

Serum Prostate-Specific Antigen as a Biomarker in Breast Carcinoma: A Hospital-Based Case-Control Study

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Abstract

Background: Breast carcinoma is the most frequently diagnosed malignancy in Indian women and continues to present at locally advanced stages. Prostate-specific antigen (PSA), historically regarded as prostate-specific, is now known to be expressed at low concentrations in female breast tissue and serum. Its diagnostic role in breast carcinoma remains contested. **Objectives:** To evaluate serum PSA (sPSA) as a biomarker for the diagnosis of breast carcinoma and to examine its association with clinical staging. **Methods:** A hospital-based observational case-control study was conducted in the Department of General Surgery, JSS Medical College and Hospital, Mysuru, between April 2024 and September 2025. Eighty women were enrolled: 40 cases with histopathologically confirmed breast malignancy and 40 controls with newly diagnosed benign breast disease. Serum PSA was estimated using a standardised chemiluminescent immunoassay (reference range 0–1.4 ng/mL). Statistical analyses included Chi-square test, independent t-test, one-way ANOVA and Pearson correlation, with $p < 0.05$ considered significant. **Results:** Cases were significantly older than controls (mean 51.58 ± 9.97 vs 25.25 ± 5.45 years, $p < 0.001$) and predominantly post-menopausal (52.5%). Invasive ductal carcinoma accounted for 85.0% of malignancies, and Stage III disease for 42.5% of cases. Serum PSA was detectable in 100.0% of cases (0.003–0.008 ng/mL) versus 55.0% of controls (0.003–0.006 ng/mL) ($\chi^2 = 20.717$, $p < 0.001$), although all values remained below the 1.4 ng/mL threshold. Detection-based diagnostic performance yielded sensitivity 100.0%, specificity 45.0%, positive predictive value 64.5%, negative predictive value 100.0% and accuracy 72.5%. No significant association was observed with clinical stage ($p = 0.576$), T stage ($p = 0.174$), N stage ($p = 0.841$) or lump size ($r = -0.128$, $p = 0.432$). **Conclusion:** Although absolute sPSA values remained below the conventional reference threshold, qualitative detectability of sPSA significantly distinguished malignant from benign breast disease. Serum PSA may serve as a minimally invasive rule-out biomarker but lacks specificity for confirmation and does not correlate with disease extent.

Keywords: Prostate-specific antigen; Breast carcinoma; Biomarker; Case-control study; Diagnostic accuracy; Kallikrein.

1. Introduction

Breast carcinoma is the most frequently diagnosed malignancy in women and the leading cause of cancer-related mortality among females worldwide. According to GLOBOCAN 2020, an estimated 2.3 million new cases of female breast cancer were diagnosed globally in 2020, accounting for 11.7% of all cancer diagnoses and surpassing lung cancer as the most commonly diagnosed cancer.^[1] Mortality from breast cancer contributed an estimated 685,000 deaths worldwide during the same year, with death rates in transitioning countries such as India remaining considerably higher than in transitioned economies (15.0 vs 12.8 per 100,000, respectively).^[1]

In India, breast cancer has emerged as the most common malignancy affecting women, with an age-adjusted incidence rate of 25.8 per 100,000 women and a mortality rate of 12.7 per 100,000.^[2] The disease demonstrates a marked urban-rural divide, with age-adjusted rates as high as 41.0 per 100,000 in Delhi, 37.9 in Chennai, 34.4 in Bangalore, and 33.7 in Thiruvananthapuram District. Statistically significant increases in age-adjusted rates over the period 1982–2014 have been documented across all major population-based cancer registries, with annual percentage changes ranging from 1.42% in Mumbai to 2.84% in Bangalore.^[2] Mortality-to-incidence ratios reach 66% in rural registries compared with 8% in urban centres, a disparity that reflects the interplay of late presentation,

limited access to screening, and constrained availability of definitive oncological treatment in resource-limited settings.^[2,3]

A striking feature of the Indian breast cancer landscape is the younger age at diagnosis compared with Western populations. Approximately half of Indian breast cancer patients are diagnosed before the age of 50, in stark contrast to the median age of 62 years reported by SEER registry data from the United States.^[3] Late-stage presentation is common; in a large cross-sectional study of 569 Indian breast cancer patients, Saxena et al. reported that stage IIIB was the most frequent stage at diagnosis (35.2%), with lymph node positivity in 80.2% of evaluable cases and invasive ductal carcinoma comprising 88.2% of histological diagnoses.^[4] The consistent observation of locally advanced disease at diagnosis underscores the pressing need for accurate, minimally invasive biomarkers that could facilitate earlier detection, particularly in populations where population-based mammography screening is neither feasible nor cost-effective.^[3]

Prostate-specific antigen (PSA), also designated human kallikrein 3 (hK3, KLK3), is a 33-kDa glycoprotein serine protease of the kallikrein family, initially identified in seminal plasma and subsequently established as the single most useful serum marker for the detection and monitoring of prostate cancer in men. The name is, however, a historical misnomer. Diamandis and Yu, in a landmark review, catalogued non-prostatic sources of PSA and demonstrated that female breast tissue and its secretions constitute the second-richest reservoir of PSA in the human body, with nipple aspirate fluid containing up to 5000 µg/L and lactational milk up to 300 µg/L.^[5] The expression of PSA in female tissues is governed by steroid hormone receptors, with androgens, glucocorticoids, and progestins acting as up-regulators of PSA gene transcription, while oestrogens exert a suppressive influence on androgen-induced PSA production.^[5]

