Research Article

Breast Cancer Subtypes And Prognosis: Answers To Subgroup Classification Questions, Identifying The Worst Subgroup In Our Single-Center Series

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Purpose: Because of advances in treatment, long life is now possible even in patients with metastatic BC. In many studies, the triple-negative breast cancer (TNBC) subgroup is stated to have the worst prognosis, as such patients are deprived of antihormonal and Herceptin therapy. While HER2 overexpression was interpreted as a poor prognostic factor before Herceptin, it was reported to be the worst prognostic subgroup of TNBC. In this study, we aimed to find the worst prognostic subgroup.

Methods: We reviewed the records of patients with BC who were admitted to our department between July 1999 and December 2019. We grouped the patients into four main groups (Luminal A, Luminal B, triple-negative, and HER2-enriched) and we recorded patient and treatment characteristics and oncological results. Survival curves were generated using the Kaplan–Meier method, and the significance of survival differences among the selected variables was compared by using the log-rank test.

Results: A total of 2474 patients with BC and after exclusions, statistical analysis was performed on 2017 patients with BC. The HER2 positivity rate was 23.7% and the TNBC patient rate was 11.7% (n = 236). The distribution of the four main groups was 47.1% for Luminal A, 34.1% for Luminal B, 7.1% for HER2-enriched, and 11.7% for the TN subgroup. Age (<35 years), no axillary surgery, Ki67≥15, high tumor grade, high mitotic index, the presence of skin infiltration, advanced T/N stage, the presence of metastasis, nontreatment with chemotherapy, less than 5 years of using TMX or AI, and being in the HER2-enriched subgroup were determined to be negative factors for overall survival as a result of multivariate analysis.

 doi.org/10.32388/UB8V1E

Conclusions: The HER2-enriched subgroup had the worst prognosis despite receiving targeted therapy. However, treatment with trastuzumab increased survival 1.5-fold over that of the HER2-enriched subgroup that did not receive it.

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Introduction

Breast cancer (BC) remains the most common cancer in women, and it is the second leading cause of cancer-related death in women after lung cancer[1]. However, because of advances in treatment, long life is now possible even in patients with metastatic BC, whereas certain groups of patients survive for a very short time despite being diagnosed at an early stage[2]. Every day, we are getting closer to understanding this differential clinical course of BC, and we have the opportunity to define heterogeneity in BC, thanks to the detection of molecular receptors that play a role in breast carcinogenesis and the detection of pathways indicative of rapid proliferation[3][4][5][6].

Immunohistochemical (IHC) staining and in situ fluorescent hybridization (FISH) methods are currently used methods for identifying tumor subtypes to achieve better treatment choices and survival. Since the St. Gallen International consensus panel in 2011, four main robust subtypes (Luminal A, Luminal B, triple-negative, and HER2-enriched) have proven to be a good classification scheme. According to the presence or absence of receptors, in the classification of BC, four different molecular subtypes have been defined:

- Luminal A (ER and PR positive, HER2-negative, Ki67 low),
- Luminal B (ER and/or PR positive, HER2-positive or Ki67 high),
- HER2-enriched (Hormone (ER and PR) receptor-negative and HER2-positive)
- Triple-negative (TN) (ER- and PR- and HER2-negative).

Each subtype exhibits different oncological results and different treatment strategies[4].

Retrospective data help to identify prognostic factors as well as to measure the effectiveness of treatments and test their effects on subgroups. It is also possible to determine the best subgroup with a good prognosis and to predict the clinical course[7][8]. However, determining the subgroup with the worst prognosis and predicting the clinical course is still unclear and confusing when the literature is evaluated.

