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Breast Cancer Classification According to Immunohistochemical Markers: Clinicopathologic Features in Women Treated at Pietersburg Hospital, Limpopo, South Africa

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Abstract

Background: Breast cancer molecular subtypes can provide critical prognostic information that is useful to individualize care in addition to avoiding unwarranted adjuvant chemotherapy in some patients. Micro-array-based genetic profiling used to type breast tumours is expensive and unavailable for most low- and middle-income countries. Molecular subtyping using immunohistochemical (IHC) markers has proved to be a viable alternative. We therefore used this immunohistochemical technique to classify the breast cancer subtypes in our patient population and evaluated its association with clinicopathologic characteristics in these patients.

Patients and Methods: We retrospectively evaluated immunohistochemical marker results (oestrogen, progesterone and Human Epidermal Growth Factor Receptor-2 (HER2) receptors) of 254 breast cancer patients treated at a tertiary referral hospital from 01 January 2010 to 31 December 2011 against their clinicopathological variables.

Results: Out of 329 patients with invasive breast cancer treated within the study period 254 who had complete IHC marker test results were evaluated. Of these 136(54%) were luminal A, 69(27%) triple negative breast cancer (TNBC), 29(11%) HER2- enriched, and 20(8%) luminal B. The mean age at diagnosis was 55.6, 54.4, 54.2, and 52.3 years for luminal A, TNBC, HER2, and luminal B, respectively. The rate of lymph node involvement was 80%, 66.7%, 66.2%, and 58.6% for luminal B, TNBC, luminal A, and HER2, respectively.

There was no association between molecular subtypes and clinical stage (p=0.578) or distant metastasis (p = 0.940). Grade 3 tumours were most prevalent in TNBC (59.4%) followed by luminal B (40%), HER2 (37.9%), and luminal A (37.5%). Five-year survival was 27.2%, 25%, 24.1% and 23.2% for luminal A, luminal B, HER2 and TNBC respectively (p=0.104).

Conclusion: A large proportion or our patients (22.8%, n=75) with missing IHC test results could not be classified into breast cancer molecular subtypes and may have missed an opportunity for an individualized care. There is need for IHC tests in every breast neoplasm biopsy specimen to include ER, PR, HER2, and Ki67 markers for classification of the lesion according to molecular subtypes.

Keywords: oestrogen receptor; progesterone receptor; HER2 molecular subtype

Introduction

Breast cancer is the most commonly diagnosed cancer in females worldwide with an estimated 2.3 million new cases annually [1]. In South Africa, breast cancer contributed to 23.22% of all histologically diagnosed neoplasms in females in 2019 [https://www.nicd.ac.za/centres/national-cancer-registry/cancer-statistics/] translating to 33.95 cases per 100 000 per year. The development of new therapies for breast cancer has resulted in significant increase in disease-free survival and reduction in cancer specific mortality [2].

However, selection and administration of therapies according to the patient and disease characteristics is critical in improving disease-free and overall survival, and also to prevent late treatment-related complications such as cardiac toxicities caused by anthracyclines, myelodysplastic syndrome, leukaemia and taxane-associated neuropathy [3].

Historically, confirmation of diagnosis and classification of invasive breast cancer was made according to the morphological appearance of malignant cells. The main histological types according to morphological classification are invasive ductal carcinoma not otherwise specified (IDC-NOS), and lobular carcinoma. However, breast cancer is now known to be a highly heterogeneous disease comprising distinct phenotypes with diverse clinical behaviour [4]. The different phenotypes present with diverse clinical history, morphological characteristics, therapeutic outcome, and prognosis [5]. The heterogeneousness of breast cancer was first reported twenty years ago with the discovery of intrinsic molecular subtypes that helped to explain the diversity of biological behavior and response to treatment [6, 7] which now demands that treatment for each patient be individualized.

Hierarchical cluster analysis of intrinsic genes associated with breast tumors reveal the existence of at least four molecular subtypes, namely: luminal A, luminal B, HER2-enriched, and basal-like or triple negative breast tumours [3]. Following the discovery of the molecular subtypes, several microarray-based gene expression profiling kits have been developed and are now used routinely to predict outcome for individual breast cancer patients with good prognosis who do not require adjuvant chemotherapy [8]. Examples of commercially available microarray-based gene testing kits include MammaPrint [MP], Oncotype DX, PAM-50 risk recurrence score, Breast Cancer Index, and Endo Predict [3].

