



Original Research

Do elastography values correlate with molecular subtype of breast cancer or is morphology the key player?

Rupa Renganathan¹, Prema Subramaniam¹, Vinita Thakur², Divya Vishwanatha Kini¹, P. Suganya¹, Adrija Mandal¹

¹Division of Breast and Women's Imaging and Interventions, Department of Diagnostic and Interventional Radiology, Kovai Medical Center and Hospital, Coimbatore, Tamil Nadu, ²Department of Radiology, Aastha Super Speciality Hospital, Opposite Bharat Petrol Pump, Siliguri, West Bengal, India

***Corresponding author:**

Rupa Renganathan,
Lead Consultant, Division
of Breast and Women's
Imaging and Interventions,
Department of Diagnostic and
Interventional Radiology, Kovai
Medical Center and Hospital,
Coimbatore, Tamil Nadu, India.

drupa@kmchhospitals.com

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ABSTRACT

Objectives: We evaluated the association between shear wave elastography (SWE) parameters and the molecular subtypes and other prognostic factors of breast cancer. We also examined the influence of morphological characteristics and other prognostic factors of breast cancers on elastography values.

Materials and Methods: This is a retrospective observational study on 148 patients with biopsy-proven 152 breast cancers who underwent both B-mode ultrasonography and SWE. The analysis involved calculating the correlation between the five quantitative parameters on SWE (mean, minimum, maximum, standard deviation, and ratio) and the molecular subtyping and other prognostic factors of breast cancer. We analyzed the influence of morphology and other prognostic factors on SWE parameters.

Results: The mean, minimum, and maximum values in SWE were significantly lower ($P < 0.05$) in cases where the lesion displayed non-mass abnormality, circumscribed margins, posterior acoustic enhancement, and non-ductal histopathology. Hormone positive tumors frequently exhibited spiculated margins and higher ($P < 0.05$) elastography values, although within this category, circumscribed masses demonstrated lower elastography values compared to the remaining cases. Triple negative breast cancers often displayed circumscribed margins and lower ($P < 0.05$) elastography values; however, among this subgroup, masses with spiculated margins exhibited higher values compared to the rest.

Conclusion: Our findings indicate that elastography independently do not correlate with molecular subtypes in breast cancer. Morphological features and histopathological subtypes appear to have strong influence on elastography values.

Keywords: Shear wave elastography, Morphology, Molecular subtyping

INTRODUCTION

Breast cancer exhibits heterogeneity in clinical, imaging, and prognostic factors.^[1] Through gene expression profiling, the understanding of breast cancer heterogeneity has been significantly advanced, and classified into four molecular subtypes, namely Luminal A, Luminal B, HER2 enriched, and Basal-like Triple Negative Breast Cancers (TNBC). These subtypes differ significantly in terms of incidence, clinical and imaging characteristics, treatment response, disease progression, and survival rates.^[2] The determination of molecular subtypes is based on percutaneous biopsy samples and relies on the presence or absence of estrogen receptor (ER), progesterone receptor (PR), HER2 receptor, and Ki-67 levels.^[3] Molecular subtyping not only

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enhances our understanding of breast cancer characteristics but also plays a crucial role in the treatment decision-making and the development of targeted therapies. Luminal A cancers demonstrate the most favorable prognosis among all subgroups, whereas basal-like TNBC represent the most aggressive subtype with a high likelihood of metastasis and recurrence.

There have been numerous studies conducted on the prediction of molecular subtypes of breast cancer using various imaging modalities and techniques. Previous research has firmly established that tumors that are positive for ERs typically exhibit spiculated margins and posterior shadowing.^[4-7] Conversely, TNBC often display circumscribed margins accompanied by posterior acoustic enhancement.^[4-9] Additionally, HER2 positive cancers tend to present with microcalcifications and multifocal involvement.^[5,6,8]

Over the past decade, ultrasound elastography has emerged as a significant addition to the repertoire of sonographic techniques due to its ability to assess tissue stiffness *in vivo*. The literature contains a limited number of studies that assess the utility of SWE in predicting the molecular subtypes of breast cancer. The findings of these studies exhibit considerable variation.^[10-17] Some studies demonstrate no correlation between elastography values and molecular subtypes of breast cancer,^[10,12,14,15] while others indicate an association between molecular subtypes and elasticity values, albeit with conflicting outcomes.^[11,13,16,17]

This study aims to analyze if elastography values correlate with molecular subtypes of breast cancer and to observe further the influence of morphology and other prognostic factors on SWE parameters in different molecular subtypes of breast cancer.