The first systematic investigation of serum PSA in women with breast cancer was reported by Gjai, Yu, Diamandis and colleagues, who evaluated serum PSA in women with and without breast cancer using a highly sensitive time-resolved immunofluorometric assay.^[6] The authors reported that in women aged 50 years or older, serum PSA levels did not differ significantly between healthy controls and breast cancer patients, but that most circulating PSA in women existed in its free (33-kDa) molecular form, in contrast to the predominantly α1-antichymotrypsin-complexed form observed in male serum.^[6] This observation set the stage for the hypothesis that molecular sub-forms of PSA, rather than absolute concentrations, might carry diagnostic value in breast disease.

The introduction of ultrasensitive PSA immunoassays with lower analytical detection limits (approximately 1 ng/L) transformed the ability to detect PSA in female biological samples. Ferguson, Yu, Diamandis and colleagues reported that ultrasensitive time-resolved immunofluorometric assay and third-generation chemiluminescent immunoassay platforms detected PSA immunoreactivity above 15 ng/g of total protein in 28% and 23%, respectively, of breast tumour cytosols from 315 patients, with significant associations between PSA positivity and progesterone and oestrogen receptor status ($p < 0.001$).^[7] These findings suggested that PSA expression in breast tumours is under hormonal regulation and might carry prognostic significance.

The clinical significance of tumour PSA expression was subsequently addressed by Foekens et al., who examined PSA levels in the cytosols of primary breast tumours from 1,516 patients with primary breast cancer and 434 patients with recurrent disease treated with tamoxifen.^[8] Low tumour PSA levels were more frequently observed in larger tumours, in tumours of older and post-menopausal patients, and in steroid hormone receptor-negative tumours. In multivariate analysis corrected for age, disease-free interval, site of relapse, and steroid hormone receptor status, low PSA emerged as an independent predictor of poor response to first-line tamoxifen and shorter progression-free and overall survival, positioning tumour PSA as a potential marker for the selection of women likely to benefit from systemic endocrine therapy.^[8]

Evidence from serum-based case-control studies has been notably inconsistent. Romppanen et al. used a highly sensitive chemiluminescent enzyme immunoassay to evaluate serum total PSA in a case-control cohort of 90 women with histologically confirmed breast cancer, 94 women with benign breast disease and 100 healthy controls, matched for age and long-term place of residence.^[9] Serum total PSA levels did not differ significantly between the three groups, and no relation was found between serum PSA and histological type, grade, or clinical stage. In benign breast disease, all patients with mastopathy had normal total PSA, whereas an elevation was noted in 7% of women with fibroadenoma. The authors concluded that serum total PSA alone could not discriminate healthy women from those with breast cancer or benign breast disease.^[9]

A different picture emerges when molecular sub-forms of PSA are considered. In a review of the diagnostic and prognostic utility of PSA in breast disease, Black and Diamandis emphasised that although the level of serum PSA is elevated in both benign and malignant breast disease, the molecular form of circulating PSA differs between women

with and without breast cancer.^[10] In a follow-up analytical study, Black et al. demonstrated that free PSA constituted the predominant molecular form of serum PSA in a significantly higher proportion of breast cancer patients (approximately five times the rate observed in healthy women or those with benign breast disease), and that PSA levels declined after surgical removal of the tumour. The diagnostic performance of the free-PSA-predominant pattern showed limited sensitivity (approximately 20%) but high specificity (approximately 96%) compared with women without breast cancer or with benign disease.^[11]

The literature from resource-limited settings has generally supported a diagnostic role for serum PSA. Mashkoo et al., in a case-control study from Iraq involving 55 women with breast cancer and 82 healthy female controls, found that both total and free PSA were significantly elevated pre-operatively in breast cancer patients compared with healthy women (both $p < 0.001$), and that both parameters declined significantly after surgical removal of the tumour ($p < 0.001$).^[12] Notably, 83.6% of pre-operative breast cancer patients had free PSA as the predominant molecular form, compared with 0% of controls and 1.8% of post-operative samples ($p < 0.001$), findings that reinforce the potential diagnostic and monitoring utility of the free PSA molecular fraction.^[12]

More recently, Hanamura et al. examined serum PSA in 144 Japanese women with breast cancer and 132 healthy controls using a chemiluminescent enzyme immunoassay.^[13] In the pooled cohort, PSA detection rates did not differ significantly (29.2% vs 25.8%, $p = 0.53$); however, when analysis was restricted to post-menopausal women, PSA was detected in 27.4% of breast cancer patients compared with 11.3% of healthy controls ($p = 0.009$). Within post-menopausal metastatic breast cancer, higher serum PSA values correlated weakly with serum testosterone and androgen receptor positivity, suggesting that PSA in this subgroup partially reflects tumour androgen receptor signalling.^[13]

Taken together, the published evidence indicates that (i) PSA is expressed at low concentrations in the serum of women, (ii) the diagnostic signal in breast cancer is more likely to reside in molecular sub-form patterns and qualitative detection than in absolute concentrations exceeding the conventional male reference threshold, and (iii) there is substantial heterogeneity across studies attributable to differences in assay platform, calibration, patient selection, hormonal milieu and ethnicity.^[5,9–13] Indian data on serum PSA in breast carcinoma remain sparse, and existing studies have been limited by modest sample sizes, variable assay methodology and inconsistent subgroup analyses. The present study was designed to address this evidence gap in an Indian tertiary care setting by evaluating serum PSA as a biomarker for breast carcinoma in a formally powered case-control cohort and by examining its relationship with clinical staging.