Microarray-based tumour profiling using the 70-gene MammaPrint was introduced in South Africa in 2007 followed by the introduction of local referral criteria for payment by medical insurance companies [5, 9]. However, gene testing kits are expensive and not universally available to a majority of eligible patients in South Africa and other low- and middle-income countries [5, 10]. Additionally, most kits require fresh frozen tissue samples that make it necessary to repeat a biopsy to obtain fresh tissue sample for testing [10]. Initially in South Africa, analysis was performed on fresh tissue only, until 2012, when the use of formalin fixed paraffin embedded (FFPE) tissue became available and is now the only method in use [5]. This has made gene testing available to more patients than before thereby facilitating a limited degree of individualized patient care.

The process of individualizing therapy starts with the assessment of clinicopathologic characteristics such tumour size, grade, lymph node involvement, patient demographics, and several molecular markers found within the tumour. The most significant molecular markers commonly considered in treatment decision-making in a resource–constrained setting are oestrogen receptor (ER), progesterone receptor (PR), human epidermal growth factor receptor 2 (HER2) and cytokeratin 5/6, usually identified by conducting immunohistochemistry (IHC) studies on biopsy specimen of a lesion [10].

IHC uses antibodies specific for each biomarker, with the estimated number of positively staining cells in the tumour correlating to positive or negative result. IHC is considered adequate for testing ER and PR. However, IHC alone for HER2 protein expression is not perfect and leads to approximately 10% false negative rates [11] making it necessary to perform fluorescent or chromogenic in situ hybridization (FISH or CISH) to confirm HER2 gene amplification, or to establish its presence in inconclusive results. IHC and ISH show high concordance with microarray-based gene profiling and are often used as surrogates for gene expression analyses as they reflect the genetic subtypes in a nearly similar way to advanced genetic testing techniques for molecular markers. Thus, laboratory results of IHC and ISH can be used to identify histologic subtype or molecular phenotype in an accessible, affordable and easier way [12].

Even in the absence of microarray-based gene profiling, classification of breast cancers by immunohistochemistry into molecular subtypes is still crucial because the subtypes markedly influence the kind of treatment a patient is likely to respond to and the prognosis [10]. Therefore, identification of subtypes is critical in selecting the most effective therapy while minimizing unnecessary use of chemotherapy and in determining the prognosis for each patient. The aim of this study was to classify tumours in women with breast cancer treated at Pietersburg Hospital according to molecular subtypes based on immunohistochemistry markers, and to evaluate any association between molecular subtypes and clinicopathologic characteristics of these patients. The institutional ethics committee (Turfloop Research Ethics Committee) approved the study (Certificate number TREC/127/2021: PG). Permission to access patient records was provided by the Hospital administration.

Patients and Methods

We conducted a retrospective review of all breast cancer cases treated in Pietersburg Tertiary Teaching Hospital over a 2-year period from 01st January 2010 to 31st December 2011 who were subsequently followed up for up to 60 months. Cases were selected from the Medical and Radiation Oncology Clinic database.

Immunohistochemical staining

Immunohistochemical tests were performed on primary breast tumours obtained from the initial biopsy. Antibodies against ER, PR, and HER2 (Leica bond) were used according to the manufacturer's specifications. IHC analyses were also run-on Leica bond III machine. External tissue controls were used to standardize and optimize the reagents. Automation of the process guaranteed the uniformity of results and avoided variations due to human error. We collected and analysed the information as found in each patient's medical record without any alterations.

To determine IHC subtype for each patient we combined the result of ER, PR, and HER2 receptor as follows: Luminal A (ER+ and/or PR+, HER2-), luminal B (ER+ and/or PR+, HER2+), HER2-enriched (ER-, PR-, HER2 +), and TNBC/basal type [ER-, PR-, HER2). The TNBC and basal type tumours were considered together. Tumour grade was reported according to Elston-Ellis's modification of Bloom-Richardson systems [13]. Clinical staging was determined according to the American Joint Committee on Cancer (AJCC) Staging Manual 8th edition [14].

