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FULL PAPER

Shear wave elastography as a supplemental tool in the assessment of unsuspicious axillary lymph nodes in patients undergoing breast ultrasound examination

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Objectives: To define reference values for shear wave elastography (SWE) in unsuspicious axillary lymph nodes in patients undergoing breast ultrasound examination.
Methods: In total, 177 clinically and sonographically unsuspicious axillary lymph nodes were prospectively evaluated with SWE using Virtual Touch Tissue Imaging Quantification (VTIQ) in 175 women. Mean values of tissue stiffness for axillary fatty tissue, lymph node cortex, and lymph node hilus were measured. Additionally, test-retest reliability of SWE in the assessment of axillary lymph node stiffness was evaluated by repeating each measurement three times.
Results: In 177 axillary lymph nodes, the mean stiffness of lymph node cortex, hilus, and surrounding fatty tissue as quantified by SWE was 1.90 m/s (SD: 0.34 m/s), 2.02 m/s (SD: 0.37 m/s), and 1.75 m/s (SD: 0.38 m/s), respectively. The mean stiffness of cortex and hilus was significantly higher compared to fatty tissue ($p < 0.0001$). SWE demonstrated good test–retest reliability in the assessment of stiffness of the lymph node hilus, cortex, and the surrounding fatty tissue with an intraclass correlation of 0.79 (95% CI: 0.75; 0.83), 0.75 (95% CI: 0.70; 0.79), and 0.78 (95% CI: 0.74; 0.82), respectively, ($p < 0.0001$).
Conclusions: Reference values for SWE in unsuspicious axillary lymph nodes are determined. These results may help to better identify axillary lymph node metastasis for breast cancer patients when combined with other lymph node features. SWE is a reliable method for the objective quantification of tissue stiffness of axillary lymph nodes.
Advances in knowledge: This study presents physiological reference values for tissue stiffness by examining the axillary lymph nodes with SWE in 175 women with sonomorphologically unsuspicious lymph nodes.

INTRODUCTION

Axillary ultrasound (AUS) is routinely used as part of the pretherapeutic clinical evaluation of patients with early breast cancer and is recommended by national and international guidelines.[1,2] Axillary lymph node (LN) status should be determined pretherapeutically as it is an important prognostic factor for disease recurrence and overall survival, and drives the selection of therapy regimens, both systemic and surgical, especially due to the broad use of neoadjuvant chemotherapy in high-risk patients.[3] Pretherapeutic AUS aims to identify patients who will not benefit from extensive axillary surgery and may undergo sentinel LN biopsy.[4] Although AUS is widely used in clinical routine, its diagnostic accuracy is low as criteria for LN positivity have been proposed but not yet standardized.[5]

Sonomorphological criteria for physiological axillary LNs are the presence of a slim cortex and a hyperechoic hilus. Color Doppler imaging can be used to determine arterial flow through the hilus. A lobulated oval shape is also considered a physiological feature for a LN.[6,7] A cortical thickness of >2–3 mm, a focal cortical bulge or the loss of the hyperechoic hilus are associated with metastatic involvement but have a low positive-predictive value as these signs are unspecific.[8] Overall, conventional sonographic B-mode axillary staging is usually associated with a sensitivity of 45–87% and a specificity of 49–97%.[9–11] Even when considering needle biopsy in sonographically suspicious

LN, there is still a high rate of false-negatives (21–29%).[10–15] Additional techniques are needed to improve the diagnostics of axillary LN status. One potential technique might be 2D shear wave elastography (SWE) that provides sonographic quantification of tissue stiffness based on shear wave velocity.[16] Increased tissue stiffness is a known predictor for malignancy and is an established complementary tool in breast cancer diagnostics.[17–20] Also, as a method for response assessment during neoadjuvant systemic therapy, SWE has shown the potential to improve sensitivity compared to standard clinical assessment.[21] Correspondingly, SWE could be utilized to assist in axillary staging in breast cancer patients. In this context, recent studies have hypothesized that measuring tissue stiffness by SWE could be utilized as a complementary tool to improve presurgical detection of axillary LN metastasis.[22,23]

However, literature on the evaluation of axillary LNs of breast cancer patients with SWE is still limited, especially on the elastography of physiologic axillary LN tissue. This study aims to quantify the distribution of physiological tissue stiffness values in unsuspicious axillary LNs as measured by 2D SWE. Additionally, the technical feasibility and reliability of 2D SWE for axillary staging in a clinical routine setting is accessed.

