Stepping forward in personalizing breast cancer management

Added value of diagnostic assets

Moorman, A.M.

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A.M. Moorman

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aan de Universiteit van Amsterdam

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ten overstaan van een door het College voor Promoties ingestelde commissie,

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op woensdag 20 september 2023, te 11.00 uur

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Chapter 1.

Introduction and aims of thesis

General introduction

Breast cancer is the most common type of cancer worldwide according to statistics released by the international agency for research on cancer (IARC) in December 2020[1], taking over the first place position previously held by lung cancer. It is also the most diagnosed cancer among women, accounting for 1 in 4 cancer cases. In 2020, there were 2.3 million new cases of breast cancer with an estimated number of deaths of 684,996. In the Netherlands 16,975 women are diagnosed with breast cancer every year and 2,300 are diagnosed with an in situ carcinoma [2]. One in every 7 women will be diagnosed with breast cancer during her lifetime and one in 27 women will die of breast cancer. The number of women alive after the diagnoses however has doubled in the last 20 years[2]. Since the introduction of screening mammography programs, the proportion of small cancers is higher and consequently the incidence of axillary metastases has decreased. At the same time, with the increased use of breast conserving therapies and widespread use of chemo- and hormone therapy, breast cancer outcomes have significantly improved. The improved outcomes are implied to be due to the extended indications and applications of the above-mentioned treatment modalities and herein lies the potential to overtreatment. In order to control overtreatment, de-escalation demands a careful selection process.

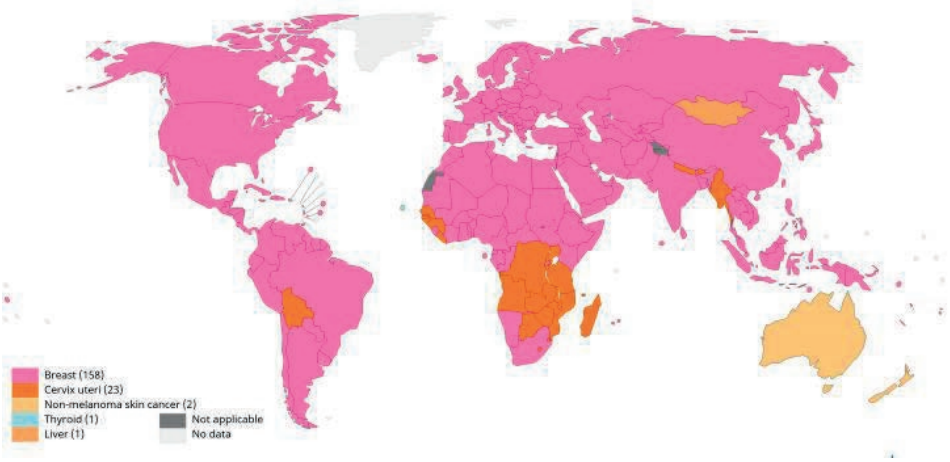

Figure 1. Incidences of cancer in females, per country in 2020 according to the World Health Organisation (WHO).

History of breast cancer.

We have come a long way since one of the first well documented mastectomies was performed in the 16th century by a German surgeon, William Fabry. He used an instrument to compress and fix the base of a breast during mastectomy which allowed for rapid excision in a time prior to the development of anaesthetics[3], see figure 2. Although the earliest reports of breast cancer were found in the Edwin Smith Egyptian papyrus[4], originating 3,000-2,500 B.C, possible attributable to Imhotep (Egyptian physician-architect) and later several references dating back to ancient Greece during the time of Hippocrates (c. 400 B.C.). The first major contributions in lymph node mapping was described in the 18th century by Henri Le Dran who wrote that 'cancer begins as a local disease but spreads via lymph'[5]. It wasn't until the 19th century, after the introduction of general anaesthesia, that the field of surgery dramatically changed. Seishu Hanaoko, a Japanese surgeon performed the world's first procedure under general anaesthesia in 1804, which was a mastectomy[6].

Although he was not the first to describe a wide extirpation of the breast, William S. Halsted became worldly known after publishing his paper in 1894 on the recommended surgical procedure for breast cancer. This procedure has been the standard operation for a long time, in which the emphasis lay on en-bloc resection of the breast with all suspected tissue to prevent spread and removal of the pectoralis major to prevent recurrence[7].

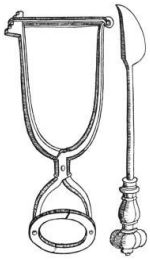

Figure 2. Instruments used by William Fabry to perform mastectomies in the 16th century.

In 1895 the first X-ray was taken by Wilhelm Conrad Röntgen[8] and within one year X-rays were used to treat a patient with breast cancer[9]. Due to its local toxicity and de novo cancer development, radiotherapy wasn't very popular and widely used then, but it created the possibility for a more multidisciplinary approach. By the 1930s radiotherapy was used as an alternative to the invasive and mutilating radical mastectomy.

In 1949 the modified radical mastectomy was introduced. In that same year a study presented their results on the different treatment modalities in patients treated with simple, radical or modified radical mastectomies with or without radiotherapy and the results were similar[10]. Over the next century the

development in radiotherapy became more targeted and effective. By the 1970s the advances in cancer biology and disease understanding led scientist to begin to experiment with ways of combining treatments to improve outcomes, such as less invasive and more targeted treatments. By this time the widespread use of mammography allowed for earlier detection and gave insight into the development and thereby management of breast cancer.

In 1981 an article was published from Veronesi et al[11], in which they compared the Halstead mastectomy with a quadrantectomy, axillary dissection and radiotherapy in patients with small cancers of the breast and found no differences in recurrence or overall survival after initial follow-up of 5 years. Similar results were seen in the National Surgical Adjuvant Breast and Bowel Project (NSABP) B-04 trial by Fisher et al., comparing patients with radical mastectomy with segmental mastectomy, axillary dissection with or without radiotherapy[12]. Breast conserving therapy became the standard for stage I/II breast cancer.

Aside from the advances in surgical treatment, other treatment options emerged during these years. With its origin in the second world war, the 1960s and 70s led to the next big development in breast cancer treatment with the introduction of chemotherapy and hormone therapy.

Current treatment strategies of breast cancer.

The current landscape of breast cancer treatment has changed a lot over the years. Breast cancer is a very heterogeneous disease encompassing an extraordinary diverse group of diseases in terms of presentation, morphology, biology, clinical behaviour and response to therapy. Whereas surgery was the primary treatment in the past, the current treatment is multidisciplinary combining the expertise of oncologic surgeons, radiologists, medical oncologist, radiation oncologist, pathologist, plastic surgeons and specialized nurses. Surgical management of breast cancer has transformed from a radical mutilating procedure to a less invasive and oncologic safe operation with a variety of breast reconstruction options.

Not only breast surgery itself, but also the treatment of the axilla became less invasive over the years. Giuliano et al. first reported the sentinel lymph node procedure in 1994[13]. Nowadays the sentinel lymph node biopsy (SLNB) is the method of choice for axillary staging in patients with clinically node negative disease. It is a safe and accurate method with substantially less post-operative morbidity compared to the traditional axillary lymph node dissection[14–16]. The SLB principle has paved the way for less invasive treatments of the axilla. For SLN negative disease or isolated tumour cells a wait and see management is fully accepted[17], but even for limited disease of the axilla most guidelines indicate

now that further axillary dissection is not necessary to improve axillary control and survival[18,19]. In this era of de-escalation, we feel it is important to be able to select those patients in whom 'less is more', but to keep in mind the very heterogeneous nature of the disease.

Personal experience and considerations for this thesis.

During my journey to become a surgeon, and in particular a surgical oncologist, I was early on involved in the care of breast cancer patients. Learning about the needs of these patients and the uncertainties we encounter as caregivers I became aware on a couple of issues were I felt there were clinical unmet needs. In 2011, during my early intern-ships, my attention was drawn to the consequences of the extend of axillary disease in breast cancer patients with regard to post-operative radiotherapy and reconstruction options. We felt it was important to identify those patients with a low probability of extended axillary nodal disease and the standard use of pre-operative ultrasonography appeared to be a good tool. However this was the same year in which the results of the ACOZOG Z0011[20] were published diminishing the role of axillary lymph node dissection (ALND) and therefore the data we analysed became less relevant and we decided to extend our database first before publication. During our research however, we also became aware of other aspects in breast cancer management we wanted to proceed.

Outline of this thesis.

In this thesis we focused on several aspect of breast cancer, including the heterogeneous nature of the disease and the de-escalation of axillary treatment of breast cancer.

Can we improve prognostication of the individual cancers aside from the standard clinical and pathological variables as age, size, grade and nodal status. **Chapter 2** discusses the value of tumour-stroma ratio in triple negative breast cancer as a parameter to help optimize risk stratification in this subgroup of patients for whom no definite prognostic parameters are available.

Are we able to better stage the axillary lymph nodes aside from physical examination before proceeding to surgery, can we even omit axillary surgery in those patients at low risk of lymph node involvement? Particularly also taking primary cancer parameters into account. **Chapter 3** evaluates the use of ultrasonography of the axilla to identify patients preoperatively with a minimal risk of axillary disease in whom ALND might not be necessary. **Chapter 4** describes the utility and diagnostic accuracy of axillary ultrasonography and ultrasonography with fine-needle aspiration cytology in detecting axillary lymph node metastases in breast cancer patients. In **Chapter 5** we analysed the variables associated with axillary nodal disease and used them to build a nomogram in predicting nodal disease to select those patients with a less than 5% chance of macrometastases of the axilla in whom SLNB might be omitted.

On occasion we encountered a patient with a 'special type' or rare breast cancer and learned that little is known on the clinical outcomes. This brought me to set up a database combining alle the rare cancers, to try and learn more about them. **Chapter 6** reports data on the so called 'special types' in breast cancer to better understand the different characteristics of these rare and strongly heterogeneous group of breast cancers in order to optimize treatment strategies.

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Chapter 2

The prognostic value of tumour-stroma ratio in triple-negative breast cancer.

A.M. Moorman, R. Vink, H.J. Heijmans, J. van der Palen, E.A. Kouwenhoven

European Journal of Surgical Oncology. 2012 Apr;38(4):307-13.

ABSTRACT

Background

Triple-negative cancer constitutes one of the most challenging groups of breast cancer given its aggressive clinical behaviour, poor outcome and lack of targeted therapy. Until now, profiling techniques have not been able to distinguish between patients with a good and poor outcome. Recent studies on tumour stroma, found it to play an important role in tumour growth and progression

Objective

To evaluate the prognostic value of the tumour-stroma ratio (TSR) in triple-negative breast cancer.

Methods

One hundred twenty four consecutive triple negative breast cancer patients treated in our hospital were selected and evaluated. For each patient the Haematoxylin-Eosin (H&E) stained histological sections were evaluated for percentage of stroma. Patients with less than 50% stroma were classified as stroma-low and patients with ≥ 50% stroma were classified as stroma-high.

Results

Of 124 triple-negative breast cancer patients, 40% had a stroma-high and 60% had a stroma-low tumour. TSR was assessed by two investigators (kappa 0,74). The 5-years relapse-free period (RFP) and overall survival (OS) were 85% and 89% in the stroma-low and 45% and 65% in the stroma-high group. In a multivariate cox-regression analysis, stroma amount remained an independent prognostic variable for RFP (HR 2.39; 95% CI 1.07-5.29; p=0.033) and OS (HR 3.00; 95% CI 1.08-8.32; 0.034).

Conclusion

TSR is a strong independent prognostic variable in triple-negative breast cancer. It is simple to determine, reproducible and can be easily incorporated into routine histological examination. This parameter can help optimize risk stratification and might lead to future targeted therapies.

INTRODUCTION

Breast cancer is the most frequently diagnosed cancer in women worldwide with 1.4 million new cases and 458 400 deaths in 2008[1]. While these numbers are alarming, we must keep in mind that breast cancer is a heterogeneous disease encompassing an extraordinarily diverse group of diseases in term of presentation, morphology, biology, clinical behavior and response to therapy[2]. Focusing on treatment options, breast cancer patients fall into three main groups: 1) patients with hormone-receptor positive tumours (the Luminal A and B); 2) the Her2 positive patients; and 3) those patients with hormone receptor negative breast cancer. Worse outcomes are traditionally seen among woman with triple-negative breast cancer, which accounts for 10-17% of all breast carcinomas[3–12]. They primarily affect younger women, are more prevalent in African-American women, often present as interval cancers and are significantly more aggressive than tumours of the other molecular subtypes[3–5,13]. The peak risk of recurrence is between the first and third year following therapy and the majority of deaths occur in the first 5 years following therapy[4]. Triple-negative cancer represents one of the most challenging groups of breast cancer that currently lacks the benefit of a targeted therapy[11]. Molecular profiling techniques and prognostic algorithms, like Adjuvant Online, are unable to distinguish patients with low and high risk profiles[8,14–16]. In an attempt to make a more accurate assessment, we focused on the complex tumour microenvironment.

Recently, evidence suggests that the tumour-associated stroma and cancer-associated fibroblasts (CAF) may play an important role in tumour growth, angiogenesis and progression[17–19]. Stroma is the connective tissue that supports the deeper layer of breast tissue and if normal can in fact be protective in delaying or preventing tumour formation. In case of an invasive carcinoma, the epithelium has changed genetically, and as a result the stromal changes creating a permissive and supportive environment for tumour growth[20,21]. In more advanced stages the reactive stroma even stimulates invasion and metastases which inevitably results in diminished overall survival and relapse-free period[22]

The amount of stroma has only recently been linked to a worse prognosis in cancer in a few studies. In a series of 122 colon cancer patients, the carcinoma-stromal composition appeared to be an independent prognostic variable. Patients with a high percentage of stroma had a worse overall survival and disease free period[23,24]. In a subsequent investigation of adenocarcinoma of the oesophagus[25] and in breast cancer patients[26] tumour-stroma ratio (TSR) also proved to be a significant prognostic variable.

The purpose of this study was to determine whether the amount of stroma is of prognostic value in triple-negative breast cancer. If the stromal component is indeed of prognostic value in this subgroup of breast cancer patients, it will not only be a candidate parameter for prognostication, but might also lead to the subsequent development of therapies targeting the stromal components.

METHODS

Patient enrolment

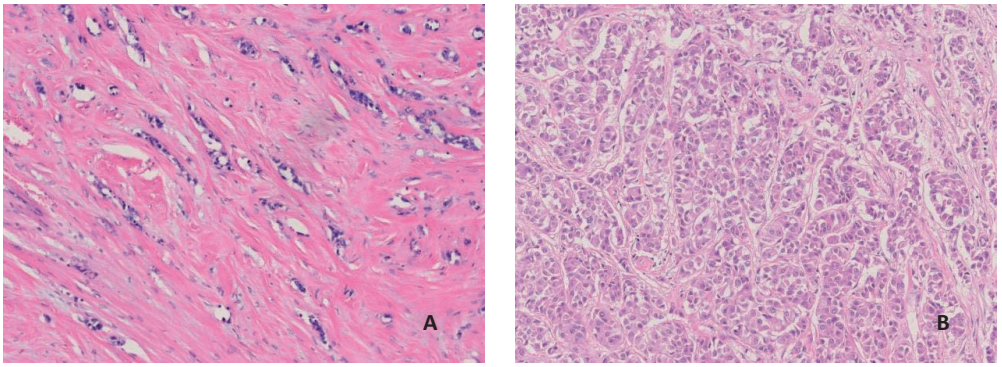
This was a retrospective cohort study. During the period of January 2004-2008 all patients with triple-negative primary breast cancer who underwent surgery at the Hospital Group Twente, location Almelo and Hengelo, were selected. Hormone status was retrieved from the original patient files. Expression of oestrogen (ER), progesterone (PR) and human epidermal growth factor receptor 2 gene (HER2) were pre-determined by immunohistochemistry on formalin-fixed paraffin-embedded tumour material according to standard diagnostic procedure. Patients treated with neo-adjuvant therapy were excluded, since accurate evaluation of the tumour-stroma ratio was not possible in the final pathology. In case of known distant metastases at the time of surgery or recurrence within one month and patients with other malignancies at the time of presentation were also excluded.

Histopathology

The H&E stained sections from the primary tumour in the surgical specimen of all patients, were retrieved from the Pathology Laboratory East Netherlands. All samples were handled in a coded fashion, according to the National ethical guidelines ('Code for Proper Secondary Use of Human Tissue', Dutch federation of Medical Scientific Societies).

All pathological specimens were independently scored by two investigators [Moorman; Vink], who were not aware of the status of the patient. Slides from 128 resected tumours, varying from 1 section to up to 20 sections per tumour, were evaluated. The amount of stroma was quantified using a 5 x objective lens to select the most invasive part of the tumour, then the 10 x objective lens was used to score. Only fields were scored where both stroma and tumour cells were present, tumour cells had to be seen on all sides of the microscopic image field. In case of tumour heterogeneity, those areas with the highest stromal percentage were decisive (see figure 1).

Figure 1. H&E stained sections of primary breast tumours.



(a) tumour with large amounts of stroma, estimated as 80% with 10x objective; (b) tumour with low amount of stroma (30% with 10x objective).

Follow-up

Follow-up data was collected until March 2011. Overall survival (OS) was defined as the time between primary surgery and death or last follow-up. Relapse-free survival (RFS) was defined as the time between primary surgery and the first recurrence, metastases, death or until date of last follow-up.

Statistics

Statistical analysis was performed using SPSS software version 17.0. The stroma was scored per tenfold percentage. A 50% cut-off was used as previously determined in colon- and breast cancer by maximum discriminative power, which was also confirmed in our breast cancer population [24,26]. Stroma-low was defined as < 50% stroma, and stroma-high as ≥ 50% stroma. The relationship between TSR (high versus low) and categorical data was assessed using the chi-square test or Fisher's exact test and the T-test or Mann-Whitney U test was used for continuous variables, depending on the distribution of the data. Variables included in multivariate analysis were the variables both related to TSR and to the outcome under investigation (both $p \leq 0.15$). Interobserver variability was analyzed using Cohen's kappa coefficient.

Analysis of the survival curves was performed using the Kaplan-Meier method and differences in survival distribution were tested with the Log Rank Statistic. The Cox proportional hazards model was used to determine the hazard ratio (HR) of explanatory variables on overall survival and relapse-free period. The results are given as hazard ratios with the 95% confidence interval (CI). P-values < 0,05 were considered statistically significant.

RESULTS

Patient demographics

One hundred twenty four consecutive triple-negative breast cancer patients were selected for this study. The mean age of patients at the time of surgery was 56 years (range 23-87). The median time of follow-up was 37 months (4-84 months). A total of 25 patients died during this study. Nine patients were still alive with disease at the time of last follow-up. Patient and tumour characteristics are listed in table 1.

Table 1. Patient, tumour, treatment and outcome characteristics grouped by tumour stroma ratio (TSR).