In many studies, the TNBC subgroup is stated to have the worst prognosis, as such patients are deprived of antihormonal therapy and trastuzumab therapy. Additionally, the main systemic treatment is chemotherapy alone in most BC patients with TN who have a poor prognosis[9][10][11][12][13][14]. The main published result in the years before targeted therapies were placed into routine clinical use was that the HER2-enriched subgroup was the worst prognostic subgroup. However, while HER2 overexpression was interpreted as a poor prognostic factor before trastuzumab treatment, it was reported to be the worst prognostic subgroup of TNBC in posttrastuzumab publications[15][16][17][18][19]. It is known according to several clinical outcomes that Luminal A is the best prognostic subgroup[7][8][9][10].

In the current study, we aimed to find the worst prognostic subgroup as far as we could by capturing the biodiversity in our series of BC patients, and for this purpose, we compared the treatment results in patients grouped according to their receptor status.

Material and Method

Following the approval of the Institutional Review Board, we reviewed the records of patients with BC who were admitted to the Radiation and Medical Oncology Department of Trakya University between July 1999 and December 2019. The Human Research Ethical Committee of the Trakya University Medical Faculty Hospital approved (TUTF-BAEK 2021/406) the use of these patients' information for the study.

We grouped the patients into four main groups (Luminal A, Luminal B, triple-negative, and HER2-enriched) according to the St Gallen International Consensus Panel and five subgroups according to the receptor status (Table 1). We recorded patient characteristics, such as age, body mass index (BMI), age at menarche, age at menopause, menstruation status, number of births, family history, breastfeeding, hormone replacement status, histological type, localization area in the breast, tumor quadrant, surgical type, axillary surgery type, tumor size, lymph node metastasis, TNM stage, grade, mitotic index, estrogen receptor (ER) status, progesterone receptor (PR) status, Human Epidermal Growth Factor Receptor 2 (HER2) status, Ki-67 level, lymphovascular invasion (LVSI), perineural invasion (PNI), extensive intraductal component (EIC), surgical margin status, skin involvement, whether or not they received chemotherapy, chemotherapy type, whether or not they received radiotherapy, radiotherapy type, tamoxifen (TMX) usage time, aromatase inhibitor (AI) usage time, and luteinizing hormone-releasing hormone (LHRH) usage time. The staging of the tumor was based on The American Joint Committee on Cancer 2013 System.

Histopathologic Evaluation

In our pathology department, receptor status assessments are made as follows:

- Primary Novocastra monoclonal antibodies (clone 6F11 for ER and clone 1 A6 for PR) were used to determine the estrogen receptor (ER) and progesterone receptor (PR) status. A positive nuclear reaction was considered "receptor-negative" in less than 1% of tumor cells.
- Immunohistochemical analyses were performed using HER2/neu (Clone 10A7, Novocastra) as the primary antibody. The DAKO Herceptest scoring system, which is also referred to in some national and international guidelines, was used. Tumors showing 3+ membranous staining immunohistochemically (IHC) for HER2/neu antibody or positive gene amplification by fluorescent in situ hybridization (FISH) were considered positive.
- Ki67 was analyzed in paraffin sections by an immunohistochemical method using the MIB-1 antibody. Our pathologist examined the stained section using a standard light microscope with a 40x objective and 10x10 graticule and defined the Ki67 score as the percentage of the total number of tumor cells with nuclear staining. This required counting at least 1000 tumor cells with nuclear staining in ten high-power fields (× 40).

Statistical Analysis

Numerical results are expressed as the mean ± standard deviation, and categorical results are shown as n (%). Survival curves were generated using the Kaplan–Meier method, and the significance of survival differences among the selected variables was compared by using the log-rank test. Univariate Cox regression analysis was used to estimate hazard ratios. Then, multivariate Cox regression analysis with the backward elimination method was used to estimate hazard ratios and to identify independent prognostic factors. All reported p values are two-sided, and a value below 0.05 was considered to indicate statistical significance. Data analysis was performed using SPSS version 20.0 (IBM SPSS Statistics for Windows, Version 20.0. Armonk, NY: IBM Corp.).