MATERIALS AND METHODS

Study design

This is a retrospective observational study that has been approved by the institutional ethical committee (EC/AP/732/07/2019) and informed consent was waived. All consecutive patients with biopsy-proven breast cancer from July 2019 to June 2020, who underwent both grayscale and SWE before the biopsy, were included in the study. As an institutional protocol, in our institute, SWE is performed for all the solid breast masses. Search from our hospital Picture Archiving and Communication System (PACS) database revealed that 194 patients with 198 breast cancers had undergone both grayscale and SWE. From this cohort, 46 lesions were excluded because of technical limitations in elastography images or measurements (5/46), mass >5 cm in size (22/46), or unavailability of pathology/immunohistochemistry (IHC) data (19/46) as shown in

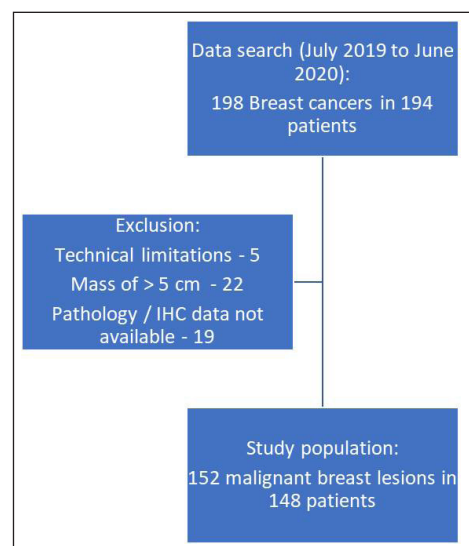


Figure 1: Patient flow in our study. IHC: Immunohistochemistry.

Figure 1. The final study population consisted of 148 patients with 152 breast cancers.

The patients underwent ultrasound and SWE using SuperSonic® Aixplorer ultrasound device (SuperSonic Imagine, Aix-en-Provence, France) equipped with a linear array transducer (4-15 MHz). The ultrasound and SWE were performed by one of two breast imaging specialists who possessed a minimum of five years' experience in this field and who follow World Federation of Ultrasound in Medicine and Biology (WFUMB) on optimal performance of SWE.^[18] Bilateral Digital Breast Tomosynthesis (DBT) and Full Field Digital Mammography (FFDM) were also performed on all patients using the Mammomat Inspiration system (Siemens Healthcare, Erlangen, Germany). Before the evaluation of the images for various criteria, such as correct placement of ROI, Q box, and ROI size of 1-3 mm, documentation of all B-mode features on ultrasound was assessed using images from the PACS.

Ultrasound data

The B-mode parameters, as delineated by the ACR BIRADS (fifth edition) Lexicon, were recorded.^[19] These parameters encompass size, shape, margins, posterior features, and the presence or absence of calcifications. Traditionally, in SWE the hardest area is coded as red and the softest area is coded as blue.^[18] The initial region of interest (ROI) was positioned in the most rigid section of the lesion identifiable by its red color. Conversely, the second ROI was placed in the adjacent normal parenchyma, which appeared as blue, representing the softest portion within the field of view (FOV). The quantitative aspects of SWE, including mean, maximum, minimum, standard deviation (SD), and ratio, were documented.

Pathological data

In addition to the stage of the tumor, molecular subtyping, and proliferation index (Ki-67), several pathological prognostic factors were observed. These factors comprised pathological type, grade, lymphovascular invasion, and the presence of lymph nodal involvement. The tumors were further categorized into four subtypes: Luminal A (ER+, PR+, HER2-, Ki-67 < 14%), Luminal B (ER+, PR+, HER2-, Ki-67 > 14%), Luminal B-like or triple positive (ER+, PR+, HER2+, Ki-67 any value), and basal-like triple negative (ER-, PR-, HER2-, Ki-67 any value). In this study, a cut-off value of 20% for Ki-67 was assigned to differentiate between low and high values for statistical analysis with reference to a prior study.^[20]

Statistical analysis

The calculation was performed to determine the correlation between the five quantitative parameters on SWE and various factors such as molecular subtyping, tumor grade, lymphovascular invasion, lymph node metastasis, and Ki-67, which are known to have prognostic implications. The correlation between these quantitative parameters and specific ultrasound features obtained through B-mode ultrasonography (USG), including tumor margins, posterior features, the presence of calcifications, and the presence of mass/non-mass abnormalities, was also calculated. Analysis of Variance (ANOVA) was used when there were more than two groups, while an independent sample test was employed in cases with two groups.

