Immunohistochemical Profile, Pattern of Recurrence, and Time to Progression of Non-Metastatic Breast Cancer Patients of the Department of Health-Breast Cancer Medicines Access Program

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ABSTRACT

Background. Breast cancer remains to be the leading cause of malignancy among women and survival rates vary worldwide. Molecular and immunohistochemical (IHC) profiling of breast cancer has emerged to improve treatment, which led to 6 different breast cancer subtypes luminal-A, luminal-B, Her-2 enriched, basal-like, claudin low, and normal breast. Essentially, this guides clinicians as to the choice of treatment and prognostication of disease. This study evaluates the characteristics of the different IHC subtypes of breast cancer among Filipinos as to pattern of recurrence and time to progression (TTP) within their 1st 2 years of follow-up.

Methods. This is a retrospective cohort study, approved by the University of the Philippines Manila Research Ethics Board (UPMREB). Study population included breast cancer patients enrolled in the DOH-BCMAP and managed at the medical oncology clinics of the Philippine General Hospital (PGH) and Jose R. Reyes Memorial Medical Center (JRRMMC) from 1 May 2011 to 31 December 2013. Patients' demographics, disease and treatment profile were gathered from the medical charts. Patients were grouped into 12 different IHC subtypes utilizing only IHC staining results of Her2neu, ER and PR. Disease progression/ relapse and time to progression (TTP) were primary outcomes analyzed and compared between subtypes using SPSS.

Results. There were 368 eligible patients; 50% were >50 years old, 48% postmenopausal, 34% stage IIA, and 94% had invasive ductal carcinoma. About 88% completed their chemotherapy regimen, mostly AC-T. At 1 to 2 years follow-up, 18% had disease progression, mostly distant metastasis, with HER2neu(-)/ER(-)/PR(-), HER2(+), and HER2neu(-)/ER(+)/PR(+) subtypes having the most number of disease progression. The HER2neu(-)/ER(-)/PR(-) subtype had the shortest median TTP (11 months $\pm$ 9sd). HER2(+) subtype had median TTP of 14$\pm$8 sd, while HER2neu(-)/ER(+)/PR(+) had median TTP at 11.6 $\pm$7.41 sd. The median TTPs among the different IHC subtypes were statistically comparable.

Conclusion. Filipinas with non-metastatic breast cancer after surgery and mainly on adjuvant chemotherapy started to develop disease progression/ relapse within the first 2 years of follow-up; 82% had no relapse. At these early years of follow-up, the median TTPs among the different breast cancer IHC subtypes who went into relapse were comparable, although HER2neu(+) regardless of ER/PR subtype tended to have more disease progression, followed by HER2neu(-)/ ER(-)/ regardless of PR subtype, and then HER2neu(-)/ER(+)/ regardless of PR subtype. IHC resultant HER2neu(+) regardless of ER/PR and HER2neu(-)/ER(-)/PR(-/+) subtypes can serve as early prognosticators of breast cancer relapse.

Key Words: breast cancer, IHC subtypes, TTP

Introduction

Breast cancer remains to be the leading cause of malignancy among women worldwide. It is commonly prevalent among the developed world but incidence in developing countries is noticeably increasing. Survival rates vary greatly worldwide, ranging from 60-80% in the middle to high-income countries and below 40% in low-income ones. This low survival maybe attributed to the lack of early detection programs, resulting in a high proportion of women presenting with late-stage disease, as well as by the lack of adequate diagnosis and treatment facilities.[1]

Similarly, breast cancer is also the leading cause of malignancy in the Philippines among women (28%) and even for both sexes combined (15%). It has the highest registered incidence rate in Asia at 58.9% among 187 countries over a 30-year period from 1980 to 2010.[2] Incidence rate starts rising steeply at age 30 with an average annual percentage change of 0.9%. At least 1 of 13 Filipino women

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will develop breast cancer in her lifetime and 3 of 100 women would have a likelihood of getting breast cancer before age 75. In 2010, breast cancer ranked as the 3rd leading cause of cancer deaths in both sexes (8%), and the highest among women (18%). The estimated national age-standardized mortality rate was 11.9 per 100,000 women. One (1.2%) of 100 women would have died from breast cancer before age 75.[2]

Several efforts have been made in the past in order to improve management and survival among breast cancer patients; but epidemiological data still showed comparable trend in treatment outcomes and still pose great challenges. Until in the recent time, gene signature studies in breast cancer have paved the way in defining peculiar characteristics of breast cancer through molecular sub-typing analysis. Investigations of these subtypes via immunohistochemical (IHC) markers have given insight into the heterogeneous nature of breast cancer, which are biologically discrete and display different behaviors. IHC markers have been found to be predictors for survival, response to systemic therapy, and locoregional or distant recurrences. The analysis of estrogen receptor (ER), progesterone receptor (PR), and Her2neu oncogene expression in breast tissue sub-classifies breast cancer patients, and this is considered standard of care in the approach to breast cancer treatment.

