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Research Article

Molecular classification of breast cancers using immunohistochemical surrogates; the Sri Lankan experience

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Abstract

Background: Breast cancer diagnosis has evolved over the past decades. Today, it is vital to classify breast cancers according to genetic expression. Immunohistochemical surrogates have been identified as a cost-effective routine method to address the genetic diversity of breast cancers.

Aim: To describe the survival pattern of a cohort of breast cancer patients in the Sri Lankan setting, according to the molecular classification using IHC surrogates.

Method: Breast cancer (BC) patients investigated in our laboratory from 2006-2015 were included. Tissue microarrays were constructed using their archived BC tissue. Immunohistochemical assessment of hormone receptors, Her2, Ki67, CK5/6 and EGFR were done. The Pearson chi-square test, Kaplan-Meier model and Cox-regression model were used for analysis.

Results: The study cohort comprised 1122 patients. The complete molecular classification could be performed only for 939 patients with 27.7% -Luminal - A, 10.5% - luminal - B (Her2-), 9.1%-luminal -B (Her2+), 14.6% - Her2 enriched, 9.9% - triple negative and 8.2% - basal-like BC.

Molecular subtypes had a significant association with age (p=0.045), tumour size (p=0.001), Nottingham grade (p<0.000), lymph node stage (p=0.001) and prevalence of lympho-vascular invasion (p=0.003). Five-year BC specific survival (BCSS) of the study cohort was 75.5% (92.3% in luminal-A, 54.2% in Her2-enriched, 72.2% in triple-negative and 64.4% in basal-like groups; p<0.001). The molecular subtype (p<0.000, p=0.003) and lymph node stage (p<0.001) had an independent effect on the BCSS and RFS respectively.

Conclusion: The molecular classification using immunohistochemical surrogates, classify breast cancers into clinically useful groups with distinctively different survival rates.

Key words: breast cancer, molecular classification, immunohistochemical surrogates

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Introduction

Breast cancer, the most common cancer among females worldwide has significant morbidity and mortality. It is now accepted as a heterogeneous disease at biological, morphological, and clinical levels. Breast cancers with similar morphology may have varying prognostic attributes resulting in varying response to treatment and prognosis. This necessitates developing a classification to identify homogenous subsets of patients to offer tailor made therapy. The general rule for such classification is that it should be scientifically sound, clinically useful, easily applicable, and widely reproducible [1].

Breast cancer research in the last decade turned a corner when the molecular classification was published by Perou et al in the year 2000 based on a study which uncovered the genotypic diversity of breast cancers captured by cDNA micro arrays [2]. Four molecular subtypes were identified; estrogen receptors (ER)-positive luminal-like, basal-like, human epidermal growth factor receptor2 (Her2)-positive and normal-like breast cancers. Subsequently two subclasses of luminal-like breast cancers; luminal-A and luminal-B were identified [3]. Normal-like subtype was later removed as identifying this subgroup and its consequences are not clear. It was assumed this subtype represented samples of low tumor cell content with normal tissue components in genetic expression analysis [4].

The main drawback identified with the molecular classification of breast cancer is its cost and difficulty in carrying out gene expression on a routine basis. In an attempt to bring the molecular classification of breast cancer into clinical practice, antibodies identified by immunohistochemistry (IHC) were considered as surrogate markers [5].

In 2013, the St Gallen guidelines recommended the IHC-based molecular classification of breast cancer for clinical decision making, recognizing molecular technologies as the most precise method for identification of molecular subtypes. Where such assays are unavailable, IHC surrogates obtained by using ER, progesterone receptors (PgR), Her2 and Ki67expression were recommended [6]. Basal-like breast cancer (BLBC) can be defined using IHC surrogates, ER, PgR, Her2, CK5/6, and EGFR [5]. Although the St Gallen–guidelines identify an 80% overlap between triple negative and intrinsic BLBC, the use of basal IHC markers to identify BLBC on a routine basis is not recommended [6]. These guidelines further suggested the addition of chemotherapy based on the molecular subtype emphasizing the value of molecular classification in the management of breast cancer patients.