1.1 Aims and objectives

1.1.2 Primary objective:

To evaluate serum prostate-specific antigen (sPSA) levels as a biomarker for the screening and diagnosis of breast carcinoma in women presenting to a tertiary care centre in southern India.

1.2 Secondary objective:

To analyse the association of serum PSA with clinical staging (TNM), T stage, N stage and tumour burden (clinical lump size) among histopathologically confirmed cases of breast carcinoma.

2. Materials and methods

2.1 Study design and setting

This was a hospital-based observational case-control study conducted in the Department of General Surgery, JSS Medical College and Hospital, JSS Academy of Higher Education and Research, Mysuru, in collaboration with the Department of Biochemistry. The total study duration was 18 months, extending from April 2024 to September 2025. Ethical clearance was obtained from the Institutional Ethical Committee before commencement of the study, and written informed consent was obtained from every participant in a language they were comfortable with.

2.2 Sample size calculation

The sample size was computed using the standard formula for comparison of means between two independent groups: $N = (Z\alpha + Z\beta)^2 \times \sigma^2 / d^2$, where $Z\alpha$ (1.96) corresponded to a two-sided level of significance of 0.05, $Z\beta$ (0.84) corresponded to a study power of 80%, σ (0.273) represented the population standard deviation derived from

published data, and d (0.12) represented the clinically meaningful effect size to be detected. Substitution yielded a minimum of 40 participants per group, giving a total sample size of 80.

2.3 Sampling technique and eligibility

Simple random sampling was employed. All women presenting to the Departments of General Surgery and Surgical Oncology with a breast lump during the study period were screened against the eligibility criteria and enrolled consecutively until the required numbers were achieved. The inclusion criteria comprised women above 18 years of age with a newly diagnosed benign or malignant breast disease; those with malignant disease were assigned to the case group, and those with benign disease to the control group. Women without any breast disease, women below 18 years of age, women previously treated for breast disease, women with current or recent (within 3 months) oral contraceptive use, and male patients with breast cancer were excluded.

2.4 Clinical evaluation

A structured proforma was used to record demographic details, menstrual and obstetric history, family history of breast or ovarian malignancy, and history of hormonal or oral contraceptive use. A complete general and local examination was performed in every participant, documenting the side and quadrant of the lump, its size, consistency, mobility, associated skin changes, nipple-areolar complex abnormalities, and regional (axillary and supraclavicular) lymph node status. Body mass index was calculated using measured height and weight, with Asian-Pacific BMI thresholds applied for categorisation.

2.5 Imaging and histopathological confirmation

All enrolled women underwent bilateral breast and axillary ultrasonography, which was used to characterise the lesion in terms of size, shape, margins, internal echotexture, posterior acoustic features, vascularity and axillary lymphadenopathy, with lesions categorised according to the Breast Imaging Reporting and Data System (BI-RADS). Mammography was performed in women aged 40 years and above, and in selected younger women with clinically suspicious lesions, and reported using the same BI-RADS classification. Fine needle aspiration cytology and/or core needle biopsy were performed in every participant to obtain a definitive histopathological diagnosis. Women with confirmed malignancy were clinically staged according to the prevailing American Joint Committee on Cancer (AJCC) TNM classification.

2.6 Serum PSA estimation

A venous blood sample of approximately 5 mL was drawn from every participant under aseptic precautions and collected in a plain vacutainer. After allowing the sample to clot at room temperature, it was centrifuged and the separated serum was transported to the Department of Biochemistry, where serum PSA was estimated using a standardised chemiluminescent immunoassay platform following the manufacturer's protocol. The reference range applied was 0–1.4 ng/mL, and the assay's lower limit of detection allowed reporting of ultra-low female serum values. All measurements were performed in a single laboratory to minimise inter-assay variability.

2.7 Statistical analysis

Data were entered into a Microsoft Excel spreadsheet and analysed using the Statistical Package for the Social Sciences (SPSS) version 28. Continuous variables were expressed as mean $\pm$ standard deviation (SD) and compared between groups using the independent-samples t-test. Categorical variables were expressed as frequencies and percentages and compared using the Chi-square test, with Fisher's exact test applied when expected cell counts were below five. Comparison of serum PSA values across ordered categories of clinical stage, T stage and N stage was performed using one-way ANOVA, and the relationship of serum PSA with continuous tumour size was assessed using Pearson correlation. The diagnostic performance of serum PSA detection was expressed in terms of sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV) and overall diagnostic accuracy. A two-sided p-value of less than 0.05 was considered statistically significant for all analyses.