Eroles et al[3] classified breast cancer into six different subtypes utilizing ER, PR, HER2, and CK5/6 EGFR IHC and molecular analysis, into luminal-A [ER(+) PR(+) HER2neu(-) CK5/6 EGFR(-)]; luminal-B [(ER(+/-) PR(+/-) HER2neu(-/+) CK5/6 EGFR(-)]; HER2neu-enriched [ER(-) PR(-) HER2neu(+) CK5/6 EGFR(+/-)]; basal-like [ER(-) PR(-) HER2neu(-) CK5/6 EGFR(+)]; claudin-low [ER(-) PR(-) HER2neu(-) CK5/6 EGFR(+/-)], and normal breast [ER(-/+) HER2(-) CK5/6 EGFR(+)]. Basal-like [ER(-) PR(-) HER2neu(-) CK5/6 EGFR(+)] and claudin-low [ER(-) PR(-) HER2neu(-) CK5/6 EGFR(+/-)] are collectively called triple negative breast cancer, considering only negative HER2neu/ER/PR results.

Luminal-A has a better prognosis and is the most common subtype (50-60%), with a relapse rate of 27.8% and median survival from the time of relapse of 2.2 years, significantly better than other subtypes with a distinct pattern of recurrence more of bone metastases (18.7%).[4] Luminal B (10 to 20%; with ER(-)/HER2neu(+) combinations) has a more aggressive phenotype, higher histological grade and proliferative index, and worse prognosis than luminal A; survival from time of relapse is lower (1.6 years) with bone (30%) as the most common site of distant metastasis; another site is liver (13.8%).[3] HER2neu-enriched subtype (15-20%) is characteristically poor in prognosis and highly proliferative with high histological grade (75%), but with the advent of anti-HER2 treatment, survival has substantially improved.[3] Triple negative breast cancer (10-20%) of mainly basal-like type, has worse prognosis than luminal subtypes

with higher relapse rate in the first 3 years of follow-up.[5] These triple negative breast tumors have high mitotic index, tumor necrosis, expanding margins and a clear stromal lymphocytic response[6]; the pattern of metastatic relapse is aggressive with predominance for visceral organs, mainly lung, central nervous system and lymph nodes.[7] Normal breast subtype has 5-10% occurrence.[3]

Several studies[8-17] have shown numerous advantages and usefulness of IHC sub-typing in breast cancer in terms of approach in the management of patients and prognostication. However, there are reports showing that the prognostic and etiologic importance of being a heterogeneous disease is complicated by many factors, including the observation that differences in clinical outcomes often correlate with race. Previous studies have shown preliminary evidences and high probabilities of molecular differences across ethnic and geographic groups, and some studies have showed that racial/ethnic differences have been observed among the different molecular subtypes in breast cancer in terms of incidence, prevalence, and treatment outcomes.[14,18-21] This study describes the characteristic and behavior of the different IHC subtypes among Filipinos with breast cancer, particularly in terms of patterns of- and time to- disease progression within the first 1-2 years follow-up after treatment with surgery and chemotherapy, with hormone therapy and radiotherapy as indicated. How do HER2neu/ ER/ PR IHC sub-types of breast cancer prognosticate the Filipino breast cancer patient, given a Philippine government hospital setting, where resources may only accommodate IHC assays, without the luxury of molecular profiling including gene amplification and CK5/6 EGFR?

Methods

This is a retrospective cohort study among Filipinos with histologically proven primary breast carcinoma stages I to III-B, who underwent definitive surgery, chemotherapy, and had radiotherapy and hormonal therapy once indicated. They were patients of the medical oncology clinics of the Philippine General Hospital (PGH) and Jose R. Reyes Memorial Medical Center (JRRMMC), two of six satellite institutions of the DOH–Breast Cancer Management Access Program (DOH-BCMAP), which provides fully subsidized medicines for chemotherapy within 15 weeks from definitive surgery, as well as IHC testing for ER, PR, and HER2neu. Philippine Cancer Society provided the patient navigation program for the DOH-BCMAP. This study included those patients enrolled since the program started in JRRMMC (1 May 2011) and PGH (1 January 2012) and followed-up for at least 1 year (31 December 2013 as data-lock).

The University of the Philippines Manila Research Ethics Board (UPMREB) approved the protocol; data confidentiality was observed.

The database and records of the eligible breast cancer patients were reviewed. Data gathered included birth date, sex, date of enrollment, disease profile, cancer stage, IHC results for HER2-neu (positive, negative, or equivocal), ER (positive or negative), and PR (positive or negative), treatment profile such as type of surgical procedure done (modified radical mastectomy/ MRM or breast conservation surgery/ BCS), chemotherapy regimen and status (complete or incomplete), radiotherapy (if any, given after completion of chemotherapy cycles), and hormonal therapy (if any, given after completion of chemotherapy cycles). Disease progression/ relapse data included progression site (local and distant) and date of recurrences. Recent survival status of patient was gathered (alive, dead, or lost-to-follow-up); for deceased patients, date and cause of death were gathered.

Classification of breast cancer patients into different subtypes was by IHC staining results for HER-2neu, ER, and PR. The IHC tests were all done at Medical Laboratory of the National Kidney and Transplant Institute, East Avenue, Quezon City and interpreted as follows: 1) HER2neu protein over-expression: 0 to +1 as negative, 2+ as equivocal, and 3+ as positive (uniform intense membrane staining of >30% of invasive tumor cells); 2) ER/PR receptor assay: total score ≤ 2 as negative, and total score ≥ 3 as positive. The subjects were classified into different IHC subtypes of breast cancer (Table 1).