To determine the correlation between B-mode features (i.e., margins and posterior features) and SWE parameters, specifically within the hormone positive and TNBC subtypes, an independent sample test was conducted.

In the HER2 positive subgroup, the correlation between the presence or absence of hormone positivity and the presence of calcifications as well as SWE parameters was calculated using an independent samples test.

RESULTS

We conducted an analysis on 152 malignant breast lesions in 148 patients. The baseline characteristics of the study population are described in Table 1. The most prevalent histopathological type was invasive ductal carcinoma, not otherwise specified (NOS) 84.2% (128/152), and the most common molecular subtype was luminal B type 43.4% (66/152).

SWE based on histopathological subtypes

Invasive ductal carcinomas, NOS revealed significantly higher mean, minimum, and maximum values on SWE (240, 189,

Table 1: Baseline characteristics of the study population.

Parameter	Value n (%)
Age	
Mean	54.8 years
Range	30-84 years
Morphological abnormality in USG	
Non-mass abnormality	11 (7.2%)
Mass	141 (92.8%)
Mass margins	
Circumscribed	22 (15.6%)
Microlobulated	40 (28.36%)
Indistinct	26 (18.43%)
Angular	13 (9.92%)
Spiculated	40 (28.36%)
Posterior features	
Shadowing	39 (25.7%)
No features	82 (53.9%)
Enhancement	31 (20.4%)
Calcification	
Absent	103 (67.8%)
Present	49 (32.2%)
Histopathology	
DCIS with microinvasion	5 (3.3%)
Invasive ductal carcinoma, Not otherwise Specified	128 (84.2%)
Invasive lobular carcinoma	8 (5.3%)
Medullary carcinoma	4 (2.6%)
Papillary carcinoma	7 (4.6%)
Molecular subtypes	
Luminal A	12 (7.9%)
Luminal B	66 (43.4%)
Luminal B-like	19 (12.5%)
HER2 enriched	25(16.4%)
Triple negative	30 (19.7%)
Grade	
I	16 (10.5%)
II	70 (46.1%)
III	66 (43.4%)
Lymphovascular invasion	
Absent	87 (57.2%)
Present	65 (42.8%)
Lymph-node metastasis	
Absent	64 (42.1%)
Present	88 (57.9%)

DCIS: Ductal Carcinoma In Situ.

and 268 kPa, respectively) compared to other histological subtypes (187, 153, and 215 kPa, respectively) with a P value of < 0.05.

SWE based on other prognostic factors

No significant difference in mean elastography values were noted between low and high Ki-67 levels (mean values of 224 and 235 kPa, respectively, P value of 0.239), presence or absence of lymphovascular invasion (mean values of 233 and

Table 2: Table showing correlation of elastography values with margins and posterior features of the mass.

Morphological features	SWE parameters				
	Mean (kPa) Mean (SD)	Minimum (kPa) Mean (SD)	Maximum (kPa) Mean (SD)	SD Mean (SD)	Ratio Mean (SD)
Non-mass abnormality	191 (25.23)	157 (25.92)	216 (43.86)	16 (14.05)	9 (3.76)
Mass	235 (52.92)	186 (51.98)	263 (52.96)	21 (13.43)	17 (11.79)
P value	0.000	0.005	0.005	0.265	0.000
Circumscribed masses	191 (59.39)	143 (45.44)	224 (72.78)	22 (13.04)	21 (17.12)
Non-circumscribed masses	243 (47.62)	193 (49.44)	270 (45.20)	21 (13.54)	16 (10.45)
P value	0.000	0.000	0.008	0.74	0.09
Posterior enhancement	206 (65.02)	163 (57.85)	234 (70.41)	19 (13.05)	19 (13.50)
No posterior features	233 (46.01)	184 (43.01)	264 (47.92)	22 (14.04)	16 (9.83)
Posterior shadowing	247 (48.59)	199 (56.18)	271 (43.86)	19 (13.05)	15 (13.11)
P value	0.004	0.011	0.008	0.262	0.218
Calcifications absent	231 (54.78)	184 (53.32)	258 (55.56)	20 (14.2)	17 (12.43)
Calcifications present	231 (48.28)	182 (46.37)	262 (49.93)	21 (11.94)	15 (9.53)
P value	0.991	0.762	0.693	0.640	0.342