	Stroma < 50% (n=74) No. pt. (%)	Stroma ≥ 50% (n=50) No. pt. (%)	Chi-square test P-value
Age (y)			
< 50	31 (41.9)	13 (26.0)	0.070
≥ 50	43 (58.1)	37 (74.0)	
Palpable tumour			
No	16 (21.6)	11 (22.0)	0.991
Yes	57 (77.0)	39 (78.0)	
unknown	1 (1.4)	0 (0.0)	
Operation type			
Breast conserving surgery	38 (51.4)	22 (44.0)	0.422
Total mastectomy	36 (48.6)	28 (56.0)	
Histological type			
IDC	65 (87.8)	42 (84.0)	0.431
ILC	2 (2.7)	4 (7.8)	
Others	7 (9.3)	4 (7.8)	
Pathologic tumour stage			
pT 1	32 (43.2)	17 (34.0)	0.515
pT2	37 (50.0)	30 (60.0)	
pT3 of 4	5 (6.8)	2 (4.0)	
unknown	0 (0.0)	1 (2.0)	
Pathologic tumour grade			
1 (well)	0 (0.0)	2 (4.0)	0.024
2 (moderate)	10 (13.5)	13 (26.0)	
3 (poorly)	64 (86.5)	34 (68.0)	
unknown	0 (0.0)	1 (2.0)	
Nodal status			
pN0	52 (70.3)	26 (52.0)	0.075
pN1	19 (25.7)	18 (36.0)	
pN2 or 3	3 (4.1)	6 (12.0)	
Family history			
Negative	8 (10.8)	12 (25.0)	0.100
Positive	22 (29.7)	10 (20.8)	
Unknown	44 (59.5)	26 (54.2)	
Extracapsular extension			
No	53 (96.4)	32 (88.9)	0.209

Yes	2 (3.6)	4 (11.1)	
Multifocality			
No	70 (94.6)	39 (78.0)	0.005
Yes	4 (5.4)	11 (22.0)	
Lymphovascular invasion (LVI)			
No	63 (85.1)	28 (56.0)	≤ 0.001
Yes	11 (14.9)	22 (44.0)	
Tumor free margin			
No	72 (97.3)	47 (94.0)	0.392
Yes	2 (2.7)	3 (6.0)	
Presence Ductal carcinoma in situ	47 (63.5)	23 (46.0)	0.054
No	27 (36.5)	27 (54.0)	
Yes			
Postmenopausal			
No	28 (38.4)	13 (27.1)	0.225
Yes	42 (57.5)	32 (66.7)	
Unknown	3 (4.1)	3 (6.3)	
Necrosis			
Absent	21 (28.4)	25 (50.0)	0.004
< 30% necrosis	36 (48.6)	23 (46.0)	
≥ 30% necrosis	17 (23.0)	2 (4.0)	
Mitotic activity index (MAI)			
0-19/2 mm^2	27 (37.0)	22 (46.8)	0.059
20-39/2 mm^2	27 (37.0)	21 (44.7)	
>39/2 mm^2	19 (26.0)	4 (8.5)	
Local therapy			
BCS – Radiotherapy	3 (4.1)	5 (10.0)	0.278
BCS + Radiotherapy	34 (45.9)	17 (34.0)	
MST – radiotherapy	29 (39.2)	10 (20.0)	
MST + radiotherapy	7 (9.9)	1 (2.0)	
Chemotherapy			
No	24 (32.4)	18 (36.0)	0.720
Yes	49 (66.2)	32 (64.0)	
unknown	1 (1.4)	0 (0.0)	
Event relapse			
No	64 (86.5)	30 (60.0)	0.001
Yes	10 (13.5)	20 (40.0)	
Death of disease			
No	67 (90.5)	36 (72.0)	0.006
Yes	7 (9.5)	14 (28.0)	

Abbreviations: Event relapse defined as recurrence, distant metastasis or death. BCS: breast conserving therapy; MST: mastectomy.

Histopathology

TSR was assessed by two investigators. In 12 cases (9,5%) there was no agreement in TSR at first individual assessment (kappa 0,74; 90% concordance in classification): after re-evaluation by both investigators together there was total agreement.

Correlations TSR with prognosis

Fifty patients (40%) were classified as stroma-high (≥ 50% stroma) and 74 patients (60%) were classified as stroma-low (< 50% stroma). In the stroma-low group 13,5% of patients (10/74) had a relapse and 9,5% (7/74) died of breast cancer during follow-up. In the stroma-high group 40% (20/50) had a relapse and 28,0% (14/50) died of metastasized disease following a relapse (both p≤0.006). Treatment and outcome characteristics are shown in table 1. The 5-years RFP and OS were 85% and 89%, respectively, in the stroma-low group and 45% and 65%, respectively in the stroma-high group. Survival analyses showed that stroma-high patients had a significantly worse RFP (HR 2.93; 95% CI 1.37-6.26; p=0.004) and OS (HR 2.56; 95% CI 1.03-6.35; p=0.035) compared to stroma-low patients (figure 2a/b).

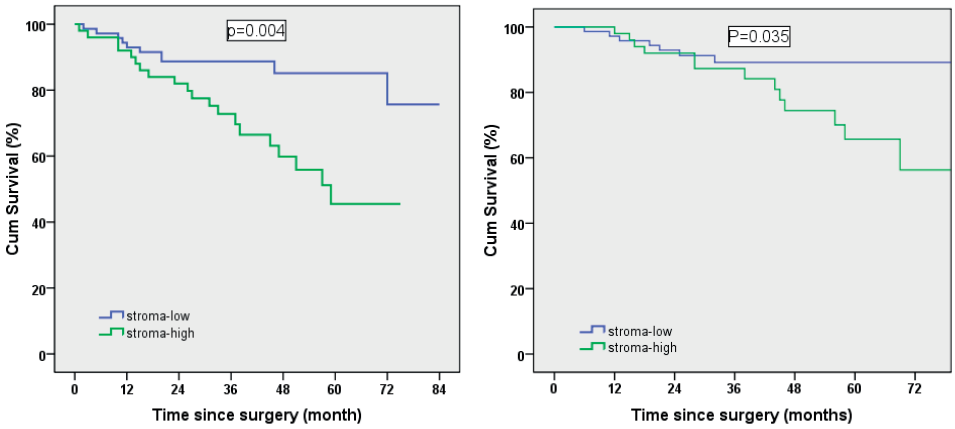


Figure 2 Kaplan-Meier curves for tumour-stroma ratio. Patients with stroma-high showed a significant worse relapse free period, RFP **(a)** and overall survival, OS **(b).**

Age, nodal status, family history, multifocality, Lymph vascular invasion (LVI), ductal carcinoma in situ, necrosis and Mitotic Activity Index (MAI) were all related to TSR (all p<0,15; see Table 1). In univariate Cox-regression analysis, nodal status, multifocality and LVI were also significantly related to RFP, as were nodal status, multifocality and necrosis to OS (all p<0.15; see table 2). In a multivariate Cox-regression analysis TSR remained an independent prognostic variable for both RFP (HR 2.39; 95% CI 1.07-5.29; p=0.033) and OS (HR 3.00; 95% CI 1.08-8.32; 0.034). Multifocality remained an independent prognostic variable for RFP. For overall survival, nodal status and the presence of necrosis were

independent prognostic variables (see table 2). Multifocality showed a trend towards a worse survival, but was not significant in OS.

Table 2 Univariate cox regression analysis for RFP and OS for the variables significantly related to tumor stroma ratio (p<0.15), and below the multivariate cox regression analysis.

	Univariate analysis					
	Relapse free period (RFP)			Overall survival (OS)		
	HR	95% CI	P-value	HR	95% CI	P-value
TSR						
Stroma-low	1.00			1.00		
Stroma-high	2.93	1.37-6.26	0.004	2.56	1.03-6.35	0.035
TSR*chemo						
Stroma-low	1.00			1.00		
Stroma-high	2.73	1.27-5.86	0.010	2.31	0.929-5.75	0.072
Chemotherapy						
Yes	1.00	0.86-3.70		1.00		
No	1.79		0.120	3.71	1.50-9.18	0.005
Age						
≤50 years	1.00			1.00		
> 50 years	1.72	0.76-3.87	0.182	1.79	0.65-4.92	0.253
pN status			0.006			0.011
pN0	1.00			1.00		
pN1	1.43	0.68-3.01	0.338	2.09	0.86-5.07	0.092
pN2/3	3.38	1.27-9.00	0.010	2.62	0.76-9.00	0.111
Family history						
Negative	1.00			1.00		
Positive	1.06	0.96-1.16	0.210	1.08	0.95-1.22	0.186
Multifocality						
No	1.00			1.00		
Yes	3.39	1.54-7.47	0.001	3.16	1.25-8.00	0.011
LVI						
No	1.00			1.00		
Yes	2.461	1.19-5.07	0.012	1.57	0.62-3.95	0.326
In situ comp						
No	1.00			1.00		
Yes	1.34	0.65-2.75	0.416	1.45	0.60-3.51	0.401
Necrosis			0.726			0.142
Absent	1.00			1.00		
<30% necrosis	0.648	0.31-1.34	0.242	0.494	0.19-1.22	0.121
≥30% necrosis	1.28	0.49-3.36	0.608	2.35	0.85-6.51	0.088
MAI			0.547			0.769
0-19/2 mm2	1.00			1.00		
20-39/2 mm2	0.842	0.29-2.43	0.751	0.77	0.30-1.96	0.584
>39/2 mm 2	0.85	0.40-1.83	0.693	1.43	0.473-4.37	0.519
Multivariate analysis						
TSR						
≤50%	1.00			1.00		
>50%	2.39	1.07-5.29	0.033	3.00	1.08-8.32	0.034

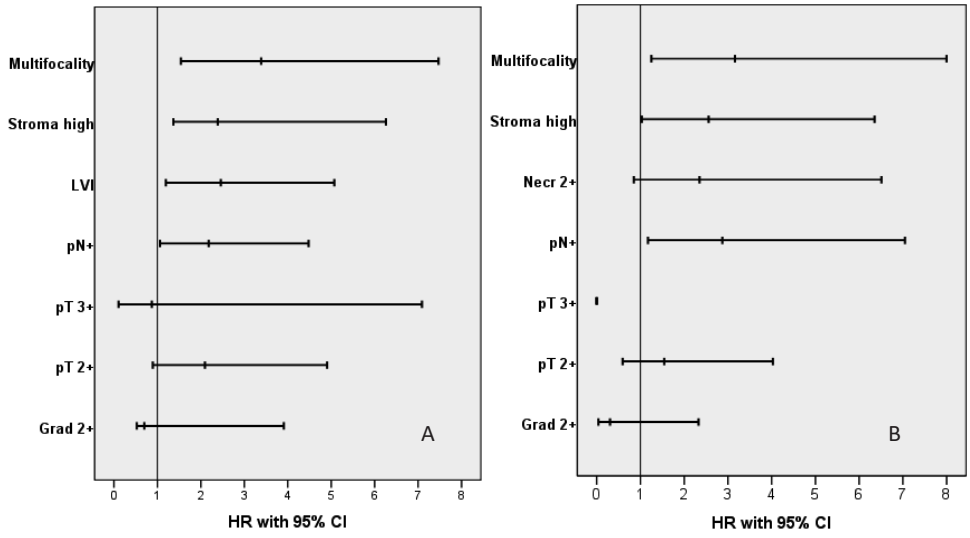
Multifocality						
No	1.00					
Yes	2.47	1.08-5.66	0.032			
pN-status				1.00		0.054
pN1				3.22	1.13-9.14	0.028,
pN2 and pN3				4.24	0.97-18.56	0.055
Necrosis				1.00		0.016,
<30% necrosis				0.86	0.30-2.41	0.774,
≥30% necrosis				5.37	1.37-21.02	0.016

Abbreviations: HR: hazard ratio; CI: confidence interval; TSR: tumour stroma ratio; TSR*chemo: analyses adjusted for chemotherapy; pN: pathological nodal status; LVI: lymph vascular invasion; MAI: mitotic activity index.

Strength of tumour-stroma ratio

The strength of the TSR is best illustrated, when compared to the routinely used variables for treatment strategies nowadays like multifocality, LVI and necrosis. The hazard ratios are respectively 2.39 (95% CI; 1.37-6.26) for stroma-high versus stroma-low, 2.18 (95% CI; 1.06-4.48) for nodal status pN1+ versus pN0, 2.10 (95% CI 0.89-4.91) for tumour size pT≥2 versus pT1 and 0.53 (95% CI 0.70-3.91) for tumour grade 2 or 3 versus tumour grade 1 for RFP. Similarly, for OS the HR for stroma high is 2.56 (1.03-6.30), for nodal status 2.87 (1.17-7.05), 1.54 for tumour size (0.59-4.03) and 0.31 (0.04-2.33) for tumour grade (Fig. 3).

Figure 3 Strength of the tumour-stroma ratio, Multifocality and LVI/necrosis compared to the routinely used variables nodal status, tumour size and tumour grade on univariate cox regression analysis for relapse free period (a) and overall survival (b).



Abbreviations: HR: hazard ratio; CI: confidence interval; LVI: lymph vascular invasion; Necr 2+:≥ 30% tumour necrosis; pN+: positive nodes on pathology; pT3+: Tumour size ≥ 20mm; pT2+: tumour size ≥ 10mm; Grad 2+: tumour grade 2 or 3.

*statistics for pT3 for overall survival were not possible, since no events occurred in that subgroup.

Effect modification of chemotherapy

We formally investigated effect modification by chemotherapy in a multivariate cox regression with TSR, chemo and the interaction term TSR*chemo. The interaction was not statistically significant (p=0.32).

DISCUSSION

Our study shows that tumour-stroma ratio is an independent prognostic variable for patients with triple-negative breast cancer. Patients with a stroma-high tumour had a significantly worse relapse free period (RFP) and overall survival (OS) in comparison with patients with stroma-low tumours. These results correspond to those found in other studies that investigated the TSR in cancer patients. Just recently, de Kruijf et al [26] found TSR to be a significant prognostic variable for RFP (HR 1.87) in triple-negative breast cancers. Their results are similar to those found in our study. For stage I-II colon cancer, the TSR also discriminated between patients with a poor and a better outcome[23], which was further validated in a subsequent study[24]. A comparable study in oesophageal cancer gave similar results, with a hazard ratio of 2.00 for overall survival and 1.55 for RFP of stroma-high tumours compared to stroma-low[25].

Aside from TSR, multifocality also appeared to be an independent prognostic variable for RFP, as were nodal status and presence of necrosis for OS. Compared to these variables and others, TSR proved to be a strong indicator. Despite being a relatively new variable, which only recently has been studied in cancer patients, it seems promising.

Determination of Tumour-stroma ratio

Determination of TSR proved to be a relatively quick and simple procedure that can easily be included in the routine pathological examination. It can be done on routine H&E sections without the necessity for further staining. The interobserver agreement kappa value was high (0.74). In the cases without agreement at first assessment, almost all involved tumours with extensive central sclerosis leaving little tumour margin to evaluate. Despite this, the kappa was still substantial. Other studies also prove that it is a reproducible method. For colorectal cancer, the interobserver agreement varied between 0.60 and 0.70 [23,24], in oesophageal cancer the kappa was 0.86 [25] and in a previous breast cancer study the kappa was 0.85 [26]. In addition to being quick, simple and reproducible, it does not lead to additional costs [24].

Risk stratification

The purpose of performing a risk assessment in breast cancer patients, is to differentiate between patients with good and poor prognosis, ultimately allowing for optimal therapy decisions. Various classification systems are available nowadays to estimate the risk for locoregional relapse, distant metastasis and death in breast cancer patients. Most common are the Nottingham Prognostic Index [27], the Sankt Gallen classification[28] and Adjuvant Online[29]. The last one is the most commonly used and has the advantage of giving an estimate of the survival benefit and prevention of relapse given the standard therapy[30]. The estimations have proven to be fairly accurate, except for certain subtypes including our own[31]. A recent phase II study on Poly ADP ribose polymerase (PARP) inhibitors reported significant improvements in response rate and RFP[32], but no results are available from subsequent studies. To this day, there are no specific guidelines for triple-negative breast cancer. Since these tumours commonly have an aggressive clinical behavior and lack the benefit of a targeted therapy, it is a subgroup of great interest. Since triple-negative breast cancer constitutes a heterogeneous group with different pathological and clinical features[11], a better understanding of these features might enable us to better select patients for future specific therapies.

Tumour micro-environment

In order to understand the growth and progression of cancer, research has focused on the complex microenvironment of the tumour. One of the components of the microenvironment is the stroma, the connective tissue of the breast [19,33–36]. Stroma is thought to promote tumour activity by multiple mechanisms including an increased number of fibroblasts, manipulation of the extracellular matrix, enhanced capillary density, recruitment of inflammatory cells, and alterations in stromal regulatory pathways[37,38]. The overall effect of these mechanisms is still not fully understood, but it strongly suggests that tissue architecture is a significant participant in tumour growth and progression. Especially the so-called cancer-associated fibroblasts (CAFs) were found to have a predominant role in tumour growth and progression[38–40]. This was also found in genetic mouse models, where the contribution of stromal fibroblasts led to tumour initiation and progression[41,42]. Another study investigated cultured primary breast epithelial cells in combination with stromal elements. The addition of the stromal elements caused the tumour to spread and become invasive, with a proportional effect on tumour growth with increasing concentrations of stromal elements[37]. Focusing on the stroma might therefore lead to better prognostication in cancer patients and provide new targets for therapy.

As far as we know, our study is the second performed in breast cancer patients, and in particular triple-negative breast cancer. More studies and larger study populations are necessary to further validate this parameter. Other variables, like growth pattern and lymphocytic infiltrate might also be of influence on the prognosis and TSR. Though, before these parameters can be properly evaluated, better definitions and assessment strategies are warranted. Current strategies are too vulnerable to subjectivity.

CONCLUSION

TSR is a strong independent prognostic variable in triple-negative breast cancer. Patients with a stroma-rich tumour were found to have an almost 2.5 fold increased chance of relapse or distant metastasis and a 3 fold increased chance of death when compared to patients with a stroma-low tumour. The TSR is easy to determine, reproducible and does not lead to additional costs. It can easily be incorporated into routine histological examination. This parameter can help optimize risk stratification in this subgroup of patients, where no definite prognostic parameters are available yet.

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Chapter 3

Axillary ultrasonography in breast cancer patients helps in identifying patients pre-operatively with limited disease of the axilla.

A.M. Moorman, R.L.J.H. Bourez, H.J. Heijmans, E.A. Kouwenhoven.

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ABSTRACT

Background

The sentinel lymph node biopsy (SLNB) procedure is the method of choice for the identification and monitoring of regional lymph node metastases in patients with breast cancer. In the case of a positive SLN, additional lymph node dissection is still warranted for regional control, although 40-65% have no additional axillary disease. Recent studies showed that after breast-conserving surgery, SLNB and adjuvant systemic therapy there is no significant difference between recurrence-free period and overall survival if there are ≤ 2 positive axillary nodes.

Purpose

Preoperative identification of patients with limited axillary disease (≤ 2 macrometastases) using ultrasonography (US).

Methods

Data from 1103 consecutive primary breast cancer patients with tumours smaller than 50 mm, no palpable adenopathy and a maximum of 2 SLNs with macrometastases were collected. The variable of interest was ultrasonography of the axilla.

Results

Of the 1103 patients included, 1060 remained after exclusion criteria. Of these, 102 (9.6%) had more than 2 positive axillary nodes on ALND. Selected by unsuspected US, the chance of having > 2 positive LN is substantially lower (4.2%). This is significant on univariate and multivariate analysis. After excluding the patients with ECE of the SLN, the chance of having > 2 positive LNs is only 2.6%. For pT1-2 this is 2.2%.

Conclusion

The risk of more than 2 positive axillary nodes is relatively small in patients with cT1-2 breast cancer. Ultrasonography of the axilla helps in further identifying patients with a minimal risk of additional axillary disease, putting ALND up for discussion.

INTRODUCTION

Sentinel lymph node biopsy (SLNB) has revolutionized the management of clinically node-negative women with breast cancer. It is a safe and accurate method for axillary staging and it causes substantially less post-operative morbidity than axillary lymph node dissection (ALND)[1–5]. The recommended management for patients with sentinel lymph node (SLN) metastases is still ALND in cases of SLN metastases larger than 0.2mm[6]. However, the need for ALND has recently been questioned [7–18] since the SLN has shown to be the only positive lymph node in 40% to 65% of these patients [12,19–24]. For these patients ALND offers no additional diagnostic, prognostic or therapeutic benefit, while subjecting them to a significant risk of additional morbidity. The incidence of nodal metastases is lower since the introduction of routine screening mammography[25]. The widespread use of chemotherapy, radiation therapy and endocrine therapy may also diminish the added benefit of ALND [14]. In addition, the AMAROS trial showed that the absence of knowledge of axillary status did not modify postoperative treatment planning[10].

A number of reports have suggested that in selected patients with SLN metastases ALND may be omitted [8,13,15,26]. Numerous mathematical models have been developed to predict non-SLN disease in patients with SLN metastases. Validation studies have demonstrated a reasonable accuracy, although limited [27–34].

This led to the development of the American College of Surgeons Oncology Group (ACOSOG) Z0011 phase III trial, in which patients with nodal metastases were randomly assigned to ALND or no ALND to assess differences in axillary recurrence and survival. They concluded that ALND did not significantly affect overall or disease-free survival in patients with clinical T1-2 breast cancer, no palpable adenopathy and a maximum of 2 SLN metastases, who were treated with lumpectomy, adjuvant systemic therapy and tangential-field whole breast radiation therapy[35]. In this study patients underwent operation before inclusion since the SLN status was known. Plus, the ACOSOG trial did not routinely perform an axillary ultrasonography. Our main interest was to try selecting those patients with a better prognosis before operation.

The aim of this study was to determine if we are able to identify patients pre-operatively (so even before SLNB) with limited disease of the axilla using axillary ultrasonography. In recent literature and also recommended by the current Dutch Guidelines[6], it appears safe to avoid ALND in patients with a

maximum of 2 positive SLNs, when they undergo breast conserving therapy and adjuvant systemic therapy[6,35–37]. So as cut-off point we used the value of 2 positive lymph nodes.