Table 1. IHC HER2neu/ER/PR subtypes of breast cancer

Subtype	HER2neu status	ER status	PR status
A	+	+	+
B	+	-	+
C	-	+	+
D	+	+	-
E	+	-	-
F	-	+	-
G	-	-	-
H	-	-	+
I	E	+	+
J	E	+	-
K	E	-	+
L	E	-	-

Legend: (+) positive; (-) negative; (E) equivocal.
Compared to standard subtypes luminal A [ER(+) PR(+) HER2(-) CK5/6 EGFR(-)]; luminal B [(ER(+/-) PR(+/-) HER2(-/+) CK5/6 EGFR(-)]; HER2-enriched [ER(-) PR(-) HER2(+) CK5/6 EGFR(+/-)]; basal-like [ER(-) PR(-) HER2(-) CK5/6 EGFR(+)];claudin-low [ER(-) PR(-) HER2(-) CK5/6 EGFR (+/-)]; and normal breast [ER(-/+) HER2(-) CK5/6 EGFR(+)].

Disease progression (recurrence or distant metastasis) during the 90-day post-surgery period disqualified the patient from study. A disease progression event after 90 days was considered as a study event.

Descriptive statistics was used to describe patient's profile, disease profile, and treatment profile of the study population. Times to progression (local recurrence and/ or distant metastasis) among the different IHC breast cancer subtypes were the primary outcome. Time to progression (TTP), disease-free survival rate (DFS), and overall survival (OS) rate (secondary study outcomes) during the 1-2 years follow-up, from the time of first definitive treatment were evaluated. Risk ratios were computed to compare the time to recurrence/progression (TTP) during the first 1-2 years follow-up; chi-square test was used to determine the significance of the association. Kaplan-Meier survival analyses with log-rank test to assess for significance were done for the DFS and OS rates. Data analysis used SPSS statistical program; a p<0.05 was considered statistically significant.

A limitation of this study is the non-accessibility of FISH, CISH or DISH laboratory tests to verify equivocal IHC HER2neu assays; hence the patient groups with IHC equivocal results were part of the IHC subtypes studied. There were also 42 patients with unspecified IHC results; and 2 patients with incomplete data set. Priority stages to include in the DOH-BCMP were stages IIIA or better, hence stage IIIB is of small proportion.

Results

There were 368 eligible non-metastatic breast cancer patients enrolled in the DOH-BCMAP from 1 May 2011 to 31 December 2013 in both institutions from JRRMMC and UP-PGH (259 and 109, respectively). As shown in Table 2, 49% were >50 years old; 50% were menopausal; 67% had no documented family history of cancer; and the average BSA was 1.57m².

Table 2. Baseline characteristics

Demographic Characteristics	Total (N=368)	
	Frequency	Relative Frequency (%)
Age Group		
• <35 year	25	6.79%
• 35-50 year	162	44.02%
• >50 year	181	49.18%
Menopausal status		
• Premenopausal	39	10.60%
• Peri-menopausal	145	39.40%
• Postmenopausal	176	47.83%
• Surgical menopause	8	2.17%
Family History of Cancer		
• None	179	67.04
• Breast	16	5.99
• Ovary	10	3.75
• Colorectal	3	1.12
• Lung	2	0.75
• Fibrosarcoma	2	0.75
• Colon	1	0.37%
• Prostate	1	0.37%
• Liver	1	0.37%
• Cancer, NOS	51	19.10%
BSA (Body Surface Area)	Median = 1.57 m²	

The most common histopathology was invasive ductal carcinoma (93%; 343/368); 33% (123/368) were at stage IIA, followed by stage IIIA (27%) (Figures 1 and 2).
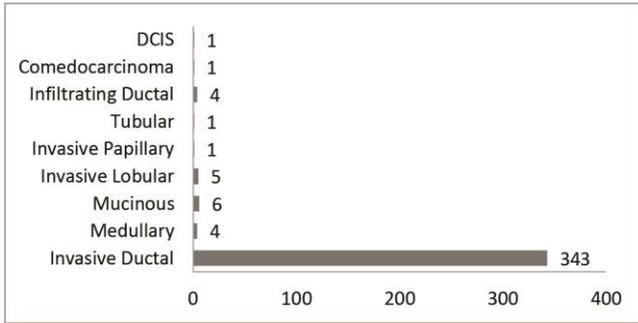
Figure 1. Histopathology, breast cancer patients, DOH-BCMAP in PGH and JRRMMC
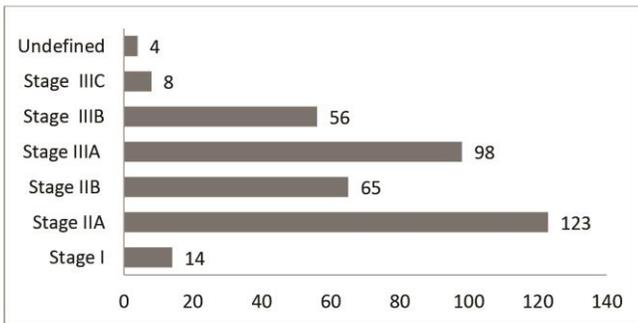