SD: Standard Deviation, Numbers in bold represent P values.

230 kPa, respectively, P value of 0.668), presence or absence of lymph-nodal metastasis (mean values of 233 and 229 kPa, respectively, P value of 0.645), and the grade of the tumor (mean values of 241, 233, and 227 kPa in grade 1, 2, and 3 tumors, respectively, P value of 0.533).

SWE based on morphology

Lesions appearing as non-mass abnormalities exhibited considerably low mean, minimum, and maximum elastography values and SWE ratio in contrast to lesions presenting as masses (as depicted in Table 2 and Figure 2). This difference was found to be statistically significant.

Among the masses, the correlation between morphological features such as margins, posterior features, calcifications, and SWE parameters were analyzed [Table 2]. Masses with circumscribed margins and masses showing posterior

enhancement had significantly lower mean, minimum, and maximum elastography values compared to masses with non-circumscribed margins and no posterior enhancement. Mean elastography values did not demonstrate a significant difference with the presence or absence of calcifications.

SWE based on molecular subtypes

Hormone receptor-positive tumors exhibited markedly elevated mean, minimum, and maximum values in comparison to hormone receptor-negative tumors [Table 3, Figure 3]. TNBC had lower mean, minimum, and maximum values than non-triple-negative tumors [Table 3, Figure 4].

Influence of morphology on SWE values within the molecular subtypes

With regards to hormone positive tumors (luminal A and B), the most prevalent margin type was spiculated (33/97 [34%]), while the least prevalent margin observed was circumscribed (8/97 [8.2%]). In the hormone positive subgroup, masses with circumscribed margins exhibited notably lower mean, minimum, and maximum elastography values in comparison to masses with non-circumscribed margins [Figure 5].

Conversely, among basal-like TNBC, the most common margin observed was circumscribed (10/30 [33.3%]) and spiculated margins were observed in 5 out of 30 cases (16.7%). The spiculated margins group displayed significantly higher mean and minimum elastography values compared to masses without spiculated margins [Table 4, Figure 6].

In the basal-like triple negative tumors, there existed a statistically significant disparity in the mean and minimum

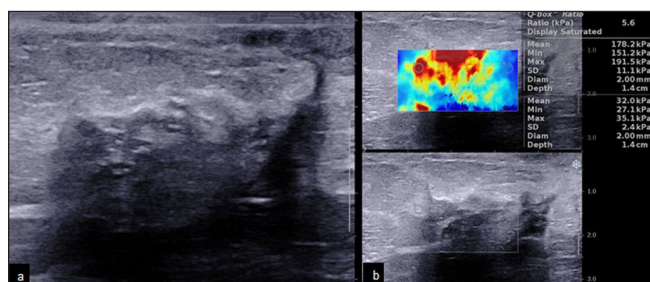


Figure 2: A 55-year-old with ductal carcinoma in situ (DCIS) presenting as non-mass abnormality and low elastography values. (a) B-mode 2D ultrasound image showing a hypoechoic non-mass abnormality with posterior acoustic shadowing. (b) Elastography image showing peripheral hardness with lower elastography values (mean: 178 kPa, minimum: 151 kPa, and maximum: 192 kPa).

Table 3: Table showing difference in elastography values in triple negative and non-triple negative breast cancers.