3. Results

3.1 Demographic and reproductive characteristics

A total of 80 women were enrolled: 40 cases with histopathologically confirmed malignant breast disease and 40

controls with newly diagnosed benign breast disease. All 80 participants were female, consistent with the protocol-defined exclusion of male breast cancer. The mean age of cases (51.58 ± 9.97 years) was significantly higher than that of controls (25.25 ± 5.45 years; $t=14.656$, $p<0.001$). Cases were almost entirely aged above 40 years, with 75.0% clustering in the 41–60 year age band, whereas 85.0% of controls were aged 30 years or below ($p<0.001$). Body mass index did not differ significantly between the two groups (24.00 ± 1.39 vs 23.94 ± 1.34 kg/m², $p=0.845$), and the majority of participants in both groups fell within the overweight Asian-Pacific category. Cases were significantly more likely to be post-menopausal (52.5% vs 0.0%, $p<0.001$) and multiparous (60.0% vs 15.0%, $p<0.001$), while mean age at menarche (13.28 ± 1.18 vs 13.75 ± 1.28 years, $p=0.087$) and mean duration of breastfeeding among parous women (16.05 ± 8.55 vs 15.13 ± 7.86 months, $p=0.677$) were comparable. A positive family history of breast cancer was more frequent among cases (25.0%) than controls (7.5%), although this trend did not reach statistical significance ($p=0.066$). The full demographic and reproductive profile is summarised in Table 1.

Table 1. Demographic and reproductive characteristics of cases and controls.

Parameter	Cases (n=40)	Controls (n=40)	p-value
Age, mean $\pm$ SD (years)	51.58 ± 9.97	25.25 ± 5.45	$<0.001^*$
Age, median (years)	51	24	—
Age range (years)	32–73	18–37	—
≤ 30 years, n (%)	0 (0.0)	34 (85.0)	$<0.001^*$
31–40 years, n (%)	5 (12.5)	6 (15.0)	—
41–50 years, n (%)	15 (37.5)	0 (0.0)	—
51–60 years, n (%)	15 (37.5)	0 (0.0)	—
>60 years, n (%)	5 (12.5)	0 (0.0)	—
Post-menopausal, n (%)	21 (52.5)	0 (0.0)	$<0.001^*$
Multiparous, n (%)	24 (60.0)	6 (15.0)	$<0.001^*$
Positive family history, n (%)	10 (25.0)	3 (7.5)	0.066

3.2 Clinical and imaging findings

Cases presented with significantly larger lumps than controls (4.33 ± 2.18 vs 1.76 ± 1.04 cm, $p<0.001$) and with characteristics indicative of malignancy: 75.0% of case lumps were hard in consistency (vs 7.5% of control lumps, $p<0.001$), and 65.0% exhibited restricted or fixed mobility (vs 17.5%, $p<0.001$). Skin changes were confined to cases, with dimpling in 45.0% and peau d'orange in 30.0%, while ulceration was not documented in any participant. Palpable axillary lymphadenopathy was present in 90.0% of cases (predominantly fixed or mobile nodes), compared with 32.5% of controls in whom only small discrete axillary nodes were noted ($p<0.001$). Nipple discharge was more frequent among cases (20.0% vs 5.0%) but did not reach statistical significance ($p=0.091$). The upper outer quadrant was the most frequent site of involvement in both groups, with no significant difference in quadrant distribution ($p=0.444$). Ultrasonographic lesion size mirrored clinical size (4.39 ± 2.28 vs 1.76 ± 1.06 cm, $p<0.001$), and BI-RADS 4 or 5 categorisation was recorded in 75.0% of cases compared with 7.5% of controls ($p<0.001$). Mammography was performed in 97.5% of cases, with BI-RADS 4 in 35.0%, BI-RADS 5 in 30.0% and BI-RADS 3 in 32.5%; controls did not undergo mammography in keeping with their younger age. Clinical and imaging comparisons are summarised in Table 2.

Table 2. Clinical examination and imaging findings among cases and controls.

Parameter	Cases (n=40)	Controls (n=40)	p-value
Mean lump size, cm ($\pm$ SD)	4.33 ± 2.18	1.76 ± 1.04	$<0.001^*$
Hard consistency, n (%)	30 (75.0)	3 (7.5)	$<0.001^*$
Restricted/fixed mobility, n (%)	26 (65.0)	7 (17.5)	$<0.001^*$
Skin dimpling, n (%)	18 (45.0)	0 (0.0)	$<0.001^*$
Peau d'orange, n (%)	12 (30.0)	0 (0.0)	$<0.001^*$
Nipple discharge, n (%)	8 (20.0)	2 (5.0)	0.091
Palpable axillary nodes, n (%)	36 (90.0)	13 (32.5)	$<0.001^*$
USG lesion size, cm ($\pm$ SD)	4.39 ± 2.28	1.76 ± 1.06	$<0.001^*$
BI-RADS 4/5 on USG, n (%)	30 (75.0)	3 (7.5)	$<0.001^*$