Figure 2. Stage of disease, breast cancer patients, DOH-BCMAP in PGH and JRRMMC

By IHC results (Figure 3), HER2neu(-)/ER(-)/PR(-) disease was the most common IHC subtype at 20% (n=72/366) followed by the HER2neu(-)/ER(+)/PR(+) at 16% (57/366) and Her2neu(+)/ER(-)/PR(-) at 12% (45/366).
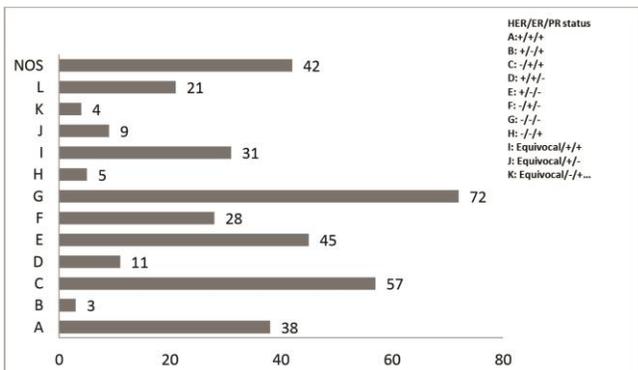

Figure 3. IHC subtypes, breast cancer patients, DOH-BCMAP in PGH and JRRMMC

Among the three most common IHC subtypes (Figures 4 to 6), HER2neu(-)/ER(-)/PR(-) was noted to be common among ages 35-50 years old, in postmenopausal patients, and mostly in stage IIIA; HER2neu(-)/ER(+)/PR(+) was seen mostly among >35 years old, in peri-menopausal period, and

in stage IIA; and HER2neu(+)/ER(-)/PR(-) subtype was seen in >50 years old, among postmenopausal, and in stage IIA.
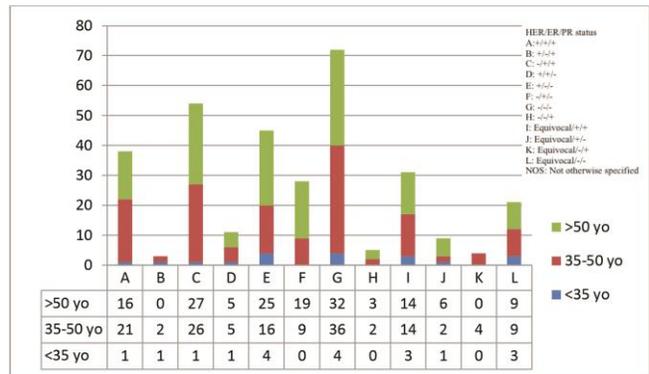

Figure 4. IHC subtypes by age group, breast cancer patients, DOH-BCMAP in PGH and JRRMMC

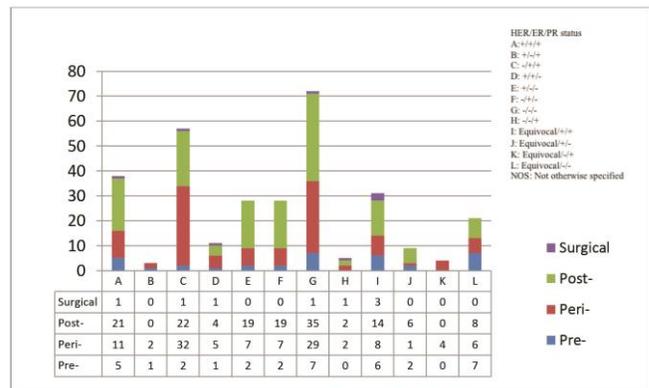**Figure 5.** IHC subtypes by menopausal status, breast cancer patients, DOH-BCMAP in PGH and JRRMMC
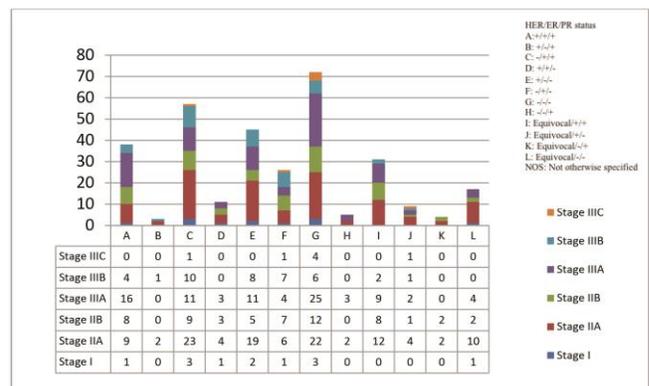

Figure 6. IHC subtypes by stage, breast cancer patients, DOH-BCMAP in PGH and JRRMMC

Modified radical mastectomy (MRM) was done in 87% and 10% had a biopsy-only procedure (these patients were Stage IIIB patients managed with neo-adjuvant chemotherapy prior to the planned definitive surgery)

(Tables 3 and 4). Majority completed the prescribed chemotherapy (87.77%), mainly adjuvant. The most common chemotherapy regimen was AC-T (adriamycin and cyclophosphamide, followed by docetaxel) in 59% of cases. Five patients who were given tamoxifen alone had stage I, ER-positive disease.