Breast cancer is the commonest cancer in Sri Lanka since the year 2000 with an increase in the age standardized breast cancer mortality trends [7-12]. It has been found that high grade and hormone receptor negative breast cancers are more prevalent in Sri Lankan females in keeping with Asian trends [13-15]. Breast cancer patients are managed according to established guidelines at special cancer-care units in Sri Lanka. Although IHC is used routinely to identify ER, PgR and Her2 status of the tumour, Ki67 or basal markers are not routinely performed to classify molecular subtypes. The available literature confirms that most studies on molecular classification of breast cancer were conducted in western populations, with only few studies conducted on Asians.

This paper aims to describe the distribution of molecular subtypes and document the survival pattern according to the molecular classification in a cohort of breast cancer patients in Sri Lanka.

Materials and method

This was a cohort study done using both retrospective and prospective data. All breast cancer patients who sought the services of our IHC laboratory from 2006 to 2015 were enrolled as study subjects with informed written consent. Patients whose archived paraffin wax blocks contained perished tissue and patients who had no traceable information were excluded. Tissue microarrays (TMAs) were constructed using archived breast cancer tissue blocks for IHC analysis. Details of ER, PgR and Her2 expression of each tumour and other clinico-pathological data were retrieved from the laboratory records.

Construction of Tissue Micro Arrays (TMA)

Tissue blocks were first examined for their physical suitability and a histopathologist reviewed the Haematoxylin and Eosin (H&E) stained slides of each case. The best representative tumour region with minimum fixation artifacts was selected. A core of 2mm diameter tissue from each of these donor blocks, was extracted using TMA builder™(ThermoFisher). The cores were transposed into the recipient TMA wax mold

prepared previously which contained 24 pits to hold 23 breast cancer cores. A core of brain tissue was transposed into the 24th pit in the mold as a guide to identify the rows and the columns of the TMA. A map for each TMA block was prepared to link the biomarker score to clinico-pathological data of each case.

Immunohistochemical staining and assessment

Primary antibodies, ER α clone 1D5 (Dako-M7047), PgR (Dako-M3569) and Her2 (Dako-A0485) had been used with the secondary antibody (Dako Real EnVision™) for IHC staining of all breast cancers. Ki67 (Dako M7240) in 1/75 dilution, CK 5/6 (Dako M7237) in 1/50 dilution and EGFR (Dako M3563) in 1/100 dilution were used for the corresponding markers on the TMA sections. EGFR antigen retrieval was done using proteinase. For CK5/6, microwave antigen retrieval with pH 9 buffer was done while pH 6 buffer was used for antigen retrieval by pressure cooking for the rest of the antibodies. IHC staining was done manually with a positive control.

A score of ≤2 out of 8 for ER and PgR with a score of 0 or +1 or +2 for Her2 together was considered the criterion for categorizing as triple negative breast cancer (TNBC) for this analysis. ER and PgR were considered positive when the Allred score for each was >2. Patients with Her2 equivocal staining (+2) was excluded when FISH results were not available. When FISH results were available, breast cancers were classified as Her2 positive or negative according to the result. If ≥5% of tumour cells in a TMA showed cytoplasmic staining for CK5/6 and membrane staining for EGFR, they were considered positive for the respective antibodies. More than 14% tumour cell nuclear staining with Ki67 was considered positive staining for Ki67.

Each breast cancer was assigned a molecular subtype depending on the expression of the mentioned IHC markers based on the criteria specified in the St Gallen Guidelines 2013. Complete molecular classification was done when ER, PgR, Her2 and Ki67 results were available. Since IHC markers for identifying basal-like sub type are not specified, TNBCs positive for either CK5/6 or EGFR were considered basal-like [5,16]. When a non-TNBC showed positivity for one of these basal markers, they were not classified as basal subtype but classified according to their ER, PgR, Her2 and Ki67 expression.

Follow-up and outcomes

The follow-up details of all patients were retrieved from the clinic files. Mean follow-up time was 36.9 (SD±24.1) months. Estimated median survival time was 102±11.42 months (95% confidence interval 79.61-124.38). Data for more than five years of follow-up after diagnosis was available for 27.6% of patients and at least four years of follow up was available for 40.4% of the cohort which included those who had five or more years of follow up as well. Similarly, 87.2% of the cohort had at least two years and 63% had at least three years of follow up data. There were no patients with less than one year follow up after diagnosis.