METHODS

Patients and procedures

This study is a retrospective analysis. Between January 2007 and August 2011, data from 1103 consecutive primary breast cancer patients who underwent surgery in our hospital were collected into a single database. Patients with clinically apparent nodal involvement, tumours clinically larger than 5 cm, known distant metastases and patients who underwent neo-adjuvant therapy were excluded. All patients underwent a routine mammography, ultrasonography of the breast and ipsilateral ultrasonography of the axilla. Suspected axillary lymph nodes (thickness of cortex > 2.3mm), were further analysed by US guided fine needle aspiration cytology (FNAC). In case of a positive US guided FNAC (i.e. malignant cells on cytology), an ALND was performed. Patients with unsuspected axillary US and those with negative US guided FNAC on pathology or insufficient material for diagnosis, were scheduled for SLNB.

Patients underwent breast-conserving surgery or mastectomy, a decision made by the attending physician. SLNs were harvested after Scintigraphy and Patent Blue Dye injection during or prior to surgery. A sentinel node was identified as any blue staining node, hot node or node with at least 10% of the highest hot node count. Pathologic examination of the SLN was classified as macrometastases (>2 mm), micrometastases (0,2-2 mm), or isolated tumour cells (<0,2 mm). Complete ALND was routinely performed when one micro- or macrometastases was present in the SLN according to the Dutch Guidelines.

Patients and tumour characteristics were retrieved from the original patient files and are listed in table 1. We focused on the association between having more than two positive lymph nodes and the patients and tumour characteristics. In particularly within the selection of patients with clinical tumours smaller than 50mm, no palpable adenopathy and a maximum of 2 SLNs containing macrometastases identified by frozen section, touch preparation, or hematoxylin-eosin staining on permanent section. With the exception of a positive SLN, this is the same selection of patients previously investigated by Giuliano[35,37]. Patients with radiological unsuspected axillary lymph nodes formed the cohort of interest in this study and were further analysed. Patients with negative pathology after initially suspected axillary US are not included.

Statistical analyses

Statistical analysis was performed using SPSS software version 17.0. The relationship between having more than two positive lymph nodes on SLNB or ALND and categorical data was assessed using the chi-square test or Fisher's exact test and the T-test or Mann-Whitney U test was used for continuous variables, depending on the distribution of the data. Variables included in multivariate analysis were those significant in univariate analyses ($p < 0.15$). The Cox proportional hazards model was used to determine the hazard ratio (HR) of explanatory variables on having more than two positive lymph nodes. The results are given as hazard ratios with the 95% confidence interval (CI). P-value < 0.05 were considered statistically significant.

RESULTS

Patients and tumour characteristics

Among the 1103 patients, 127 had more than two positive lymph nodes (11.5%) with SLNB or ALND. Selected by clinical tumour size smaller than 50mm, a maximum of 2 positive lymph nodes with SLNB and no palpable adenopathy (initial selection), a group of 1060 patients remained and was further analysed. The number of patients having more than two positive lymph nodes was 102 (9.6%). Mean patient age at time of surgery was 61 years (range 24-93). Patient and tumour characteristics are listed in table 1. Patients with more than 2 positive lymph nodes were more likely to be younger, 32.4% of the women were under the age of 50 compared with 16.3% in the 2 or less positive lymph nodes group. They also had larger tumours both clinical and pathological, 18.6% had a clinical T2 tumour compared with only 1.3% T2 tumours in the 2 or less positive lymph nodes group. Lymph vascular invasion (LVI), multifocality, histological grade, axillary ultrasonography (US), number of SLNs and extra capsular extension (ECE) of the SLN were all significantly different between both groups. Histology and tumour receptor status were not significantly different between both groups. As mentioned in the methods section, US of the axilla was the variable of interest. The ultrasonography was negative in 842/958 (87.9%) of the patients in the 2 or less positive lymph nodes group, compared to 37/102 (36.3%) in the other group. This difference was significant in univariate and multivariate analysis.

Table 1. Baseline patient and tumour characteristics.

	≤ 2 positive LNs (*n*= 958) *n* (%)	**> 2 positive LNs (*n*=102), *n* (%)**	***p*-value < 0.15**
Age (years)			
< 50	156 (16.3)	33 (32.4)	< 0.001
≥ 50	802 (83.7)	69 (67.6)	
Palpable tumour			

No	417 (43.7)	15 (14.7)	< 0.001
Yes	538 (56.3)	87 (85.3)	
Clinical T-stage rounded up			
1a-c	675 (70.5)	35 (34.3)	< 0.001
2	283 (29.5)	67 (65.7)	
Ultrasonography of the axilla	842 (87.9)	37 (36.3)	< 0.001
Negative	116 (12.1)	65 (63.7)	
Positive			
Operation type			
Excision biopsy	1 (0.1)	0 (0.0)	< 0.001
BCT+SLNB	528 (55.1)	18 (17.6)	
BCT+ALND	10 (1.0)	12 (11.8)	
MST+SLNB	389 (40.6)	25 (24.5)	
MST+ALND	30 (3.1)	47 (46.1)	
ECE SLNB			
No	940 (98.2)	79 (78.2)	< 0.001
Yes	17 (1.8)	22 (21.8)	
Number of macrometastases with SLNB			
1	111 (90.2)	79 (78.2)	< 0.001
2	12 (9.8)	22 (21.8)	
Histology			
IDC	775 (80.9)	91 (89.2)	0.254
ILC	113 (11.8)	7 (6.9)	
Ducto-lobular	14 (1.5)	1 (1.0)	
Other	56 (5.8)	3 (2.9)	
Histologic grade			
1	296 (31.2)	15 (14.7)	< 0.001
2	435 (45.8)	42 (41.2)	
3	219 (23.1)	45 (44.1)	
Multifocality			
No	848 (88.5)	84 (82.4)	0.078
Yes	110 (11.5)	18 (17.6)	
LVI			
No	851 (88.8)	60 (58.8)	< 0.001
Yes	107 (11.2)	42 (41.2)	
Radicality			
Yes	876 (91.4)	95 (93.1)	0.586
No	82 (8.6)	7 (6.9)	
ER			
Negative	151 (15.8)	17 (16.7)	0.879
Positive	801 (83.6)	85 (83.3)	
Missing	6 (0.6)	0 (0.0)	
PR			
Negative	281 (29.3)	32 (31.4)	0.879
Positive	671 (70.0)	70 (68.6)	
Missing	6 (0.6)	0 (0.0)	
Her2/Neu			
Negative	860 (89.8)	88 (86.3)	0.361
Positive	92 (9.6)	14 (13.7)	
Missing	6 (0.6)	0 (0.0)	
Postmenopausal			
Pre	181 (18.9)	34 (33.3)	< 0.001
Post	745 (77.8)	65 (63.8)	

Missing	32 (3.3)	3 (2.9)	
Pathologic T-stage			
1a	28 (2.9)	0 (0.0)	< 0.001
1b	151 (15.8)	0 (0.0)	
1c	448 (46.8)	24 (23.5)	
2	308 (32.2)	64 (62.7)	
3	22 (2.3)	13 (12.7)	
4	1 (0.1)	1 (1.0)	
Pathologic T-stage rounded up			
1a-c	627 (65.4)	24 (23.5)	< 0.001
2	308 (32.2)	64 (62.7)	
3	22 (2.3)	13 (12.7)	
4	1 (0.1)	1 (1.0)	

Abbreviations: LN, total number of lymph nodes either by SLN or ALND; BCT, breast conserving therapy; MST, mastectomy, SLNB, sentinel lymph node biopsy; ALND, additional lymph node dissection; ECE, extra capsular extension; LVI, lymph vascular invasion; ER, oestrogen receptor; PR, progesterone receptor;

Selection by axillary ultrasonography (US)

The group of interest in this study were those patients with unsuspected axillary lymph nodes during US alone (n=879). Of these patients 37 had more than 2 positive lymph nodes on SLND or ALND. That is 4.2%, less than half the value of 9.6% in the unselected group. Patients and tumour characteristics within this selected population are shown in table 2. Patients with more than 2 positive lymph nodes are again more likely to be younger, have palpable tumours during clinical examination of the breast, have a tumour larger than 20mm, have ECE of the SLN, LVI of the tumour and the patients are more likely to be premenopausal.

On univariate analyses especially ECE of SLN and pathological tumour size larger than 50mm were of great value, with respectively HR of 43.962 (95% CI; 18.214-105.950, p=0.000) and 13.988 (95% CI; 3992-49.015, p=0.000). These also remained significant when analysis was adjusted for differences in clinical pathologic variables with multivariate analysis, see table 2.

Table 2 Univariable and multivariable associations of patient and tumour characteristics and having more than 2 positive lymph nodes.

	Univariable *p*-value	HR (95% CI)	Multivariable *p*-value	HR (95% CI)
Age (years)				
< 50	< 0.001	4.098 (2.084-8.060)	-	-
≥ 50				
Palpable tumour				
No	< 0.001	2.741 (1.278-5.880)	0.614	1.305 (0.463-3.678)
Yes				
Clinical T-stage				
1a-c	< 0.001	4.732 (2.392-9.359)	0.004	3.847 (1.546-9.575)

2				
ECE SLNB				
No	< 0.001	43.962 (18.241-	<0.001	54.157 (18.797-156.038)
Yes		105.950)		
LVI				
No	< 0.001	4.416 (2.174-8.971)	0.137	1.981 (0.804-4.880)
Yes				
Postmenopausal				
Pre	< 0.001	3.767 (1.892-7.498)	0.013	3.031 (1.266-7.261)
Post				
Pathologic T-stage				
1a-c	< 0.001	4.616 (2.205-9.663)		
2			-	-
3	< 0.001	13.988 (3.992-49.015)		

Abbreviations: HR; hazard ratio with 95% confidence interval. ECE; extra capsular extension SLNB. LVI; lymph vascular invasion.

Further selection by excluding ECE of the SLN

Selected by unsuspected axillary US, the chance of having more than 2 positive lymph nodes is substantially lower (4.2%) than in the unselected population. When we select this subgroup even further by excluding the patients with ECE on SLN, the chance of more than 2 positive lymph nodes is only 2.6% (22/851). Subdivided by clinical tumour size (see table 3) the chance of more than 2 positive lymph nodes is 0.96% in case of cT1 tumour and 7.0% in case of a cT2 tumour. For pathological T-status the numbers are respectively 0.87% for pT1, 5.0% for pT2 and 22.2% for pT3 tumours. For pT1-2 this is 2.2%.

Table 3. Subdivision by clinical and pathological tumour status.

	Clinical T status		Pathological T status		
	cT1 (%)	cT2 (%)	pT1 (%)	pT2 (%)	pT3 (%)
≤ 2 positive lymph nodes	617 (99)	212 (93.0)	568 (99.1)	247 (95.0)	14 (77.8)
> 2 positive lymph nodes	6 (1.0)	16 (7.0)	5 (0.9)	13 (5.0)	4 (22.2)

Abbreviations: T, tumour; cT, clinical tumour size; pT, pathological tumour size

DISCUSSION

Standard axillary ultrasonography with fine needle aspiration cytology (FNAC) or core biopsy of suspected nodes is being incorporated into the routine clinical assessment of breast cancer patients in our hospital since 2007. It is non-invasive, widely available and easily incorporated into the diagnostic examination. Axillary ultrasonography (US) has improved the preoperative characterization of axillary lymph nodes, being moderate sensitive (48,8-87,1%) and fairly specific (55,6-97,3%) depending of the

reference standard. With the use of ultrasound guided biopsy, the specificity increases up to 100%[38–40].

By implementing routine US of the axilla in the selection procedure we are able to select patients pre-operatively, in which the chance of more than two positive lymph nodes is very low. The risk of having more than 2 positive lymph nodes in our unselected population is 9.6%. Adding ultrasonography of the axilla to the standard work-up this chance will be more than halved to 4.2*%.* This value is even smaller in T1 tumours: 0.87% for pT1 and 0.96% for cT1. For T2 tumours the percentages are respectively 5.0% and 7.0%. Although low, more research is required to see if these percentages are significant for locoregional disease and overall survival.

Breast cancer is currently diagnosed much earlier than in the past. If we look to our own population the mean tumour size is 1.3cm. The incidence of axillary nodal disease in these small tumours is low and the extent is often limited. Even in case of a positive SLN, as analysed by Giuliano et al[37], the recurrence rates are small. The mean tumour size in their population group was 1.7cm. Within the whole population of Giuliano 27.3% had additional axillary lymph nodes, compared to 18.2% in our population if we would have made the selection by positive SLN. Another difference between both our population groups is the fact that we did not exclude patients with ECE of the SLN as we were most interested in selecting patients based on variables known before operation. Nonetheless, the risk of additional disease of the axilla seems to be smaller in our population. On top of that, the wide use of adjuvant systemic therapy is also known to diminish locoregional recurrence in breast cancer patients. Given all the above, the fear of locoregional disease of the axilla decreases.

Over the past decade, physicians and patients have been confronted with the issue of whether to do ALND after identification of nodal metastases by SLNB. Recent data from the ACOSOG Z0011 trial suggests avoiding ALND in case of a positive SLN where treatment strategy includes whole breast radiation alone or combined with adjuvant therapy following lumpectomy of T1-2 breast cancer. They found no significant benefit in locoregional control or overall survival with completion ALND[35].

Other recent studies have also questioned the additional value of ALND in patients with SLN metastases, and more importantly questioned why not to do a routine ALND after positive SLN. Firstly, ALND is associated with considerable morbidity, when compared to SLNB alone[4,10,41,42]. Secondly, several retrospective studies have been published reporting low axillary recurrence rates in patients with positive SNs who did not have ALND. The axillary recurrence rate was less than 2% [8,9,13–16,18]. In a review by Rutgers the 2 to 3-year risk of axillary recurrence was even lower: 0-1.4% in the untreated axilla[43]. As the recurrence rates in foregoing studies were approximately similar between the 2 groups it also suggests that not all non-SN metastases develop into clinically detectable disease. However,

most of these studies are limited by their small population size, limited knowledge of the reason why no ALND was performed and patient selection bias. Thirdly, most patients receive whole breast radiotherapy, which includes a portion of the axilla and/or adjuvant systemic therapy. The 25-years follow-up of the NSABP B04 trial shows that patients with mastectomy and locoregional radiotherapy have a better locoregional control compared with mastectomy and ALND[11,44]. Louis-Sylvestre et al[45] reported in their 15-year follow-up no differences between locoregional control. Finally there is the question of postoperative treatment planning. The AMAROS trial reports that the absence of knowledge regarding the extent of nodal involvement appears to have no major impact on the administration of adjuvant therapy[10].

In conclusion, the risk of having more than 2 positive axillary nodes is relatively small in patients with cT1-2 breast cancer, and especially the cT1 tumours. Selecting patients pre-operatively by using US of the axilla, we are able to even halve this percentage. Ultrasonography of the axilla helps in further identifying patients with a minimal risk of additional axillary disease, putting ALND up for discussion in this selected group of breast cancer patients. Larger studies and the effect of long-term results have to be seen.

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Chapter 4

Pre-operative ultrasonographic evaluation of axillary lymph nodes in breast cancer patients: for which group still of additional value and in which group cause for special attention?

A.M. Moorman, R.L.J.H. Bourez, D.M. de Leeuw, E.A. Kouwenhoven

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ABSTRACT

Introduction

A non-invasive and widely available method for pre-operative evaluation of the axilla is axillary ultrasonography (US).

Purpose

The purpose of this study was to evaluate the diagnostic accuracy of axillary US and fine-needle aspiration cytology (FNAC) in a large cohort of breast cancer patients.

Results

The sensitivity and specificity of US and FNAC in our cohort of 1124 patients were 42.2% and 97.1% respectively. With increasing number of axillary nodes, the sensitivity increased. The percentage of false-negative US was 18.9%, patients in this subgroup were significantly younger, had larger tumours, showed more often lymph vascular invasion (LVI) and were more likely to have oestrogen (ER) positive tumours.

Conclusion

Ultrasonography in combination with FNAC is useful in the pre-operative work-up of breast cancer patients, especially within the group of patients with ≥ 3 nodal metastases. Special attention should be paid to the younger woman with larger tumours in which a higher percentage of false-negatives is found.

INTRODUCTION

Over the years there has been growing interest in the development of clinical prediction tools to estimate the risk of having nodal metastases in the axilla of patients with breast cancer, and thereby making it possible to plan specific therapies. Sentinel lymph node biopsy (SLNB) has become the standard method of axillary lymph node staging in patients with invasive breast cancer. It has replaced axillary lymph node dissection (ALND) as it is associated with a significantly lower morbidity[1]. However, SLNB is still an invasive method and has a 4-14% rate of complications such as lymphoedema, seroma, paraesthesia, chronic pain and immobility[2]. When node metastases are found with SLNB, ALND is still warranted, which means that the patient has to undergo a second operation. This is not only an inconvenience for the patient, but also results in more operation time, space and costs[3].

A non-invasive and widely available screening method is axillary ultrasonography (US). Pre-operative axillary US, with or without fine needle aspiration cytology (FNAC) of lymph nodes suspicious for metastases, is now routinely performed in many breast cancer centres[4]. The utility of axillary US in detecting nodal metastases has been studied extensively. The results vary widely, especially in patients with early-stage breast cancer[5–7]. Sensitivity and specificity of axillary US range between 40-92%, and 56-100%. The specificity increases to 100% with the use of FNAC. However, as with all US procedures, the sensitivity and specificity of axillary US depends strongly on the experience of the ultrasonographer and the reference standard for malignancy used. The majority of previous studies on axillary US and FNAC have studied small patient groups (< 500 patients) and have used different morphologic criteria for detecting nodal metastases: palpable vs non palpable nodes, inclusion or exclusion of micrometastases and differences in the prevalence of axillary nodal burden[7–16].

The aim of this study was to evaluate the utility and diagnostic accuracy of axillary US and US with FNAC in detecting axillary lymph node metastasis in a large cohort of breast cancer patients.

PATIENTS AND METHODS

Patients

This retrospective cohort study was conducted in the Hospital Group Twente, a large teaching hospital located in Almelo and Hengelo, the Netherlands. Approval from the institutional review board (IRB) was not required since this was a non-interventional retrospective study using known data. From January 2007 until July 2011, 1124 consecutive primary breast cancer patients were selected. These patients were both screen detected and/or symptomatic. All patients underwent pre-operative axillary

ultrasonography (US) and subsequent surgery with sentinel lymph node biopsy (SLNB) and/or complete axillary lymph node dissection (ALND) according to current Dutch guidelines. Patients with palpable axillary disease, clinical and radiological T4 status, ipsilateral recurrent breast malignancy and neo-adjuvant chemotherapy were excluded.

Pre-operative ultrasonography and fine-needle aspiration

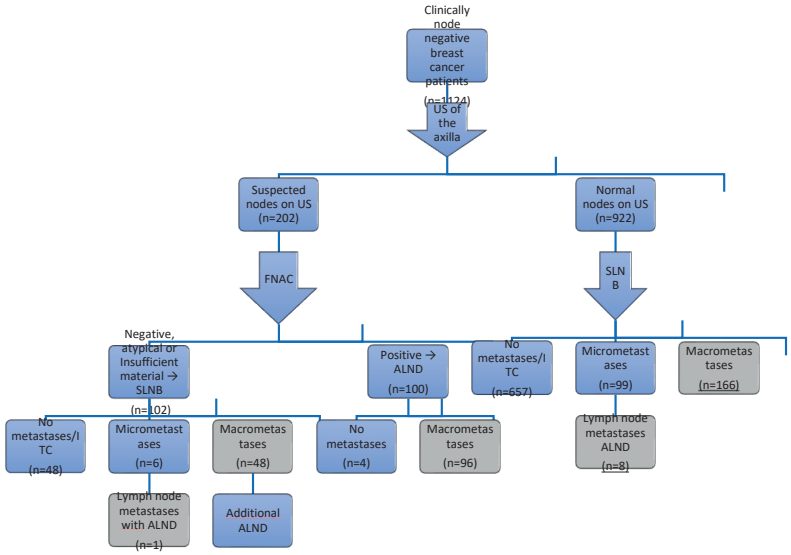
All patients underwent routine mammography, ultrasonography of the breast and ipsilateral ultrasonography of the axilla by a trained radiologist or a radiology resident under the supervision of a trained radiologist. Two commercial ultrasound scanners were used; the Acuson X300, VF13-5 transducer (Siemens, Seongnam, south Korea) with a frequency bandwidth of 4.4-13.0 MHz and a maximum field of display of 61 mm; and the Aloka Prosound Alpha 7, UST-5412 transducer (Aloka, Tokyo, Japan) with a frequency bandwidth of 5-13 MHz and maximum field of display of 60 mm. These where located at different sites, so the ultrasound scanner used was the one available in the hospital where patients presented. Lymph nodes were classified as suspicious if the cortical thickness was > 2.3 mm or if it had an irregular nodular cortex and/or a diminished or absent hilum[17]. When suspicious nodes were found, US-guided FNAC was performed using a 21-gauge needle and the aspirate was sent to the pathology department for cytological analysis. If needed a second attempt was made. FNAC analysis was carried out after Giemsa and PAP staining (Surepath).