METHODS AND MATERIALS

Study design and enrollment

This is a prospective, single-center observational study. The study protocol was approved by the local ethics committee and written informed consent was obtained from each patient (S-396/2019). The study was conducted according to the Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) guidelines. Supplementary Material 1

Shear wave elastography (SWE)

B-mode ultrasound (US) and SWE were performed using 2D-SWE systems Siemens Acuson S2000 or S3000 equipped with virtual touch tissue imaging quantification (VTIQ) software utilizing a 9MHz probe (Siemens Healthineers). The VTIQ algorithm estimates the velocity of the induced shear waves in meters per second (m/s) which is correlated with tissue stiffness. A quality map indicated if an accurate measurement was obtained or was compromised due to the compression or movement of the patient.[24] In these cases, the measurement was repeated under more optimized conditions. As our study was performed during clinical routine at the breast unit, residents with and without experience in SWE as well as senior physicians experienced in SWE were involved in the study exam. Experience in SWE was defined as regular use of VTIQ in the breast unit for >6 months. After randomly choosing one representative LN, the physician performed SWE. The LN was chosen in level I or II of the axilla by the same physician, who performed SWE. Criteria for LN selection were good accessibility as well as the absence of malignancy signs such as cortical thickening, rounded LN with complete or partial effacement of the fatty hilus or, pathological color doppler images.[25]

Patients were positioned identically for imaging as in standard breast US with the ipsilateral arm elevated. Long-axis, short-axis diameter of each LN was measured. SWE was performed with minimum compression induced by the transducer. Elasticity values were measured in m/s, whereby a range from 0 to 10 m/s can be calculated by the device. Velocities higher than 10 m/s are given as "high". Elasticity values from the regions of interest, namely, hilus and cortex of the LN as well as from the surrounding fatty tissue were documented. In each measured area, three repetitive measurements were obtained. As internal control, SWE velocities measured in LNs of the right and left axilla were compared against each other.

Statistical analysis

This is an exploratory study. Statistical tests and resulting *p*-values can therefore only be interpreted descriptively. The study cohort was described by the measures of empirical distribution. Depending on the level of measurement, mean and standard deviation (SD) as well as absolute and relative frequencies were calculated. To compare the study cohort and sonomorphology of the LNs with respect to their location in the right and left axilla as well as experienced and

Table 1. Patient's characteristics

	Study cohort	Physician with experience > 6 months	Physician with experience < 6 months	p-value[a]
Age in years	46.6 (SD[b]: 13.5)	49.6 (SD: 14.5)	44.4 (SD: 12.4)	$p = 0.01$
Side				
Left	91 (51.4%)	36 (50.7%)	55 (51.7%)	
Right	86 (48.6%)	35 (49.3%)	51 (48.3%)	$p = 0.99$
Depth of the measurement in cm				
Cortex	1.55 (SD: 0.54)	1.55 (SD: 0.56)	1.55 (SD: 0.53)	$p = 0.34$
Hilus	1.66 (SD: 0.55)	1.67 (SD: 0.57)	1.65 (SD: 0.54)	$p = 0.17$
Fatty tissue	1.37 (SD: 0.56)	1.36 (SD: 0.51)	1.38 (SD: 0.59)	$p = 0.11$
Diameter of the LN in mm				
Long-axis	13.18 (SD: 4.70)	13.1 (SD: 4.48)	13.2 (SD: 4.87)	$p = 0.91$
Short-axis	6.86 (SD: 3.35)	6.6 (SD: 2.49)	7.1 (SD: 3.92)	$p = 0.36$