The recurrence free survival (RFS) time was calculated from the date of surgery/ commencement of neo-adjuvant chemotherapy to the date of death or the date of diagnosis of the recurrence (local / distant metastasis) [17]. Radiological and histopathological evidence was used to confirm the recurrence. The date of the said investigation was considered the date of recurrence. Patients who did not experience the relevant end point were censored at the last follow-up [17].

Breast cancer specific survival (BCSS) time was calculated from the date of diagnosis of the disease to the date of death. Patients who died of breast cancer or who died with breast cancer (progression/metastasis) were included [17]. Patients who died of other causes or from unknown causes were censored to the date of death. The cause of death of the patient was obtained from the death certificate issued by the Department of Registrar General for Births, Marriages and Deaths.

Statistical Analysis

The Pearson chi-square test was used to determine the association between molecular subtypes and clinico-pathological features. Kaplan-Meier model was used to estimate the BCSS and RFS. The log-rank test was used to compare the survival of different groups. The Kaplan-Meier model for univariate analysis and Cox-regression model with backward stepwise factor and retention method for multivariate analyses were used to estimate the predictors of survival.

Ethical approval was obtained from the Ethical Review Committee of our institution, before commencing the study.

Results

A total of 1122 patients were included. Due to the unsuitability of archived breast cancer tissue and the TMA tissue loss during staining, 183 breast cancers could not be classified. However, identifying luminal (53.4% n=595) and non-luminal (46.6%, n=519) could be done for 1114 with hormone receptor staining results alone. Follow up details were available in only 845 patients. The total cohort comprised 1121 women and one man. Most patients were between 36 to 60 years of age (65.2%, n=729) while 28.3%(n=316) were more than 60 years. Young breast cancer patients (≤35 years) comprised 6.5% (n=73). The most common molecular subtype was TNBC while the least common was basal-like (Table 1).

Table 1: Distribution of the molecular subtypes in the study cohort

Molecular type		Frequency	Percent	Valid Percent
Luminal-A		260	23.2	27.7
Luminal-B	ER+Her2- Ki67 >14% or PR-low	99	8.8	10.5
	ER+Her2+ any Ki67 any PR	85	7.6	9.1
Her2-enriched		137	12.2	14.6
TNBC		281	25.0	29.9
Basal-like		77	6.9	8.2
Total		939	83.7	100.0
Missing data*		183	16.3	
Total		1122	100.0	

*Missing data due to the unsuitability of breast cancer tissue for TMA construction and the TMA tissue loss during staining

Clinicopathological profile of the study cohort

Invasive duct carcinoma comprised 97.4% (1081/1122) of all breast cancers included. Invasive lobular carcinoma was encountered in 1.5% (17/1122). Mucinous carcinoma and invasive papillary carcinoma were present in 0.4% (4/1122) and 0.2% (20/1122) of patients, respectively. A large proportion had high Nottingham grade tumours of grade 2 and 3. (grade 1-13.5%, grade 2- 48.4%, grade 3-38.1%).

The majority (57.2%) had tumours measuring 2-5cm (T2) and only 9.1% had tumours larger than 5cm (T3). Lymph node metastases were present at the time of diagnosis, in 51.6% of patients (lymph node stage 1-24.4%, 2-16.2%, 3-11%) and lymph node metastases were not present in 48.4% of patients. Most cancers were at TNM stage II or above at the time of diagnosis (stage I-15.4%, II-46.5%, III-36.7, IV-0.8%). Lympho-vascular invasion was evident in 40.3%. Hormone receptor expression was noted in less than half of the cohort (ER-46.8%, PgR-46.1%).

The patients included in the study have been treated/are on treatment for breast cancer according to the current guidelines. There were 344 (89.5%) who received hormone therapy out of the 384 patients who had hormone receptor positive breast cancers. In the study cohort 91.8% (783/853) received chemotherapy. Only 31% of those who were positive for Her2 (68/219) have received Trastuzumab. Mastectomy with level II axillary clearance has been done for 91.7% (1031/1122) of patients. Post mastectomy radiotherapy was given to 69.6% (592/851).