SLNB and ALND protocol

The study protocol is summarized in Fig. 1. Patients with non-suspicious nodes after axillary US and those with no malignant cells after FNAC (or in whom insufficient material was obtained for diagnosis after several attempts), were scheduled for SLNB. Sentinel lymph nodes (SLNs) were harvested after Scintigraphy and Patent Blue Dye injection during or immediately prior to surgery by one of our experienced breast surgeons or by a surgical trainee under their strict supervision. A sentinel node was identified as any blue staining node, hot node or node with at least 10% of the highest hot node count. Pathologic examination of the SLN were classified as macrometastases (>2 mm), micrometastases (0,2-2 mm), or isolated tumour cells (<0,2 mm). If US guided FNAC proved positive for malignant cells, ALND was performed. Complete axillary lymph node dissection (ALND) was routinely performed when a metastases was present in the SLN. In this study we focused on macrometastases, since micrometastases do not normally alter the morphology of the lymph node and are thereby difficult to detect [18].

Patients and tumour characteristics were retrieved from the original patient files. The final pathology results, based on SLNB and/or ALND, were correlated with axillary US alone or US in combination with FNAC.

Figure 1. Study algorithm



Abbreviation: Grey coloured boxes = total of patients with lymph node metastases; Underlined boxes = total of patients with false-negative ultrasonography.

Statistical analysis

Sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) were calculated for axillary US alone and axillary US in combination with FNAC with the final pathological findings with SLNB and/or ALND as gold standard. The utility of US and US with FNAC was assessed by determining the positive and negative likelihood ratios. The correlation between clinic- and pathological variables and false-negativity of axillary ultrasonography was analysed using the Chi-square test. A p-value of < 0.05 was considered statistically significant.

RESULTS

Patient and tumour characteristics

During the observational period from January 2007 until July 2011, 1178 patients were treated for primary invasive breast cancer in the Hospital Group Twente, the Netherlands. Of these patients, 20 had palpable axillary lymph nodes and 34 patients did not undergo the routine work-up for other reasons, leaving 1124 patients for further analysis. All patients had solitary tumours. The median age of the patients was 61 years (ranging 24-93 years). The mean primary breast cancer tumour size was 20.46 mm (range 1-130 mm). Subdivided by tumour stage, 59.5% were T1 tumours, 35.3% were T2 tumours, 4.9% were T3 and 0.2% were pathological T4 tumours. There were 910 invasive ductal carcinomas, 138 invasive lobular carcinomas, 15 with mixed ductal and lobular carcinoma, and 61 other carcinomas diagnosed (total of n=1124). The median number of nodes removed with sentinel node biopsy was 2 (range: 1-13).

US in relation to the number of axillary nodal metastases

The overall percentage of axillary (macro)-metastases was 28.4%. Of the 1124 patients, a total of 922 (82.0%) had no suspicious axillary lymph nodes on ultrasonography (US), 202 (18%) did have suspicious nodes. The sensitivity of US to determine nodal involvement was 45.5% in case of 1 nodal metastases, with a specificity of 92.9%. The positive predictive value (PPV) and the negative predictive value (NPV) were 71.8% and 82.0% respectively. The positive likelihood ratio (positive LR) was 6.41 and the negative likelihood ratio (negative LR) was 0.59. In case of 2 nodal metastases, US sensitivity was 56.9%, specificity 92.9%, PPV 64.4%, NPV 90.6%, positive LR 8.01 and negative LR 0.46. In case of 3 or more nodal metastases, sensitivity was 60.8%, specificity 92.9%, PPV 58.1%, NPV 93.6%, positive LR 8.56 and negative LR 0.42 (see Table 1). Total number of patients decreased with increasing number of nodal metastases as we compared the patients with one or more, two or more and three or more macrometastases with no metastases.

Table 1. Accuracy and utility of US and US in combination with FNAC subdivided by number of axillary nodal metastases.

	N	Sensitivity %	Specificity %	PPV %	NPV %	+ LR	-LR
Positive US*							
Overall	1124						
1 metastases	319	45.5	92.9	71.8	82.0	6.41	0.59
(minus micro†)	311	46.6	92.5	73.2	79.8	6.21	0.58

2 metastases	181	56.9	92.9	64.4	90.6	8.01	0.46
3 metastases	130	60.8	92.9	58.1	93.6	8.56	0.42
Positive US with positive FNAC[‡]							
Overall	1071						
1 metastases	301	42.2	97.1	85.2	81.2	15.6	0.59
(minus micro[†])	293	43.3	97.3	87.6	79.8	16.0	0.58
2 metastases	175	55.4	97.1	81.5	90.6	19.1	0.46
3 metastases	127	59.8	97.1	77.6	93.6	20.6	0.41

Abbreviation: US = ultrasonography, FNAC = fine needle aspiration, PPV = positive predictive value, NPV = negative predictive value, +LR = positive likelihood ratio, -LR = negative likelihood ratio. Metastases = number of micro- or macrometastases found with SLNB or ALND.

[*] Positive US = axillary US suspicious for malignancy versus axillary US with no suspicious nodes (negative US)

[†] If micrometastases would have been excluded from the population. Only 8 patients had additional metastases after ALND of which only 1 had > 3 metastases.

[‡] Positive US + positive FNAC/atypical cells versus negative US.

US in combination with FNAC

FNAC was performed on all nodes that were considered suspicious with axillary US. The positive US/positive FNAC group consisted of those patients who were found to have suspicious nodes on axillary US and were proven to have a malignancy after FNAC. Also included in this group were patients in whom atypical cells were found with FNAC and patients whose cytologic specimens were inadequate for evaluation. The sensitivity of US with FNAC in determining nodal involvement was 42.2%, specificity 97.1%, PPV 85.2%, NPV 81.2%, positive LR 15.6 and negative LR 0.59. The respective numbers for two and three nodal metastases are listed in table 1. The total number of patients within this group also decreased with increasing number of nodal metastases, as we compared patient with a positive US and positive FNAC with those with negative US. In analysis of sensitivity for two nodal metastases, patients with only one nodal metastases are not automatically placed in the negative group, but were removed from further analysis.

False negative axillary ultrasonography

Of the 922 patients with a negative US, 18.9% had a false-negative ultrasound. These patients were found to have positive nodes after SLNB and/or ALND. Patient and tumour characteristics were compared between the true-negative and false-negative groups, and results are summarized in table 2. Patients with false-negative axillary US were younger, had larger tumours, more often had lobular carcinomas, were found to have lymph vascular invasion (LVI) and were more likely to have oestrogen (ER) or progesterone (PR) positive tumours. Univariate and multivariate analysis are outlined in table 3. Age, LVI, ER status and pathological tumour size remained significant after multivariate logistic

regression analysis. For a patient < 50 years of age who has a tumour ≥ 20mm, the chance of a false-negative US is 45.9%, almost one in two patients.

Table 2. Differences of patient and tumour characteristics between false-negative and true negative axillary ultrasonography.

	True negative N= 748 (%)	**False negative N=174 (%)**	**p-value < 0.15**
Age (years)			
< 50	97 (13.0)	56 (32.2)	< 0.001
≥ 50	651 (87.0)	118 (67.8)	
Clinical tumour size			
0-19 mm	551 (75.1)	100 (59.2)	< 0.001
≥ 20 mm	183 (24.9)	69 (40.8)	
Histology			
IDC	606 (81.0)	139 (79.9)	0.011
ILC	84 (11.2)	30 (17.2)	
Other	58 (7.8)	5 (2.9)	
Histologic grade			
1	252 (34.0)	42 (24.1)	0.025
2	326 (44.0)	94 (54.0)	
3	163 (22.0)	38 (21.8)	
Multifocality			
No	665 (88.9)	142 (81.6)	0.008
Yes	83 (11.1)	32 (18.4)	
LVI			
No	679 (90.8)	127 (73.0)	< 0.001
Yes	69 (9.2)	47 (27.0)	
ER			
Negative	120 (16.2)	12 (6.9)	0.001
Positive	622 (83.8)	162 (93.1)	
PR			
Negative	222 (29.9)	34 (19.5)	0.006
Positive	520 (70.1)	140 (80.5)	
Her2/Neu			
Negative	666 (89.8)	161 (92.5)	0.320
Positive	76 (10.2)	13 (7.5)	
Pathologic T-stage			
1a-c	524 (70.1)	78 (44.8)	< 0.001
2	213 (28.5)	78 (44.8)	
3	11 (1.5)	17 (9.8)	
4	0 (0.0)	1 (0.6)	

Abbreviation: LVI = lymph vascular invasion, ER = oestrogen receptor, PR = progesterone receptor, Her2/Neu = Human Epidermal Growth Factor *Receptor* 2.

The influence of micrometastases in the population

Sentinel lymph node biopsy revealed micrometastases in 103 patients. Of these patients, only 8 were found to have macrometastases after ALND and only 1 patient had three or more additional axillary nodes. Of the 4 patients found to have suspicious nodes with axillary ultrasonography, none had any

additional axillary disease after ALND. If this population would have been excluded from the study group, the values of sensitivity, specificity and so on would have differed slightly (see Table 1). Since ALND is still advised after micrometastases according to the Dutch guidelines, we did not exclude this group from our analysis.

Table 3 Univariable and multivariable associations of patient and tumour characteristics and false-negative ultrasonography.

	Univariable *p*-value	**HR (95% CI)**	**Multivariable *p*-value**	**HR (95% CI)**
Age (years)				
< 50				
≥ 50	< 0.001	3.185 (2.172-4.671)	< 0.001	3.077 (2.006-4.719)
Clinical T-stage rounded				
1a-c				
2	< 0.001	2.078 (1.465-2.946)	0.436	1.190 (0.768-1.842)
Histology				
IDC				
ILC	0.057	1.557 (0.987-2.456)	0.151	1.459 (0.871-2.444)
Rest	0.040	0.376 (0.148-0.954)	0.164	0.505 (0.193-1.321)
Tumour grade				
1				
2	0.007	1.730 (1.161-2.579)	-	
3	0.171	1.399 (0.865-2.263)		
Multifocality				
No				
Yes	0.009	1.806 (1.156-2.821)	0.340	1.281 (0.770-2.131)
LVI				
No				
Yes	< 0.001	3.642 (2.402-5.522)	< 0.001	3.028 (1.872-4.895)
ER				
Yes				
No	0.002	2.605 (1.404-4.832)	< 0.001	4.592 (2.304-9.149)
PR				
Yes				
No	0.007	1.758 (1.171-2.639)	-	
Pathologic T-stage				
1a-c				
≥ 2	< 0.001	2.879 (2.055-4.034)	< 0.001	2.224 (1.454-3.402)

Abbreviation: LVI = lymph vascular invasion, ER = oestrogen receptor, PR = progesterone receptor, Her2/Neu = Human Epidermal Growth Factor *Receptor 2*

DISCUSSION

In the present study, we assessed the accuracy and clinical utility of routine pre-operative axillary US in combination with FNAC in patients with breast cancer in The Netherlands. The prevalence of nodal

metastases (macrometastases) in our study was 28.4%. Our study suggests that specificity and PPV in detecting axillary metastases were higher for patients who underwent axillary US in combination with FNAC, especially those with gross nodal disease, than for patients who underwent US alone. Tahir et al[19] reported an increase in sensitivity from 47.1% to 80% when two or more nodal metastases were found. Sensitivity, on the other hand, was lower compared for US + FNAC than for US alone, as also reported in the literature[20,21].

Ultrasonography of the axilla is a useful diagnostic technique for the evaluation of axillary lymph nodes because it is non-invasive, widely available and easily incorporated in the standard work-up of breast cancer patients. Its sensitivity and specificity vary greatly in literature, ranging from 40-92% and 56-100% respectively[7–15]. Specificity increases when US is combined with FNAC, but sensitivity continues to vary greatly. There are a number of reasons for this discrepancy. First, the prevalence of axillary metastases in the study populations differs greatly, ranging between 25 and 58%[20]. In our study, the prevalence of nodal metastases was 28.4%, which is relatively low compared with values in the literature. Because PPV and NPV are directly proportional to the prevalence of the disease, these would increase with higher prevalence. To overcome this problem we also calculated the positive and negative LR, as these are not influenced by the prevalence.

Second, patient selection differs. Our selection of patients had relatively early breast cancers, with nearly 60% T1 tumours. A third reason for the observed differences is the exclusion criteria. Some studies include both palpable and non-palpable nodes, which are of great significance in analysing accuracy. If we would have included palpable nodes and cT4 tumours, our sensitivity would have increased. Finally, the criteria used for node morphology and needle biopsy differ. In our study we performed FNAC on suspicious nodes, whereas some studies performed FNAC on all lymph nodes ultrasonographically visualized independently of their appearance or size [7,9,22–28]. Some studies used nodal size as a criterion for malignancy and others used morphological criteria [29,30]. In our study the cut-off point for malignant nodal size was 2.3 mm; interobserver variation is to be expected with a cut-off behind the decimal point.

Results from the American College of Surgeons Oncology Group (ACOSOG) Z0011 [31] randomised controlled trial, however, revealed equivalent survival and regional control in patients with T1-T2 tumours , a maximum of two macrometastases with SLNB and additional radiation therapy with or without additional ALND. These observations would suggest a diminishing role of pre-operative US and FNAC in this population. In this study, pre-operative US and FNAC were of special of value in patients with gross nodal disease, the subgroup that was excluded with the ACOSOG trial. Within this subgroup, ALND is still of additional value.

Because sensitivity is often found to be low, we also investigated the clinical-pathological variables that are associated with patients with false negative US. The total rate of false-negative results in our series was 18.9%. Patients with false-negative axillary US were significantly younger, had larger tumours, more often had lymph vascular invasion and were more likely to have ER positive tumours. Lobular histology and PR positive tumours were more often seen within this subgroup, although did not remain significant after multivariate analysis. In the literature, the percentage of false-negative US and FNAC range between 28.1 and 31% [20,32,33]. These patients were younger, had larger tumours, tumours that were lobular (although not always significant) and had lymph vascular invasion [32–34]. The reasons are not always clear. Johnson et al. [32] state that a false-negative US is more likely in larger tumours and tumours with lymph vascular invasion because of the higher pre-test probability of metastatic disease. The correlation of ER positive tumours and false-negativity is unknown. The prevalence of false-negativity and PR positive tumours might be correlated by a higher risk of node metastases in PR positive tumours [35,36]. Although not always significant, there was a tendency toward lobular carcinomas in the false-negative group. Lobular carcinomas are known to be harder to diagnose with US; perhaps the same also supplies for the detection of nodal metastases[37].

CONCLUSION

Ultrasonography in combination with FNAC is useful in the pre-operative work-up of breast cancer patients. Patients with early stage breast cancer are unlikely to have heavy axillary disease burden, and in this subgroup the value of ALND has recently been up for discussion. Within the group of patients with three or more nodal metastases, however, the accuracy of US and FNAC is much higher and will be of additional value. Special attention should be paid to the younger woman with larger tumours in whom a larger percentage of false-negatives results are obtained. US and FNAC are of lower sensitivity, and direct SLNB might be preferred.

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Chapter 5

Incidence, clinical features and outcomes of special types in breast cancer in a single institution population.

A.M. Moorman, R. Vink, E.J.Th. Rutgers, E.A. Kouwenhoven.

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ABSTRACT

Introduction

The low incidence of special types of breast cancer hinders adequate clinical research efforts. As such, collecting sufficient data to develop well-established therapy strategies is difficult.

Purpose

The aim of our study was to obtain more data on these special types in order to better understand the different characteristics and optimize therapy strategies.

Patients and methods

A single institution retrospective cohort study from January 2007 until September 2015.

Results

One hundred and five patients remained after excluding the patients with invasive ductal and lobular carcinoma. The percentage of these so called special types in this population was 4%. Tubular carcinoma, cribriform carcinoma, carcinoma with medullary features, carcinoma with apocrine differentiation, secretory carcinoma, mucinous carcinoma and invasive papillary carcinoma had a good or excellent prognosis, while invasive micropapillary carcinoma, adenoid cystic carcinoma, metaplastic carcinoma and carcinoma with neuro-endocrine features had a worse prognosis.

Conclusion

Special types of breast cancer form a heterogeneous group. Submitting them all to the same treatment modality may lead to both over- and under-treatment. We need to combine our data to optimize treatment strategies for the different special types.

INTRODUCTION

Invasive breast cancer is a heterogeneous disease characterized by differences in clinical behaviour, response to treatment and prognosis. Most common are the invasive breast cancer of no special type (IBC-NST)[1]. The second most common is invasive lobular carcinoma (ILC) which encompasses 12.1% according to the NABON Breast Cancer Audit (NBCA) data[2]. The prognosis of breast cancer is traditionally predicted by clinical and pathological criteria. In recent years, many studies were conducted on gene expression profiling with promising results in addition to the standard clinical-pathological criteria. Current decisions on treatment of patients with breast cancer are based on well-established international guidelines from large randomized controlled trials and meta-analyses almost completely focussing on IBC-NST and ILC [3]. The remainder of breast cancers are much less common and will be referred to as 'special types' in the rest of this article. Recently the World Health Organization (WHO) published their 5th edition, but we will refer to the 4th edition in the rest of the article since the pathology reports were based on the 4th edition [4]. Optimal treatment strategies are not fully established for these subgroup of cancers due to their rarity. This makes conducting prospective studies unrealistic. The limited information we have is from case reports and small series. Management of these special types remains a challenge to the practicing clinician. Mounting outcome evidence from different patient cohorts may improve management strategies for these special types of breast cancer.

The aim of our study is to obtain more data on these special types from a large cohort of breast cancer patients in order to better understand the different characteristics and optimize therapy strategies. We compared our data with previously published results.

PATIENTS AND METHODS

Patients

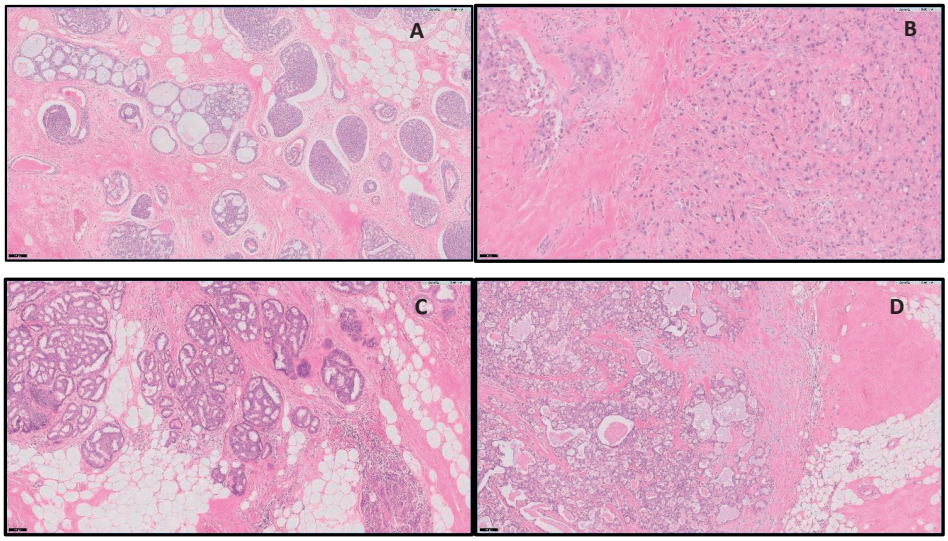
This is a single institution retrospective cohort study, conducted in a large teaching hospital in the Netherlands. From January 2007 until September 2015, 2473 consecutive primary breast cancer patients were registered. All patients were discussed in a multidisciplinary team before and after surgery. They underwent pre-operative axillary ultrasonography (US) and subsequent surgery with sentinel lymph node biopsy (SLNB) and/or complete axillary lymph node dissection (ALND) according to prevailing Dutch guidelines. After surgery, adjuvant treatment was given if so advised according to the guidelines. Patients with known distant metastasis at the time of surgery, recurrence within one month or other malignancies at the time of presentation were excluded for this analysis.

Pathology

Patients and tumour characteristics were retrieved from the original patient files. The diagnoses of the special types were made by pathologists from the Laboratory of Pathology East Netherlands according to the 4th WHO classification.