Data collection

We collected demographic data of each patient such as age at diagnosis, menopausal status, histological type, ER, PR, HER2 results, tumour grade, tumour size (T stage), involvement of regional lymph nodes (N), and metastasis (M) status. We also recorded the time interval from diagnosis to the last recorded hospital visit or up to 60 months of follow-up. The last recorded date in the clinical record before 60 months was used as a surrogate for survival time in case of patients who did not complete 60-months follow-up.

Data analysis

Data were analysed on SPSS software version 27 [SPSS, Inc. Chicago IL, United States of America]. Descriptive statistics (mean, proportions and frequency) were used to analyse the variables. Analysis of variance (ANOVA) was used to assess the association between breast cancer subtypes and continuous variables such as age and menopausal status. The Chi-squared test was used to determine the association between clinicopathological features and breast cancer subtypes [discontinuous variables]. A p-value of 0.05 was considered statistically significant. Time interval from initial diagnosis until last recorded date, or 60 months follow-up was used to calculate overall survival by the Kaplan-Meier method. Data is presented as graphs and charts.

Results

Patient characteristics are shown in Table 1.

Characteristic	Total (n=329)	TNBC n=69(21.0%)	HER2/neu n=29(8.8%)	Luminal A n=136(41.3%)	Luminal B n=20(6.1%)	Unknown n=75(22.8%)	P value
Age at diagnosis							
Mean ± SD	55.3 ±14.2	54.4 ±13.5	54.2 ±13.0	55.6 ±15.4	52.3 ±14.9	56.1±12.8	0.755
Age by group							
< 40	42 (12.8%)	7 (10.1%)	5 (17.2%)	19 (14.0%)	5 (25.0%)	6 (8.0%)	
40 - 49	81 (24.6%)	21 (30.4%)	4 (13.8%)	33 (24.3%)	4 (20.0%)	19 (25.3%)	

50 - 59	94 (28.6%)	23 (33.3%)	9 (31.0%)	31 (22.8%)	5 (25.0%)	26 (34.7%)	0.991
60 - 69	52 (15.8%)	5 (7.2%)	7 (24.1%)	26 (19.1%)	4 (20.0%)	10 (13.3%)	
± 70	60 (18.2%)	13 (18.8%)	4 (13.8%)	27 (19.9%)	2 (10.0%)	14 (18.7%)	
Age by group							
< 50	123(37.4%)	28 (40.6%)	9 (31.0%)	52 (38.2%)	9 (45.0%)	25 (33.3%)	
50 - 69	146(44.4%)	28 (40.6%)	16 (55.2%)	57 (41.9%)	9 (45.0%)	36 (48.0%)	0.783
± 70	60 (18.2%)	13 (18.8%)	4 (13.8%)	27 (19.9%)	2 (10.0%)	14 (18.7%)	
Menopausal Status							
Premenopausal	179(54.4%)	40 (58.0%)	16 (55.2%)	74 (54.4%)	12 (60.0%)	37 (49.3%)	
Postmenopausal	149(45.3%)	29 (42.0%)	13 (44.8%)	62 (45.6%)	8 (40.0%)	37 (49.3%)	0.578
Undetermined	1 (0.3%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	1 (1.3%)	
Tumor stage T							
T1	27 (8.2%)	6 (8.7%)	3 (10.3%)	9 (6.6%)	0 (0%))	9 (12.0%)	
T2	15 (4.6%)	3 (4.3%)	2 (6.9%)	7 (5.1%)	0 (0%)	3 (4.0%)	
T3	55 (16.7%)	13 (18.8%)	3 (10.3%)	24 (17.6%)	6 (30.0%)	9 (12.0%)	0.647
T4	64 (19.5%)	14 (20.3%)	9 (31.0%)	27 (19.9%)	2 (10.0%)	12 (16.0%)	
Tx	168(51.1%)	33 (47.8%)	12 (14.4)	69 (50.7%)	12 (50%)	42 (56.0%)	
Oestrogen receptor							
Positive	166(50.6%)	0 (%)	0 (0%)	135 (99.3%)	20 (100%)	11 (14.9%)	
Negative	103(31.4%)	69 (100%)	29 (100%)	1 (0.7%)	0 (0%)	4 (5.4%)	0.000
Unknown	59 (18.0)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	59 (79.7%)	
Progesterone receptor							
Positive	99 (30.1%)	1 (1.4%)	1 (3.4)	78 (57.4%)	12 (60%)	7 (9.3%)	
Negative	166(50.5%)	68 (98.6%)	27 (93.1)	58 (42.6%)	8 (40%)	5 (6.7%)	0.000
Unknown	64 (19.5%)	0 (0%)	1 (3.4)	0 (0%)	0 (0%)	63 (84%)	
HER2							
Positive	47 (14.3)	0 (0%)	28 (96%)	0 (0%)	19 (95%)	0 (0%)	
Negative	205(62.3%)	69 (100%)	1 (3.4%)	135 (99.3%)	0 (0%)	0 (0%)	0.000
Unknown	77 (23.4%)	0 (0%)	0 (0%)	1 (0.7%)	1 (5%)	75 (100%)	