There was a statistically significant difference in the distribution of molecular subtypes within the three age groups (p= 0.045). The molecular subtypes were significantly different with regard to the tumour stage (T) (p=0.001), Nottingham grade (p<0.001), lymph node stage (N) (p=0.001) and lympho-vascular invasion (p=0.003). The stage group did not significantly differ between the subtypes (p= 0.772) (Table 2). The frequency of recurrences also was significantly different between the subtypes (p=0.001). Recurrences were less frequent in luminal A (9.9%) and luminal B (Her2-) (12.5%) subtypes compared to the other subtypes. Luminal B (Her2+) had a recurrence rate of 27.1% while the percentage of patients with recurrences in Her2 enriched, TNBC and Basal like breast cancers were 26.0%, 24.2% and 18.9% respectively.

Table 2: Clinico-pathological characteristics of each molecular subtype

Molecular subtype	luminal A	luminal B (Her2-)	luminal B (Her2+)	Her2 + group	TNBC	Basal
Breast Cancer Specific Survival (BCSS)						
Nottingham grade	**0.007**	0.874	0.958	0.566	0.053	0.188
Tumour size	0.334	0.746	0.684	0.198	0.128	0.881
Lymph-node metastasis	0.616	**0.011**	0.499	**0.019**	**0.004**	**<0.001**
TNM stage	**<0.001**	0.191	0.063	**0.003**	**<0.001**	**0.004**
Lympho-vascular invasion	0.604	0.229	0.901	0.245	0.167	**0.015**
Recurrence Free Survival (RFS)						
Nottingham grade	0.183	**0.029**	**0.019**	0.356	0.946	0.870
Tumour size	0.788	0.807	0.475	0.130	0.410	0.553
Lymph-node metastasis	0.196	0.867	0.526	**0.008**	**0.008**	**0.010**
TNM stage	0.342	0.151	**0.026**	**0.040**	**<0.001**	**0.048**
Lympho-vascular invasion	0.533	0.881	0.300	0.760	0.910	**0.035**

Survival according to the molecular subtypes

Five-year BCSS and the five-year RFS of the whole study cohort were 75.5% and 69.7% respectively. There was a statistically significant difference in the BCSS and RFS among the molecular subtypes of breast cancer (p<0.001; RFS, p=0.001) (Figure 1 and 2). RFS curve of the Luminal-B (Her2+) was similar to the Her2-enriched group showing the worst RFS compared to all the other types. The cohort was again reclassified as luminal and non-luminal and the BCSS and RFS curves of these two categories were significantly different (BCSS p<0.001, RFS p=0.003) (Figure 3 and 4).

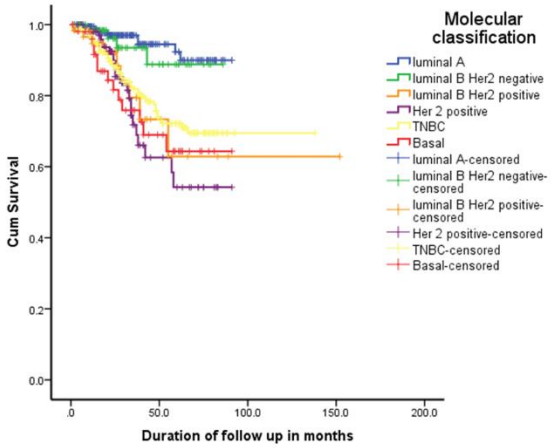

Figure 1: Breast cancer specific survival of the molecular subtypes of the cohort. (BCSS: Total=715; Luminal-A=195, Luminal-B(Her2-)=72, Luminal-B(Her2+)=60; Her2–enriched subtype=104, TNBC=230; Basal-like=54; Total events=107; log-rank P <0.001)

To eliminate the effect of advanced stage on this analysis, all patients with operable breast cancers were selected and BCSS and RFS were re-analysed. The difference between the molecular subtypes remained significant for both BCSS and RFS of operable breast cancer patients as well.