A total of 105 patients remained after excluding al the patients with IBC-NST and ILC. The different subtypes of the special types breast cancer other than ILC were: tubular carcinoma, cribriform carcinoma, mucinous carcinoma, carcinomas with medullary features, carcinoma with apocrine differentiation, invasive micropapillary carcinoma, metaplastic carcinomas of no special type, adenoid cystic carcinoma, carcinomas with neuroendocrine features, secretory carcinoma and invasive papillary carcinoma. See figure 1 for H&E stained sections of some of the special types.

Figure 1. H&E stained sections of special types of primary breast tumours.



Abbreviations: (a) Adenoid cystic carcinoma with 100x objective; (b) carcinoma with apocrine differentiation with 200x objective; (c) Cribriform carcinoma with 100x objective; Secretory carcinoma with 100x objective.

Follow-up

Patient follow-up was defined as time between surgery and last outpatient contact. Disease free survival (DFS) was defined as the period between the first surgery and time of local recurrence,

metastasis, death or last follow-up. Overall survival (OS) was defined as the period between surgery and death or last follow-up. The last follow-up for a patient in our study was conducted on November 16th, 2016. Five patients were lost to follow up. Our mean follow-up was 50 months and 38 (36%) patients had a minimal follow-up of five years.

Statistical analysis

Statistical analysis was performed using SPSS Statistics, Version 24.0. Clinicopathologic factors were mostly evaluated using descriptive statistics. For the different histological types, we calculated the disease-free survival and overall survival using the Kaplan-Meier method. Differences were considered statistically significant if $p < 0.05$.

RESULTS

A total of 2473 consecutive patients were diagnosed with breast cancer in the period of January 2007 until September 2015. After excluding the patients with IBC-NST and ILC, 105 patients remained. Table 1 shows the different special types with their characteristics. The percentage of so called special types in this population was 4% with a mean age of 63 years (range 31-93) and median tumour size of 16 mm.

Table 1. Summary of characteristics of special breast types.

Type	N	Median age (y)	Median tumour size (mm)	ER (%)	PR (%)	HER-2 (%)	Axillary lymph nodes (%)	DSF (%)	OS (%)	Mean follow-up (m)
Tubular carcinoma	15	53	9	100	80	0	7	100	100	41
Cribriform carcinoma	3	50	7	100	67	0	0	100	100	91
Mucinous carcinoma	29	68	15	100	79	0	3	86	93	52

Carcinoma with medullary features	13	48	24	0	0	0	23	100	100	47
Carcinoma with apocrine differentiation	4	60	27	50	50	50	25	100	100	45
Invasive micropapillary carcinoma	5	64	25	100	100	20	40	80	80	58
Metaplastic carcinoma	12	63	22	0	0	0	0	75	92	61
Adenoid cystic carcinoma	2	63	19	0	0	0	0	50	50	20
Carcinoma with neuroendocrine features	8	70	19	75	63	13	0	63	63	38
Secretory carcinoma	1	67	19	0	0	0	0	100	100	12
Invasive papillary carcinoma	13	70	12	92	92	0	8	92	92	52

Abbreviations: ER, oestrogen receptor expression; PR, progesterone receptor expression; HER-2, Human Epidermal growth factor Receptor 2 expression; DSF, disease-free survival; OS, overall survival.

Tubular carcinoma

Fifteen patients had tubular carcinoma (0.6%), these were all low grade, had no lymph vascular invasion (LVI) and low incidence of nodal involvement. In 73% the patients were over 50 years of age. One patient had nodal metastasis after SLNB which was suspected on axillary ultrasonography. All the tumours were oestrogen positive and HER-2 negative, with most also being progesterone positive. With the exception of one, all tumours were smaller than 20 mm. Ten patients underwent breast conserving therapy with SLNB and radiotherapy and four patients underwent mastectomy with SLNB. Only one patient underwent additional lymph node dissection after positive SN and mastectomy and was treated with adjuvant chemotherapy.

Cribriform carcinoma

Only 3 patients were diagnosed with cribriform carcinoma. Patients were between 49 and 72 years of age, had well differentiated tumours smaller than 10 mm and no axillary nodal involvement. Tumours were all oestrogen positive and HER-2 negative, while two were also progesterone positive. They all underwent breast conserving therapy with SLNB and adjuvant radiotherapy.

Mucinous carcinoma

Invasive mucinous carcinoma was the most common special type carcinoma, with an incidence of 1.2% (n=29). The mean age was 65 years. Only one patient had axillary nodal involvement after ALND. Over half of the tumours were grade 1, multifocality was seen in 14%, LVI in 10% and 28% of tumours were larger than 20 mm. All tumours were oestrogen positive, HER-2 negative and progesterone positive in 79%. Overall, 41% of patients underwent breast conserving therapy with additional radiotherapy (except for one), none received chemotherapy and 45% received hormone therapy. Within this group, one patient had recurrent disease, one patient had a distant metastasis and two patients died during follow-up due to reasons not related to breast cancer.

Carcinoma with medullary features

Carcinoma with medullary features represents 0.5% of the breast carcinomas in this population with a mean patient age of 50 years (range 36-74 years). The tumours were poorly differentiated in all but one. Multifocality and LVI were rarely seen. All the tumours were triple negative. Axillary nodal involvement was seen in 3 patients. The tumour size varied, with 69% being smaller than 50 mm. Patients underwent both breast conserving therapy (31%) and mastectomy (69%) with SLNB or ALND. Twelve patient received adjuvant chemotherapy and one patient received hormone therapy.

Carcinoma with apocrine differentiation

The incidence of carcinoma with apocrine differentiation was 0.2% (n=4) with a mean patient age of 60 years (range 49-70 years). Although axillary involvement was suspected in two patients, only one had true axillary nodal involvement. The tumours were moderately or poorly differentiated. One tumour showed multifocality, but none showed LVI. Two patients were triple negative. Two underwent breast conserving therapy with adjuvant radiotherapy, one chemo and one hormone therapy. None of the patients developed recurrence or distant metastasis.

Adenoid cystic carcinoma

Only 2 patients (56 and 70 years) with adenoid cystic carcinoma were encountered. Both patients had well differentiated tumours, no LVI, were both triple negative with a median tumour size of 19 mm and node negative. One patient developed liver metastasis and died during follow-up.

Invasive micropapillary carcinoma

There were 5 (0.2%) patients with invasive micropapillary carcinoma with a mean age of 65 years (range 56-75 years). Three patients had suspicious lymph nodes for which axillary ultrasonography was performed and showed that two of them had actual nodal involvement. LVI was seen in 3 of the patients. All tumours were oestrogen and progesterone positive, and one patient had a HER-2 positive cancer. Four of them received hormone therapy and one chemotherapy. One patient developed a distant metastasis and died.

Metaplastic carcinoma

The incidence of metaplastic carcinoma was 0.5% (n=12). Except for one, all patients were over 50 years with a mean of 65 years. One patient had clinically suspicious nodes, but eventually none of the patients had axillary nodal involvement. One patient had a tumour with LVI. All tumours were triple negative, except for one in which hormone receptor status was unspecified. Median tumour size was 22 mm. Half underwent breast conserving therapy and adjuvant radiotherapy, the other half mastectomy. Five patients received adjuvant chemotherapy. One patient developed local regional recurrence and one developed distant metastasis. During the mean follow-up period of 61 months one patient died of unknown cause.

Carcinoma with neuroendocrine features

Eight patients (0.3%) had a carcinoma with neuroendocrine features. The mean age at diagnosis was 69 years, with 7 of the patients being over 50 years. The median tumour size was 19 mm and only one patient had a micro metastasis. The tumours showed no multifocality or LVI. Oestrogen was positive in 75%, progesterone in 63% and HER-2 receptor expression was negative in 88%. Most patients underwent breast conserving therapy with SLNB followed by radiotherapy. Only one received

chemotherapy and one patient received hormone therapy. None of the patient developed recurrent disease or distant metastasis. However, three patients died during follow-up with a mean follow-up period of 38 months giving an OS of 63%. The cause of death was unknown for two patients, one died of cardiac failure.

Secretory carcinoma

There was only one patient diagnosed with a secretory carcinoma. She was 67 years and had a well differentiated triple negative tumour of 19 mm without nodal involvement. At 12 months of follow-up she was still free of disease.

Invasive papillary carcinoma

Thirteen patients (0.5%) were diagnosed with an invasive papillary carcinoma. The mean age of these patients was 71 years and all patients were over 50 years. The median tumour size was 12 mm (range 7-100 mm). Two patients had a micro metastasis in the SN. Tumour grade varied, DCIS was found in 46%, multifocality and LVI in 8% of tumours. All tumours were oestrogen and progesterone receptor positive and HER-2 negative. For one patient this information was not available. Seven of the patients underwent breast conserving therapy, SLNB and radiotherapy. Hormone therapy was given to 4 patients. None of the patients developed recurrent disease or distant metastasis. During a mean follow-up period of 52 months, one patient died of unknown cause.

Good versus worse prognosis in special types.

Tumours with a good or excellent prognosis (n=80) were as follows: tubular carcinoma, cribriform carcinoma, carcinomas with medullary features, carcinoma with apocrine differentiation, secretory carcinoma, mucinous carcinoma and invasive papillary carcinoma. Tumours with a worse prognosis (n=25) were as follows: invasive micropapillary carcinoma, metaplastic carcinomas and carcinomas with neuro-endocrine features. The median tumour sizes were 14 mm and 21 mm for the good and worse prognosis group respectively. The groups are significantly different with respect to only two parameters: tumour grade and oestrogen status, with higher tumour grade and oestrogen receptor negative breast cancers in worse prognosis group.

DISCUSSION:

This article represents our efforts to gain more insight into the special types of breast cancer by presenting the data on these special types within our single institution patient population. We will discuss our findings in relation to available published previous studies, and thus aim to find the most reasonable treatment strategies. To this end, we have identified low-risk and more high-risk special types of breast cancer and have divided our discussion as such.

Special types with excellent prognosis.

The incidence of tubular carcinoma in this population was very low (0.6%) compared to 0.7-10.3% in literature[5] . We could confirm that this well differentiated tumour type presented in older women and were both oestrogen and progesterone positive but HER-2 negative. The incidence of nodal involvement appears to be low, ranging 3-17%[5] which raises the question if our patient with nodal involvement might not have had a purely tubular carcinoma. Nevertheless, the prognosis is very good with a 5 years DFS for node positive patients of 94%[6,7]. This raises the issue whether SLNB is at all indicated in tubular carcinoma. If ongoing studies show that SLNB does not add to prognosis, it is very likely that women with tubular breast cancers are the right candidates to omit SLNB procedures[8].

Cribriform carcinoma is very similar to tubular carcinoma with respect to biological behaviour. It is a relatively uncommon type of breast cancer which can be subdivided further into pure and mixed phenotypes. They are typically seen in older women, are well differentiated, oestrogen positive, progesterone positive in two-thirds of patients and HER-2 negative, with an excellent prognosis for the pure variant with a ten year OS of 90-100%[9]. Given the excellent prognosis, this subgroup might also benefit from a more conservative approach and treatment strategy.

Yet another type of breast cancer known for its excellent prognosis, is the mucinous carcinoma. Pure mucinous carcinoma accounts for roughly 1-4% of all breast cancers. In our population the incidence was 1.2%, which is relatively low. In accordance with literature, these tumours are mostly diagnosed in older women with a median age of 71 years[10]. The median age in our population was 65 years. Although only half of the tumours is well-differentiated and some have axillary nodal involvement, multifocality and LVI, the overall survival is good. The 10-, 15- and 20-year survival rates known from literature are 89%, 85% and 81% respectively[10]. However these results are gain after standard locoregional treatment.

Despite unfavourable histologic features, the prognosis of carcinoma with medullary features is generally good. Incidence ranges between 1-7% and they mostly affect relatively younger women. These cancers are generally triple negative, as reflected in our data. The tumour size varied greatly

and the incidence of nodal involvement was 23%, similar to the 27% reported in the literature. The prognosis is good reaching 10-year DFS of 85-95%[11,12]. In our mean follow- up of 47 months we found a 100% DFS and OS. These good results are achieved through adequate locoregional treatments. Adjuvant chemotherapy is generally advised for these cancers. One of the changes in the 5th edition of the WHO classification is that carcinoma with medullary features are now considered a morphological pattern of IBC-NST[1].

Carcinoma with apocrine differentiation is known to have a good to intermediate prognosis. The incidence ranges between 0.3-4%[13]. With regard to hormone receptor expression, studies show varying results[14,15]. In our population hormone receptor expression was equally distributed and the prognosis was excellent. Literature on carcinoma with apocrine differentiation is scarce, maybe partly explained by the lack of a clear diagnostic definition in the past. The prognosis is similar to that of IBC-NST[16] and thus so should be the treatment.

Adenoid cystic carcinoma comprises 0.1-1% of breast cancers[17]. It predominately affects postmenopausal woman with a mean age of 60 years. Pain can be the first symptom due to neural involvement. It is a tumour with low malignant potential and lymph node involvement is seen in 0-8% of patients. They are generally triple negative, although some studies also report hormone receptor positive tumours. This data correspond with our findings. The prognosis is generally good with an OS of 85-88%, 75% and 60% after respectively 5-, 10- and 15 years of follow-up. Even after metastasis are diagnosed, patients may live for many years. The most frequent site of metastasis are the lungs[18,19]. A wide local excision without breast irradiation is in most cases adequate for local control. In absence of suspicious lymph nodes on US, SLNB procedure could be omitted. Systemic therapy is of uncertain value [20].

Exceptionally rare are the secretory carcinomas, we observed an incidence of 0.04% compared to 0.15% in literature. Although it mostly affects younger women, our patient was older. She did have a low grade, triple negative tumour. Nodal involvement occurs in around 15%. Prognosis is generally good, especially in children and young adolescents, but the tumour tends to be more aggressive in older women[21,22].

According to the WHO classification invasive papillary carcinoma of the breast is considered a differentiated adenocarcinoma with otherwise no distinguishing clinical, genetic or prognostic features[23]. The terminology in literature is somewhat confusing and most case reports of invasive papillary carcinoma are actually about encapsulated papillary carcinoma (EPC) and solid papillary carcinoma (SPC), which mostly behave as an in situ lesion or an indolent form of invasive carcinoma[24,25]. Based on our data with excellent DSF and OS, we should be careful in making assumptions as our

population might not be pure. In case of EPC and SPC we should be careful not to overtreat if we are certain no invasive component is present. If an invasive component is present this is often an IBC-NST. Particularly the EPC can be treated with wide local excision alone. Extra care should be taken in the diagnosis of actual invasive papillary carcinoma.

High risk special types.

Invasive micropapillary carcinoma is mostly seen in patients around menopause and is characterized by extensive LVI (35-91%) and nodal involvement (44-100%)[26–28]. In our study, the numbers were 60% and 40% respectively with a mean follow-up of 58 months. This tumour is often thought of having a poor prognosis, but varying outcomes have been published. The most recent meta-analysis including 1888 micropapillary carcinomas across 14 studies revealed a higher rate of local recurrences than IBC-NST patients, but no significant differences in DFS or OS[29]. Our population is far too small to draw any conclusions, however, we did observe that the patients with extensive nodal involvement developed distant metastasis within a year and died shortly after that. Due to its rather grim outcome, micropapillary carcinoma's are in general treated with adjuvant chemo and hormone therapy, next to optimal locoregional staging and treatment.

Metaplastic carcinoma is a heterogeneous group of invasive breast cancers. They are mostly poorly differentiated, larger, often triple negative, presents in older women and has a low incidence of nodal metastasis. The incidence ranges between 0.2-0.6% with a relatively high proportion in African-American and Hispanic women. The median tumour size in our population was 22 mm, smaller than the 34-44 mm that is reported in literature[30,31]. Metaplastic carcinoma has a higher potential for hematogenous metastasis compared to IBC-NST. One study found a median overall survival of 37 months. Patients with metaplastic carcinomas receive similar treatment to IBC-NST patients, but have a poorer prognosis[32]. This suggests that a more aggressive treatment approach might be warranted for certain metaplastic carcinomas.

Carcinoma with neuroendocrine features is a very rare malignant tumour, although some degree of neuroendocrine differentiation occurs in 10-30% of IBC-NST. In the 5th edition of the WHO, carcinomas with neuroendocrine differentiation are also considered a morphological pattern of IBC-NST[33]. Wang et al[34] reported in a review of 142 SEER cases that they mostly occur in older women (> 60 years), with larger tumours (> 20mm), higher histologic grade and higher clinical stage. The tumours often express both oestrogen and progesterone receptors. HER-2 status is typically negative, which was also observed in our study. Axillary involvement is seen in 43%. Mean tumour size in our population was only 18 mm and only one patient developed micro metastasis in the axilla. This might be an explanation

for the relatively good prognosis in our small group which is in contrast with previously published results. Younger patients with larger tumours and nodal involvement, are more likely to have a poorer prognosis[35]. In general treatment is in line with that of IBC-NST[36].

Comparing the tumours with a good prognosis to tumours with a worse prognosis, we found that the groups showed mostly similar baseline characteristics except for grade and oestrogen receptor status. Although histopathology has been our standard, recent research into the deeper understanding of the biology of breast cancer has led to a more specific classification of tumours according to their genetic expression [37–39]. Gene expression profiling might give us an even better understanding of the biological pathway, which in turn could improve treatment allocation.

This study has a number of limitations in addition to its retrospective nature. Special types of breast cancer are difficult to classify and in the new 5th edition of the WHO breast classification several changes have been made. The data was achieved from the original patient files, they have not been revised. Because of the small numbers, clinicopathologic factors were mostly evaluated using descriptive statistics. Making assumptions is difficult considering these small numbers.

CONCLUSION

Special types of breast cancer form a heterogeneous group of tumours with differences in tumour behaviour and prognosis. Submitting them all to the same treatment modality may lead to both over- and under-treatment. Although it is important to realize that these results are based on the effect achieved after standard care. The poor prognosis of some reflect the need for a better understanding, not only by their histological appearance, but also intrinsic molecular biology.

This study was a conscious effort to expand the limited evidence on special types of breast carcinoma that is currently available. Additionally, we hope the publication of our data will inspire other institutions to undertake similar efforts to report their data on special type breast carcinomas. A more comprehensive study on both the clinicopathological and genomic aspect of rare breast cancer types is needed to provide specific pathways and targets to develop standardized treatment algorithms and guidelines to ultimately provide optimal treatment.

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Chapter 6

Omitting SLNB in breast cancer: is a nomogram the answer?

A.M. Moorman, E.J.Th. Rutgers, E.A. Kouwenhoven

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ABSTRACT

Background

SLNB is standard care as a staging procedure in patients with invasive breast cancer. The axillary recurrence rate, even after positive SLNB, is low. This raises serious doubts regarding the clinical value of SLNB in early breast cancer. The purpose of this study was to select those patients with low suspected axillary burden in whom we might omit SLNB.

Methods

We retrospectively analysed 2,015 primary breast cancer patients between 2007-2015, with 982 patients allocated to the training and 961 to the validation cohort. Variables associated with nodal disease were analysed and used to build a nomogram in predicting nodal disease.

Results

A total of 32.8% of patients had macro-metastatic disease. A predictive model was constructed based on age, cN0, morphology, grade, multifocality and tumour size with an AUC of 0.83. Implicating a false-negative rate of 5%, 32.8% of patients could be spared axillary surgery. In a sub-analysis of patients with relatively favourable characteristics, 26.8% had a less than 5% chance of macrometastases.

Conclusion

We present a model with excellent predictive value in which we can select one third of patients in whom SLNB is deemed not necessarily due to less than 5% chance of nodal involvement. Whether missing 1 in 20 patients with macro-metastatic disease is worthwhile balanced against preventing side-effects of the SLN procedure, remains to be established. A number of ongoing large prospective trials evaluating the outcome of omitting SLNB have to be awaited. In the meanwhile, this nomogram may be used for individual decision-making.

INTRODUCTION

In breast cancer management knowledge of the axillary nodal status is considered relevant for prognostic information and guidance to elective treatment of affected lymph nodes to improve regional control and guiding adjuvant systemic therapy decisions. Sentinel lymph node biopsy (SLNB) is the method of choice and is considered standard care in the identification of nodal disease in patients with clinically unsuspected axillary nodes. SLNB is a safe alternative for the traditional axillary lymph node dissection, with significantly less morbidity [1–3].