Results

A total of 2474 patients with BC who were treated between July 1999 and December 2019 were evaluated. Patients who did not have the examined parameters were excluded from the study. A total of 131 patients with ductal carcinoma-in-situ and lobular carcinoma in situ, 9 patients with phyllodes tumors, and 244 patients whose ER, PR, HER2, and Ki67 information could not be completely obtained were excluded from the analysis. After exclusions, statistical analysis was performed on 2017 patients with BC

(Figure 1). The mean age was 52.07 years, the mean menopausal age was 48.35 years, and the mean menarche age was 13.15 years. The mean BMI was 29.9. The HER2 positivity rate was 23.7%.

The TNBC patient rate was 11.7% (n = 236), and there were no statistically significant differences between the two groups when comparing DFS (190.37±7.19 (176.27-204.46) for TNBC, 218.23±3.68 (211.01-225.44) for NTNBC, p=.739) and OS (221.68±7.92 (206.14-237.21) TNBC and 231.77±3.29 (225.32-238.22) p=.252) (Table 2, Figure 2a, b).

The distribution of the four main groups was 47.1% for Luminal A, 34.1% for Luminal B, 7.1% for HER2-enriched, and 11.7% for the TN subgroup. The worst prognostic main group comprised HER2-enriched patients, with 113.70±7.17 months DFS and 125.45±3.03 months OS (Table 3, Figure 3a, b).

The DFS was 101.50±6.4 (88.77-114.23), the OS was 118.14±6.16 (106.06-130.22) in the Herceptin group, and the DFS was 92.79±18 (57.44-128.13); the OS was 94.44±15.23 (64.58-124.30) in the non-Herceptin group (Table 4). The HER2-enriched Herceptin subgroup did not have the lowest DFS and differed from the TNBC, Luminal A, and Luminal B subgroups at the level of statistical significance. However, DFS did not differ statistically significantly between HER2-enriched patients who received Herceptin and HER2-enriched patients who did not receive Herceptin. OS was the lowest survival time at the statistical significance level among the TNBC-, Luminal A-, Luminal B-, and HER2-enriched Herceptin subgroups (Table 4, Figure 4a, b).

The DFS was 163.796±5.78 (152.45-175.13) months, and the OS was 178.95±5.15 (168.83-189.06) months in Luminal B HER2-positive patients. The DFS was 101.23±2.35 (96.61-105.86) months, and the OS was 114.16±2.01 (110.20-118.11) months in Luminal B HER2-negative patients. The Luminal B HER2-positive subgroup had a longer DFS and OS than the Luminal B HER2-negative subgroup. However, this difference was not statistically significant (Table 5, Figure 5a, 5b). There was a statistically significant difference in the DFS and OS times with HER2-enrichment in the Herceptin, triple-negative, Luminal A, Luminal B-HER2-positive, and Luminal B-HER2-negative groups (Table 5, Figure 5a, b).

HER2 positivity was separated according to the status of negativity and treatment or nontreatment with Herceptin or not. While the HER2-negative subgroup showed the best survival, it did not differ statistically from the survival of the TNBC, Luminal A, and Luminal B subgroups. However, the HER2-negative subgroup had a significantly better time than the HER2-enriched subgroup in terms of both DFS and OS times. The HER2-negative subgroup had a significantly better outcome than the HER2-enriched subgroups in terms of both DFS and OS times. The best DFS and OS were detected in the HER2-negative subgroup. Additionally, while the HER2-negative subgroup showed the best survival, the survival of the

Luminal B Herceptin subgroup was not statistically different from that of the Luminal B non-Herceptin subgroup (Table 6, Figure 6a, b).

The worst subgroup for DFS and OS was the HER2-enriched subgroup, whether receiving Herceptin or not (Table 7). In all pairwise comparisons, only the DFS duration in the HER2-enriched subgroup was not statistically significant. In comparison with all other subgroups, both DFS and OS times were significantly different. In pairwise comparisons of the HER2-enriched subgroup, the strongest difference was found in the order of Luminal A, Luminal B, and TNBC.