Table 3. Treatment profile, breast cancer patients, DOH-BCMAP in PGH and JRRMMC

Treatment Category	Total (N= 368)	
	Frequency	Relative Frequency (%)
Surgery		
• MRM	323	87.77%
• Lumpectomy	9	2.45%
• Biopsy	36	9.78%
Chemotherapy Regimen		
• CMF	1	0.27%
• AC	28	7.61
• AC-T	217	58.97%
• TC	35	9.51
• FAC	82	22.28%
• Tamoxifen only	5	1.36%
Completion of Chemotherapy		
• Complete	323	87.77%
• Incomplete	45	13%
Hormone therapy		
• Yes	186	51.81%
• No	173	48.19%

Legend: MRM-Modified radical mastectomy; CMF-cyclophosphamide, methotrexate, 5-FU; AC-adriamycin, cyclophosphamide; AC-T-adriamycin, cyclophosphamide, then docetaxel; TC-docetaxel and cyclophosphamide; FAC-5-FU, adriamyxcin, cyclophosphamide.

Table 4. Chemotherapy regimen, by stage, breast cancer patients, DOH-BCMAP in PGH and JRRMMC

Stage	Chemotherapy Regimen						Total
	CMF	AC	AC-T	TC	FAC	TAM	
Stage I	0	0	7	1 (7.14%)	6	0	14
		(50%)			(42.86%)		(100%)
Stage IIA	0	13	68	15	24	3	123
		(10.57%)	(55.28%)	(12.20%)	(19.51%)	(2.44%)	(100%)
Stage IIB	0	4	51	3	6	1	65
		(6.15%)	(78.46%)	(4.62%)	(9.23%)	(1.54%)	(100%)
Stage IIIA	0	5	64	11	17	1	98
		(5.10%)	(65.31%)	(11.22%)	(17.35%)	(1.02%)	(100%)
Stage IIIB	1	5	19	4	27	0	56
	(1.79%)	(8.93%)	(33.93%)	(11.43%)	(48.21%)		(100%)

Legend: CMF-cyclophosphamide, methotrexate, 5-FU; AC-adriamycin, cyclophosphamide; AC-T-adriamycin, cyclophosphamide, then docetaxel; TC-docetaxel and cyclophosphamide; FAC-5-FU,adriamycin, cyclophosphamide; TAM-tamoxifen.

There was a total of 67 (18%) patients with disease progression. Progression to distant sites was most common at 54%, followed by local recurrences at 36%. Site of disease progression was not specified in 10%.

As seen in Table 5, Figures 7 and 8, HER2neu(-)/ER(-)/PR(-) disease had the most disease progression or relapse at 24%, followed by HER2neu(+)/ER(-)/PR(-) at 22%, and then HER2neu(-)/ER(+)/PR(+) at 13%. Relapse noted

among HER2neu(-)/ER(-)/PR(-) subtype were mostly at distant sites, with metastases to the liver as the most common; likewise, HER2neu(+)/ER(-)/PR(-) had mainly distant metastatic locations with lung as the most common; HER2neu(-)/ER(+)/PR(+) subtype had mainly local recurrence, and if with distant relapse frequently this was noted in the bone.

Other HER2neu (+) subtypes such as subtypes A and D demonstrated disease progression more on the distant sites in lung and liver, respectively. Subtype-L [equivocal HER2neu/ER(-)/PR(-), which can be HER2neu(+) or HER2neu(-) group] had 8.95% disease progression mainly in distant sites.
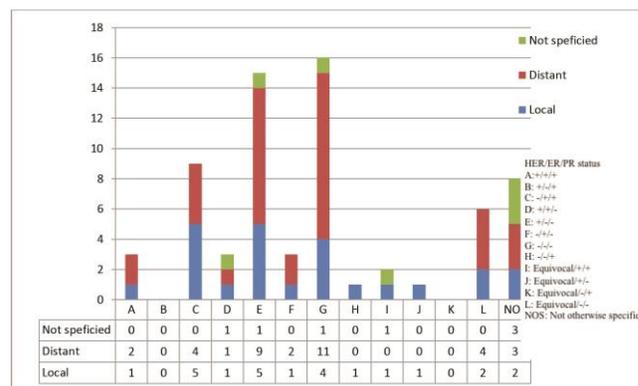

Figure 7. Disease progression site by IHC subtypes, breast cancer patients, DOH-BCMAP in PGH and JRRMMC
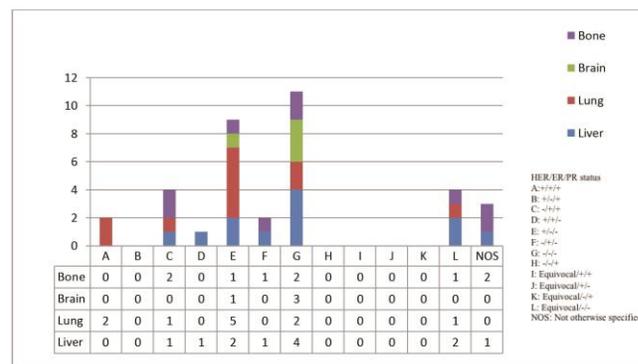

Figure 8. Distant metastatic sites by IHC subtypes of breast cancer patients, DOH-BCMAP in PGH and JRRMMC

TTP by IHC subtypes is shown in Table 6. HER2neu(-)/ER(-)/PR(-) had the shortest median TTP at 11 months $\pm$ 9 sd; HER2neu(+)/ER(-)/PR(-) showed a median TTP of 14 months $\pm$ 8 sd; while HER2neu(-)/ER(+)/PR(+) had median TTP at 11.6 months $\pm$ 7.41 sd. Within the first 1-2 years follow-up, however, the differences in the median TTP across all IHC subtypes were statistically comparable.

