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PhD THESIS

**PREDICTIONS IN ESTABLISHING BREAST
MALIGNANCY: THE IMPACT OF DEMOGRAPHIC
FACTORS, CONVENTIONAL ULTRASOUND AND
REAL-TIME ELASTOGRAPHY**

ABSTRACT

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GENERAL PART

Breast cancer is a major public health disease worldwide but one of the most curable if diagnosed in an incipient state. According to Globocan 2020, breast cancer is the leading cause of cancer mortality among women in Romania, with a proportion of approximately 27% of all cancer-related deaths. Breast cancer is a well-known multifactorial disease, its development being influenced by a combination of genetic, environmental, hormonal, and lifestyle factors. The understanding of its complexity may improve targeted screening and detection and can aid in developing more effective prevention and treatment strategies. The screening and early detection have a crucial role because it may greatly improve the outcomes and the quality of life for the affected patients. Attention must be focused on improving and adjusting noninvasive diagnostic methods in order to minimize unnecessary surgery.

Breast ultrasound elastography is an advanced imaging technique that gained much ground lately, adding additional diagnostic value to conventional ultrasound by assessing the mechanical properties of breast tissue. It helps in differentiating benign from malignant lesions based on tissue stiffness, with malignant tumors being generally stiffer than benign ones and normal breast tissue.

Sonoelastography is a valuable tool, with many advantages, and great diagnostic performance. It was primarily used to differentiate solid from cystic lesions, rapidly evolved, nowadays being a mandatory support of mammography in the evaluation of dense breasts. A special concern is breast cancer in younger women where unique challenges and considerations appear when compared to cases in older women. Breast sonoelastography is a primary screening tool for female patients under 40, where mammography is not recommended. Other advantages of ultrasound elastography examination include its cost effectiveness, its non-irradiating and painless nature allowing routine use, its real-time character allowing for an immediate interpretation. Another advantage is brought to pregnant and lactating women, here being the method of choice. Additionally, it can improve differential diagnosis for breast masses and last but not least, it can be used as an aid in performing guided biopsy. Despite all of its advantages, and due to being routinely used, it can have the negative side effect of increasing benign findings requiring the adaption and improvement of imagistic screening and diagnostic tools. Optimal diagnostic performance is here the goal in order to avoid causing patient supplemental anxiety and distress.

SPECIAL PART

1 GENERAL OBJECTIVES

The present thesis is comprised of three studies that are gradually and consecutively meant to reach the final aim: ***to assess the best strategy in the evaluation of solid breast nodules by real time ultrasound elastography and to identify most relevant demographic reproductive and lifestyle risk factors predicting malignancy risk***

The three consecutive studies have the following objectives:

1. The first aim was to establish the best risk stratification algorithm in predicting malignancy risk for breast cancer, using the added value of real time strain elastography to conventional ultrasound in order to provide a better selection of nodules for biopsy or follow up.
2. The second aim was to establish which patients are at higher risk of breast cancer, analyzing the most relevant demographic reproductive and lifestyle risk factors representative for our geographical region
3. The third aim was to establish which factors could be associated to false positive results when assessing solid breast lesions by ultrasound elastography. Morphological characteristics of the nodules, alongside patient related factors, are to be evaluated in order to optimize the outcomes as to minimize benign biopsies without reducing the likelihood of potential malignancy identification.

2 PREAMBLE: RISK STRATIFICATION FOR BIOPSY REFERRAL IN THE CASE OF PATIENTS WITH SOLID BREAST LESIONS – CAN IT BE IMPROVED?

2.1 AIM OF THE STUDY

A better risk stratification approach, by using any additional available patient and nodule characteristics, prior to biopsy, is welcomed. The aim of this thesis is to evaluate if improvements to the biopsy referral strategy can be made by including both patient anamnestic data and nodule characteristics, as gathered during breast ultrasound elastography.

2.2 MATERIALS AND METHODS

2.2.1 COHORT AND DATA COLLECTION

The thesis is structured as three retrospective studies, evaluating patient data in the January 2017 to January 2022 period, gathered from the “Dr. D” medical center database. Inclusion criteria for the study were the following: any female patient, regardless of age and reason for presentation (subjective accuses, follow-up or screening) that underwent breast ultrasound examination where solid breast nodules were identified was included. Patients with prior breast cancer diagnostics were excluded. A database was established using the statistical software IBM SPSS version 23 (IBM Statistics, USA) where all relevant patient data was input based on the evaluation reports of the medical center.