In univariate analysis, age (<35 years), early age at menarche, postmenopausal status, advanced T/N stage, no surgery on the breast and/or axillary node, high tumor grade, high mitotic index, the presence of skin infiltration, multifocal tumor, ER, PR negativity and HER2 positivity, positive EIC, positive LVI, Ki67≥15, the presence of metastasis, nontreatment with chemotherapy and radiotherapy, less than 5 years of using TMX or AI, less than 2 years' use of LHRH, and being in the HER2-enriched subgroup were determined to be negative factors for OS. No axillary surgery, T and N stage, not receiving radiotherapy, using TMX for less than 5 years, and LHRH for less than 2 years were statistically significant negative factors for OS in multivariate analysis (Table 8).

In univariate analysis, age (<35 years), postmenopausal status, advanced T/N stage, no surgery on the breast and/or axillary node, high tumor grade, high mitotic index, presence of skin infiltration, multifocal tumor, ER, PR negativity and HER2 positivity, presence of metastasis, positive EIC, positive LVI, Ki67≥15, positive surgical margin, nontreatment with chemotherapy and radiotherapy, less than 5 years of using TMX or AI, less than 2 years' use of LHRH, and being in the HER2-enriched subgroup were determined to be negative factors for OS. Age (<35 years), no axillary surgery, Ki67≥15, high tumor grade, high mitotic index, presence of skin infiltration, advanced T/N stage, presence of metastasis, nontreatment with chemotherapy, less than 5 years of using TMX or AI, and being in the HER2-enriched subgroup were determined to be negative factors for OS as a result of multivariate analysis (Table 9).

Discussion

Retrospective data measure the efficacy of treatments while also helping to test their impact on prognostic factors and subgroups. Similar to old age, its strength comes from experience, from knowing what might happen in the future. The need to group our series in this way and to find the subgroup with the worst prognosis indicated the inconsistency of our patient-specific experiences and the literature information. Patient follow-up in our series was carried out meticulously and regularly by the same physicians. Since the patient-file information was reliable and complete in our study, it is remarkable in

terms of its results, although it comprised retrospective data. Although HER2-targeting antagonists have revolutionized the treatment of HER2-overexpressing BC and have produced a better clinical outcome for the HER2-enriched subgroup, it was still identified as the subgroup with the lowest DFS and OS in our series.

Herceptin reduced the risk of the event 1.5 times in the HER2-enriched subgroup, which we determined to be the subgroup with the worst prognosis (p=.223, HR 1.515 (95% CI 777-2.954)). The HER2-enriched subgroup had a 10-fold increased risk of overall survival compared to the Luminal A subgroup.

Foulkes WD et al., in the TNBC article by[9], stated that the subgroup with the lowest survival was TNBC, even though the lowest survival subgroup was seen as HER2-enriched in the survival curve. In the article, it was stated that the HER2-enriched subgroup had the lowest survival rate, and the curve was the subgroup with the lowest survival since these patients did not use targeted therapies. However, the subgroup that showed the lowest survival despite receiving targeted therapy in our series was the HER2-enriched subgroup. In addition, while there was no statistically significant difference between Luminal A and Luminal B in both DFS and OS times of patients with TNBC in our series, we found that the survival times were significantly better than those of the HER2-enriched subgroup.

Overexpression of HER2 accounts for 20-30% of all BCs. The rate in our series was 23.7%. Activation of the HER2 receptor via tyrosine phosphorylation[20] results in increased proliferation, which is associated with increased relapse rates and increased mortality. Although HER2 expression is a critical event in the etiology of HER2-positive BC, the molecular mechanisms that regulate disease progression and how and why drug resistance develops in a short time are still not fully understood[15][16][17].

In 1987, Slamon et al.[18] reported that patients with BC in whom HER2 amplification was detected had a significantly shorter relapse and overall survival times[15][18]. Moreover, amplification was also associated with negative ER or PR status[15].