[a]p-values are calculated with independent t-test and chi-square test
[b]SD: standard deviation

unexperienced physicians independent t-test and chi-square test were used. To assess the test–retest reliability of three measurements for each LN taken on cortex, hilus, and fatty tissue intraclass correlation coefficient (ICC) with 95% confidence interval (CI) were calculated and interpreted according to Koo et al.[26] To compare velocities with respect to cortex, hilus and fatty tissue two-sided t-test for dependent samples was used. To investigate the influence of LN size and LN localization on LN velocity linear regression was used. Statistical analysis was performed with R (version 4.1.0 – © 2021 The R Foundation for Statistical Computing).

RESULTS

Description of study population

Analysis was based on 177 LN in 177 axillae of 175 patients. Mean age was 46.6 years (SD: 13.5) years. 91 (51.4%) of all LN were in the left axilla, 86 (48.6%) where in the right axilla (Table 1).

Thirty-three (18.6%) of all patients had a history of breast cancer on the contralateral breast and was scheduled in the breast unit for follow-up. Seventy-one (40.1%) patients had preventative medical check-up or follow-up as part of the high-risk program due to a familial history of breast cancer or the detection of a pathological genetic mutation. Eighty-two (46.3%) of all patients had breast findings categorized as BI-RADS 3 in an external clinic and visited the breast unit for specific diagnostics. Retrospectively, 25 of 33 patients (76%) with history of breast cancer of the contralateral breast had a follow-up in the breast unit with a median follow-up of 26 months (minimum 5 months, maximum 32 months). No patient was diagnosed with a second breast cancer or contralateral axillary metastases. Mean size of the chosen LN was 13.2 mm (SD: 4.7 mm) x 6.8 mm (SD: 3.3 mm). SWE was performed in 106 axillary LN of 104 patients by residents with little experience in SWE, while 71 LN of 71 patients were measured by senior physicians or experienced residents. The two groups showed a statistically significant difference regarding the age of the patients ($p = 0.01$), all other clinical and sonomorphological parameters were not statistically different ($p > 0.05$). (Table 1)

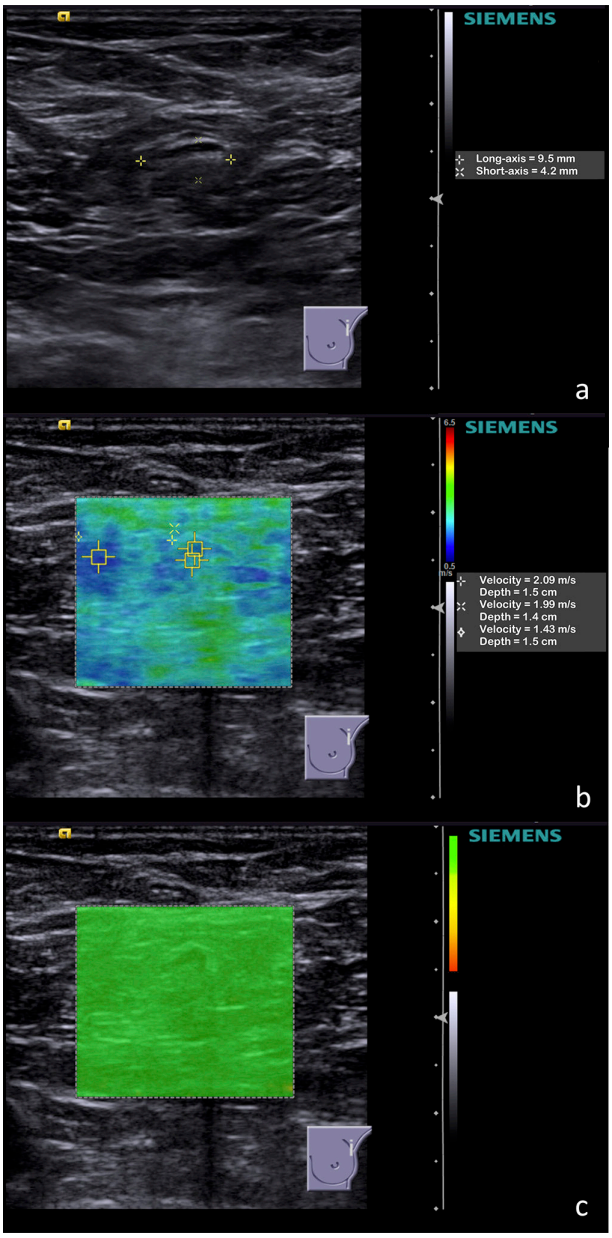
A representative measurement is shown in Figure 1.