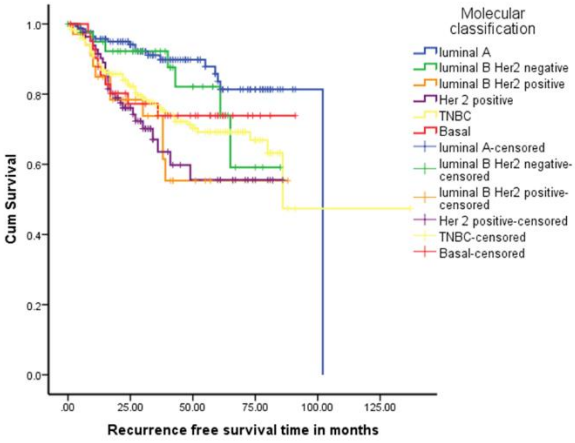

Figure 2: Recurrence free survival of the molecular subtypes of the cohort. (RFS: Total=543; Luminal-A=141, Luminal-B(Her2-) =42, Luminal-B(Her2+) =34; Her2-enriched subtype=83, TNBC=201; Basal-like=42; Total events=125; log-rank P 0.001)

The five-year BCSS of luminal-A was 92.3%. It was least in the Her2-enriched group (54.2%). TNBC had a five-year BCSS of 72.2% and for the basal-like cancers it was 64.4%. The two luminal-B types had small samples and the five-year BCSS was not calculated. Survival of patients with TNBC was compared against the basal-like breast cancer patients but no statistically significant survival difference was observed.

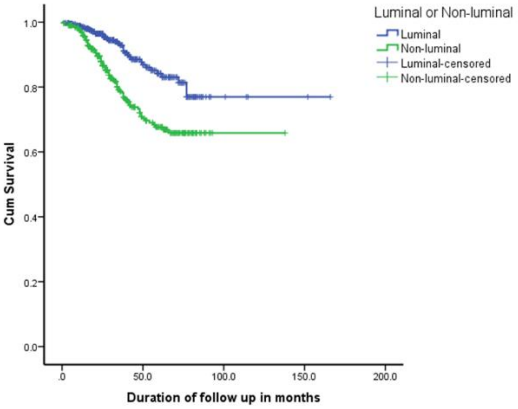

Figure 3: Breast cancer specific survival of the luminal vs non-luminal subtypes of the cohort. (BCSS: Total=840; Luminal= 433; Non-luminal=407; Total events=126; log-rank P <0.001)

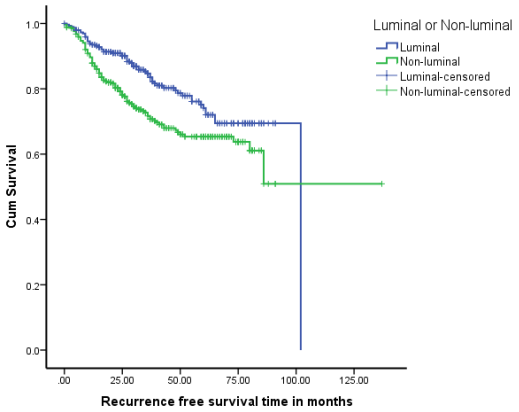

Figure 4: Recurrence free survival of luminal vs non-luminal subtypes of the cohort
(RFS: Total=638; Luminal=296, Non-luminal=342; Total events=158; log-rank P =0.003)

Factors which determined the survival of each molecular subtype was analysed separately (Table 3). TNM stage and lymph node stage were the prime factors affecting both BCSS and RFS in most of the molecular subtypes. The results of the univariate and multivariate analysis of BCSS and RFS are given in the table 4. Multivariate analysis revealed that the molecular subtype (p<0.000, p=0.003) and lymph node stage (p<0.001) had an independent effect on the BCSS and RFS of the cohort. TNM stage was not included in the multivariate analysis as the three components of TNM stage were included separately.

Table 3: Factors which affect the BCSS and RFS of each molecular subtype.