2.2.2 EQUIPMENT AND IMAGING DIAGNOSIS

For conventional ultrasound, color Doppler scanning and real-time strain elastography evaluation, a Hitachi Preirus machine was used. The machine was equipped with “Hitachi Realtime Tissue Elastography” (HI-RTE) real-time elastography and color Doppler software. All ultrasound investigations were performed by equipping the machine with a 920 mm wide breast probe (EUP-L53L) adapting a dedicated water bag device for improved superficial tissue (skin, fascia and ligament) evaluation. Real-time strain elastography was performed using a small, 50 mm wide, EUP-L74M, 5-18 MHz linear multifrequency probe oriented perpendicular to the skin. Conventional B mode high-resolution images were collected alongside color Doppler and elastograms. All imagistic evaluations were carried out by the same experienced operator.

2.2.3 CONVENTIONAL GRAY SCALE ULTRASOUND AND RISK STRATIFICATION

Bilateral breast evaluation was performed using a ducto-radial ultrasound scanning technique following the lobar approach. All lesions were evaluated on two perpendicular planes by measurements performed in a radial and anti-radial orientation. After identifying and evaluating each solid lesion by ultrasonography, the following characteristics were recorded for each nodule: dimension, morphological characteristics according to the ACR guidelines, alongside color Doppler results assessing lesion vascularity and evaluation of abnormal lymph nodes. Afterwards, a BIRADS score was given based on the presence of malignancy markers, on a 1 to 5 scale, increasing with the likelihood of malignancy.

2.2.4 ULTRASOUND REAL-TIME ELASTOGRAPHY

After conventional ultrasound evaluation was performed, real-time strain elastography including both qualitative and semi-quantitative techniques. Tsukuba elasticity scores were attributed on the scale of 1 to 5 based on the color balance inside and around the examined nodule and split into three categories: TES 1 and 2 (low stiffness), TES 3 (intermediate stiffness), TES 4 and 5 (high stiffness). A TES value of 4 or 5 was chosen as indicative for malignancy. The FLR values were automatically calculated by comparing the average strain measured inside a lesion with the average strain of the adjacent fatty tissue (ROI). FLR values were split into three risk categories as follows: $FLR < 2.8$ (low stiffness), $FLR \geq 2.8$ and < 4.5 (intermediate stiffness), $FLR \geq 4.5$ (high stiffness)

2.2.5 BIRADS REGRADING BY ADDITION OF REAL-TIME STRAIN ELASTOGRAPHY

The initially attributed BIRADS score (from conventional ultrasound) was regraded after elastography to account for nodule stiffness. Downgrading was performed in the case of nodules with low stiffness (TES = 1 or 2 and $FLR < 2.8$) and BIRADS scores of 3 and 4A, while upgrading was performed for nodules with high stiffness (TES > 4 and $FLR > 4.5$) and BIRADS scores 3, 4A or 4B. According to EFSUMB guidelines, lesions characterized as BIRADS scores 4B, 4C and 5 were never downgraded. The final BIRADS score, determined the course of action for each patient as follows: BIRADS 3 and 4A were referred to 6 -12 month follow-up, BIRADS 4B, 4C and 5 were referred to biopsy

2.2.6 STATISTICAL ANALYSIS

Statistical analysis was performed using a combination of the R programming language (R Core Team Vienna, Austria) and Microsoft Excel (Microsoft Corporation, Redmond, Washington, USA) as needed. A 95% confidence interval was used for statistical significance as shown by the p-value < 0.05 . Normal distribution was evaluated with the Shapiro-Wilk test in the case of continuous variables and subsequently, these were reported as means coupled with standard deviation, while non-normally distributed variables were reported as median and interquartile range. Categorical variables were characterized as

frequency distributions and proportions in percentages. Based on the particular requirements of each independent study, other statistical methods like the Mann-Whitney U test, the Pearson's chi-squared test, logistic regression models, Nagelkerke's R-squared and Receiver Operating Characteristic curves were used.

2.3 RESULTS

A total number of 3227 nodules were collected. Out of these, cystic lesions were excluded as not being in the scope of the study. After exclusion of the cystic lesions, a total of 2688 solid breast nodules, belonging to 1423 female patients were analyzed. The cohort was split into patient characteristics (age, provenience(rural/urban),BMI, age at menarche, age at menopause, type of menopause (physiological/surgical/drug induced), number of births, age at first birth, breastfeeding history) and nodule characteristics (size, location(left breast/right breast), lobar clockwise position of the nodule) and lesion characteristics including morphological aspects by ACR guidelines alongside presence of vascularity and presence of lymphadenopathy.

2.4 DISCUSSIONS

In the past few years, elastography has gained ground as a complementary method to ultrasonography in noninvasive breast cancer screening. Despite positive outcomes, there is a further need to refine the method, especially regarding BIRADS category 4 where the distinction between benignancy and malignancy is established. Currently, the criteria for referral to biopsy, subsequent imagistic evaluation, is the presence of any breast lesion categorized as BIRADS score 4B and above as recommended by ACR guidelines. When evaluating our cohort, we can see that a total number of 527 breast lesions (or 15.43%) were asserted as BIRADS 4B and above by conventional ultrasound. It is in this group that the likelihood for false diagnostic and subsequently unnecessary biopsy is the highest. This thesis aimed to identify the best approach of solid breast lesions by assessing ultrasound and real-time elastography outcomes alongside patient anamnestic characteristics in order to obtain a better patient management.