Shear wave elastography values	TNBC (30) Mean (SD)	Non-TNBC (122) Mean (SD)	P value	HR positive cancers (97) Mean (SD)	HR negative cancers (55) Mean (SD)	P value
Mean (kPa)	211 (64.36)	236 (48.36)	0.020	239 (48.69)	218 (56.83)	0.016
Minimum (kPa)	164 (52.50)	188 (49.71)	0.018	164 (52.50)	188 (49.71)	0.034
Maximum (kPa)	239 (69.59)	265 (47.95)	0.017	239 (69.59)	265 (47.95)	0.028
SD	20 (13.39)	21 (13.55)	0.666	20 (13.39)	21 (13.55)	0.741
Ratio	16 (11.36)	16 (11.67)	0.812	16 (11.36)	16 (11.67)	0.404

SD: Standard Deviation, TNBC: Triple Negative Breast Cancer, HR: Hormone Receptor.

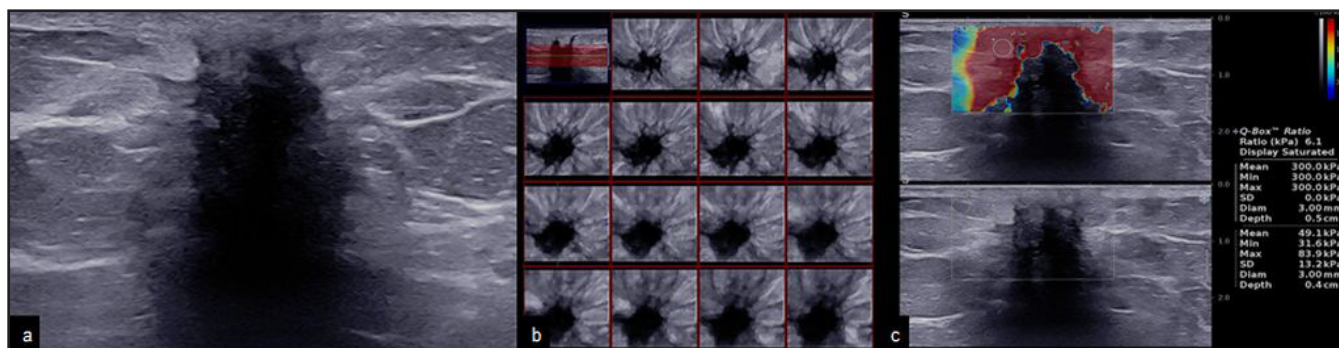


Figure 3: A 59-year-old with hormone receptor-positive breast cancer. The mass shows spiculated margins and high elastography values. (a & b) B-mode 2D and 3D ultrasound images showing an irregular hypoechoic mass with spiculated margins and posterior acoustic shadowing. (c) Elastography image showing peripheral hardness of the lesion with high elastography values (mean: 300 kPa, minimum: 300 kPa, and maximum: 300 kPa).

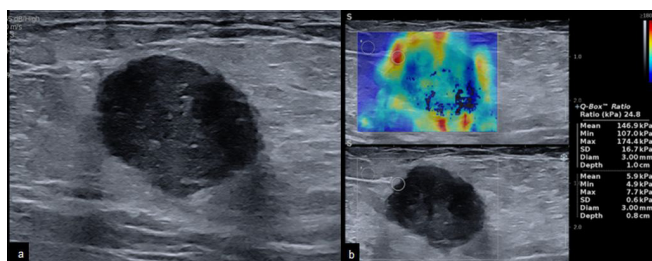


Figure 4: A 40-year-old with triple negative breast cancer. The mass shows circumscribed margins and low elastography values. (a) B-mode 2D ultrasound image showing a round hypoechoic mass with circumscribed margins and posterior acoustic enhancement. (b) Elastography image showing peripheral hardness of the lesion with lower elastography values (mean: 147 kPa, minimum: 107 kPa, and maximum: 174 kPa).

elastography values among tumors with posterior acoustic enhancement where it was low (175 and 132 kPa, respectively) in comparison to the higher value in tumors without posterior enhancement (230 and 180 kPa, respectively).