Velocity of cortex, hilus and fatty tissue

Measuring velocity of cortex, hilus, and fatty tissue showed a mean shear wave velocity of 1.90 m/s (SD: 0.34 m/s), 2.02 m/s (SD: 0.37 m/s), and 1.75 m/s (SD: 0.38 m/s), respectively (Table 2). As an internal control, velocity of the LN measured in the left axilla was compared to the respective velocities measured in the right axilla. There was no significant difference with a p-value of 0.77, 0.09, and 0.92, respectively (Table 3). Comparing mean velocity of cortex and fatty tissue as well as hilus and fatty tissue the differences were statistically significant ($p < 0.0001$). The comparison of the mean velocity of cortex and hilus was also significantly different with $p < 0.01$ (Figure 2). The mean depth of the measurements was 1.55 cm (SD: 0.54 cm), 1.66 cm (0.55 cm), and 1.37 cm (0.56 cm), respectively, for the three localizations.

The quality map indicated a good quality of measurement in all patients.

Figure 1. Virtual Touch Imaging Quantification (VTIQ) of an unsuspicious axillary lymph node (a). *conventional ultrasound documentation of an unsuspicious lymph node (b). VTIQ on cortex, hilus and surrounding fatty tissue (c). quality map*[a] [a]*green is high quality; yellow is lower quality; red is poor quality*



Univariate linear regression showed no correlation of size of LN on velocity ($r^2<0.1$ in cortex and hilus, respectively). The depth of the measurement showed no influence on the velocity as well ($r^2<0.1$ in cortex and hilus, respectively).

Reliability of VTIQ

Within each of the 177 LN, three individual measurements were taken on cortex, hilus, and fatty tissue, respectively. Therefore, nine measurements were performed on each LN. ICC were obtained from each localization and were 0.79 (95% CI: 0.75; 0.83), 0.75 (95% CI:

Table 2. Shear Wave Elastography

	Cortex	Hilus	Fatty tissue
Mean stiffness (SD) in m/s	1.90 (SD[a]: 0.34)	2.02 (SD: 0.37)	1.75 (SD: 0.38)
Minimum stiffness in m/s	1.03	1.17	0.80
Maximum stiffness in m/s	2.77	3.37	3.03

[a]SD: standard deviation

0.70; 0.79), and 0.78 (95% CI: 0.74; 0.82), respectively, indicating a good test–retest reliability.

LN measured by senior physicians or experienced residents showed a mean velocity of 1.86 m/s (SD: 0.36), 2.02 m/s (SD: 0.41), and 1.66 m/s (SD: 0.39) in cortex, hilus, and fatty tissue. When measured by unexperienced residents the mean velocity was 1.93 m/s (SD: 0.32), 2.02 m/s (SD: 0.37), and 1.82 m/s (SD: 0.35), respectively, for each localization. ICC of the measurements by experienced physicians were 0.81 (95%CI: 0.75; 0.86), 0.68 (95%CI: 0.58; 0.76), and 0.84 (95%CI: 0.79; 0.89) for cortex, hilus, and fatty tissue, respectively. ICC of the measurements by unexperienced physicians were 0.77 (95%CI: 0.71; 0.82), 0.81 (95%CI: 0.76; 0.85), and 0.72 (95%CI: 0.66; 0.78) for cortex, hilus, and fatty tissue (Figure 3).