2.5 CONCLUSIONS

The collected, 5 year data, comprising a significant number of solid breast lesions alongside their ultrasound, elastographic and patient anamnestic characteristics is considered a solid foundation to be used in the proposed evaluation of potentially improved patient risk stratification approaches for breast malignancy risk evaluation and subsequent referral to biopsy.

3 STUDY I: DIAGNOSTIC VALUE OF DIFFERENT RISK-STRATIFICATION ALGORITHMS IN SOLID BREAST LESIONS

3.1 AIM OF THE STUDY

The following study's' aims are to evaluate the best risk stratification system for characterizing solid breast nodules with regards to malignancy based on sonoelastographic evaluation. Two new risk stratification reporting systems ("BIRADS TM" and "BIRADS worst case") are proposed and their diagnostic performance is subsequently evaluated

3.2 MATERIALS AND METHODS

The inclusion criterion for our retrospective study was the presence of any solid breast mass in women of all ages (mean, 40.85 _ SD 27.11), detected during ultrasound examination. The Breast Imaging Reporting and Data System (BIRADS)–US criteria were used in the assessment of each nodule by conventional US (gray-scale mode) and Doppler evaluation. The Ueno score and strain ratio were also measured for all the described lesions. We considered multiple algorithms for the risk reassessment of solid breast nodules: classical BIRADS–US, EFSUMB BIRADS, worst-case scenario BIRADS and BIRADS TM.

3.3 RESULTS

The Sensitivity (Se), Specificity (Sp), Positive Predictive Value (PPV), Negative Predictive Value (NPV) and Accuracy (Acc) were obtained for all the proposed risk-stratification reporting systems: conventional BIRADS-US (Se, 74.23%; Sp, 63.95%; PPV, 13.53%; NPV, 97.79%; Acc, 65%); EFSUMB BIRADS (Se, 71.23%; Sp, 81.55%; PPV, 22.68%; NPV, 97.99%; Acc, 81%); worst-case scenario BIRADS (Se, 84.23%; Sp, 58.23%; PPV, 13.29%; NPV, 98.84%; Acc, 60%); BIRADS TM (Se, 81.23%; Sp, 75.84%; PPV, 20.35%; NPV, 98.81%; Acc, 77%).

3.4 CONCLUSIONS

The most efficient risk-stratification reporting system was the proposed one, BIRADS TM, which considers both upgrading and downgrading the conventional BIRADS-US.

4 STUDY II: BREAST CANCER RISK FACTORS AMONG WOMEN WITH SOLID BREAST LESIONS

4.1 AIM OF THE STUDY

The study was designed to analyze the impact on breast cancer risk, of a set of independent factors (age, BMI, menarche, menopause, years of exposure to estrogen, number of births, breastfeeding length, use of oral combined contraceptives, smoking, family medical history with regards to breast cancer, and living environment urban / rural).

4.2 MATERIALS AND METHODS

From January 2017 to December 2021, 1161 patients with solid breast lesions, as detected by sonoelastography, were divided into two groups: patients with benign lesions (1019, 87.77%) and patients with malignant nodules (142, 12.23%). The malignancy group was confirmed by a histopathological result. Variables including age, BMI, menarche, menopause, years of exposure to estrogen, number of births, breastfeeding period, use of oral combined contraceptives, smoker status, family medical history and living area (rural-urban) were recorded.

4.3 RESULTS

It was evidenced by our study that the main risk factors for malignancy were elevated age (OR = 1.07, 95% CI 1.05–1.08), BMI (OR = 1.06, 95% CI 1.02–1.10), living area (rural) (OR = 1.86, 95% CI 1.13–2.85) and family medical history (negative) (OR 3.13, 95% CI 1.43–8.29). The other proposed risk factors were not found to be statistically significant.

4.4 CONCLUSIONS

Age and BMI were observed to be the most significant factors for breast cancer risk increase, followed by living in a rural area. A family history of breast cancer was shown to be inversely correlated with cancer risk increase.

5 STUDY III: FACTORS ASSOCIATED WITH FALSE POSITIVE BREAST CANCER RESULTS IN THE REAL-TIME SONOELASTOGRAPHY EVALUATION OF SOLID BREAST LESIONS

5.1 AIM OF THE STUDY

The following study aims to find false positive predictors for cancer diagnosis and their influence on the quality of ultrasound elastography evaluation in the case of solid breast nodules, with an increased focus on lesions categorized as BIRADS 4B, 4C and 5.