Table 1: Patient characteristics.

The median age was 55 years (±14.2 standard deviation (SD), range 26 to 96 years). The most common WHO histological type was IDC-NOS (81.8%, n=269) followed by mucinous (colloid) carcinoma (4.3%, n=14), medullary carcinoma (3%, n=10), papillary carcinoma (2.7%, n=9), and other histological types (3.9%, n=13). Histology was unknown in 14 cases (4.3%) [Table 2].

WHO histological type	Number of patients (%)
Infiltrating ductal carcinoma (NOS)	269 (81.8%)
Mucinous (colloid) carcinoma	14 (4.3%)
Medullary carcinoma	10 (3%)
Invasive papillary carcinoma	9 (2.7%)
Others	13 (3.9)
Unknown	14 (4.3)

Table 2: WHO histological types

Characteristics of 254 patients with complete information for subtype classification who were further analysed are shown in Table 3.

Characteristic	Total (n=254)	Basal cell n=69(%)	HER2/neu n=29 (%)	Luminal A n=136 (%)	Luminal B n=20 (%)	P value
Lymph node status						
Positive (N1-N3)	169(66.5%)	46 (66.7%)	17 (58.6)	90 (66.2%)	16 (80.0%)	
Negative (N0)	63 (24.8%)	16 (23.2%)	10 (34.5%)	33 (24.3%)	4 (20.0%)	0.886
Not determined	22 (8.7%)	7 (10.1%)	2 (6.6%)	13 (9.6%)	0 (0%)	
Distant metastasis at diagnosis						
No metastasis (M0)	170(66.9%)	47 (68.1%)	18 (62.1%)	92 (67.6%)	13 (65%)	
Metastasis (M1)	84 (33.1%)	22 (31.9%)	11 (37.9%)	44 (32.4%)	7 (35%)	0.940
AJCC stage						
Stage I	7 (2.8%)	1 (1.4%)	1 (3.4%)	5 (3.7%)	0 (0%)	
Stage II	38 (15.0%)	13 (18.8%)	4 (13.8%)	18 (13.2%)	3 (15%)	
Stage III	119 (46.9)	31 (44.9%)	12 (41.8%)	65 (47.8%)	11 (55%)	0.578
Stage IV	85(33.5%)	22 (31.9%)	11 (37.9%)	46 (33.8%)	6 (30%)	
Unknown	5 (2.0%)	2 (2.9%)	1 (3.4%)	2 (1.5%)	0 (0%)	
Histologic grade						
Grade 1-2	109(42.9%)	25 (36.2%)	13 (44.8%)	60 (44.1%)	11 (55.0)	0.587
Grade 3	111(43.7%)	41 (59.4%)	11 (37.9%)	51 (37.5%)	8 (40.0%)	
Unknown	34 (13.4)	3 (4.3%)	5 (17.2%)	25 (18.4%)	1 (5.0%)	
Surgery						
Yes	143(56.3%)	36 (52.2%)	19 (65.5%)	75 (55.1%)	13 (65.5%)	0.997
No	107(42.1%)	32(46.40%)	10 (34.5)	58 (42.6%)	7 (34.5%)	
Unknown	4 (1.6%)	1 (1.4%)	0 (0%)	3 (2.2%)	0 (0%)	
Chemotherapy						
Yes	215(84.6%)	57 (82.6%)	25 (86.2%)	114 (83.8%)	19 (95.0%)	0.870
No	39 (15.4%)	12 (17.4%)	4 (13.8%)	22 (16.2%)	1 (5%)	
Adjuvant hormone therapy						
Yes	124(48.8%)	6 (8.7%)	5 (17.2%)	98 (72.1%)	15 (75%)	0.000
No	130(51.2%)	63 (91.3%)	24 (82.8%)	38 (27.9%)	5 (25.0%)	
Radiation therapy						
Yes	110(43.3%)	26 (37.7%)	13 (44.8%)	63 (46.3%)	8 (40.0%)	0.183
No	139(54.7%)	41 (59.4%)	15 (51.7%)	72 (52.9%)	11 (55.0%)	
Unknown	5 (2.0%)	2 (2.9%)	1 (3.4%)	1 (0.7%)	1 (5.0%)	
Survival						
0 – 12 months	79(31.1%)	27 (39.1%)	7 (24.1%)	39 (28.7%)	6 (30.0%)	
13 – 24 months	55 (21.7%)	18 (26.1%)	8 (27.6%)	25 (18.4%)	4 (20.0%)	
25 - 36 months	31 (12.2%)	6 (8.7%)	3 (10.3%)	19 (14.0%)	3 (15.0%)	0.104
37 – 48 months	15 (5.9%)	1 (1.4%)	2 (6.9%)	11 (8.1%)	1 (5.0)	
49 – 60 months	9 (3.5%)	1 (1.4%)	2 (6.9 %)	5 (3.7%)	1 (5.0%)	
61+ months	65 (25.6%)	16 (23.2%)	7 (24.1)	37 (27.2%)	5 (25%)	