We know that the estrogen receptor activates the HER2 receptor signaling pathway[17][20][21][22][23]. This may make trastuzumab treatment more effective, as it brings with it the use of antiestrogen (TMX, AI)[22][23]. In the HER2-enriched subgroup, in which estrogen and progesterone receptors are negative and only HER2 is overexpressed, the efficacy of treatment was limited to only trastuzumab, which may cause the HER2-enriched group to have a worse prognosis. While treatments for HER2 have revolutionized the treatment of HER2-overexpressing BC, the HER2-enriched subgroup still had the lowest survival rate in our series.

In addition, when the parameters used to create the subgroup were taken into the Cox regression analysis one by one, we found that the Ki67≥15 level negatively affected overall survival; in the multivariate analysis, the HR was 2.627 (1.478-4.670) p=.001, which is consistent with the literature[24][25]. Another remarkable point in our series is that the use of TMX for more than 5 years reduced both relapse and mortality and the risk of death in AI[26][27][28].

Currently, as personalized treatments based on the principle that the patient, not the disease, should be treated are discussed and recommended, we can predict that subtyping classifications in BC will assume a much higher place in our future treatment plans and will continue to be a guide for clinicians in the long term. Our results show that subtyping captures most of the biodiversity occurring in BC.

Conclusion

Retrospective data measure the efficacy of treatments while also helping to test their impact on prognostic factors and subgroups. In our series, the HER2-enriched subgroup had the worst prognosis despite receiving targeted therapy. The belief that targeted therapies solve all problems may prevent clinicians from identifying patients with the worst prognosis. However, treatment with trastuzumab increased survival 1.5-fold over that of the HER2-enriched subgroup that did not receive it. Therefore, the HER2-enriched subgroup is a subgroup that needs to be followed carefully, and new treatment options are needed.

Tables

Groups Name	How is the classification made?	Group Branches
Subtyping 1	Subtype Triple-Negative	Triple-Negative None-Triple Negative
Subtyping 2	Original Subtype	Triple-Negative Luminal A Luminal B HER2-enriched
Subtyping 3	Subtype HER2-enriched (received Herceptin)	Triple-Negative Luminal A Luminal B HER2-enriched (received Herceptin) HER2-enriched (did not receive Herceptin)
Subtyping 4	Subtype HER2 positive-negative	Triple-Negative Luminal A Luminal B HER2 positive Luminal B HER2 negative HER2-enriched (received Herceptin) HER2-enriched (did not receive Herceptin)
Subtyping 5	Subtype received Herceptin	Luminal B (received Herceptin) Luminal B (did not receive Herceptin) HER2-enriched (received Herceptin) HER2-enriched (did not receive Herceptin) HER2 negative

Table 1. The step-by-step parameters according to which the groups are created

doi.org/10.32388/UB8V1E

		Subtyping 1		p-value (Log-rank test)
		Triple-Negative (TNBC)	Non-Triple-Negative (NTNBC)	
Disease-free survival	**Mean ± SD**	190.3 ± 7.1	218.2 ± 3.6	0.739
	95% Confidence Interval	176.2-204.4	211.0-225.4	
Overall survival	**Mean ± SD**	221.6 ± 7.9	231.7 ± 3.2	0.252
	95% Confidence Interval	206.1-237.2	225.3-238.2	

Table 2. Disease-free survival, and overall survival times, comparative Log-rank test, p-values obtained using the Kaplan-Meier method of Triple-Negative Breast Cancer and Non-Triple-Negative Breast Cancer subgroups forming Subtyping 1

SD: Standard deviation, CI: Confidence Interval

	Subtyping 2	Mean ± Std. Error (Months)	95% Confidence Interval		p values (Log-rank test)			
			Lower Bound	Upper Bound	Triple-Negative	Luminal A	Luminal B	HER2-enriched
Disease-free survival	Triple-Negative	190.3 ± 7.1	176.2	204.4				
	Luminal A	226.7 ± 4.3	218.3	235.2	0.139			
	Luminal B	168.3 ± 4.3	159.8	176.9	0.971	0.016		
	HER2-enriched	113.7 ± 7.1	99.6	127.7	**<0.001**	**<0.001**	**<0.001**	
Overall survival	Triple-Negative	221.6 ± 7.9	206.1	237.2				
	Luminal A	237.4 ± 3.8	229.9	244.9	**0.002**			
	Luminal B	180.2 ± 4.0	172.3	188.2	0.160	0.450		
	HER2-enriched	125.4 ± 3.0	112.0	138.9	**<0.001**	**<0.001**	**<0.001**	