Table 5. Disease progression by IHC subtypes, breast cancer patients, DOH-BCMAP in PGH and JRRMMC

| Subtypes: Her2/ER/PR	Type and Site of Recurrences			Total Frequency	Percentage
	Local	Distant Recurrence	Not specified		
A: +/+/+	1	2	-	3	4.52%
B: +/-/+	0	0	-	0	0
C: -/+/+	5	4	-	9	13.43%
D: +/+/-	1	1	1	3	4.47%
E: +/-/-	5	9	1	15	22.38%
F: -/+/-	1	2	-	3	4.47%
G: -/-/-	4	11	1	16	23.88%
H: -/-/+	1	0	-	1	1.49%
I: E/+/+	1	0	1	2	2.98%
J: E/+/-	1	0	-	1	1.49%
K: E/-/+	0	0	-	0	0
L: E/-/-	2	4	-	6	8.95%
NOS	2	3	3	8	11.94%
Total	24	36	7	N = 67	100%

Legend: (+) positive; (-) negative; (E) equivocal; NOS, not otherwise specified

Table 6. TTP within 1-2 years follow-up by IHC subtypes, breast cancer patients, DOH-BCMAP in PGH and JRRMMC

| Subtypes (Her-2/ER/PR); (number who went into relapse)	TTP (DAYS/MONTHS)		Cox Hazard Ratio (Exponential coefficient)	p-value
	Median,+SD (days/months)	Minimum and Maximum days (days/months)		
A: +/+/+; (n=3)	260 days + 188 days (8.2mos + 6.26 mos)	43 to 583 days (1.38 mos to 19.43 mos)	0.935	0.870
B: +/-/+	0	0	1.457	0.717
C: -/+/+; (n=9)	348 days + 222.39 days (11.6 mos+ 7.41 mos)	55 to 787 (1.83 mos to 26.2 mos)	0.722	0.502
D: +/+/-; (n=3)	497 days + 245.65 days (17 mos+ 8.18 mos)	43-612 (1.4 mos to 20.4 mos)	0.294	0.237
E: +/-/-; (n=15)	415 days + 226.79 days (14 mos+ 8 mos)	4 to 867 (0.13 mo to 29 mos)	0.379	0.211
F: -/+/-; (n=3)	493 days + 89.98 days (16 mos+ 3 mos)	351-568 (11.7 mos-19 mos)	0.794	0.575
G: -/-/-; (n=16)	325.5 days + 253.9 days (11 months + 9 months)	114- 863 days (3.8 mosto 29mos)	0.000	0.975
H: -/-/+; (n=1)	163 (6 months)	-	0.570	0.208
I: E/+/+; (n=2)	340 days + 250.14 days (11 mos + 8 mos)	104 -793 days (3.46 mos to 26.43 mos)	1.142	0.834
J: E/+/-; (n=1)	205 days (6.83 mos)	-	0.725	0.405
K: E/-/+	0	0	0.580	0.263
L: E/-/-; (n=6)	123.5 days + 277.01 days (4 mos+ 9 mos)	51-811 days (1.7 mos to 27 mos)	0.638	0.188

Legend: (+) positive; (-) negative; (E) equivocal

Over the first 1-2 years follow-up, across all IHC subtypes (Table 7, Figures 9 and 10), the DFS and OS rates were comparable. The overall DFS was at 69% and overall OS rate at 74% (Table 7).
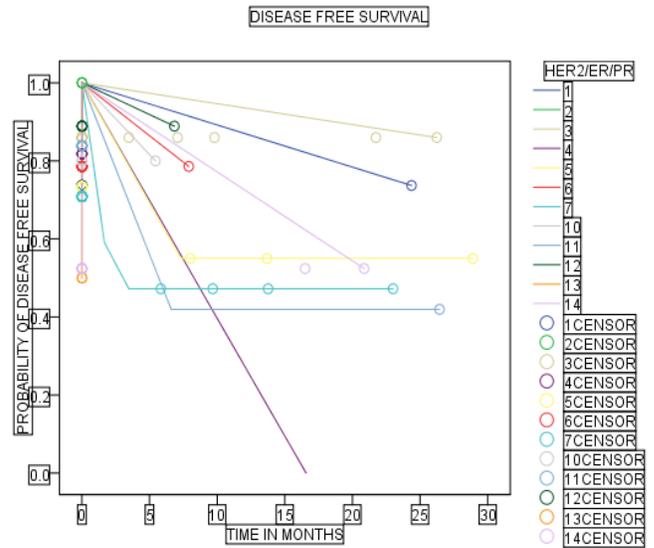
Parameter	Chi-Square	df	Sig.
Log Rank (Mantel-Cox)	14.485	11	0.207
Breslow (Generalized Wilcoxon)	15.456	11	0.163
Tarone-Ware	15.407	11	0.165