3.3 Histopathology, staging and grade

Among cases, invasive ductal carcinoma dominated the histological spectrum, accounting for 85.0% (n=34) of malignancies, followed by invasive lobular carcinoma in 12.5% (n=5) and other malignant subtypes in 2.5% (n=1). Among controls, fibroadenoma constituted 70.0% (n=28) and breast abscess 30.0% (n=12) of benign diagnoses. Clinical stage distribution among cases revealed Stage III as the most frequent stage at diagnosis (42.5%, n=17), followed by Stage II (30.0%, n=12), Stage I (20.0%, n=8) and Stage IV (7.5%, n=3). T2 was the commonest tumour size category (35.0%), N1 the predominant nodal status (35.0%), and distant metastasis (M1) was documented in 7.5% of cases. Histological grading using the Modified Scarff-Bloom-Richardson system showed Grade II tumours in 50.0%, Grade III in 30.0% and Grade I in 20.0% of cases, and lymphovascular invasion was identified in 42.5% of cases. The distribution of histopathological, staging and prognostic parameters is presented in Table 3.

Table 3. Histopathological diagnosis, clinical staging and prognostic parameters.

Diagnosis	Frequency (n)	Percentage (%)
Cases (n=40)		
Invasive ductal carcinoma	34	85.0
Invasive lobular carcinoma	5	12.5
Other malignant	1	2.5
Controls (n=40)		
Fibroadenoma	28	70.0
Breast abscess	12	30.0
Clinical stage among cases		
Stage I	8	20.0
Stage II	12	30.0
Stage III	17	42.5
Stage IV	3	7.5
Histological grade (Modified SBR)		
Grade I	8	20.0
Grade II	20	50.0
Grade III	12	30.0
Lymphovascular invasion present	17	42.5

3.4 Serum PSA — descriptive analysis and detection

Serum PSA values in both cases and controls remained well within the conventional reference range of 0–1.4 ng/mL, with no participant in either group exhibiting an 'elevated' value by conventional male clinical criteria. Mean serum PSA in cases was 0.0054 ± 0.0016 ng/mL (range 0.003–0.008 ng/mL, median 0.005 ng/mL), and in controls 0.0025 ± 0.0024 ng/mL (range not detected to 0.006 ng/mL, median 0.003 ng/mL). Comparison of raw values by independent t-test yielded $t=6.262$, $p<0.001$, though the absolute magnitudes remained orders of magnitude below the diagnostic threshold. Serum PSA was detectable in all 40 cases (100.0%) and in 22 of 40 controls (55.0%), with the remaining 18 controls (45.0%) demonstrating undetectable PSA. This difference in detection rate was highly statistically significant ($\chi^2 = 20.717$, $df = 1$, $p < 0.001$; Fisher's exact $p < 0.001$). The descriptive comparison and detection frequencies are summarised in Table 4.

Table 4. Serum PSA levels and detection frequency among cases and controls.

Parameter	Cases (n=40)	Controls (n=40)	p-value
sPSA, mean $\pm$ SD (ng/mL)	0.0054 ± 0.0016	0.0025 ± 0.0024	$<0.001^*$
sPSA, median (ng/mL)	0.005	0.003	—
sPSA range (ng/mL)	0.003–0.008	ND–0.006	—
sPSA detected, n (%)	40 (100.0)	22 (55.0)	$<0.001^{*\dagger}$
sPSA undetectable, n (%)	0 (0.0)	18 (45.0)	—
Above 1.4 ng/mL threshold, n (%)	0 (0.0)	0 (0.0)	—

3.5 Diagnostic performance of serum PSA

Treating detectability of serum PSA (rather than an absolute concentration cut-off) as the diagnostic criterion produced a 2x2 contingency of 40 true positives, 0 false negatives, 22 false positives and 18 true negatives. The corresponding diagnostic parameters were sensitivity 100.0%, specificity 45.0%, positive predictive value 64.5%, negative predictive value 100.0% and overall diagnostic accuracy 72.5% (Table 5). The absence of any false negative case indicates that undetectable serum PSA effectively excluded breast carcinoma in this cohort, giving the test a strong rule-out property. However, the specificity of 45.0% and the moderate positive predictive value reflect that just over half of women with benign breast disease also exhibited detectable serum PSA, limiting its role as a confirmatory test when used in isolation.

Table 5. Diagnostic performance of serum PSA detection for breast carcinoma.

Diagnostic parameter	Value	Interpretation
True positives (TP)	40	Cases with detectable sPSA
False negatives (FN)	0	Cases with undetectable sPSA
False positives (FP)	22	Controls with detectable sPSA
True negatives (TN)	18	Controls with undetectable sPSA
Sensitivity	100.0%	All cases detected
Specificity	45.0%	Limited discrimination
Positive predictive value (PPV)	64.5%	—
Negative predictive value (NPV)	100.0%	Strong rule-out value
Diagnostic accuracy	72.5%	—