The SLN principle has paved the way for less invasive treatments of the axilla. For SLN negative disease or isolated tumour cells a wait and see management is fully accepted [4]. In the past several studies have been published reporting low axillary recurrences even in women with positive SLNs who did not have ALND for various reasons [5–9]. Bilimoria et al. [10] reported no differences in axillary recurrence rate for patient with or without ALND after a positive SLNB, which compromised 20.8% of the 97,314 patients included in their retrospective review of the national cancer data base (NCDB). Three important trials have helped us to further diminish the need for ALND for patients with SLN positive breast cancer. The ACOZOG Z0011 trial randomly assigned 891 patients with cT1-2N0 breast cancers and one or two positive axillary SLNs to either ALND or no surgical treatment of the axilla. The results reported no benefit for ALND in terms of local control, disease free survival (DFS) and overall survival (OS) in this subgroup of patients who were treated with breast conserving surgery, adjuvant systemic therapy and radiation therapy [11,12]. Similar result were seen in the IBCSG 23-01 trial in a population of only micro-metastatic nodal disease, confirmed after 10 years follow-up [13]. In the AMAROS trial, 681 patients with a positive SLN were treated with axillary and supraclavicular radiotherapy and 744 patients underwent ALND. They reported no significant difference between the groups (cT1-2N0) regarding DFS and OS, but there was significantly less morbidity in the irradiation group [14]. Present-day, most guidelines indicate that further ALND is not necessary to improve axillary control and survival in patients with low volume nodal disease [15,16].

In all, strong evidence indicates that axillary clearance in SLN positive breast cancer offers no disease free or overall survival benefit in patients with early-stage breast cancer. To take this even further, one could raise doubts on the role of the SLNB itself. What is the advantage of performing a SLN biopsy in clinically node negative early breast cancer? Is there any impact on treatment and consequently survival? In comparison with ALND, SLNB substantially reduces the axillary morbidity but does not completely eliminate them. Lymphedema still occurs in approximately 5-8%, paraesthesia in 6-10.2%, arm and shoulder impairment is reported in 0-57.7% and pain in 5.1-51.1% [17–19].

These considerations have led to the initiation of a number of trials investigating the clinical outcome of women with early cN0 breast cancer with or without a SLN procedure such as the SOUND, POSNOC, INSEMA and BOOG 13-08 trial[20–23]. The results of these trials are expected in the next 4-5 years. In the meantime, are we able to safely select patients with a very low chance of SLN macro-metastatic disease or do keep performing SLNB in all patients? This information could enable us to discuss the additional value of the SLNB on individual basis.

To that end, we aimed to develop a model to predict the probability of axillary lymph node metastases based on variables pre-operatively known and validate the model on an independent subset of the population in order to select those patients with an expected less than 5% chance of having nodal disease in which SLNB might be omitted. The chosen 5% corresponds with the accepted false negative rate (FNR) of the SLNB itself [24–26].

METHODS

Study population

This is a single institution retrospective cohort study of patients with cT1-3 primary breast cancer with or without clinically suspected axillary nodes between January 2007 and November 2015. Data was retrieved from the original patient files. All patients underwent breast surgery and axillary staging according to the current Dutch guidelines. Patients treated with neo-adjuvant therapy and patients with known distant metastasis at the time of surgery were excluded. Only patients without missing data were further analyzed. The patients that remained were randomized to a training and a validation cohort. Distribution between the two cohorts was performed using the statistical analyzing program SPSS version 24.0 selecting a random sample of cases.

Predictors and model development

Data collection was based on the following variables: age at diagnosis, palpable tumour, tumour size, clinical nodal status (physical examination combined with ultrasonography of the axilla followed by FNAC if suspicious; in case of metastasis consecutive ALND followed), type of surgery, morphology, tumour grade, number of axillary metastases on final pathology, lymph vascular invasion, multifocality (two or more separate foci of tumour irrespective of the distance between them), oestrogen (ER) and progesterone (Pgr) hormone receptor and Her2neu status. We focused primarily on variables pre-operatively known in the final analyses. The tumour size was defined as size in millimeters measured

at definitive pathology since the absolute size on imaging was not available for all patients in the database. Of note, we could not find a relevant difference between the clinical and pathological tumour stage ($p < 0.001$). Therefor we used the tumour size based on the definitive pathology result for further analyses as surrogate for the clinical tumour size.

In the analyses we compared patient with one or more macrometastases (after SLNB or ALND) with patients with no axillary disease. Patients with micrometastases were excluded to minimize bias and ITCs were considered no axillary disease.

Statistical analyses

Univariate analyses was conducted to examine individual risk factors for nodal disease. The distribution of continuous variables was analysed using the Mann-Whitney U test, and the X^2 or Fishers Exact test were used for categorical variables. Univariate logistic regression analysis was used to explore the variables significantly associated with having nodal disease. We focused on the variables pre-operatively known as described above. All variables were categorical except for age and tumour size.

All variables with a P value less than 0.15 in the univariate analyses were included in a binary logistic regression analysis using both the manual and backward stepwise likelihood ratio method. If two or more variables were highly correlated, only one of them was included in the final nomogram to minimize the risk of multicollinearity. Variables with a P value less than 0.05 were included in the final predictive model. Internal validation was assessed using a bootstrap procedure resampling with 1000 replicates from the training cohort to estimate the accuracy of the prediction model. The resulting multivariate predictive model was then validated using the separate validation cohort. We assessed the discriminatory ability of the model by the area under the receiver operating characteristic curve (AUC). The sensitivity and specificity were analysed for various cut-off values. Statistical analyses was performed by SPSS statistics version 24 software.

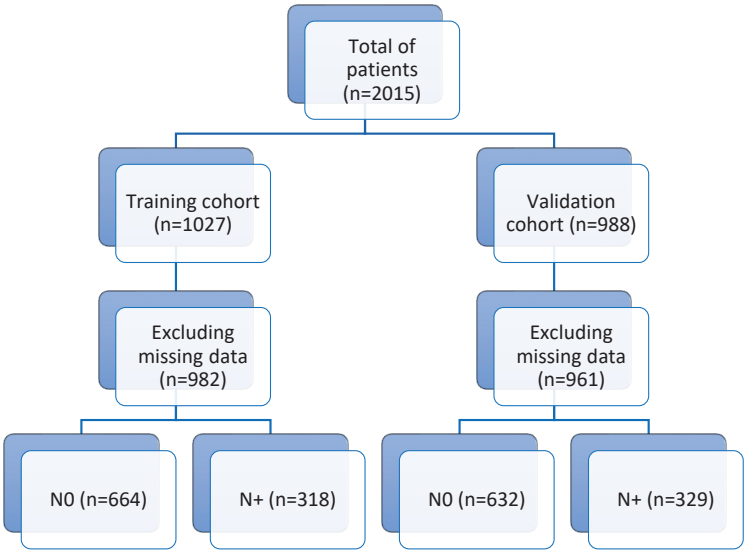
RESULTS

Training cohort

Between January 2007 and November 2015, 2,015 consecutive patients were treated for primary invasive breast cancer in the Hospital Group Twente, The Netherlands. Of the 2,015 patients, 982 were randomly assigned to the training cohort and 961 to the validation cohort. See figure 1. The mean age

of the 982 patients in the training cohort was 61 years with a mean tumour size of 20.9 mm and 318 (32.8%) had nodal disease. The baseline characteristics are shown in table 1.

Figure 1. Study design



Abbreviations: N0, no nodal disease; N+, nodal disease (macrometastases (>2 mm)).

Table 1. Baseline characteristics training cohort.

	N0	**N+**	**N=982**
	N=664 (%)	N=318 (%)	*P* < 0.15
Age			
Mean	61.7	58.79	< 0.001
Range	31-93	24-90	
Palpable tumour			
No	327 (49.2)	80 (25.2)	< 0.001
Yes	337 (50.8)	247 (75.5)	
cN			
No suspicious nodes	660 (99.4)	227 (71.4)	< 0.001
Suspicious nodes	4 (0.6)	91 (28.6)	
Operation type			
Breast conserving	364 (54.8)	94 (29.6)	< 0.001
Mastectomy	300 (45.2)	224 (70.4)	
Morphology			
NST	542 (81.6)	260 (81.8)	< 0.001
Lobular	79 (11.9)	57 (17.9)	
Other	43 (6.5)	1 (0.3)	
Grade			
1	231 (34.8)	61 (19.2)	< 0.001
2	284 (42.8)	160 (50.3)	

3	149 (22.4)	97 (30.5)	
Multifocality			
No	586 (88.3)	259 (81.4)	0.004
Yes	78 (11.7)	59 (18.6)	
LVI			
No	373 (90.8)	124 (67.4)	< 0.001
Yes	38 (9.2)	60 (32.6)	
ER			
Positive	559 (84.2)	266 (83.6)	0.829
Negative	105 (15.8)	52 (16.4)	
Pgr			
Positive	469 (70.6)	230 (72.3)	0.583
Negative	195 (29.4)	88 (27.7)	
Her2-neu			
Positive	61 (9.2)	38 (11.9)	0.178
Negative	603 (90.8)	280 (88.1)	
Tumour size			
Mean	17.03	29.07	< 0.001
Range	0-100	5-110	
pT stadium			
1	476 (71.7)	125 (39.3)	< 0.001
2	176 (26.5)	151 (47.5)	
3	11 (1.7)	38 (11.9)	
4	1 (0.2)	4 (1.3)	

Abbreviations: cT, clinical tumour stadium; cN, clinical nodal status (based on physical examination and ultrasonography with FNAC if suspicious); NST, invasive breast cancer of no special type; Multifocality, two or more separate foci of tumour irrespective of the distance between them; ER, oestrogen; Pgr, progesterone; Her2-neu, human epidermal growth factor receptor 2; pT, pathological tumour status

Prediction model for nodal disease in the training cohort

Only pre-operative available variables were analyzed. To minimize multicollinearity, palpability was excluded and the more objective tumour size remained for further analyses. Increasing age, cN0, presence of a special type morphology, lower grade, unicentric cancers and a smaller tumour size were associated with no axillary disease after multivariate logistic regression using the backward selection analyses. See table 2. The nomogram was subjected to bootstrap resampling (n=1000) for internal validation with similar results. The discriminatory ability of the nomogram for predicting nodal disease was investigated using ROC curves. The area under the curve (AUC) was 0.83.

The following mathematical equation was produced from the logistic regression analysis to predict the presence of axillary disease, with *p* denoting the probability of axillary nodal macrometastases; *a* denotes age in years; *b* denotes the tumour size in mm; *c* denotes suspicious clinical nodal disease by physical examination and axillary US (1 if N+, 0 is N0); *d* denotes morphology (lobular vs rest); *e* denotes morphology (special types vs the rest); *f* denotes grade (grade 1 vs grade 2 and 3) and *g* grade (grade 2 vs grade 1 and 3); and *h* denotes multifocality (1 is positive, 0 if negative).

Ln(p/(1-p)) = -0.791 – 0.031 X a + 0.060 X b + 4.066 X c - 0.127 X d – 3.060 X e + 0.307 X f + 0.566 X g + 0.421 X h.

Table 2. Multivariate logistic regression for prediction of nodal disease using backward analysis (training cohort) resulting in a nomogram for predicting axillary nodal disease.

	Training cohort N0 vs N+ model					
	Coefficient	SE.	Wald	*p*	Odds ratio	95% CI
Age*	-.031	0.007	19.987	< 0.001	0.969	0.956-0.983
Tumour size**	0.060	0.007	68.891	< 0.001	1.062	1.047-1.077
cN						
Yes	4.066	0.550	54.671	< 0.001	58.330	19.852-171.390
Morphology				0.026		
ILC vs rest	-0.127	0.253	0.254	0.615	0.880	0.536-1.445
Special types vs rest	-3.060	1.150	7.084	0.008	0.047	0.005-0.446
Grade				0.033		
1 vs 2/3	0.307	0.251	1.488	0.222	1.359	0.830-2.224
2 vs 1/3	0.566	0.221	6.549	0.010	1.761	1.142-2.717
Multifocality						
Yes	0.421	0.447	3.314	0.069	1.523	0.968-2.396
Constant	-0.791					

Abbreviations: ILC, Lobular carcinoma; special types, rare types of breast cancer according to the 4th edition of WHO.
* The odds ratio for age is interpreted as the odds on having axillary disease for the difference of one year in age;
** odds ratio for tumour size is interpreted as the odds of having axillary disease with increasing tumour size (in mm).

Clinical value of the nomogram

We aimed to select those patients in whom the chance of macro-metastatic SLN involvement is very low and therefor SLNB might be omitted. A high sensitivity is important for the nomogram as it refers to the nomograms ability to correctly detect nodal disease in patients with actual axillary disease. In other words, a low false-negative rate (FNR). A false-positive outcome is of no concern since additional SLNB is still standard care. Currently we accept a FNR of 5% for the SLNB[24–26]. If we would accept a FNR of 5% in our nomogram (sensitivity 95%), the cut-off value will be 0.148 in the ROC curve. If we use this cut-off value, the number of patients in whom no axillary surgical staging is warranted is 218/664, that is 32.8% of all patients who do not have nodal disease. With this chosen model we would have missed 16/318 patients with actual macro-metastatic disease.

Sub-analyses and variations in the training cohort

When multivariate logistic regression for prediction of nodal disease is performed using manual forward stepwise analysis, multifocality would not be included in the prediction model. Nonetheless this model has a similar discriminatory ability of 0.82. With a cut-off value of 0.151 (sensitivity 95%) we would have missed 16 patients with macro-metastatic disease, but 230/664 (34.6%) patients could have been spared axillary surgery.

Further sub-analyses was performed for the selection of cN0 patients (n=887) with a mean age of 61 years and a mean tumour size of 19.8 mm. Age, tumour size, morphology and ER status were included in the prediction model using the backward selection analyses. The AUC was 0.77. With a cut-off at 95%, 170/660 (25.8%) patients could have been spared axillary surgery although we would have missed macro-metastatic disease in 11 patients.

Pre-operative ultrasonography is standard care in the Hospital Group Twente since 2007, though specific information (other than cN) was not available in the dataset for the whole population. Therefore, we performed a sub-analysis on the group with additional information on US and FNAC of suspicious lymph nodes (n=595) to analyze potential bias. The model was based on age, cN, morphology and tumour size. With a sensitivity of 95%, corresponding to a cut-off point of 0.156 and AUC of 0.83, 151/411 (36.7%) patients with no nodal involvement could have been spared axillary surgery based on this nomogram, but we would have missed macro-metastatic disease in 9 patients.

After selecting patients based on relatively favorable characteristics (tumour size (≤ 20 mm), cN0, no lobular carcinoma or multifocality), 72/460 (15.7%) had macro-metastatic disease in the SLN. Age, tumour size, morphology and oestrogen receptor status were included in the prediction model after backward selection analyses. A total of 104 patients (26.8%) could have been spared axillary surgery (cut-off point: 0.077; AUC of 0.73), but we would have missed 3 patients with macro-metastatic disease.

Validation cohort

The mean age of the patients in the validation cohort was 62 years with a mean tumour size of 21.5 mm. Of the total of 988 patients, 961 remained after excluding the patients with missing data. A total of 329 (34.2%) patients had macrometastases in the SLN.

Prediction model for nodal disease in the validation cohort

Age, cN, morphology, tumour size, multifocality and ER remained significant after multivariate logistics regression using the backward selection analysis. The current model differs from the training cohort as grade is now excluded and ER status is included in the nomogram. See table 3. The AUC was 0.81. At a cut-off value of 0.169 (95% sensitivity), 154/632 (24.4%) patients could have been spared axillary surgery but 16 patients with macro-metastatic disease would have been missed.

If we implement the nomogram constructed in the training cohort to the validation cohort, the results are quite similar with the exception of clinical nodal status in which the odds ratio is even higher. See table 3. Using the same variables and cut-off point in our validation cohort as in the training cohort, 95/632 (15%) patients with no nodal involvement could have been spared axillary surgery while we would have missed metastatic disease in 7 patients.

Table 3. Multivariate logistic regression for predicting nodal disease using backward stepwise analysis (validation cohort) resulting in a nomogram in predicting nodal disease. In addition, the results of applying the nomogram of the training cohort on the current validation cohort.

	Nomogram validation cohort N0 vs N+		**Nomogram training cohort applied on the validation cohort N0 vs N+**				
	Odds ratio	P	Coefficient	SE.	Wald	p	Odds ratio (95% CI)
Age*	0.989 (0.976-1.001)	0.078	-.012	0.006	3.665	0.056	0.988 (0.975-1.000)
Tumour size**	1.045 (1.033-1.058)	< 0.001	0.043	0.006	46.753	< 0.001	1.044 (1.031-1.057)
cN Suspicious	266.247 (44.226-1602.846)	< 0.001	5.362	0.879	37.249	< 0.001	213.133 (38.092-1192.536)
Morphology		0.004				0.004	
ILC vs rest	0.871 (0.548-1.385)	0.559	-0.096	0.236	0.166	0.684	0.908 (0.571-1.444)
Special types vs rest	0.060 (0.011-0.325)	0.001	-2.866	0.861	11.085	0.001	0.057 (0.011-0.308)
Grade						0.033	
1 vs 2/3			0.150	0.243	0.381	0.537	1.162 (0.721-1.873)
2 vs 1/3			0.504	0.217	5.425	0.020	1.656 (1.083-2.532
Multifocality Yes	1.485 (0.972-2.269)	0.068	0.399	0.215	3.447	0.063	1.490 (0.978-2.269)

ER	0.398 (0.240-0.660)	< 0.001	
Constant	-1.105		-1.444

Abbreviations: ILC, Lobular carcinoma;
* the odds ratio for age is interpreted as the odds on having axillary disease for the difference of one year in age;
** odds ratio for tumour size is interpreted as the odds of having axillary disease with increasing tumour size (in mm).

DISCUSSION

The SLNB procedure is standard care in the identification of lymph node metastases in patients with clinical unsuspected axillary nodes. Giuliano et al. [27] first reported the SLNB procedure in 1994 with a false-negative rate (FNR) of 7%. Since its introduction multiple trials have investigated the prediction value of the SLNB, which is found in 95% of the cases with a mean sensitivity of 95% (ranging 84-100%) [24–26,28]. With time and experience the identification rate reaches 100%.

The axillary recurrence rate (ARR) in patients with a negative SLN was found to be low, 0.3% with a median time-interval of 20 months (range 4-63) in a large systematic review of van der Ploeg et al [28]. Similar low percentages were seen in a systematic review of Pepels et al. [29], with a ARR of 0.3-0.4% after a negative SLN and a ARR of 1.7% after a positive SLN. The ACOZOG Z0011 trial reported a ARR rate of 0.9% in patients who were treated with SLNB alone and a ARR of 0.5% in those treated with ALND [12]. A large review of Francissen et al. analysed thirty articles published between 2001 and 2010 with a total of 7,151 patients with SLN-positive breast cancer and reported low axillary recurrences. The rates varied between 0-0.9% for micro-metastatic disease and 0.2-1.2% for macro-metastatic disease. The rates were comparable to that of patients in whom ALND was performed (0.2-1.0%). A recent systematic review of Huang et al. [30] included the Z0011 trial and six cohort studies between 2011 and 2019 from different areas of the world, with a total of 8,864 patients. They concluded that performing SLNB alone in patients with early-stage breast cancer and one or two SLN metastases had equivalent survival and recurrence outcomes to those receiving SLNB and ALND.

What are the reasons for these low ARRs when a back-up axillary dissection is not performed while we know the SLNB has a FNR of 5%? First, breast-conserving surgery is followed by whole breast irradiation which includes the lowest portion of the axilla. In 76-94% of patients, the clip marker placed after SLNB was located within the standard tangential fields of the whole breast irradiation [31–33]. A systematic review of van Wely et al. [34] reported that patients with a negative SLN who received breast irradiation had a significant lower ARR compared to patients who were not treated with breast irradiation. Secondly, the increasing use of adjuvant systemic therapy which is known to diminish locoregional

recurrences, both chemo- and hormone therapy [35]. The current guideline's advice adjuvant chemotherapy based on tumour characteristics, patient's characteristics and nodal status. Treatment recommendations are rarely altered by the additional information gained by ALND [36]. The MIRROR trial studied the effects of systemic therapy in patients with ITC's or micrometastases, and demonstrated an improvement in DFS in the patients who were treated with adjuvant systemic therapy [37]. In the Z-0011 and AMAROS trials, 95% of patients received adjuvant systemic therapy and this will have contributed to the low incidence of axillary recurrences. We can also extrapolate data from the neo-adjuvant therapy trials, showing high rates of nodal pathologic complete response after systemic therapy among patients with Her2 positive and triple negative disease[38–41]. Thirdly, not all axillary metastases ultimately progress to become clinically evident. Patients in the NSABP B-04 study reported low axillary recurrences, but did not receive routinely adjuvant chemotherapy [42]. The same conclusion was drawn in several retrospective and prospective studies suggesting that only a small proportion of breast cancer metastases in the axilla develop into clinically relevant disease [5,43,44].