In HER2 strong positive tumors, the presence or absence of calcifications (mean values of 228 and 236 kPa, respectively) did not exhibit any significant correlation (P value of 0.590). Similarly, the hormone receptor status also did not exhibit any significant correlation (P value of 0.244) with mean

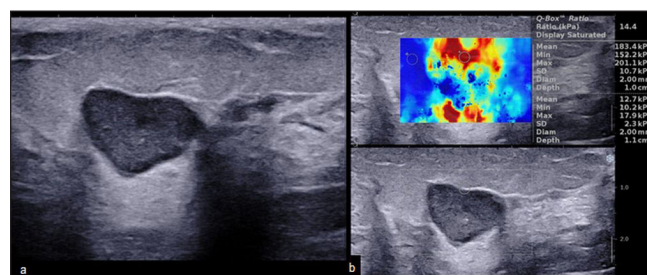


Figure 5: A 45-year-old with hormone receptor-positive breast cancer. The mass shows circumscribed margins and low elastography values. (a) B-mode 2D ultrasound image showing an oval hypoechoic mass with circumscribed margins and posterior acoustic enhancement. (b) Elastography image showing peripheral hardness of the lesion (suspicious lesion despite the benign morphology) with lower elastography values (mean: 183 kPa, minimum: 152 kPa, and maximum: 201 kPa).

elastography values (242 and 225 kPa in ER positive and ER negative tumors, respectively).

DISCUSSION

The role of SWE in the assessment of solid masses and in distinguishing between benign and malignant masses has been established in multiple studies in the literature.^[21–23] However, its role in molecular subtyping and other unfavorable prognostic factors, like Ki-67, LVI, LN status, and tumor

Table 4: Correlation of elastography features with margins in hormone positive and triple negative subgroups.

Shear wave elastography values	Hormone positive breast cancers: masses (89/97)		P value	Triple negative breast cancers (30)		P value
	Mean (SD)			Mean (SD)		
Margins	Circumscribed margins (8)	Non-circumscribed margins (81)		Spiculated margins (5)	Non-spiculated margins (25)	
Mean (kPa)	190 (31.47)	249 (45.91)	0.001	264 (59.37)	201 (61.01)	0.042
Minimum (kPa)	148 (31.40)	197 (48.88)	0.007	214 (72.94)	154 (42.69)	0.017
Maximum (kPa)	222 (49.48)	276 (42.83)	0.001	274 (58.89)	232 (70.49)	0.22
SD	21 (10.49)	21 (13.76)	0.918	13 (15.01)	21 (12.99)	0.24
Ratio	15 (11.8)	16 (10.57)	0.687	16 (6.39)	16 (12.21)	0.94

SD: Standard Deviation

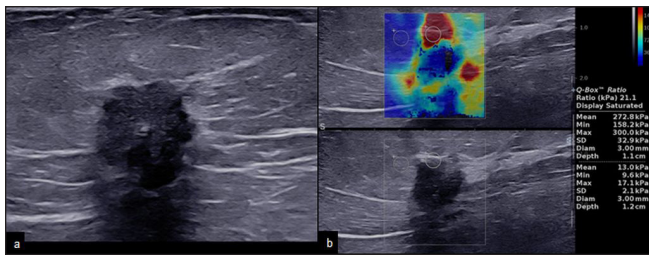


Figure 6: A 65-year-old with triple negative breast cancer. The mass shows spiculated margins and high elastography values. (a) B-mode 2D ultrasound image showing an irregular hypoechoic mass with spiculated margins and posterior acoustic shadowing. (b) Elastography image showing peripheral hardness of the lesion with higher elastography values (mean: 273 kPa, minimum: 158 kPa, and maximum: 300 kPa).

grade, has yielded varying results in existing literature.^[10-17] The reason for variable results in different studies may be due to the lack of studies investigating a correlation between B-mode features (morphology) and the elastography values within different molecular subtypes, constant development in the shear wave technology, and variations across different SWE units.

In our study, hormone receptor-positive tumors exhibited higher elastography values and basal-like TNBC had lower elastography values [Figure 7]. Numerous studies have supported this finding.^[13,24-28]

We noted that the morphology and the pathological subtypes of the tumor appear to have a significant impact on