5.2 MATERIALS AND METHODS

The study was designed as a retrospective analysis of patient data, collected during the January 2017 and January 2022 period. The cohort was made up of 1432 female patients exhibiting 1625 solid nodules as confirmed by ultrasound. The inclusion criteria was the presence of any breast nodule characterized as BIRADS 4B, 4C or 5 subsequent sonoelastographic evaluation. The exclusion criteria were BIRADS 3 and 4A (referred for follow-up), BIRADS 1 and 2 (normal breast or cystic breast lesion), any patient with previous history of breast cancer and respective breast surgery and any patient with prosthetic breast implants. For each patient, a set of data was collected as follows: Age, BMI, Number of births, History of breastfeeding, Affected breast (left/right), Nodule position in the breast (defined with the clockwise lobar approach). For each solid nodule, a BIRADS score was attributed subsequent evaluation by conventional grayscale ultrasound, color Doppler scanning and real-time strain elastography. Histopathological results from biopsy or surgery were used as the gold standard for diagnostic performance evaluation.

5.3 Results

Our study showed that most Sonoelastography factors linked to incorrect and overdiagnosis were due to a nodule dimension (OR = 1.02 per unit increase), posterior acoustic shadowing (OR = 12.26), reactive adenopathy (OR = 6.35), and an increased TES score (TES3 OR = 6.60; TES4 OR = 23.02; TES5 OR = 108.24). Regarding patient characteristics, age (OR = 1.09 per unit increase), BMI, (OR = 1.09 per unit increase), and breastfeeding history (OR = 3.00) were observed to increase the likelihood of false positive results. On the other hand, the nodules less likely to be part of the false positive group exhibited the following characteristics: a regular shape (OR = 0.27), homogenous consistency (OR = 0.42), and avascularity (OR = 0.22).

5.4 Conclusions

Older age, high BMI, patients with a breastfeeding history, and those who exhibit the following specific nodule characteristics were most often linked to false positive results: large tumors with posterior acoustic shadowing and high elasticity scores, accompanied by reactive adenopathy. On the other hand, homogenous, avascular nodules with regular shapes were less likely to be misdiagnosed.

FINAL CONCLUSIONS

1. The best risk stratification algorithm in malignancy prediction of solid breast lesions, assessed by real time ultrasound elastography

Both semiquantitative and qualitative methods proved to be significantly accurate (ACC, 94%).by our study A semiquantitative threshold value of 4,5 and greater was determined to be optimal for predicting malignancy, confirming that increased stiffness is associated with an increased likelihood of malignancy. The best diagnostic performance showed by our study was including real time elastography as a parameter of the BIRADS score in solid breast lesion assessment. By upgrading (BIRADS 3,4A,4B) and downgrading (BIRADS 3,4A) strategy, a diagnostic performance of 81.23% sensitivity, 75.84% specificity, 77% accuracy, 20.35% PPV and 98.81% NPV was achieved. Our results confirm the significant additional value of strain elastography in the evaluation of solid breast nodules, encouraging its use as an additional tool to conventional ultrasound.

2. The most relevant demographic reproductive and lifestyle risk factors for breast cancer.

The definition of women at high risk for breast cancer development, as shown by our study, and representative for our geographical region, are obese menopausal women with a median age of 52,5 living in rural areas.

3. Morphological nodule characteristics, elastography and patient related factors that influence the false positive results in ultrasound real time elastography breast nodule examination

Our study showed that the most specific nodule characteristics linked to false positive outcomes were benign nodules with US morphological appearance mimicking malignant lesions, namely large tumors with calcifications, posterior acoustic shadowing and or presenting high elasticity scores with a median value

of 4.5 for FLR and elasticity score of 3. Here, there was an approximately 560% higher likelihood of false diagnosis of malignancy. On the other hand, homogenous, avascular nodules with regular shape were shown to be less likely to be associated with a false positive diagnostic. Of the patient characteristics, older age (greater than 52) with each year adding approximately a 9% increase in false positive risk, obesity with its technical difficulties and potential adipose breasts, alongside patients with breastfeeding history or active breastfeeding were shown by our study to be most often linked to false diagnosis when assessing solid breast lesions by sonoelastography. The advisable strategy is to employ all adequate imaging techniques, in addition to clinical examination, prior to biopsy referral. The goal is to minimize the benign biopsy rates but at the same time not to miss any potential malignancies

To conclude, breast cancer, as a complex disease, requires the corroboration of multiple aspects in the complex management of patients at risk. This thesis, considering the significant number of solid nodules examined, shows that real-time strain elastography considerably improves the diagnostic performance of conventional ultrasound breast evaluation. Additionally, an optimal risk stratification method was proposed for augmenting conventional ultrasound results with the addition of real-time elastography, while on the other hand the risk factors for sonoelastographic false positive results were found. Another contribution is the identification of patient related characteristics increasing the risk for malignancy.