Figure 9. 1-2 years DFS Kaplan Meier Curve by IHC subtypes, breast cancer patients, DOH-BCMAP in PGH and JRRMMC
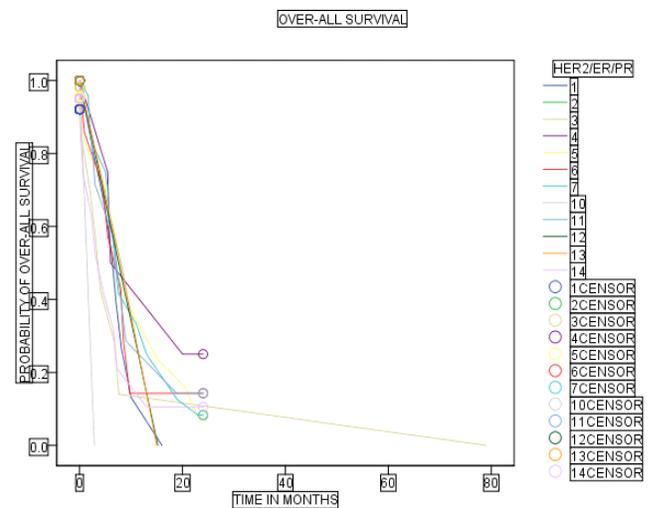
Parameter	Chi-Square	df	Sig.
Log Rank (Mantel-Cox)	7.904	11	0.722
Breslow (Generalized Wilcoxon)	12.730	11	0.311
Tarone-Ware	11.146	11	0.431

Figure 10. 1-2 years OS Kaplan Meier Curve by IHC subtypes, breast cancer patients, DOH-BCMAP in PGH and JRRMMC

Table 7. DFS and OS rates over 1st 2-years of Follow-up by IHC subtypes, breast cancer patients, DOH-BCMAP, PGH and JRRMMC

IHC Subtypes	Total patients enrolled (N)	PD	NED	Dead	LTF	Alive	DFS rate at 1-2 years (NED/N*100)	OS rate at 1-2 years (Alive/N*100)
A: +/+/+	38	3	28	5	5	28	73.68%	73.68%
B: +/-/+	2	0	2	0	0	2	100%	100%
C: -/+/+	57	9	48	1	7	49	84.21%	85.96%
D: +/+/-	11	3	8	2	1	8	72.72%	72.72%
E: +/-/-	45	15	30	8	5	32	66.67%	71.71%
F: -/+/-	28	3	19	0	6	22	67.85%	78.57%
G: -/-/-	72	16	41	8	15	49	56.94%	68.05%
H: -/-/+	5	1	4	0	1	4	80%	80%
I: E/+/+	31	2	24	1	5	25	77.42%	80.64%
J: E/+/-	9	1	7	0	1	8	77.78%	88.88%
K: E/-/+	4	0	2	2	0	2	50%	50%
L: E/-/-	21	6	9	4	6	11	42.85%	52.38%
Total	323	59	222	31	52	240	Overall DFS rate:68.73%	Overall OS rate: 74.30%

Legend: (+) positive; (-) negative; (E) equivocal; PD, progressive disease; NED, no evidence of disease;
LTF, lost to follow-up; DFS, disease free survival; OS, overall survival. Subtype not otherwise specified were

Discussion

Breast cancer is known to be a heterogeneous disease and the biology has been defined into groups of unique biological subtypes. Gene expression studies using DNA microarray and IHC staining have identified these several distinct breast cancer subtypes with each subtype presenting a particular clinical, pathological and molecular phenotype associated with diverse natural histories, therapeutic implications and prognoses. Molecular and/or IHC sub-typing has been considered as standard of care for breast cancer management, which classify breast cancer into six different subtypes according to the Her2-neu, ER, PR, and CK5/6 EGFR expression status.[3] Luminal-A [HER2neu(-)/ER(+)/PR(+)/CK5/6 EGFR(-)] is the most predominant subtype and has the better prognosis among subtypes.[4] Luminal B [(ER(+/-) PR(+/-) HER2neu(-/+) CK5/6 EGFR(-)] has a more aggressive phenotype, higher histological grade and proliferative index, and worse prognosis compared to luminal A.[3] Her2-enriched [ER(-)/PR(-)/HER2neu(+)/CK5/6 EGFR(+/-)] and triple-negative or basal-like [ER(-)/PR(-)/HER2neu(-)/CK5/6 EGFR(+)] and claudin-low [ER(-)/PR(-)/HER2neu(-)/CK5/6 EGFR (+/-)] are considered to have a poor prognosis with the latter having a higher relapse rate in the first 3 years[5], and an aggressive pattern of metastatic relapse with preponderance for visceral organs, mainly lung, central nervous system and lymph nodes.[7]

In this study, the ER(-)/PR(-)/HER2neu(-) (subtype-G) was the most frequent subtype. This is similar in some ethnic groups.[2] A study done among Asian patients treated in two New York City institutions in 2012 found that Chinese and Japanese had a higher proportion of good-prognosis luminal-A cancers compared to Filipinos and Koreans (p = 0.001); Koreans had a higher proportion of triple negative cancers compared to other ethnic groups (p = 0.06);[20] Filipinos were noted to have a higher proportion of

HER2neu positive cancers (45.6%) compared to other ethnic groups (p = 0.002).[20] In this study, if we were to take all HER2neu positive patients, this proportion would be 31% (with exclusion of equivocal results).