Since the introduction of screening mammography programs, the proportion of small cancers is higher and consequently the incidence of axillary metastases has decreased. With the increased practise of breast conserving therapies and widespread use of adjuvant therapy, based on primary cancer characteristics, nodal assessment may also become less important. Therefor we feel there is a clinical need to identify the subgroup of patients with a very low risk of axillary disease in whom SLNB might be omitted. Over the years several nomograms have been developed to predict the risk of axillary disease, each with its own limitations. The MSKCC is the best known and most widely used nomogram[45–47]. Although most of the variables in our nomogram have already been documented of value, we focused especially on the clinical nodal status with specific attention to the performance of axillary ultrasonography which also has recently received more attention in literature [48]. Ultrasonography in combination with FNAC is useful in the pre-operative work-up with a sensitivity and specificity of 42.2% and 97.1%, respectively[49]. The accepted FNR for prediction models was < 10% in previous studies [48,50,51]. We aimed to develop a model with a FNR of ≤ 5%, based on the accepted false-negative rate of the SLNB itself. With our nomogram we were able to identify a third of patients in whom SLNB could be omitted with an AUC of 0.83, 25.8% if selected for cN0 status, and 26.8% if selected for relatively favourable characteristics like a smaller tumour, no lobular carcinoma, no multifocality and cN0. These patients could have been spared axillary surgery. So, for which patients is this nomogram of additional value and do we find it acceptable to miss 5% of macro-metastatic nodal disease?

To make a well-informed decision, the treatment strategy is of great importance. If systemic therapy is indicated based on tumour characteristics, then nodal status is of no additional value if gross nodal

disease is not expected. This is endorsed by the recently presented results of the RxPONDER trial which reports no benefit of adding chemotherapy to standard hormone therapy in postmenopausal women with early-stage IBC-NST, hormone positive and Her2 negative breast cancer with one to three positive nodes and a low recurrence score (Oncotype DX)[52]. A nomogram could be of additional value so select those patients who are not directly good candidates for de-escalation, like patients with higher grade tumours that are less strongly hormone positive and Her2 negative. Care should be taken with triple negative tumours, for which there is targeted therapy available to control potential residual disease and nodal status may impact decision making even in smaller tumours. Nodal status might also impact the type of chemotherapy given in Her2 positive tumours. The type of surgical procedure is also very important in the decision-making. In case of breast conserving therapy, additional irradiation of the breast is almost always indicated in which part of the axilla is also irradiated. On the other hand, in the context of de-escalation, knowledge of the SLNB is obligatory in case partial breast irradiation or even a wait and see policy is a considered. As an example, the ongoing TOP trial in elderly patients (>70 yrs.) with early breast cancer requires a negative SLN before inclusion [53]. The National Comprehensive Cancer Network (NCCN) and the American Society of Breast Surgeons guidelines recommend against routine axillary staging in women over 70 years of age with early stage IBC-NST hormone receptor positive (HR+) and Her2 negative breast cancer [54,55]. This is indorsed by a recent study of McKevitt et al[56], reporting an excellent breast cancer specific survival in women > 70 years of age with HR+ breast cancers irrespective of nodal status as long as hormone therapy was given. On the other hand, Sun et al[57] state SLNB should not be routinely omitted in the elderly patient who might be candidates for chemotherapy based on performance score or tumour biology in whom SLNB is still of additional value. Despite these guidelines the majority of surgeons are still performing a SLNB in the elderly patient, in clinical practise recommendations are more often based of functional status than age[58]. A nomogram might be of additional value, giving the physician the extra support in making therapy decision of the axilla. If a mastectomy is performed, the possible additional potential side effects of a SLNB are insignificant compared to the mastectomy itself and therefore omitting a SLNB seems futile.

This study has several limitations aside of being a single institution retrospective study. We focused on variables pre-operatively known. Morphology, tumour grade and hormone status were retrieved from the definitive pathological specimens as the information based on core biopsy was not available in our database. According to literature however, core needle biopsy is very accurate in evaluating these tumour characteristics [59]. Tumour size was also based on the definitive pathology, though the clinical and pathological tumour status did not significantly differ, this is a relevant limitation as lobular carcinoma is sometimes difficult to measure. Additional information for axillary imaging and FNAC was not separately available for the whole population, however all patient had the same diagnostic work-

up and definitive nodal status was known. To avoid any bias, we did a sub-analysis which gave the same results.

To conclude, beware of the nomograms. So far it appears difficult to identify women in whom we could safely omit SLNB. Until the previous mentioned ongoing prospective studies on the value of SLNB present their results, nomograms might assist in the shared decision-making in the treatment strategy of the axilla. It is important to realize that omitting SLNB can affect the treatment strategy of other disciplines. In patients with small screen-detected breast cancers and no suspicious nodes in the axilla one could discuss foregoing SLN procedures as a probable safe option on individual basis. This nomogram might particularly be of use in the elderly patients or patients with comorbidities to assist in the decision-making, as in most centres a SLNB is still performed.

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Chapter 7

Summary and conclusions

In **chapter 1**, the rationale behind this thesis was outlined. Breast cancer has become the leading cause of cancer for women worldwide. Nevertheless, the outcomes have improved over the years. This is due to improved awareness of women themselves and the extended indications and applications of different modalities like: screening mammography and improvements in diagnostic imaging, pathology, surgery, radiotherapy and systemic therapy. Treatment nowadays consists of a multidisciplinary approach combining the expertise of surgical oncologists, radiologists, medical oncologist, radiation oncologist, pathologist, plastic surgeons and specialized nurses. The (surgical) management of breast cancer has transformed from a radical mutilating procedure to a less invasive and oncologic safe operation with greater attention to aesthetics.

After the introduction of breast conserving therapy, the treatment of the axilla became also less invasive with the introduction of the sentinel lymph node biopsy (SLNB). Axillary lymph node dissection (ALND) is only indicated in certain situations. In this era of de-escalation, it is important to select those patients where a 'less is more' approach is oncologic save while keeping in mind that breast cancer is a very heterogeneous disease. The aim of this thesis was to focus on both the heterogeneous aspect of breast cancer and the de-escalation of axillary treatment in breast cancer patients.

In **chapter 2** we evaluated the prognostic value of tumor-stroma ratio (TSR) in triple negative breast cancer patients to optimize risk stratification in this challenging subgroup. Stroma is the connective tissue of the breast parenchyma and it is hypothesized to have a protective function in delaying or preventing cancer if stroma is functioning as normal. On the other hand, in the presence of an invasive carcinoma, the stroma changes thus creating a permissive and supportive environment for tumour growth. To evaluate the tumor-stroma ratio we selected all triple negative breast cancer patients (n=124 pts) from our hospital-based database over a consecutive period of 4 years. The hematoxylin and eosin (H&E) stained sections of the primary tumour were retrieved from the Pathology Laboratory East Netherlands and independently scored for tumour-stroma-ratio by two investigators (kappa 0.74). Patients with less than 50% stroma were scored as stroma-low (40% of pts) and patients with ≥ 50% were classified as stroma-high (60% of pts). The 5 years relapse free period (RFP) and overall survival (OS) were 85% and 89% in the stroma-low and 45% and 65% in the stroma-high group. After multivariate cox-regression analyses, TSR remained an independent prognostic variable for RFP (HR 2.39; p=0.033) and OS (HR 3.00; p=0.034). We therefore concluded TSR to be a strong independent prognostic variable and deserves to be validated. The TSR could help optimize risk stratification and might also lead to the development of targeted therapies.

The sentinel lymph node biopsy (SLNB) procedure is the method of choice for staging the axilla in patients with clinically node negative breast cancer. Traditionally additional axillary lymph node dissection (ALND) is performed after a positive SLN. However, the role of ALND has been continuously questioned over the years, especially in patients with limited disease of the axilla. The aim of **chapter 3** was to determine if we were able to identify those patients with limited disease of the axilla pre-operatively by using axillary ultrasonography (US). Therefore 1,103 consecutive primary breast cancer patients without palpable axillary disease and a maximum of 2 positive SLNs (based on ACOZOG Z0011 trial) were analyzed. All patients underwent routine ALND if a micro- or macrometastases was found in the SLN. Patients with radiologic unsuspected axillary lymph nodes formed the cohort of interest in this study and were further analyzed. A total of 102 (9.6%) patients had more than 2 positive axillary nodes. Using ultrasonography as a selection method, the chance of having more than 2 positive axillary nodes was 4.2%, in case of a cT1-2 tumour without extra capsular extension (ECE) in the SLN the chance of more than 2 positive axillary nodes was only 2.2%. These variables (negative US; clinical tumour stage; ECE) remained significant after multivariate analysis. We therefore concluded that US of the axilla helps in identifying patients with a minimal risk of additional axillary disease, putting ALND up for further discussion.

With the growing interest in the development of clinical prediction tools to estimate the risk of axillary disease in patients with breast cancer, we evaluated the utility and diagnostic accuracy of axillary ultrasonography (US) combined with fine-needle aspiration cytology (FNAC) in detecting axillary lymph node metastases in **chapter 4**. The overall percentage of axillary metastases was 28.4%. The sensitivity and specificity of US and FNAC in our cohort of 1124 breast cancer patients were 42.2% and 97.1%, respectively. US of the axilla is non-invasive, widely available and easily incorporated into the standard work-up for breast cancer. The specificity increases when US is combined with FNAC. The low sensitivity could be explained by the high representation of early breast cancers in this population, exclusion of palpable axillary disease and the overall low prevalence of axillary disease. The sensitivity increased with increasing number of axillary metastases. As ALND is already up for discussion in early breast cancer patients with low suspected axillary disease, US combined with FNAC is especially useful in patients with extended nodal disease. The percentage of false-negative US results was 18.9%: patients in this subgroup were significantly younger, had a larger tumour, lobular carcinoma, often lymph vascular invasion (LVI) and were more likely to have a hormone receptor positive tumour. In conclusion, US in combination with FNAC is useful in the pre-operative work-up of breast cancer patients, especially patients with 3 or more nodal metastases. Special attention should be given to younger woman with a larger tumour in whom a higher percentage of false-negative results were obtained.

As mentioned previously, breast cancer is a very heterogeneous disease. Current decisions on treatment strategies are based on well-established international guidelines from large randomized controlled trials and meta-analyses. However, these are almost completely focussing on invasive breast cancer of no special type (IBC-NST) and lobular carcinoma (ILC). The remainder of breast cancers are much less common and optimal treatment strategies are not fully established due to their rarity. The aim of **chapter 5** was to analyse these so-called special types of breast cancer to better understand the different characteristics and optimize therapy strategies. Over a period of 8 years, we selected all patients with a special type of breast cancer. A total of 105 patients (4% of the total population of 2,373 pts) remained after excluding IBC-NST and ILC. Clinicopathologic factors were mostly evaluated using descriptive statistic due to the small numbers. Tubular carcinoma, cribriform carcinoma, carcinoma with medullary features, carcinoma with apocrine differentiation, secretory carcinoma, mucinous carcinoma, and invasive papillary carcinoma had a good or excellent prognosis. While invasive micro-papillary carcinoma, adenoid cystic carcinoma, metaplastic carcinoma, and carcinoma with neuroendocrine features had a worse prognosis. Submitting all these different types of breast cancer to the same treatment modality may lead to both over- and under treatment. We therefore concluded that additional data are necessary, both clinicopathological as genomic, to provide the information we need to develop standardized treatment algorithms and guidelines for these special types of breast cancer.

With the diminishing role of ALND over the years, we turned our focus to the value of the SLNB itself. The axillary recurrence rates are low, even after positive SLNB, raising doubts on the clinical utility of the SLNB in itself in patients with early breast cancer. In comparison with ALND, SLNB is associated with substantially less axillary and arm morbidity but still comes with a small but relevant percentage of axilla and arm problems (dysesthesia and edema). In **chapter 6,** we therefore aimed to develop a model to predict the probability of axillary lymph node metastases based on variables pre-operatively known. With this model we wish to select those patients with low suspected axillary burden in whom we might omit SLNB. We retrospectively analysed 2,015 primary breast cancer patients over a period of 8 years and randomized them into a training and a validation cohort. The variables associated with nodal disease were analysed and used to build a nomogram in predicting nodal disease. A predictive model was constructed based on age, clinical nodal status (cN0), morphology, grade, multifocality and tumour size with an area under the curve (AUC) of 0.83. Accepting a false-negative rate of 5%, 32.8% of patients could be spared axillary surgery. In a sub-analysis of patients with smaller tumours (<20mm), cN0, no lobular carcinoma or multifocality, 26.8% had a less than 5% chance of macrometastases. What brought us to the next dilemma: do we find it acceptable to miss 5% of macrometastatic nodal disease? We concluded that it appears difficult to identify woman in whom we

could safely omit SLNB. The chosen treatment strategy is of great importance, in terms of extend of operation, indication radiotherapy and/or systemic therapy. A number of ongoing large prospective trials evaluating the outcome of omitting SLNB have to be awaited. In the meanwhile, our nomogram may be used as a tool for individual decision-making.

Chapter 8

Samenvatting en conclusies

De motivering voor dit proefschrift wordt in **hoofdstuk 1** toegelicht. Mammacarcinoom is het meest voorkomende type kanker bij vrouwen wereldwijd. Ondanks deze toename, zijn de uitkomsten duidelijk verbeterd over de jaren. Dit is te danken aan de toegenomen bewustwording van vrouwen zelf, maar ook door screening programma's naar borstkanker, continue ontwikkelingen binnen diagnostische beeldvorming, pathologie, chirurgie, radiotherapie en systeemtherapie. De hedendaagse behandeling is multidisciplinair, waarbij de kennis en kunde van oncologisch chirurgen, radiologen, medisch oncologen, radiotherapeuten, pathologen, plastisch chirurgen en gespecialiseerde verpleegkundigen samenkomen. De (chirurgische) behandeling van het mammacarcinoom is getransformeerd van een radicaal en mutilerende procedure naar een minder invasief en oncologisch veilige operatie met aandacht voor esthetiek.

Met de introductie van de schildwachtklier (SWK) procedure zet de trend van minder invasieve behandelingen binnen het mammacarcinoom zich ook voort gericht op de behandeling van de axilla. Een aanvullende okselklierdissectie (OKD) is nog slecht geïndiceerd in bepaalde situaties. In dit tijdperk van de-escalatie is het belangrijk om de juiste patiënten te selecteren waarbij een 'less is more' benadering gerechtvaardigd is, rekening houdende met het heterogene karakter van het mammacarcinoom. In dit proefschrift concentreren we ons op zowel het heterogene karakter van het mammacarcinoom als de de-escalatie van de axillaire behandeling bij het mammacarcinoom.

In **hoofdstuk 2** hebben we de diagnostisch waarde van de tumor-stroma ratio (TSR) geëvalueerd bij patiënten met een triple negatief mammacarcinoom. Dit ten einde een inschatting te kunnen maken wat betreft de prognose bij deze patiënten met één van de meest uitdagende subgroepen van het mammacarcinoom. Stroma is het bindweefsel in de borst welke een beschermende functie heeft indien normaal, echter in de aanwezigheid van een invasief carcinoom veranderd het stroma dusdanig dat het tumorgroei juist ondersteunt en bevorderd. Om de tumor-stroma ratio te analyseren zijn gedurende 4 jaar alle patiënten (n=124 pt) die zich presenteerden in ons ziekenhuis met een triple negatief mammacarcinoom geselecteerd. De routine Haematoxyline-eosine (H&E) gekleurde coupes werden opgevraagd bij het Laboratorium Pathologie Oost Nederland en gescoord door twee onderzoekers onafhankelijk van elkaar (kappa 0.74). Patiënten met minder dan 50% stroma (40% van de patiënten) werden geclassificeerd als stroma-laag en patiënten met ≥ 50% stroma (60% van de patiënten) werden geclassificeerd als stroma hoog. De 5-jaars recidief vrije periode (relapse free period; RFP) en algehele overleving (overall survival; OS) waren 85% en 89% in de stroma-low en 45% en 65% in de stroma-high groep. Na multivariaat cox-regressie analyse bleef TSR een onafhankelijke significante prognostische variabele voor RFP (HR 2.39; p=0.033) en OS (HR 3.00; p=0.034).

Concluderend stellen we dat tumor-stroma ratio een sterke onafhankelijke significante prognostische variabele is in de subgroep van triple negatief mammacarcinoom en gevalideerd dient te worden. Deze variabele optimaliseert de risicostratificatie en zou kunnen leiden tot doelgerichte therapieën in de toekomst.

De schildwachtklier (SWK) procedure is de gouden standaard wat betreft stagering van de axilla bij patiënten met mammacarcinoom. Indien een micro- danwel macro-metastase werd gevonden bij de SWK procedure, volgde er van oudsher een aanvullende okselkliердissectie (OKD). Echter, de waarde van de OKD staat al jaren ter discussie en met name bij beperkte ziekte van de axilla. Het doel van **hoofdstuk 3** was om te bepalen of we in staat zijn om patiënten preoperatief te selecteren middels echografie op beperkte ziekte van de axilla. Derhalve zijn 1,103 achtereenvolgende patiënten met een mammacarcinoom, zonder palpabele ziekte axillair en een maximum van 2 metastasen bij de SWK-procedure (gebaseerd op de ACOZOG Z0011 studie) verder geanalyseerd. In totaal bleken 102 (9.6%) patiënten meer dan 2 positieve klieren na OKD te hebben. De kans op meer dan 2 positieve klieren axillair bij een onverdachte echo axilla, was 4.2%. In het geval van een cT1-2 tumor zonder extracapsulaire extensie (ECE) van de SWK en een onverdachte echo axilla, was de kans op meer dan 2 positieve klieren nog slechts 2.2%. Deze variabelen (onverdachte echo axilla, cT en ECE) bleven ook na multivariaat analyse significant. Concluderend kunnen we stellen dat echografie van de axilla helpt bij de identificatie van patiënten met beperkte axillaire ziekte, waardoor de waarde van een OKD ter discussie wordt gesteld.

Er is een toenemende vraag naar klinische predictie modellen om het risico van ziekte in de axilla bij patiënten met mammacarcinoom te kunnen vaststellen. In **hoofdstuk 4** evalueren we het gebruik en de diagnostische waarde van de echo axilla gecombineerd met fine needle aspiration cytologie (FNAC) om axillaire ziekte te detecteren. Het percentage axillaire metastasen in het gehele cohort was 28.4%. De sensitiviteit en specificiteit van echografie van de axilla gecombineerd met FNAC in dit cohort van 1124 patiënten waren respectievelijk 42.2% en 97.1%. Echografie van de axilla is non-invasief, zeer toegankelijk en makkelijk te introduceren in de standaard work-up van het mammacarcinoom. De specificiteit nam toe wanneer echografie werd gecombineerd met FNAC. De lage sensitiviteit zou verklaard kunnen worden door het relatief grote aantal kleine tumoren, exclusie van palpabele ziekte axillair en de algehele lage prevalentie van axillaire ziekte in deze populatie. De sensitiviteit nam toe bij toename aantal metastasen. De OKD staat al ter discussie bij het vroeg stadium mammacarcinoom met laag verwachte axillaire metastasen. Voor patiënten met uitgebreide ziekte axillair is echografie van de axilla in combinatie met FNAC met name geschikt. Het percentage vals-negatieven was 18.9%, dit waren met name jongere patiënten met grotere tumoren, vaker het lobulaire type, vaker lymf vasculaire invasie (LVI) en hormoonreceptor gevoelig. Concluderend kunnen we stellen dat echografie

van de axilla in combinatie met FNAC een goede bijdrage levert in de preoperatieve work-up van patiënten met mammacarcinoom, met name bij die patiënten met 3 of meer axillaire metastasen. Extra voorzichtigheid moet daarbij in acht worden genomen bij de jongere patiënt met een grotere tumor waarbij een hoger percentage vals-negatieven werd gezien.