Table 3: Clinico-pathologic characteristics of the molecular subtypes.

Comparison of WHO histological types and molecular subtypes are shown in Table 4.

Who Histological Type	IHC Sub-Type			
	Luminal A	Luminal B	HER2/neu	TNBC
Infiltrating ductal carcinima (NOS)	113 (83.1%)	18 (90%)	23 (79.3%)	60 (87%)
Invasive papillary carcinoma (NOS)	5 (3.7%)	1 (5%)	1 (3.4%)	1 (1.4%)
Medullary carcinoma	3 (2.2%)	-	-	4 (5.8%)

Invasive lobular carcinoma (NOS)	1 (0.7%)	-	2 (6.9%)	-
Mixed ductal and lobular	-	-	1 (3.4%)	-
Mucinous (colloid) carcinoma	8 (5.9%)	-	-	3 (4.4%)
Ductal carcinoma in situ (DCIS)	1 (0.7%)	-	-	1 (1.4%)
Others	3 (2.2%)	-	1 (3.4%)	-
Unknown	2 (1.5%)	1 (5%)	1 (3.4%)	-
Total	**136**	**20**	**29**	**69**

Table 4: Comparison of WHO histological types and molecular subtypes

Overall ER, PR and HER-2 positivity was 50.6%, 30%, and 14.3% (p=0.000) respectively. Luminal A (54%, n=136) was the most frequent molecular subtype followed by TNBC (27%, n=69), HER2-enriched (11%, 29), and luminal B (8%, n=20), respectively. Luminal A had a peak incidence in relatively younger patients (40-49 years old), whereas luminal B, TNBC, and HER2-enriched all had peak incidence at 50 – 59 years. The mean age at diagnosis for each of the molecular subtypes were 55.6±15.4 SD, 54.4±13.5 SD, 54.2±13.0 SD, and 52.3±14.9 SD for luminal A, TNBC, HER2-enriched, and luminal B, respectively (p = 0.755). The rate of lymph node involvement (N1 –N3) was 80%, 66.7%, 66.2%, and 58.6% for luminal B, TNBC, luminal A, and HER2-enriched subtypes respectively (p = 0.886). There was no association between the molecular subtype and clinical stage (p=0.578), and with distant metastasis (p = 0.940).

