significant difference between the groups ($p=0.36$). For all patients included in the study, final pathology results were consistent with frozen section findings.

Assessment of the cortical thickness of axillary LNs on preoperative US revealed statistically significant differences among the groups. A thickened cortex was observed more frequently in the SLNI-positive group (Group 1: 79.3% normal cortex, 20.7% thickened cortex; Group 2: 58.3% normal cortex, 41.7% thickened cortex; $p=0.016$).

Similarly, the presence of LVI was significantly higher in the SLNI-positive group than in the SLNI-negative group (Group 1: 73.9% absent, 26.1% present; Group 2: 44.4% absent, 55.6% present; $p=0.002$).

Tumor size, serum albumin levels, and neutrophil counts showed statistically significant differences between Group 1 (SLNI negative) and Group 2 (SLNI positive). Compared with Group 1, the SLNI-positive group demonstrated lower serum albumin levels, larger tumor size, and higher neutrophil counts (Albumin: Group 1, 4.52 ± 0.38 vs. Group 2, 4.28 ± 0.36 , $p=0.002$; Tumor size: Group 1, 13.30 ± 4.90 vs. Group 2, 15.47 ± 4.02 , $p=0.02$; Neutrophil count: Group 1, 4.31 ± 1.31 vs. Group 2, 4.90 ± 1.89 , $p=0.046$).

Comparisons of laboratory, demographic, and clinicopathological features between the groups are presented in Tables 1-3.

In univariate analysis, significant differences were observed between Group 1 (SLNI negative) and Group 2

(SLNI positive) with respect to tumor size, serum albumin levels, and neutrophil counts. Compared with the SLNI-negative group, patients with SLNI had lower serum albumin and monocyte levels, larger tumor size, a higher frequency of LVI, and higher neutrophil counts ($p=0.002$, $p=0.064$, $p=0.02$, $p=0.002$, and $p=0.046$, respectively).

In multivariate logistic regression analysis, LVI, serum albumin level, neutrophil count, and monocyte count were identified as independent variables associated with SLNI (Table 4).

Discussion

In invasive BC, SLNI is considered a critical prognostic indicator and has traditionally been evaluated together with tumor size when planning systemic and radiation therapy (6). Following the landmark studies by Giuliano et al. (11) and the ACOSOG Z0011 (Alliance) trial published in 2017 (12), routine axillary dissection has progressively been replaced by SLNB in cN0 patients with T1 and T2 BC.

Recent studies have increasingly evaluated the feasibility of omitting SLNB in selected subgroups of BC patients (13). In particular, the INSEMA and SOUND trials have demonstrated that omission of SLNB in carefully selected patients is non-inferior to standard SLNB with respect to oncological outcomes (5,6). These findings have not yet been fully integrated into existing clinical guidelines; however, they underscore the increasing significance of precisely predicting SLNI, particularly in patients with T1 ESBC. In this context, identifying clinicopathological characteristics and preoperative SI parameters associated with SLNI may support a more individualized approach to axillary management in patients with T1 ESBC and unfavorable histologic types.

Multiple clinical factors have been reported to influence disease progression and SLNI in patients with BC. In addition, accumulating evidence suggests that both disease progression and the likelihood of SLNI may be affected by the host's SI response (2,3,9,10,14,15). To more accurately evaluate the association between BC and preoperative factors potentially related to SLNI, it is important to focus on patient cohorts with homogeneous characteristics. Accordingly, the present study was restricted to patients with T1 tumors to minimize heterogeneity arising from differences in neoadjuvant and adjuvant treatment strategies for T1 versus T2 disease (16). In addition, favorable histologic subtypes, which have consistently been associated with lower rates of SLNI in the literature (17,18), were excluded from the analysis.

Low albumin levels in cancer patients have been attributed to malnutrition, increased metabolic demand, and suppressed albumin synthesis in the context of systemic inflammation. In addition, serum albumin has been proposed to function as an endogenous antioxidant

	Group 1 SLNI (-) (n=92)	Group 2 SLNI (+) (n=36)	p-value
Age (mean ± SD) (n=128)	52.58±10.98	49.55±9.16	0.145
Multifocality/multicentricity (n, %) (n=128)			0.295
Absent	72 (78.3%)	25 (69.4%)	
Present	20 (21.7%)	11 (30.6%)	
Lymph node characteristics (n, %) (n=128)			0.016
Normal finding	73 (79.3%)	21 (58.3%)	
Thick cortex	19 (20.7%)	15 (41.7%)	

Data are presented as mean±standard deviation or number (%). Continuous variables were compared using Independent Samples t-test or Mann-Whitney U test, as appropriate, and categorical variables were compared using the chi-square test.
 Bold values in the table represent statistically significant parameters.
 SLNI: Sentinel lymph node involvement, SD: Standard deviation