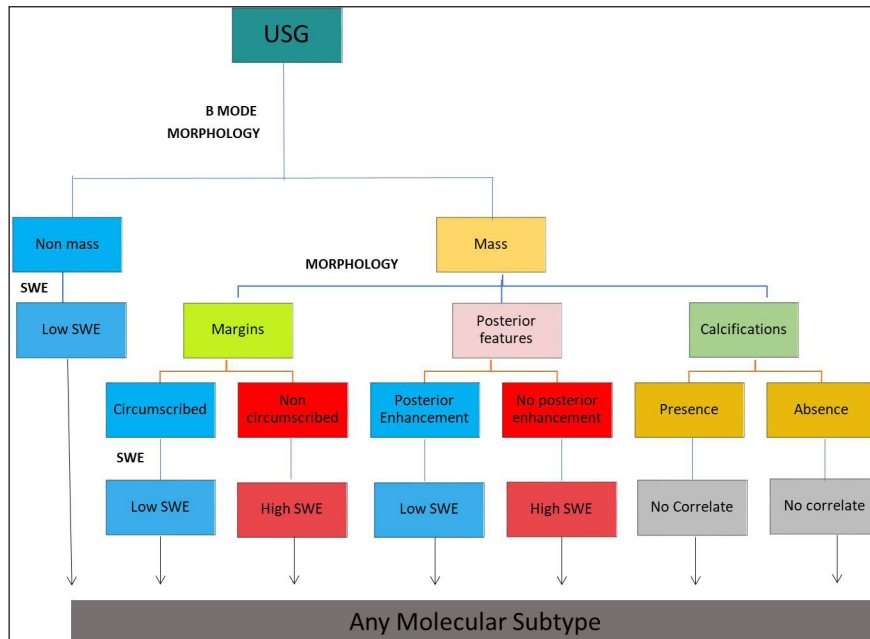


Figure 7: Flow chart demonstrating that elastography is dependent on morphological characters and plays no significant role in predicting molecular subtypes. SWE: shear wave elastography.

elastography values in different molecular subtypes. To the best of our knowledge, this correlation has not been published earlier.

Tumors presenting as non-mass abnormalities or as non-ductal and in situ cancers were found to have significantly lower elastography values even when these tumors belonged to the hormone positive subgroup. However, several differences had been reported in the literature compared to our study. Ductal carcinoma in situ demonstrating lower elastography values compared to invasive ductal carcinomas has been reported by Bae *et al.*^[29] However a study by Suvannarerg *et al.* showed no association with any tumor type,^[30] while Brkljačić *et al.* concluded that lobular carcinomas had higher elastography values.^[31]

In our study, 34% (33/97) of the HR positive cancers had spiculated margins and high SWE values and 33.3% (10/30) of basal-like TNBC had circumscribed margins and low SWE values which is consistent with prior literature.^[32] An 8.2% (8/97) of HR positive tumors had circumscribed margins and 16.7% (5/30) of basal-like TNBC had spiculated margins (atypical morphology) and had low and high SWE values, respectively.

The above subset of HR positive and basal-like TNBC that exhibited atypical morphological characteristics and non-ductal pathological subtypes also had atypical elastography values. This particular discrepancy in elastography values across various molecular subtypes of breast cancer, contingent upon the morphology, has not been documented in the published literature.

Our analysis revealed that HER2 positive cancers did not display any dominant morphological features and the presence of calcifications and hormone positivity did not have an impact on the elastography values. Figure 7 summarizes our results and demonstrates that elastography is dependent on morphological characteristics and plays no significant role in predicting molecular subtypes.

There are limitations to this study which include its retrospective nature and the fact that it was conducted at a single center. The entire study was conducted using a single shear wave unit, so the results may not be applicable to other units. Additionally, the number of non-ductal cancers in the study population was relatively low. While we have identified a few potential reasons for the discrepancy in elastography values among different molecular subtypes, it is evident that the cause is multifactorial and requires larger studies.

CONCLUSION

SWE values appear to be dominated by morphological features and the same was reflected as differences in values

within the same molecular subtype. We therefore conclude that SWE independently does not correlate with the molecular subtypes and morphology seems to play a key role in elastography values.

Further investigation using a larger dataset and different SWE units may better establish the clinical utility of SWE in the assessment of different molecular subtypes of breast cancer.

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Ethical approval

The research/study approved by the Institutional Review Board at Kovai Medical Center and Hospital Limited, number EC/AP/732/07/2019, dated 24/08/2019.

Declaration of patient consent

Patient's consent not required as there are no patients in this study.

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Conflicts of interest

There are no conflicts of interest.

Use of artificial intelligence (AI)-assisted technology for manuscript preparation

The authors confirm that there was no use of AI-assisted technology for assisting in the writing of the manuscript or editing of the manuscript and no images were manipulated using AI.

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