Molecular subtype	luminal A	luminal B (Her2-)	luminal B (Her2+)	Her2 + group	TNBC	Basal
Breast Cancer Specific Survival (BCSS)						
Nottingham grade	**0.007**	0.874	0.958	0.566	0.053	0.188
Tumour size	0.334	0.746	0.684	0.198	0.128	0.881
Lymph-node metastasis	0.616	**0.011**	0.499	**0.019**	**0.004**	**<0.001**
TNM stage	**<0.001**	0.191	0.063	**0.003**	**<0.001**	**0.004**
Lympho-vascular invasion	0.604	0.229	0.901	0.245	0.167	**0.015**
Recurrence Free Survival (RFS)						
Nottingham grade	0.183	**0.029**	**0.019**	0.356	0.946	0.870
Tumour size	0.788	0.807	0.475	0.130	0.410	0.553
Lymph-node metastasis	0.196	0.867	0.526	**0.008**	**0.008**	**0.010**
TNM stage	0.342	0.151	**0.026**	**0.040**	**<0.001**	**0.048**
Lympho-vascular invasion	0.533	0.881	0.300	0.760	0.910	**0.035**

Her2, human epidermal growth factor receptor 2; TNBC, triple negative breast cancer; BCSS, breast cancer specific survival; RFS, recurrence free survival; TNM, tumour node metastasis

Discussion

Many clinicopathological features are considered in the current management of patients with breast cancer. Age at presentation, size and grade of the tumour, vascular invasion, lymph node status, TNM stage, hormone receptor and Her2 status are being routinely used for prognostication and stratification of breast cancer patients for therapeutic decision making [17-19]. However, it has been often claimed that even within the same strata the outcome may differ. This clinical observation has led to the designing many research protocols on better classification of breast cancer. Although gene expression profiling has expanded the landscape of research on biology of breast cancer evolving towards better classification, its routine clinical utility is debatable due to its time-consuming and low cost-effective nature. Conversely the IHC surrogates for molecular classification seems much more appealing to a country like ours where the health budget of the country is borne by the government to a great extent.

Table 4: Univariate and multivariate analysis of BCSS and RFS of the study cohort.

Factors	Univariate analysis (P value)	Multivariate analysis (P value)
Breast Cancer Specific Survival (BCSS)		
Molecular subtypes	<0.001	**<0.000**
Lymph-node stage	<0.001	**<0.001**
Tumour size	0.006	0.065
Nottingham grade	0.004	0.485
Lympho-vascular invasion	0.003	0.905
Lymph-node metastasis	<0.001	
Recurrence Free Survival (RFS)		
Molecular subtypes	0.001	**0.003**
Lymph-node stage	<0.001	**<0.001**
Lympho-vascular invasion	0.030	0.985
Nottingham grade	0.057	
Tumour size	0.139	
Lymph-node metastasis	<0.001	

BCSS, breast cancer specific survival; RFS, recurrence free survival; p, significance; TNM, tumour node metastasis

Routine histopathology reporting of breast cancer in Sri Lanka reaches standards comparable to many developed countries. Immunohistochemistry services are available at identified large central histopathology laboratories in Sri Lanka in order to maintain consistency in staining procedures and to reduce the cost. There are guidelines developed for reporting of ER, PgR and Her2 by the College of Pathologists of Sri Lanka, which are almost similar to the guidelines used worldwide. However, Ki67 and EGFR are not performed routinely; hence identification of basal-like breast cancer is limited to its triple negative nature. Therefore, there are no data on the proportion of breast cancers that belong to the basal-like subgroup in our

country. Our study revealed that basal-like breast cancers defined as TNBC with positivity either with CK5/6 or EGFR comprised 8.2% of the cohort and a similar percentage can be expected in the rest of the country. Basal-like breast cancers are reported to have a poor prognosis but are found to respond to platinum-based chemotherapy [16].

The luminal A molecular type comprised 27.7% of the study cohort and does not reach the proportion seen in many other countries; 50-60% [3] and 30-40% [5] reported in two publications. Prevalence of luminal-B (19.6%) is comparable to some reports;15-20% [3] but is less compared to 20-30% reported by others [5]. The overall hormone receptor expression in our population (53.4%) is less compared to 65-75% reported by most western studies [3,5] explaining why the two luminal subtypes are less prevalent.