	Group 1 (n=92) SLNI (-)	Group 2 (n=36) SLNI (+)	p-value
Tumor size (mean ± SD) (n=128)	13.30±4.90	15.47±4.02	0.02
Grade (n, %) (n=128)			0.269
I	34 (37.0%)	8 (22.2%)	
II	39 (42.4%)	18 (50.0%)	
III	19 (20.6%)	10 (27.8%)	
LVI (n, %) (n=128)			0.002
Absent	68 (73.9%)	16 (44.4%)	
Present	24 (26.1%)	20 (55.6%)	
Ki-67 score (%) (n=95)			0.90
≤15	43 (61.4%)	15 (60%)	
>15	27 (38.6%)	10 (40%)	
Molecular subtype (n, %) (n=128)			0.872
Luminal A	45 (49.0%)	18 (50.0%)	
Luminal B	36 (39.1%)	12 (33.4%)	
Her2 enriched	5 (5.4%)	3 (8.3%)	
Triple negative	6 (6.5%)	3 (8.3%)	
Total removed SLN (mean±SD) (n=128)	2.98±1.55	3.27±1.71	0.360

Data are presented as mean±standard deviation or number (%). Continuous variables were compared using Independent Samples t-test or Mann-Whitney U test, as appropriate, and categorical variables were compared using chi-square test.
 Bold values in the table represent statistically significant parameters.
 LVI: Lymphovascular invasion, SLNI: Sentinel lymph node involvement, SLN: Sentinel lymph node, SD: Standard deviation

and may play a role in reducing cancer-related risk (19). Several previous studies have demonstrated an association between decreased serum albumin levels and poorer prognosis across different malignancies, including BC (10,19,20).

In a large population-based cohort study, Yang et al. (19) investigated the association between pre-diagnostic serum albumin levels and overall cancer risk across multiple cancer types. No significant association was observed for BC, which the authors attributed to the limited number of BC cases and to the low proportion of female participants in the study. In contrast to these

findings, the present study identified the preoperative serum albumin level as an independent predictor of SLNI in multivariate analysis among patients with T1 ESBC. We believe that this association becomes more apparent when the patient cohort is restricted to a clinically and pathologically homogeneous group, as described above.

Similarly, Fuji et al. (21) reported that reduced serum albumin levels were not associated with disease recurrence or SLNI in BC patients. However, in their subsequent study with longer follow-up, the same group demonstrated that OS and RFS were significantly shorter in patients with low serum albumin levels (10). These findings are consistent

Variables (mean ± SD)	Group 1 (n=92) SLNI (-)	Group 2 (n=36) SLNI (+)	p-value
Albumin (g/dL)	4.52±0.38	4.28±0.36	0.002
Lymphocyte (x10 ⁹ /L)	2.19±0.70	2.15±0.75	0.798
Monocyte (x10 ⁹ /L)	0.48±0.15	0.43±0.13	0.064
Neutrophil (x10 ⁹ /L)	4.31±1.31	4.90±1.89	0.046
Platelet (x10 ⁹ /L)	291.43±76.96	295.30±76.48	0.798
WBC (x10 ⁹ /L)	7.22±1.72	7.70±2.30	0.197
RDW (%)	13.91±1.44	14.28±1.56	0.209
LMR	4.75±1.61	5.25±2.06	0.147
NLR	2.19±1.10	2.51±1.36	0.167
PLR	148.34±67.87	154.86±72.49	0.663

Data are presented as mean±standard deviation. Continuous variables were compared using Independent Samples t-test or Mann-Whitney U test, as appropriate. Bold values in the table represent statistically significant parameters.
SLNI: Sentinel lymph node involvement, SD: Standard deviation, WBC: White blood cell counts, RDW: Red cell distribution width, LMR: Lymphocyte to monocyte ratio, NLR: Neutrophil to lymphocyte ratio, PLR: Platelet to lymphocyte ratio

	OR	95% CI	p-value
Tumor size (mm)	1.071	0.970-1.183	0.177
Albumin (g/dL)	0.234	0.073-0.750	0.015
Neutrophil (x10 ⁹ /L)	1.381	1.030-1.853	0.031
Monocyte (x10 ⁹ /L)	0.020	0.001-0.764	0.035
Lymphovascular invasion	2.632	1.045-6.452	0.040
Lymph node characteristics	1.647	0.640-4.255	0.301

Multivariate logistic regression analysis was performed to identify independent factors associated with SLNI. Bold values in the table represent statistically significant parameters.
OR: Odds ratio, CI: Confidence interval, SLNI: Sentinel lymph node involvement

with the prognostic trends observed in our cohort and support the potential clinical relevance of serum albumin in ESBC.