TNBC was associated with high-grade tumour with 41 out of 69 patients (59.4%) having grade 3 tumours, followed by luminal B (40%), HER2-enriched (37.9%), and luminal A (37.5%). Luminal A had the least percentage of grade 3 tumours (p=0.587). The five-year overall survival for the entire cohort was 25.5%. Five-year survival was 27.2%, 25%, 24.1% and 23.2% for luminal A, luminal B, HER2-enriched and TNBC, respectively (p=0.104) [Figure. 1]
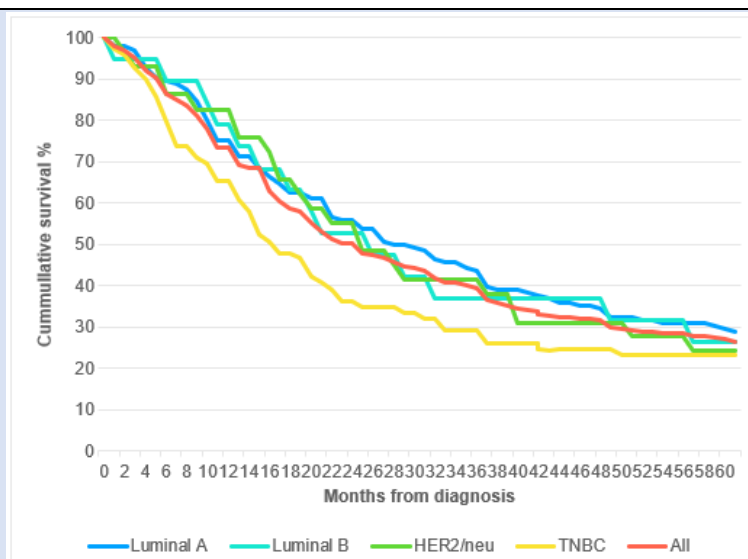

Figure 1: Overall survival according to molecular subtypes.

Discussion

The mean age of women with breast cancer in South Africa at diagnosis is 54.4 years ±14.2 SD [15]. However, median age at diagnosis is variable across different African countries and among different population groups [16]. The predominant WHO histological type reported in African women was IDC-NOS [17, 18]. Hormone receptor positivity also shows a marked regional variation as reported in a systematic review of 80 African studies published between January 1980 and April 2014 [19]. In this review, variation in hormone receptor positivity rates was explained by the diverse methods of collection, preparation and handling of the specimen whereby mean ER positivity rate was 10% (range 4 –17%) lower for studies performed on archived tumour blocks compared to those performed on freshly collected specimen.

A study in South Africa reported breast cancer molecular subtype prevalence rates of 29.5%, 24.1%, 22.4%, 22.4%, 18.1% and 6%, for luminal A, luminal B HER2-negative, triple-negative, luminal B HER2-positive, and HER2/neu-positive, respectively [18]. The main difference between this study and ours is that the former factored in proliferation index (Ki-67) in determining molecular sub-types. However, our findings are consistent with another South African study which found 53.7%, 14.6%, 20.4%, and 11% of their patients with luminal A, luminal B, TNBC, and HER2/neu, respectively [17] and with another study from Peru [10] which used similar method to ours to characterize molecular subtypes. A study comparing the prevalence of molecular sub-types in Sudanese and German women found less occurrence of luminal A (36.9% versus 68.4%), nearly the same proportion of luminal B (13% versus 10.7%), twice the proportion of HER 2/neu (15.7% versus 6.8%), and twice the proportion of TNBC (34.5% versus 14.2%) in the Sudanese and German women [20]. The authors suggest that the disparity in the proportion of molecular subtypes between the two population groups could be a result of both environmental and inherent biologic factors such as exposure to agricultural pesticides and viral and parasitic infections, which could modulate immune and tumour microenvironment and possibly induce Sudanese women to develop a more aggressive disease [20].

Lymph node involvement indicates tumour infiltration beyond the primary disease and determines treatment outcome. Our study found no association between lymph node involvement and any molecular subtype. This is consistent with other studies that found no association between molecular subtype and axillary lymph node involvement [18, 21]. Similarly, a number of studies found no difference in the prevalence of distant metastasis amongst the four molecular subtypes [18, 21, 22]. However, one study found highly significant association between molecular subtypes and axillary node status (p=0.001), and a greater prevalence of distant metastasis in HER2/neu tumours (p=0.014) compared to the other subtypes [10].