DISCUSSION

Based on the analysis of 177 LNs, proposed reference values of SWE in unsuspicious axillary LNs. The cortex, hilus, and surrounding fatty tissue of a LN showed a mean velocity of 1.90 m/s (SD: 0.34), 2.02 m/s (SD: 0.37), and 1.75 m/s (SD 0.38).

This prospective study validates in a representative cohort of patients the results of previous studies measuring SWE in benign axillary LNs, whereby no distinction was made between cortex and hilus of the LN.[22,23]

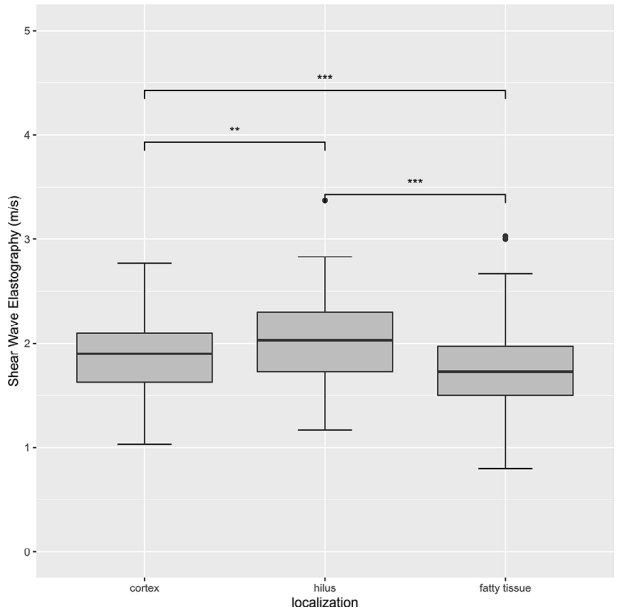
The velocity of LN hilus showed a significantly higher stiffness value compared to the cortex of the same LN ($p < 0.01$). This could be due to the increased vascularity of the LN hilus. This difference in velocity shows the importance of correct positioning of the SWE and that there is a need for precise evaluation of SWE in LNs with regards to its anatomy.

The good test–retest reliability, shown by an ICC of 0.79, 0.75, and 0.78 for cortex and hilus of a LN as well as surrounding fatty tissue, indicates that SWE is a reliable method that can be implemented in axillary diagnostics. There was no significant difference in SWE when measurements were performed by experienced or less experienced physician in LN cortex and hilus while the velocities in the surrounding fatty tissue did differ significantly ($p < 0.01$). In a sonomorphologically unsuspicious LN cortex and hilus are well defined so that ROI is clearly selectable in most cases even with little experience in SWE or sonography *per se*. But when performing SWE in the surrounding fatty tissue the physician needs to distinguish between fat, glandular tissue, tendon, muscle, or vessels inside the SWE window. Depending on the selected LN and its localization in the axilla, it might be possible that major vessels or contracted tendons/muscles are close to the ROI leading to higher velocity measurements. However, considering the good ICC as well as the SWE measurements in cortex and hilus of the LN, it can be concluded that SWE is a reliable method regardless of operator experience in a clinical routine setting. The size of the target LN as well as its depth does not seem to affect SWE in the axilla, although SWE is known to have limits in deeper tissue.[27] Since other B-mode findings are not that accurate, the addition of another feature may improve both negative and positive predictive value helping to triage patient appropriately. Another important aspect is the total duration of the ultrasound examination per patient. When performing SWE routinely in the breast, extending the examination to the axilla can be performed using the same probe and software while the patient can maintain the same position. Therefore, axillary SWE does prolong the examination for about 1–2 min per side, which is acceptable for routine clinical application.