3.6 Association of serum PSA with clinical stage and tumour burden

Mean serum PSA values did not differ significantly across the four clinical stages (Stage I: 0.0051 ± 0.0010 ; Stage II: 0.0058 ± 0.0014 ; Stage III: 0.0051 ± 0.0017 ; Stage IV: 0.0060 ± 0.0026 ng/mL; one-way ANOVA $p=0.576$). Similarly, no significant difference was observed across T stage (T1–T4 means ranging from 0.0046 to 0.0062 ng/mL; $p=0.174$) or N stage (N0–N3 means ranging from 0.0051 to 0.0056 ng/mL; $p=0.841$). Pearson correlation between serum PSA and clinical lump size in the case cohort was weak and non-significant ($r = -0.128$, $p = 0.432$). Serum PSA therefore did not track systematically with the extent of disease and could not be used as a surrogate for tumour burden or disease progression in this cohort (Table 6). Definitive surgical management was undertaken in 55.0% of cases (modified radical mastectomy 30.0%, breast conserving surgery 25.0%) and preceded by neoadjuvant chemotherapy in 45.0%, reflecting the substantial burden of locally advanced disease at presentation.

Table 6. Association of serum PSA with clinical stage, T stage, N stage and lump size among cases (n=40).

Variable	n	Mean sPSA $\pm$ SD (ng/mL)	p-value
Clinical stage I	8	0.0051 ± 0.0010	0.576
Clinical stage II	12	0.0058 ± 0.0014	—
Clinical stage III	17	0.0051 ± 0.0017	—
Clinical stage IV	3	0.0060 ± 0.0026	—
T1	10	0.0051 ± 0.0010	0.174
T2	14	0.0057 ± 0.0016	—
T3	10	0.0046 ± 0.0018	—
T4	6	0.0062 ± 0.0015	—
N0	11	0.0056 ± 0.0014	0.841
N1	14	0.0051 ± 0.0013	—
N2	11	0.0055 ± 0.0019	—
N3	4	0.0053 ± 0.0022	—
Pearson r (sPSA vs lump size)	40	$r = -0.128$	0.432

4. Discussion

The present case-control study evaluated serum PSA as a candidate biomarker in an Indian tertiary care cohort of 40 women with breast carcinoma and 40 women with benign breast disease. Three central observations emerged: (i) serum PSA was detectable in every case of breast carcinoma but in only 55.0% of controls, a highly significant qualitative difference ($p < 0.001$); (ii) absolute serum PSA concentrations in both groups remained well below the conventional reference threshold of 1.4 ng/mL and were not clinically 'elevated' by any standard male-derived criterion; and (iii) serum PSA levels did not correlate with clinical stage, T stage, N stage or tumour burden. These findings sit within a heterogeneous and often contradictory body of international literature, appraised in the sections that follow.

The demographic contrast between cases (mean age 51.58 ± 9.97 years, 52.5% post-menopausal) and controls (mean age 25.25 ± 5.45 years, 100.0% pre-menopausal) reflects a well-recognised epidemiological pattern in the Indian population, in which fibroadenoma dominates in young women and breast carcinoma clusters in peri- and post-menopausal women. Malvia et al. reported an age-adjusted incidence of 25.8 per 100,000 among Indian women, with a mean age at diagnosis approximately a decade earlier than that observed in Western populations.^[2] Saxena and colleagues, in a large cross-sectional analysis of 569 Indian breast cancer patients from New Delhi, reported a mean age at presentation of 47.8 years, with a range of 13–82 years and the highest concentration of cases in the fifth decade,^[4] broadly paralleling the age distribution observed in our case cohort. The younger age at diagnosis in the Indian context, together with the absence of population-based mammographic screening, provides the epidemiological rationale for the search for minimally invasive serum biomarkers that could complement clinical and imaging assessment.^[3]

The clinical presentation of our cases mirrored the pattern of late-stage disease described repeatedly in Indian series. The mean clinical lump size was 4.33 ± 2.18 cm, 75.0% of case lumps were hard in consistency, 65.0% demonstrated restricted or fixed mobility, and 90.0% had palpable axillary lymphadenopathy. Stage III disease accounted for 42.5% of cases in our cohort, closely paralleling the stage IIIB predominance (35.2%) reported by Saxena et al.,^[4] and consistent with the broader consensus that late presentation, rather than biological aggressiveness alone, is the principal driver of the elevated mortality-to-incidence ratios reported in Indian breast cancer.^[2,3] McDonald and colleagues emphasised that the prevalence of late-stage disease and high mortality rates in India are largely explained by the reliance on clinical and self-breast examination for detection, rather than mammography, and that the absence of population-based screening remains a major unresolved public-health challenge.^[3]

Histopathologically, invasive ductal carcinoma dominated our case series (85.0%), followed by invasive lobular carcinoma (12.5%). These proportions correspond closely with the 88.2% invasive ductal and 3.7% invasive lobular pattern reported by Saxena et al. among 569 Indian breast cancer patients,^[4] confirming that the histological distribution of our cohort is representative of the wider Indian population. Among controls, fibroadenoma was the predominant pathology (70.0%), followed by breast abscess (30.0%), a pattern consistent with the disease spectrum expected in women under 30 years of age.