Table 3. Disease-free survival, and overall survival times, comparative Log-rank test, p-values obtained using Kaplan-Meier method of Triple-Negative Breast Cancer, Luminal A, and Luminal B and HER2-enriched subgroups forming Subtyping 2

doi.org/10.32388/UB8V1E

	Subtyping 3	Mean ± Std. Error (Months)	95% Confidence Interval		Log-rank (Mantel-Cox) Chi-Square (Sig)				
			Lower Bound	Upper Bound	Triple Negative	Luminal A	Luminal B	HER2-enriched (HER-A) received Herceptin	HER2-enriched (HER-B) did not receive Herceptin
Disease-free survival	Triple-Negative	190.37±7.19	176.27	204.46					
	Luminal A	227.02±4.31	218.57	235.46	2,085 (.149)				
	Luminal B	168.12±4.36	159.57	176.67	0.000 (.977)	5.224 (**.022**)			
	HER2-enriched (HER-A) received Herceptin	101.50±6.49	88.77	114.23	9.262 (**.002**)	28.443 (**<.001**)	13.935 (**<.001**)		
	HER2-enriched (HER-B) did not receive Herceptin	92.79±18.00	57.44	128.13	10.318 (**.001**)	17.954 (**<.001**)	10.409 (**.001**)	1.665 (.197)	
Overall survival	Triple-Negative	221.68±7.92	206.14	237.21					
	Luminal A	237.44±3.83	229.92	244.97	3.100 (.078)				
	Luminal B	180.29±4.04	172.37	188.22	2.122	.387			

					(.145)	(.534)			
	HER2-enriched (HER-A) received Herceptin	118.14±6.16	106.06	130.22	4.548 (**.033**)	21.267 (**<.001**)	16.439 (**<.001**)		
	HER2-enriched (HER-B) did not receive Herceptin	94.44±15.23	64.58	124.30	16.092 (**<.001**)	32.866 (**<.001**)	30.357 (**<.001**)	5.602 (**.018**)	

Table 4. Disease-free survival and overall survival times, comparative Log-rank test, p-values obtained using Kaplan-Meier method of Triple-Negative Breast Cancer, Luminal A and Luminal B and HER2-enriched received Herceptin, HER2-enriched did not receive Herceptin subgroups forming Subtyping 3

	Subtyping 4	Mean ± Std. Error (Months)	95% Confidence Interval		Log-rank (Mantel-Cox) Chi-Square (Sig)					
			Lower Bound	Upper Bound	Triple Negative	Luminal A	Luminal B HER2 Positive	Luminal B HER2 Negative	HER2-enriched (HER-A) received Herceptin	HER2-enriched (HER-B) did not receive Herceptin
Disease-free survival	Triple-Negative	190.37±7.19	176.27	204.46						
	Luminal A	227.02±4.31	218.57	235.46	2.085 (.149)					
	Luminal B HER2 Positive	163.79±5.78	152.45	175.13	0.266 (.606)	6.980 (**,008**)				
	Luminal B HER2 Negative	101.23±2.35	96.61	105.86	0.431 (.512)	0.941 (.332)	1.387 (.239)			
	HER2-enriched (HER-A) received Herceptin	101.50±6.49	88.77	114.23	9.262 (**.002**)	28,443 (**<.001**)	8.157 (**.004**)	15.406 (**<.001**)		
	HER2-enriched (HER-B) did not	92.79±18.00	57.44	128.13	10,318 (**.001**)	17,954 (**<.001**)	7.883 (**.005**)	13.455 (**<.001**)	1.665 (.197)	