Table 3. Mean SWE in m/s of LN cortex, hilus, and surrounding fatty tissue in the left and right axilla

	left	right	*p*-value
cortex	1.89	1.91	0.77
hilus	1.97	2.06	0.09
fatty tissue	1.76	1.75	0.92

Figure 2. Mean velocity measured in cortex and hilus of a lymph node as well as surrounding fatty tissue



$^{**}p < 0.01$ $^{***}p < 0.0001$

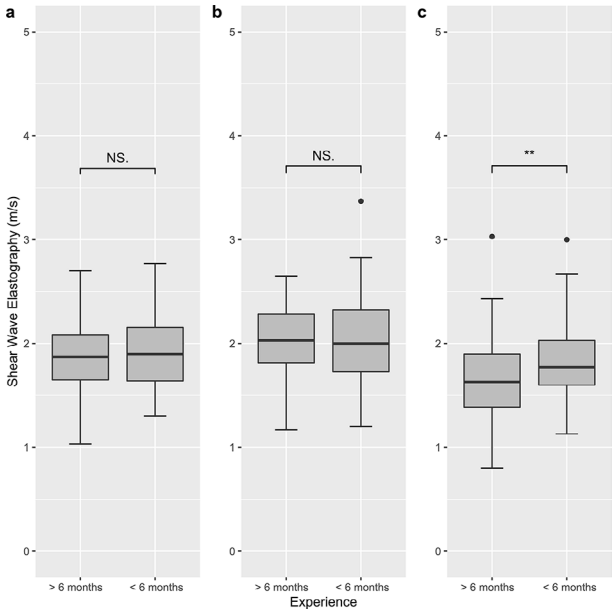
SWE is utilized in a wide range of specialties to characterize quality of LNs in a variety of anatomical regions, for example in the diagnostics of head and neck malignancies or in intramuscular LNs.[28–30] In these studies, SWE was used to differentiate between benign and malignant LNs and seems to offer a diagnostic improvement when combined with conventional B-mode US. SWE was also used to evaluate breast fatty tissue and breast lesions, especially after SWE was implemented in the latest edition of the BI-RADS classifications.[17,31,32] There are also a few studies evaluating the enhancement of sensitivity and specificity in axillary staging of patients with breast lesions with SWE in combination with B-mode sonography.[22,23,33] But to the best of our knowledge there is no study that evaluated the velocity of sonomorphologically inconspicuous axillary LNs as a reference.

There are several limitations of this study. First, it must be noted, that the selection of LN is solely based on sonomorphology in females with no clinical suspicion for malignancy. It is known that sonomorphologically unsuspicious LN may contain malignant cells as it can be seen in positive sentinel LNs.[34] This study was conducted during clinical routine examinations. Therefore, no LNs were examined by both experienced and unexperienced physicians, which limits comparability of results. Instead, LNs were randomly assigned to either an experienced or unexperienced physician and can therefore at least be considered comparable in key anatomic criteria including size, depth, and measured SWE.

Additionally, as it is true for sonography *per se*, SWE underlies subjective observer bias, which cannot be ruled out as a limitation of the present study as well. SWE was used in a pretherapeutic setting only. Using SWE in an *ex vivo* setting during surgery may be also a potential application, and potentially even allow the prediction of axillary LN metastasis intraoperatively in patients undergoing breast cancer surgery in the future.[35,36]

In the next steps SWE should be implemented in axillary staging to differentiate between benign and malignant LN, while comparing the velocity to the histopathological results with the gold standard of histopathological assessment.

Figure 3. Shear Wave Elastography (SWE) measured by experienced and unexperienced physicians in SWE in cortex (a), hilus (b), and surrounding fatty tissue (c) *NS. Not significant*



**$p < 0.01$ Experience was defined as regular use of VTIQ in the breast unit for > 6 months

In conclusion, SWE is a reliable method to measure the stiffness of unsuspicious axillary LNs. Our results show that SWE is a reliable tool in AUS regardless of depth and size of the LN as well as operator experience. Further studies are needed to confirm the diagnostic value of SWE as an additional tool in axillary staging by comparing the results with the respective histopathologic standard.

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