Two studies in the literature found significant association between molecular subtype and T-stage [10, 21]. In one study, mean tumour diameter at diagnosis differed significantly among the sub-types (P < 0.0001) with TNBC and HER 2/neu presenting with larger tumour diameter compared to either luminal A or luminal B [21]. Similarly, a higher percentage of T3

(54.4%) in HER2/neu, and T4 (38.6%) tumours in TNBC has been reported [10]. However, in our study there was no association between AJCC stage and molecular subtype as most patients had locally advanced or metastatic tumours.

Histological grade and molecular subtypes are both independent markers for outcome. The association between tumour grade and subtype was reported in the systematic review mentioned earlier [19]. TNBC is often associated with high grade tumours reflecting loss of oestrogen expression in more advanced forms of the disease. Other studies have also consistently revealed significant association between molecular subtypes and histological grade [10, 18, 21]. Spitale et al. (21) in a European study observed significant differences among molecular subtypes in which TNBC and HER2/neu cases show the highest prevalence of poorly differentiated phenotype (75.9% and 66.7%, respectively), whereas luminal A tumours are more frequently well to moderately differentiated (84.6%).

Vallejos et al. [10] found a significant association (p<.0001) between molecular subtype and histologic grade, with well to moderately differentiated tumours (grade 1 and 2) appearing most frequently in the luminal A (76.6%), whereas a greater percentage (70.3%) of poorly differentiated tumours (grade 3) occur in TNBC. In South Africa, Kakudji et al. [18] also found statistically significant association (p<0.001) where both luminal and non-luminal molecular subtypes are associated with grade 2 and 3 tumours.

Predictors of overall survival among breast cancer patients reported in a large study were age, menopausal status, AJCC stage, distant metastasis at diagnosis, histologic grade, Ki-67 proliferation index, tumour size and molecular subtype [21]. Among molecular subtypes TNBC and HER2/neu showed reduced survival probability at 2 years after diagnosis (89.4% and 91.7% respectively) compared to luminal A and B cases (96.5% and 96.7% respectively). Histologic type (ductal versus lobular carcinomas) did not convey significant differences in overall survival [21]. In another study with a 5-year overall survival of 73.5%, clinicopathological characteristics predicting survival were age, clinical stage, distant metastasis at diagnosis, histologic grade, axillary lymph node involvement, and tumour size [10]. Menopausal status and AJCC tumour stage did not influence survival. However, significant differences in overall survival according to molecular subtypes was observed with the highest probability of 5-year survival seen in luminal A tumours (81.9%),

followed by luminal B (72.8%), and TNBC (67.1%). HER2/neu had the worst probability of survival at 62.4% [10].

Limitations

This study has some limitations mainly resulting from missing immunohistochemical data in some records which made molecular typing difficult in 23% (n=75) of our patients. Secondly, patients who were lost to follow-up affected accurate estimation of overall survival thus possibly contributing to the survival gap in this hospital-based study. The significance of survival gap in many African breast cancer studies has been noted and the need for population-based cancer survival estimates underscored to not only reduce loss to follow-up but also to quantify heterogeneity by clinical and epidemiological factors [16].

Conclusion

This study has revealed the need for minimum IHC testing for all suspected breast neoplasm biopsy specimen to include ER, PR, HER2 (with FISH or CISH if equivocal), and Ki67 markers to facilitate classification of the lesion according to molecular subtypes. Physicians concerned with primary treatment of breast cancer patients, in resource-constrained LMICs especially surgeons should employ protocols whereby basic IHC tests are requested for each biopsy specimen submitted to the laboratory while the histopathology laboratory should also ensure that basic IHC tests are conducted on every specimen.

Declarations

Acknowledgement: Not applicable

Authors' contributions

F.O and R.M designed the study and collected the data and performed data analysis
M.B provided quality control of the data and reviewed the design and quality of the study
F.O wrote the manuscript
S.A provided advice on the study design and proof-read the manuscript
All the authors read and approved the final manuscript

Availability of data and material

The authors confirm that the data supporting the findings of this study are available within the article and/or its related files. Any other information that supports the findings of this study are available from the corresponding author, [F.O], upon reasonable request.

Competing interest

The authors declare that there is no conflict of interest

Consent for publication

Not applicable

Ethics approval and consent to participate

This study was approved by the institutional review board of University of Limpopo. Permission to access and use patient records was granted by the Pietersburg Hospital Administration

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