	receive Herceptin									
Overall survival	**Triple-Negative**	221.68±7.92	206.14	237.21						
	Luminal A	237.44±3.83	229.92	244.97	3.10 (.078)					
	Luminal B HER2 Positive	178.95±5.15	168.83	189.06	1.061 (.303)	0.582 (.446)				
	Luminal B HER2 Negative	114.16±2.01	110.20	118.11	2.699 (.100)	0.018 (.892)	0.259 (.611)			
	HER2-enriched (HER-A) received Herceptin	118.14±6.16	106.06	130.22	4.548 (**.033**)	21.267 (**<.001**)	11.391 (**.001**)	13.703 (**<.001**)		
	HER2-enriched (HER-B) did not receive Herceptin	94.44±15.23	64.58	124.30	16.092 (**<.001**)	32.866 (**<.001**)	25.708 (**<.001**)	30.967 (**<.001**)	5.602 (**.018**)	

Table 5. Disease-free survival and overall survival times, comparative Log-rank test, p-values obtained using Kaplan-Meier method of Triple-Negative Breast Cancer, Luminal A and Luminal B HER2 positive, Luminal B HER2 negative and HER2-enriched received Herceptin, HER2-enriched did not receive Herceptin subgroups forming Subtyping 4.

doi.org/10.32388/UB8V1E

	Subtyping 5	Mean ± Std. Error (Months)	95% Confidence Interval		Log-rank (Mantel-Cox) Chi-Square (Sig)				
			Lower Bound	Upper Bound	HER2 Negative	Luminal B Herceptin Positive	Luminal B Herceptin Negative	HER2-enriched (HER-A) received Herceptin	HER2-enriched (HER-B) did not receive Herceptin
Disease-free survival	HER2 Negative	225.237±3.89	217.60	232.86					
	Luminal B received Herceptin	126.33±5.10	116.32	136.34	3.006 (.083)				
	Luminal B did not receive Herceptin	171.69±4.90	162.09	181.30	1.945 (.163)	0.162 (.688)			
	HER2-enriched (HER-A) received Herceptin	101.50±6.49	88.77	114.23	26.736 (**<.001**)	8.466 (**.004**)	12.783 (**<.001**)		
	HER2-enriched (HER-B) did not receive Herceptin	92.79±18,03	57.44	128.13	16.902 (**<.001**)	7.133 (**.008**)	11.736 (**.001**)	1.665 (.197)	
Overall survival	HER2 Negative	235.49±3.48	228.66	242.33					

doi.org/10.32388/UB8V1E

	Luminal B received Herceptin	148.32±4.17	140.14	156.50	0.607 (.436)				
	Luminal B did not receive Herceptin	178.20±4.91	168.56	187.84	0.386 (.534)	0.999 (.317)			
	HER2-enriched (HER-A) received Herceptin	118.14±6.16	106.06	130.22	18.218 (**<.001**)	14.578 (**<.001**)	11.466 (**.001**)		
	HER2-enriched (HER-B) did not receive Herceptin	94.44±15.23	64.58	124.30	30.150 (**<.001**)	30.660 (**<.001**)	25.569 (**<.001**)	5.602 (**.018**)	

Table 6. Disease-free survival, overall survival times, comparative Log-rank test, p-values obtained using Kaplan-Meier method of TNBC, Luminal A, Luminal B received Herceptin, Luminal B did not receive Herceptin and HER2-enriched received Herceptin, HER2-enriched did not receive Herceptin subgroups forming Subtyping 5.