Vervolgens hebben we opnieuw gekeken naar het heterogene karakter van het mammacarcinoom, en hebben we onze aandacht gericht op de zogenaamde 'special types'. De huidige behandelstrategieën zijn gebaseerd op duidelijk vastgelegde internationale richtlijnen verkregen uit grote gerandomiseerde onderzoeken en meta-analyses. Echter, deze onderzoeken zijn vrijwel geheel gebaseerd op het 'invasive breast cancer of no special type' (IBC-NST) en het lobulair carcinoom (ILC). De overige typen mammacarcinoom zijn veel zeldzamer en richtlijnen en doelgerichte behandelingen bestaan niet voor deze special types. In **hoofdstuk 5** presenteren we onze data van deze zogenaamde special types mammacarcinoom ten einde een beter beeld te krijgen van deze verschillende subtypen ter optimalisatie van de behandeling. Gedurende een periode van 8 jaar zijn alle 'special types' geselecteerd. Na exclusie van IBC-NST en ILC bleven er nog 104 patiënten over (4% van de totale populatie). Klinisch-pathologische variabelen zijn geëvalueerd middels beschrijvende statistiek gezien de kleine aantallen. Tubulair carcinoom, cribriform carcinoom, carcinoom met medullaire kenmerken, carcinoom met apocriene kenmerken, secretoir carcinoom, mucineus carcinoom en invasief papillair carcinoom hadden een goed of uitstekende prognose. Daartegenover hadden het invasief micropapillair carcinoom, adenoid cysteus carcinoom, metaplastisch carcinoom en het carcinoom met neuro-endocrine kenmerken een slechtere prognose. Door al deze verschillende (zeer heterogene) subtypes van het mammacarcinoom hetzelfde te behandelen, zal dit tot zowel over- als onder-behandeling leiden. Concluderende kunnen we stellen dat meer data nodig is, zowel klinisch-pathologisch als moleculair, alvorens we in de mogelijkheid zijn om ook voor deze verschillende subtypen gestandaardiseerde behandelingen en richtlijnen op te kunnen stellen.

De indicatie voor een OKD neemt steeds verder af, derhalve hebben we onze aandacht vervolgens gevestigd op de SWK-procedure. Het aantal regionale recidieven zijn namelijk laag, zelfs bij een bewezen metastase na SWK-procedure. Dit leidt tot vragen rondom de klinische indicatie van de SWK bij vrouwen met een vroeg stadium mammacarcinoom. Ondanks dat een SWK-procedure significant minder morbiditeit geeft dan de OKD, ervaart ook een klein maar relevant percentage patiënten na een SWK procedure klachten (dysesthesie en oedeem). Het doel van **hoofdstuk 6** was om een model te ontwikkelen op basis van preoperatieve variabelen waarmee we die patiënten kunnen selecteren met laag verdachte axillaire ziekte waarbij we de SWK procedure achterwege zouden kunnen laten. Over een periode van 8 jaar zijn alle patiënten retrospectief geanalyseerd (n= 2,015) waarbij de populatie gerandomiseerd werd tot een training en een validatie cohort. De variabelen geassocieerd

met nodale ziekte werden geanalyseerd en gebruikt om een voorspellend model te ontwikkelen. Het uiteindelijke model was gebaseerd op leeftijd, klinisch nodale status (cN0), morfologie, gradering, multifocaliteit en tumorgrootte, met een 'area under the curve' (AUC) van 0.83. Bij een geaccepteerde vals-negatieve uitslag van 5%, zou bij 32.8% van de patiënten axillaire chirurgie achterwege kunnen blijven. Bij een sub-analyse van patiënten met kleine unifocale tumoren (< 20mm), cN0 en geen lobulair type, zouden we bij 26.8% van de patiënten een SWK-procedure achterwege kunnen laten. Echter, vinden we het acceptabel om 5% macrometastasen te missen? We concludeerden dan ook dat het erg lastig is om die vrouwen te identificeren waarbij we veilig een SWK procedure achterwege kunnen laten. Het gehele behandelplan is daarbij van groot belang; gelet op uitgebreidheid van de operatie, wel of niet radiotherapie danwel systeemtherapie. Een aantal grote studies zijn heden nog gaande welke het mogelijk achterwege laten van de SWK-procedure evalueren. In de tussentijd kan dit nomogram gebruikt worden bij individuele therapie planning.

Chapter 9

Future perspectives

Breast cancer is a very heterogeneous disease

Breast cancer is biologically a heterogeneous entity, encompassing a diverse group of diseases with respect to clinical behaviour, response to treatment and prognosis. To optimize individual treatment, we have to acknowledge these differences and incorporate this in our daily management of breast cancer patients. By doing so, we have to realize that the possibilities and opportunities are increasing each day. The scientific community has unravelled many secrets in (breast)cancer development and biology of the many different action pathways. Much less is known however on the microenvironment of the cancer cells, for example the connective tissue of the breast, also known as stroma. Particularly the subgroup of patients with triple negative breast cancers is in need of a better understanding of their clinical and pathological pathways. This is a subgroup of great interest as they are known for their worse prognosis and lack of targeted therapies. We observed in our study of these triple negative tumours that there must be a recognizable mechanism between stroma and the cancer tissue as we saw a different prognosis between patients with a low tumour-stroma ratio compared to those with a high tumour-stroma ratio. Firstly, this finding could help to improve prognostications of patients with triple negative breast cancer. Secondly, this finding demands for more research to find out what the biology behind this observation is.

An additional variable that has been the subject of extensive research in recent years are the stromal tumour-infiltrating lymphocytes (sTILs). Stromal TILs are defined as mononuclear host immune cells (predominantly lymphocytes) within the boundary of a tumour that are located within the stroma without directly contacting the infiltrating tumour nests. Stromal TILs are a prognostic biomarker. High levels of sTILs are associated with improved outcomes (DFS and OS) and increased response to immunotherapy, chemotherapy and other targeted therapies in both early and advanced triple-negative and HER2-positive breast cancers. Assessment of TILs may enable clinicians to assess prognosis and in the future inform therapeutic discission-making when used in multivariate prognostic models in addition to the standard variables[1]. However, validation of sTILs assessment techniques are necessary before clinical use. Machine learning techniques might be the future in evaluate TIL distribution patterns[2].

Even more complex may be the so-called special types cancers, rare types of breast cancer who are significantly less common. Current decisions on treatment are based on well-established international guidelines from large RCT's and meta-analyses, but these are almost completely focussing on IBC-NST and lobular carcinomas [3]. Large databases are non-existent for these special types of breast cancer, which is problematic in conducting research. The biology behind these special type breast cancers is very diverse. In chapter 6 we made an attempt to better understand these so-called special types

breast cancer. Particular important for a better understanding of these special types of breast cancer, is forming and analysing large databases in the near future by combining (inter)national data. An example that these kind of endeavours are really possible and fruitful is the international corporation on male breast cancer[4]. More research has to be done to enhance knowledge of the molecular biological background of these tumours. From what cells did they originate from, is there any genetic agreement in some types with more common cancers and what are the key drivers?

The prognosis for patients with breast cancer is traditionally predicted by clinical and pathologic variables like age, performance status and tumour related factors like tumour size, lymph node status, hormone-receptor status, Her2-status and tumour grade. Breast cancer can also be evaluated by intrinsic subtype classification according to the St. Gallen surrogate classification 2016[5], being the following: Luminal A (ER+ and/or PR+, HER2- and Ki67% < 30%), luminal B HER- (ER+ and/or PR+, HER2- and Ki67% ≥ 30%), luminal B HER2+ (ER+ and/or PR+ and HER2+), HER2-neu non-luminal (ER/PR− and HER2+) and basal-like (ER/PR− and HER2−). In the St. Gallen consensus of 2021[6] ER-positive cancers are sometimes classified as 'luminal A-like' (lower grade, lower proliferation (lower Ki67), strong ER/PR expression), or 'luminal B-like' (higher grade, high proliferation (higher Ki67), lower levels of ER/PR expression), subtype associations that tend to correlate with genomic markers of risk. Gene expression profiling give us an even better understanding of the biological pathway and prognosis. By combining both clinicopathological and genomic aspects of breast cancer, we will be able to provide specific pathways and targets to develop personalized treatment and guidelines to ultimately provide optimal treatment in the future addressing the heterogeneous nature of breast cancer.

Less invasive treatments of the axilla

Over the last decades, significant developments in the de-escalation of treatment of the axilla have emerged following the de-escalation of treatment of the breast itself from radical mastectomy to breast conserving therapy. The sentinel lymph node biopsy (SLNB) had already revolutionized the management of clinically node-negative disease of the axilla being a safe and accurate method with substantially less post-operative morbidity than the traditionally performed axillary lymph node dissection (ALND)[7–11]. Even in patients with a positive sentinel node the need for ALND has been increasingly questioned over the years, especially in selected patients with a low axillary burden. The authors of the ACOZOG Z0011 trial, concluded that ALND did not significantly affect overall or disease-free survival in patients with clinical T1-2 breast cancer, no palpable lymphadenopathy, and a maximum of 2 SLN metastases who were treated with lumpectomy, adjuvant systemic therapy and tangential-field whole breast irradiation[12]. Considering the before mentioned, we were wondering if

we would be able to identify patients pre-operatively with limited disease of the axilla by using axillary ultrasonography (US). In chapter 2 and 3 we studied the utility and diagnostic accuracy of axillary US and US with FNAC in detecting axillary lymph node metastases and how to implement this in selecting patients pre-operatively. US of the axilla is a non-invasive, widely available and easily incorporated diagnostic technique in the work-up of breast cancer patients. We found US of the axilla to be helpful in identifying patients with a clinical T1-2 breast cancer with minimal risk of additional axillary disease. In patients with 3 or more nodal metastases the sensitivity of US combined with FNAC is especially high. With the extended use of neoadjuvant (primary) systemic therapy and new possibilities of targeted surgery of the breast and axilla, knowledge of the axilla is still of additional value. If axillary disease is pre-operatively diagnosed, the patient can receive chemotherapy prior to the operation followed by targeted axillary dissection (TAD)[13,14]. In case residual disease is still present in patients with initial cN1 (< 4 nodal metastases), additional level 1-4 irradiation could be given, sparing the patients an ALND with its known axillary and arm morbidity. In case of a pathological complete response (pCR), the patient will not only be spared an ALND but one could consider omitting radiotherapy of the axilla as well[15]. If extensive disease is still present, ALND is warranted. Identification of residual disease after neo-adjuvant systemic therapy is also of value for additional systemic therapy decision-making. In case of triple negative and high risk luminal type breast cancer, addition of capecitabine therapy is effective in prolonging disease-free and overall survival[16,17]. In patients with a germline mutation (BRCA 1/2) additional treatment with a PARP inhibitor can be of value, when given for 1 year according to the OlympiA trial[18]. In particular for the high-risk triple negative tumours (larger than 2cm or N+; or in case no pathological complete response was seen after neo-adjuvant chemotherapy), a PARP inhibitor could then be given instead of capecitabine. Knowledge of the axillary status pre-operatively with the use of ultrasonography makes a more personalized treatment possible when up front systemic treatment is warranted.

Prediction of nodal disease in breast cancer.

The SLNB substantially reduces axillary morbidity in comparison to ALND, but it does not eliminate them. Lymphedema, paraesthesia, arm and shoulder impairment and pain still exist[19–21]. Knowledge of the axillary nodal status is traditionally considered relevant for prognostic information, to improve regional control and guiding adjuvant systemic therapy decisions. It is reassuring to see from several large studies [22–25], that axillary recurrence rates in patients with and without metastases in the SLNB without additional ALND, are low. These results questioned the role of the SLN procedure in itself in patients with clinical N0 invasive breast cancer. To assess this, several trials are investigating the

outcomes of breast cancer patients with clinical node negative early breast cancer randomising between performing or omitting the SLN procedure[26–29]. The result of these trials have to be awaited and are expected in the coming year. Meanwhile, awaiting these results we investigated if we were able to select patient in which we could already omit the SLNB. To this end, we aimed to develop a model to predict the probability of axillary lymph node metastases based on variables pre-operatively known in order to select patients with an expected less than 5% chance of having nodal disease in which SLNB could be omitted. Since we know the false negative rate of the SLNB procedure is somewhere between 5-7%, we felt a threshold of a false negative rate by missing out on a positive SLN would be from a clinical point of view. We had to conclude that it is too difficult to identify a clinically relevant proportion of woman in whom we could safely omit the SLNB based on clinicopathological variables.

Future issues in personalizing breast cancer management

Breast cancer treatment has come a long way, from being a radical mutilating procedure mainly based on the anatomy of the cancer, to a multidisciplinary treatment modality combining surgery, radiotherapy and systemic therapy mainly based on the biology.

Most patients undergo breast conserving therapy, combining surgery and whole breast irradiation which includes the lowest parts of the axilla. This in turn, will have effect on the axillary recurrence rate. In case of limited axillary metastases, we know radiotherapy of the axilla to be equal to ALND with respect to axillary recurrence rate, disease free and overall survival[22,30,31] . On the other hand, in the context of de-escalation, the SLNB is said to be mandatory in case partial breast or even an active surveillance strategy is considered. The TOP1 trial for example requires a negative SLN before a wait and see policy in elderly patients with early breast cancer can be initiated[32]. Although, when selected by age over 70 years, a low-grade tumour smaller than 2cm, ER positive, Her2 negative and unsuspected nodes with axillary ultrasonography, the chance of a positive SLN is very low. One might consider omitting a SLNB in this selection of patients based on the ultrasonography of the axilla alone. The same could be considered in case of partial breast irradiation. The next step will be awaiting the several ongoing trials investigating omitting SLNB in selected patients with clinical node negative early breast cancer. Gentilini presented their first results of the SOUND trial at the St. Gallen International Breast cancer Conference 2023 and reported no differences in survival, adjuvant therapy decision making or in local recurrences between the sentinel node and observation arm (after negative US axilla) [33].

Patients with triple negative and Her2 positive breast cancer would be the next ideal candidates for further surgical de-escalation of the axilla considering excellent responses to neoadjuvant chemotherapy (NACT). In case of cN0 at diagnoses and a radiological complete response of the breast (rCR) with MRI, the chance of nodal positivity was less than 3% according to a study by Noorda et al[34]. Tadros et al.[35] observed no disease in the axilla if a pathological complete response (pCR) was achieved among patients with triple negative and HER2 positive breast cancer. Similar results were seen in other studies[36,37] including a review by Barron et al[38], who analysed data of 30,821 patients. They reported less than 2% nodal positivity in patients with triple negative or HER2 positive disease, cN0 prior to NACT and a breast pCR. With such a low risk we should consider omitting SLNB in patients with these subtypes when pCR is achieved. One ongoing study is the EUBREAST-01 prospective non-randomized trial in which patients will be recruited for the experimental arm; no SLNB in cases of breast pCR after NACT[39]. Another ongoing prospective, single arm study is the ASICS study[40], in which no SLNB is performed in cN0 patients with triple negative and HER2 positive breast cancer who achieve rCR with post-NACT MRI. Omitting SLNB in these cN0 patients post-NACT is not expected to increase the rate of distant metastases nor mortality, while avoiding SLNB morbidity. The results will have to be awaited for some years.

The low observed axillary recurrence rates are also due to extended irradiation of the breast and axilla, as well as the increasing use of chemotherapy and hormone therapy. In the Z-11 and AMAROS trial, 95% of patients received adjuvant systemic therapy with recurrence rates of 0.9% after a positive SLN in the Z-11 trial and after 10 years follow-up of the AMAROS data, recurrences of 1.82% in the axillary radiotherapy group and 0.93% in the ALND group[41,42] were reported. They confirmed a low axillary recurrence rate for both axillary irradiation and ALND with no differences in OS, DFS and locoregional control after 10 years follow-up in sentinel node positive cT1-2 breast cancer. Neo-adjuvant chemotherapy (NACT) has also been proved beneficial in reducing tumour burden and thereby making a breast conserving surgery a safe possibility[43]. Extending these results to the axilla, led to new studies concerning targeted axillary dissection. With targeted axillary dissection (TAD), the proven metastatic lymph node is marked (MARI; for example, with an iodine seed) prior to NACT and removed in addition to the SLNB at time of surgery. A recent systematic review of Swarnkar et al [44] demonstrated that the MARI and TAD procedure had hight technical success and acceptable false-negative rates in patients responding well to NACT for node positive breast cancer. Both the MARI and SLNB had a negative predictive value (NPV) of 84%[13] . Combining the MARI with SLNB (TAD procedure), results in a false negative rate (FNR) of 2% and a NPV of 97%[14]. The preliminary results of the Dutch prospective multicentre RISAS trial published by Simons et al. at the San Antonio Breast Cancer Symposium 2020

(SABCS 2020)[45] presented an identification rate of 98%, FNR 3.5% and NPV of 93.6%. Meaning 1 in 16 patients with an ypN0 actually had ypN+. Patients with triple negative breast cancer and Her2 positive breast cancers have the highest axillary pathologic complete response rate[46]. According to the Dutch guidelines an ALND or even irradiation of the axilla can be omitted in case of a negative SLNB/MARI (TAD) when less than 4 positive lymph nodes were detected prior to NACT[47]. The next step will be to find out if omitting axillary surgery is a possibility in the exceptional responders. One of the difficulties may be the radiological evaluation of the axilla post-NACT. It is important to investigate which imaging modality can reliably predict axillary response and thus making it possible to select patients in which axillary surgery can be omitted, while not missing patients with residual disease who will benefit from additional treatment. Schipper et al[48]. concluded in their review no acceptable NPV for physical examination, ultrasonography, MRI and PET-CT in detecting residual disease post-NACT when compared to axillary surgery. Ultrasonography can exclude gross nodal disease, but is less able to differentiate between pN1 and pN2-3[49]. A more recent review of Samiei et al.[46] of 13 studies describing 2380 patients, concluded that the diagnostic performance of our currently existing non-invasive imaging modalities is still limited. It is important to identify breast cancer subgroups in which the response can reliably be evaluated. The radiolabelled fluor-18-deoxyglucose positron emission tomography (FDG PET) combined with computed tomography (PET-CT) is very promising in measuring the response in especially triple negative and Her2 positive breast cancer patients (high FDG-uptake)[50,51]. Although more research is necessary, and perhaps we should look at combining different treatment modalities, like MRI-PET which shows promising result [52] . Major advances are also made in artificial intelligence, approaching the issues in a completely different way.

So, the next step forward will be to select those patients eligible for even further reducing the extend of axillary surgery. We have to keep in mind the heterogeneous character of breast cancer. There are some important studies to be awaited before the next step in personalized management of the axilla can be taken and we can safely omit the SLNB in selected patients with breast cancer and thereby reducing the morbidity associated with axillary surgery. For the near future we might focus first on the patients with low suspected axillary disease and the triple negative and HER2 positive breast cancers with good to excellent responses to NACT. Ultrasonography of the axilla is essential in de pre-operative work-up of breast cancer, guiding the best possible treatment strategy, but more research is necessary in evaluating the post-NACT response.

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Curriculum Vitae

Yvette Moorman (volledige naam Anna Maria Moorman) werd op 29 juli 1985 geboren in Emmen als oudste kind van Henk Moorman en Yvonne Moorman-Peter en oudere zus van twee jongere broers, Bas en Rick. Haar middelbare schoolperiode bracht zij door op het Katholiek Drents College in Emmen alwaar ze in 2004 haar eindexamen VWO behaalde.

In september 2004 begon ze aan de studie Biologie aan de Rijks Universiteit Groningen en behaalde in 2005 hiervan haar propedeuse. Na een aanvankelijke uitloting werd Yvette in 2005 ingeloot voor de studie Geneeskunde aan de Rijks Universiteit Groningen welke met succes afgerond werd in 2011.

Na het afronden van de Geneeskunde studie ging Yvette werken als ANIOS bij de afdeling Heelkunde in Ziekenhuis groep Twente (ZGT), locatie Almelo. In deze periode werd ook de basis gelegd van dit huidige proefschrift. Op 1 januari 2013 begon ze met haar opleiding tot Chirurg in regio Groningen onder begeleiding van dr. van Baal in het ZGT, locatie Almelo en Hengelo alwaar ze de eerste 3 jaren van de opleiding doorbracht. In 2016 werd de opleiding vervolgd in het Universitair Medisch Centrum Groningen (opleider Prof. dr. E Heineman, opgevolgd door dr. R.J. van Ginkel). Yvette heeft haar opleiding tot oncologisch en gastro-intestinaal chirurg afgerond in het Medisch Centrum Leeuwarden (opleider dr. M. Emous) op 17 februari 2020.

In 2020 begon Yvette als waarnemend Chirurg in het Nij Smellinghe Drachten en het Medisch Centrum Leeuwarden (MCL). In het MCL werd dit opgevolgd door een fellowship oncologische chirurgie. Na een waarneemperiode in het Wilhelmina ziekenhuis te Assen, werkt ze heden als oncologisch chirurg binnen de Heelkunde Friesland groep op locatie Medisch Centrum Leeuwarden. Yvette woont samen met Tim Meys en hun drie dochters, Lise, Bo en Vere in Groningen.

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