Breast cancer in Asia including Sri Lanka is often reported to be ER negative [13-15,20]. Hence, the prevalence of luminal breast cancer which has a better prognosis with available targeted therapy is also low. This contributes to the comparatively low BCSS in our cohort.
A study done in North India has reported a very similar result where they too had a high prevalence of TNBC [21]. An Indonesian study done in 2014 too, shows a similar distribution of molecular subtypes but with a lesser percentage of TNBC [22]. Results of a study done in Pakistan deviates from our results and had reported a higher prevalence of luminal-A breast cancers (45.8%) and a significantly lesser prevalence TNBC (18.6%). The Her2-enriched (17.8%) subtypes was more frequent in their study [23]. Another study done on a large cohort of Chinese women revealed that the prevalence of the luminal-A, luminal-B, Her2-enriched and TNBC were 48.6%, 16.7%, 13.7%, and 12.9%, respectively [24]. This indicates that even in Asian countries there are variations in the prevalence of the molecular subtypes. Inter-laboratory variations in IHC staining may have contributed to this variation in the distribution of molecular subtypes to some extent.

The good prognostic features frequently seen in luminal-A tumours, namely low histological grade, a low recurrence rate and a better survival outcome [5,25] were evident in the present study cohort and had the least rate of recurrences and lympho-vascular invasion. They were mostly grade 1 or 2 and was the commonest among the elderly patients. These good prognostic attributes have resulted in luminal-A breast cancer patients having the best BCSS (Five-year BCSS=92.3%, Figure 1) out of all the other subtypes.

Luminal-B breast cancers showed a poor prognosis compared to the luminal-A justifying its identification separately in the molecular classification. It showed the highest rate of recurrences and more grade 2 and 3 tumours in our cohort. The effect of these poor prognostic features is revealed in the corresponding survival curves lying between survival curves for luminal-A and TNBCs (Figure 1and 2). Although, they are ER positive, the survival is poor, necessitating their identification for better prediction of survival and addition of chemotherapy. Addition of Ki67 to the routine prognostic marker panel for breast cancer, at least when the tumour is ER positive will allow identification of the luminal-B patients with poor prognosis.

BLBC account for 60%-90% all TNBC and comprise 15% of all invasive breast cancers [5,25] These tumors follow an aggressive clinical course and currently lack any form of standard targeted systemic therapy [26]. In our study they were 8.2% of the total cohort and showed poor BCSS (Figure 1) as they were below the survival curves for luminal and TNBC. However, there was no significant difference in survival between TNBC and BLBC in our cohort. Therefore, identification of BLBC in the current routine setting may not be cost effective. However, adding two basal markers (CK5/6 and EGFR) to the IHC prognostic panel as second line markers for a TNBC may be encouraged when cost is not a limitation.

The present study confirms that molecular subtypes have an independent effect on both RFS and BCSS in addition to the lymph node

stage. The effect of other well recognized prognostic factors on the survival of patients in each of the molecular subtype varies.

This study has a few of the limitations inherent to retrospective studies, eg. inability to include a completely consecutive sample as a result of exclusion of some cases due to perished archived tissue and absence of some of the data. TMA is well recognized as a tool for evaluation of biomarkers yet suffers loss of data due to tissue loss during microtomy and staining. However, we have limited the data loss due to tissue loss in TMAs by making extra TMAs. Detection of Ki67 expression is often claimed to be limited in archival tissue and this limitation may have affected the prevalence of the luminal B group. The re-analysis of the study group based on the two major breast cancer subtypes (luminal, non-luminal) eliminating the effect of any limitation by Ki67, also demonstrated a significant survival difference. The BCSS of luminal cancers is claimed to be better demonstrated in a follow-up of more than 10 years. Most subjects in the current study population did not have >10 year follow-up and authors consider it as a limitation in the current study.

This is the first survival outcome-based report on the molecular classification of breast cancer patients in Sri Lanka. It also gives some clarifications to the lower BCSS in our setting; very low prevalence of luminal-A cancers with available targeted therapy and high prevalence of TNBC with no targeted therapy.

Conclusions

This study confirms that the molecular classification of breast cancers based on IHC surrogates, classify breast cancers into clinically distinctive groups with significant different survival in our setting too. Adding Ki67, CK5/6 and EGFR to the routine prognostic marker panel, at least as second line markers, will enable better classification of breast cancers as establishing costly gene expression profiling on a routine basis is beyond expectations in our setting.

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