In one study,[22] the basal cell-like subtype (triple negative) was noted to be the most prevalent in the age group of <35 years old and showed an age-specific bimodal distribution with a peak at: (a) <35 years old; and (b) 51 to 65 years of age; the molecular subtypes did not differ in terms of menopausal status. In this study, HER2neu(-)/ER(-)/PR(-) disease was mostly noted between the age group of 35-50 years old and mostly postmenopausal. HER2neu(-)/ER(+)/PR(+) and HER2neu(+) subtypes were more prevalent in >35 years old, peri-menopausal and >50 years old, postmenopausal, respectively.

Breast cancer among <35 years of age was also noted in this study at 7%. According to a study[23] among young girls in Turkey, young patients had high risk features such as the luminal B subtype, moderate to high grade tumors, stage II disease, and with positive axillary and lymphovascular invasion. Another study[24] done in Korea showed unfavorable features of <35 years old patients, frequently associated with triple-negative subtypes and higher histologic and nuclear grade.

The stage of breast cancer at diagnosis also correlates with prognosis and IHC subtypes. In this study, HER2neu(-)/ER(-)/PR(-) disease was diagnosed mostly in stage IIIA disease; HER2neu(-)/ER(+)/PR(+) and HER2neu(+)/ER(-)/PR(-)tumors presented predominantly as stage IIA disease.

Since HER2neu(-)/ER(-)/PR(-) and Her2neu (+) subtypes are both considered as highly aggressive, highly proliferative, and with poor prognosis, these subtypes are expected to have the most number of early relapses, mostly at distant sites, as so commonly discussed in literature. The Her2-enriched subtype has changed in clinical outcome

nowadays with the advent and administration of trastuzumab as targeted therapy against the Her2neu receptor,[3,4,7] similar to the change in clinical outcome with the discovery of tamoxifen as a targeted therapy for ER(+) tumors. The triple negative subtype remains to have early recurrences higher in the first 3 years after diagnosis; a targeted agent against this subtype is yet to be discovered.[25] Luminal-A subtypes mostly have a better prognosis with a relapse rate of 27.8% and survival from the time of relapse (median 2.2 years) significantly better than other subtypes with a distinct pattern of recurrence more of bone metastases (18.7%).[4]

The DFS and OS of the patients enrolled in DOH-BCMAP during the first 1-2 years of follow-up were comparable among the subtypes; the overall DFS and OS rates were 68.73% and 74.30%, respectively. The HER2neu(-)/ER(-)/PR(-) subtype had the shortest median TTP (11 months $\pm$ 9 sd) among those with high number of relapse together with Her2(+)/ER(-)/PR(-) and HER2neu(-)/ER(+)/PR(+) subtypes, with median TTP of 14 months $\pm$ 8 sd, and 11.6 months $\pm$ 7.41 sd, respectively.

If subtypes were to be combined, disease progression/relapse during the first 1-2 years of follow-up would be seen mostly among the HER2neu(+) subtypes regardless of ER/PR status at 32%, compared to HER2neu(-)/ER(-)/PR(+/-) at 25%. Both groups would have predominantly distant metastasis during relapse. This was 18% among the HER2neu(-)/ER(+)/PR(+/-) group, which had predominantly local recurrence.

The TTP, DFS, and OS may change as the patients are followed-up over 5 years, particular without trastuzumab treatment for HER2neu(+) patients. The 65 (18%) patients with equivocal HER2neu status, particularly those with ER(-) disease (25 patients), could contribute to unfavorable TTP and poor prognosis among these patients if HER2neu was positive.

The determination of gene amplification among the equivocal HER2neu patients plus the CK5/6 EGFR status (molecular profile) of all patients would perhaps add more information to the prognostication of this disease, but the specific HER2neu/ER/PR IHC profiles can define breast cancer prognosis and predict treatment outcomes given HER2neu(+), HER2neu(-)/ER(-), and HER2neu(-)/ER(+) disease.

Conclusion

Filipino patients enrolled in the DOH-BCMAP with non-metastatic breast cancer after surgery and mainly on adjuvant chemotherapy started to develop disease progression/ relapse within the first 2 years of follow-up; 82% had no relapse. At these early years of follow-up, the median TTPs, DFS rate, OS rate among the different breast cancer IHC subtypes were comparable, although HER2neu(+) regardless of ER/PR subtype trended to have more disease progression, followed by HER2neu(-)/ ER(-)/ regardless of PR subtype, and then HER2neu(-)/ ER(+)/ regardless of PR subtype. IHC resultant HER2neu(+) regardless of ER/PR and HER2neu(-)/ER(-)/PR(-/+) subtypes can serve as early prognosticators of breast cancer relapse.

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