The central finding of a differential detection rate of serum PSA (100.0% in cases vs 55.0% in controls; $\chi^2 = 20.717$, $p < 0.001$) with absolute values remaining well below the male-derived reference threshold requires careful interpretation. Diamandis and Yu, in their comprehensive review of non-prostatic sources of PSA, established that PSA is expressed in a range of female hormonally regulated tissues, particularly the breast, and that its production in these tissues is regulated by androgens, progestins and glucocorticoids acting through their respective nuclear receptors, with oestrogens exerting a suppressive influence on androgen-induced expression.^[5] The biological plausibility of a detection-based signal in breast carcinoma therefore rests upon differences in the hormonal milieu, receptor expression and ductal architecture of breast tumours relative to healthy or benign breast tissue.

Historical serum-based case-control studies have been mixed in their conclusions. Gai, Yu, Diamandis and colleagues, in the first substantive serum PSA study in breast cancer, reported that in women aged 50 years or above, serum PSA concentrations did not differ significantly between breast cancer patients and healthy controls, and that circulating PSA in women existed predominantly in its free molecular form.^[6] The authors nevertheless noted that tumour PSA (as opposed to serum PSA) had prognostic implications, an observation confirmed and extended by Ferguson, Yu, Diamandis and colleagues, who found PSA immunoreactivity above 15 ng/g of total protein in 28% of breast tumour cytosols from 315 patients using ultrasensitive time-resolved immunofluorometric assay, with significant associations between tumour PSA positivity and oestrogen and progesterone receptor status ($p < 0.001$).^[7] Foekens and colleagues, in the largest series to date, examined tumour PSA in 1,516 patients with primary breast

cancer and 434 patients with recurrent disease treated with tamoxifen.^[8] Low tumour PSA levels were more frequently observed in larger tumours, in older and post-menopausal patients, and in steroid-receptor-negative tumours. In multivariate models, low PSA emerged as an independent predictor of poor response to first-line tamoxifen and shorter progression-free and overall survival. Although this study focused on tumour PSA rather than serum PSA, it provides mechanistic support for the concept that PSA expression in breast tumours carries clinical information related to hormone-receptor biology and endocrine responsiveness. Our finding that serum PSA was universally detectable in cases, without correlating with clinical stage or tumour burden, is consistent with the Foekens observation that PSA expression relates more closely to receptor biology than to gross anatomical extent of disease.^[8]

A directly contrasting result was reported by Romppanen et al., who used a highly sensitive chemiluminescent enzyme immunoassay to compare serum total PSA in 90 women with breast cancer, 94 women with benign breast disease and 100 healthy controls matched for age and long-term place of residence.^[9] Serum total PSA did not differ significantly between the three groups, and no relation was found with histological type, grade or clinical stage; elevation of serum PSA was observed in only 7% of fibroadenoma patients. The authors concluded that serum total PSA alone could not discriminate healthy women from those with breast cancer or benign breast disease. The apparent discrepancy between this negative result and our positive detection-based finding likely reflects the different analytical endpoints used: Romppanen and colleagues assessed whether absolute serum PSA concentrations differed, whereas our study assessed whether PSA was detectable at all. As Black and Diamandis emphasised in their systematic review of the field, the diagnostic utility of serum PSA in women is more likely to reside in molecular form and qualitative detection than in quantitative elevation above the male reference range.^[10] The molecular-form hypothesis is directly supported by Black et al., who showed that free PSA constituted the predominant molecular form of serum PSA in a significantly higher proportion of breast cancer patients than in healthy women or those with benign breast disease (approximately five-fold higher), and that this pattern showed high specificity (approximately 96%) despite limited sensitivity (approximately 20%).^[11] Serum PSA also declined after surgical removal of the tumour, hinting at a tumour-related contribution to the circulating pool. Mashkoo et al., in an Iraqi case-control study of 55 women with breast cancer and 82 healthy controls, similarly reported that both total and free serum PSA were significantly higher pre-operatively in breast cancer patients than in controls ($p<0.001$), that both declined significantly after surgery ($p<0.001$), and that free PSA was the predominant molecular form in 83.6% of pre-operative breast cancer patients compared with 0% of controls ($p<0.001$).^[12] Our study did not distinguish between free and complexed forms of serum PSA, and this is a methodological limitation; nevertheless, the qualitative detection pattern we observed is consistent with the broader picture that emerges when molecular-form data are considered.

The most recent and methodologically robust serum PSA study is that of Hanamura et al., who examined 144 Japanese women with breast cancer and 132 healthy controls using a chemiluminescent enzyme immunoassay.^[13] In the whole cohort, PSA detection rates did not differ significantly (29.2% vs 25.8%, $p=0.53$); however, in the post-menopausal subgroup, PSA was detected in 27.4% of breast cancer patients compared with 11.3% of healthy controls ($p=0.009$). Higher serum PSA values in post-menopausal metastatic disease correlated weakly with serum testosterone and androgen receptor positivity, consistent with the notion that PSA in this subgroup reflects tumour androgen receptor signalling. The Hanamura result reinforces two points relevant to our study: first, that menopausal status is a critical stratifying variable in the serum PSA-breast cancer relationship; and second, that the analytical sensitivity of the assay and the definition of 'detectable' are decisive determinants of the apparent diagnostic performance.^[13] Our observation that 52.5% of cases were post-menopausal, and that serum PSA was detectable in every case, is broadly consistent with this literature when the higher analytical sensitivity of our assay platform is taken into account.