The literature presents inconsistent findings regarding preoperative inflammatory parameters. Several studies have reported an association between decreased lymphocyte counts, LMR and NLR; increased PLR; elevated monocyte and platelet counts; and unfavorable prognosis in patients with BC (14,15,22,23). In addition, some studies, including those with heterogeneous disease stages and ESBC populations, have demonstrated an association between PLR and SLNI (2,3,23).

In the present study, PLR values were higher in patients with SLNI, although this association did not reach statistical significance. This discrepancy may be related to the relatively small sample size of our cohort and to the inclusion of a more pathologically restricted population, specifically patients with unfavorable histologic subtypes of BC. In their meta-analysis, Hu et al. (14) reported that

low LMR was not significantly associated with SLNI in BC patients, which is consistent with our findings. Similarly, Goto et al. (22) found no significant differences in SLNI when comparing low and high LMR and NLR levels in BC patients receiving neoadjuvant chemotherapy. In line with these studies, no significant association was observed among NLR, LMR, and SLNI in our cohort.

In our multivariate analysis, decreased monocyte levels and increased neutrophil counts were identified as independent predictors of SLNI. These findings suggest that alterations in specific components of the SI response may be associated with nodal involvement in ESBC. Monocytes play an important role in tumor-host interactions and have been associated with tumor progression and metastatic potential in various malignancies. Consequently, fluctuations in circulating monocyte levels may indicate a compromised antitumor immune response that promotes lymphatic dissemination (24).

Consistent with previous studies, our findings indicate that LVI is significantly associated with SLNI (25,26). LVI is regarded as a biological prerequisite for systemic dissemination and metastatic spread, and its presence has been consistently associated with a poorer prognosis in BC patients. Kuhn et al. (27) emphasized that LVI reflects an aggressive tumor phenotype and is closely associated with nodal involvement. Furthermore, Wei et al. (28) demonstrated that LVI is an independent predictor of non-SLN metastases in BC patients with one or two positive SLNs, highlighting its potential role in guiding adjuvant treatment decisions. In line with these data, our results confirm LVI as a significant predictor of SLNI within a pathologically homogeneous cohort.

In the literature, distinct cut-off values for serum albumin have been reported across different patient populations, largely reflecting tumor burden or the

presence of an SI tumor microenvironment. Albumin levels below 4 g/dL are generally considered markers of poor prognosis and malnutrition in various malignancies. However, in ESBC, systemic effects of the disease are often limited, and albumin levels are therefore expected to remain within normal reference ranges. This characteristic of ESBC limits the ability to establish reliable cutoff values for SI markers such as albumin, neutrophil, and monocyte levels, which were identified in our study as significant predictors of SLNI. Future studies involving larger and more homogeneous cohorts may help define more precise cut-off values and enhance the clinical applicability of these parameters.

Study Limitations

This study has several limitations. Due to the retrospective design, access to detailed information on certain preoperative variables that may influence SI markers was limited. In addition, the single-center setting may limit the generalizability of the findings. Furthermore, the relatively small sample size resulted in limited variability in monocyte and albumin levels between groups, which may have hindered the identification of definitive cut-off values for these parameters.

Despite these limitations, the present study has notable strengths. The analysis was restricted to a clinically and pathologically homogeneous cohort consisting exclusively of patients with T1 ESBC and unfavorable histologic subtypes. This approach minimized heterogeneity related to tumor biology. Additionally, the evaluation focused on objectively measured preoperative parameters and the pathological assessment of SLN, which may contribute to the consistency of findings and provide a basis for prospective studies.

Conclusion

This study demonstrated that LVI and selected preoperative SI markers, including albumin, are associated with SLNI in patients with T1 ESBC and unfavorable histologic subtypes. These findings were derived from a pathologically homogeneous cohort and specifically reflect early-stage disease. The result may contribute to future studies aimed at improving preoperative assessment of SLNB and supporting clinical decision-making in axillary management.

Ethics

Ethics Committee Approval: Ankara Bilkent City Hospital Clinical Research Ethics Committee No. 1 approved the study (approval no.: E1/2236/2020, date: 15.12.2021).

Informed Consent: Waived the requirement for informed consent due to its retrospective nature.

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This research was presented as an oral presentation at the 23rd Turkish National Surgical Congress (April 2024, Antalya), and the abstract was included in the congress proceedings.

Footnotes

Authorship Contributions

Surgical and Medical Practices: E.G.D., E.M., Concept: E.G.D., E.M., Design: K.C., E.G.D., E.M., Data Collection or Processing: F.C., M.B.A., S.K.O., Analysis or Interpretation: H.P.O., E.M., Literature Search: K.C., H.P.O., E.M., Writing: K.C., H.P.O., E.M.

Conflict of interests: The authors declare that they have no conflict of interest related to this study.

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