The diagnostic performance of serum PSA detection in our cohort — sensitivity 100.0%, specificity 45.0%, PPV 64.5%, NPV 100.0%, accuracy 72.5% — is characteristic of a marker with strong rule-out but limited rule-in capability. The very high sensitivity and NPV imply that absence of detectable serum PSA effectively excluded breast carcinoma in the population studied, whereas the moderate specificity reflects that just over half of women with benign breast disease also had detectable serum PSA. This profile is qualitatively similar to the sensitivity–specificity trade-off reported by Black et al. for free-PSA-predominant patterns (20% sensitivity, 96% specificity),^[11] with the balance reversed because our study used simple detectability as the discriminator rather than a molecular-form or absolute-concentration cut-off. The choice of analytical threshold therefore has decisive effects on the apparent performance of the test and must be reported explicitly in any future work in this field.

The absence of any statistically significant association between serum PSA and clinical stage ($p=0.576$), T stage ($p=0.174$), N stage ($p=0.841$) or tumour size ($r=-0.128$, $p=0.432$) in our cohort is consistent with the report by Hanamura and colleagues, who found no significant correlation between PSA levels and tumour size, nodal status or pathological stage in the overall cohort, although weak associations emerged in post-menopausal metastatic subgroups.^[13] The likely explanation is that when serum PSA values remain close to the assay's lower limit of detection, stochastic measurement variability dominates any underlying biological signal of tumour burden, rendering stage-wise differences statistically undetectable. This has an important practical implication: serum PSA in our cohort should not be interpreted as a marker of disease extent or of therapeutic response.

Strengths and limitations

The principal strengths of this study include the use of a formally computed sample size, prospective enrolment, uniform application of a single ultrasensitive chemiluminescent immunoassay, and blinded serum PSA estimation performed independently of clinical and radiological assessment. The main limitations warrant explicit acknowledgment. First, the modest sample size of 40 cases and 40 controls, although sufficient for the primary comparison, limits statistical power to detect subtle gradients across molecular subtypes or hormone receptor status. Second, the cases and controls differed substantially in age and menopausal status; although this reflects the underlying epidemiological reality of the two conditions, it means that age-related and menopause-related determinants of serum PSA could not be fully disentangled from disease-related effects. Third, serum PSA was measured only in its total form, without differentiation of free and complexed molecular sub-forms, which precludes direct comparison with the free-PSA molecular-form literature. Fourth, hormone receptor and androgen receptor status were not systematically recorded, restricting exploration of the receptor-linked mechanistic pathway. Fifth, the cross-sectional design precludes evaluation of serum PSA as a marker of treatment response or recurrence surveillance, which remains an important unexplored frontier.

Implications and future directions

The present findings support cautious optimism regarding the ancillary diagnostic value of serum PSA in breast carcinoma while advising against uncritical adoption of the marker as a quantitative diagnostic test. Future work should employ larger, multicentre, prospectively enrolled case-control cohorts with standardised ultrasensitive assay platforms and WHO-calibrated reference materials, incorporate paired measurement of free and complexed PSA, and stratify analyses by menopausal status, hormone receptor status, androgen receptor expression and molecular subtype. Longitudinal serial measurements across the treatment continuum, and integration of serum PSA into multivariable diagnostic algorithms alongside CA 15-3, CEA and clinical-imaging parameters, may allow the field to move from single-marker performance metrics towards clinically useful composite risk models.

5. Conclusion

This hospital-based case-control study of 80 Indian women demonstrated that although absolute serum PSA concentrations in both breast cancer patients and women with benign breast disease remained well below the conventional reference threshold of 1.4 ng/mL, the qualitative detectability of serum PSA showed a highly significant difference between the two groups (100.0% vs 55.0%, $\chi^2=20.717$, $p<0.001$). The corresponding diagnostic performance was characterised by 100.0% sensitivity, 45.0% specificity, 64.5% positive predictive value, 100.0% negative predictive value and 72.5% overall accuracy. Serum PSA showed no significant association with clinical stage, T stage, N stage or tumour burden, indicating that it cannot serve as a marker of disease extent or progression. Taken together, these findings suggest that serum PSA, when assessed on a detection basis using an ultrasensitive chemiluminescent immunoassay, may serve as a useful minimally invasive rule-out biomarker for breast carcinoma but lacks the discriminative capacity required of a confirmatory test. Larger multicentre studies with standardised ultrasensitive assays, molecular sub-form differentiation, and stratification by hormone and androgen receptor status are required before serum PSA can be incorporated into routine diagnostic algorithms for breast carcinoma.

Declarations

Ethical approval and consent to participate: The study was approved by the Institutional Ethics Committee of JSS Medical College and Hospital, Mysuru. Written informed consent was obtained from every participant.

Consent for publication: Not applicable.

Availability of data and materials: The datasets analysed during the current study are available from the corresponding author on reasonable request.

Competing interests: The authors declare that they have no competing interests.

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Authors' contributions: Both authors contributed to the conception and design of the study, acquisition and analysis of data, drafting and critical revision of the manuscript, and approval of the final version submitted for publication.

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