			Disease-free survival		Overall survival	
			Pearson Chi-Square	Asymptotic Significance (2-sided) p-value	Pearson Chi-Square	Asymptotic Significance (2-sided) p-value
Subtyping 1			0.207	0.649	1.375	0.252
Subtyping 2		**HER2-enriched vs Luminal A** **HER2-enriched vs Luminal B** **HER2-enriched vs TNBC**	39.820 19.845 12.876	**<0.001** **<0.001** **<0.001**	39.518 29.819 9.715	**<0.001** **<0.001** **0.002**
Subtyping 3	**Received Herceptin**	**HER2-enriched vs Luminal A** **HER2-enriched vs Luminal B** **HER2-enriched vs TNBC**	26.563 12.484 8.652	**<0.001** **<0.001** **0.003**	17.540 12.787 3.437	**<0.001** **<0.001** **0.064**
	Did not receive Herceptin	**HER2-enriched vs Luminal A** **HER2-enriched vs Luminal B** **HER2-enriched vs TNBC**	17.954 10.271 10.391	**<0.001** **0.001** **0.001**	32.866 29.516 16.155	**<0.001** **<0.001** **<0.001**
Subtyping 4	**Received Herceptin**	**HER2-enriched vs Luminal A** **HER2-enriched vs Luminal B HER2 positive** **HER2-enriched vs Luminal B HER2 negative** **HER2-enriched vs TNBC**	24.648 9.208 9.410 7.911 1.805	**<0.001** **0.002** **0.002** **0.005** 0.179	17.784 10.882 9.072 3.544 6.231	**<0.001** **0.001** **0.003** 0.060 **0.003**

doi.org/10.32388/UB8V1E

			Disease-free survival		Overall survival	
		HER2-enriched vs HER2-enriched did not receive Herceptin				
	Did not receive Herceptin	**HER2-enriched vs Luminal A** **HER2-enriched vs Luminal B HER2 positive** **HER2-enriched vs Luminal B HER2 negative** **HER2-enriched vs TNBC**	17.954 9.068 13.214 10.391	**<0.001** **0.003** **<0.001** **0.001**	32.866 26.983 27.485 16.155	**<0.001** **<0.001** **<0.001** **<0.001**
Subtyping 5	**Received Herceptin**	**HER2-enriched vs HER2 negative** **HER2-enriched vs Luminal B received Herceptin** **HER2-enriched vs Luminal B did not receive Herceptin**	22.721 6.715 10.613	**<0.001** **0.010** **0.001**	14.808 12.398 9.278	**<0.001** **<0.001** **0.002**
	Did not receive Herceptin	**HER2-enriched vs HER2 negative** **HER2-enriched vs Luminal B received Herceptin** **HER2-enriched vs Luminal B did not receive Herceptin**	16.761 6.904 11.633	**<0.001** **0.009** **0.001**	29.920 30.298 25.437	**<0.001** **<0.001** **<0.001**
		HER2-enriched received Herceptin vs HER2-enriched did not receive Herceptin	1.805	0.179	6.333	**0.012**

Table 7. Comparison of HER2-enriched subgroup with other subgroups

doi.org/10.32388/UB8V1E

		Univariate analysis		Multivariate analysis	
Patients Descriptions	**Events/Total (%)**	**Hazard ratio (95% CI)**	**p**	**Hazard ratio (95% CI)**	**p**
Age group < 35 years 35–50 years > 50 years	23/84 (27.3) 138/756 (18.2) 254/1177 (21.5)	1 (Reference) 0.627 (0.403 - 0.974) 0.826 (0.539 - 1.266)	**.038** .381	1 (Reference) 1.218 (.747-1.986) 1.466 (.836-2.570)	.429 .182
BMI < 25 ≥ 25	73/373 (19.5) 342/1644 (20.8)	1.059 (0.823 - 1.364)	.656		
Menopause Age (mean) Events 48.36 years None Events 48.30 years	260/2017 (12.8)	1.010 (0.984 - 1.037)	.470		.
Menstruation Age (mean) Events 13.04 years None Events 13.17 years	415/2017(20.5)	0.915 (0.850 - 0.986)	**.019**	.959 (.851-1.080)	.486
Menstruation situation Premenopause Postmenopause	149/787 (18.9) 260/1217 (21.3)	1.221 (.998-1.495)	.052	1.053 (.746-1.487)	.769
Number of births No birth 1-2 birth 3 and more	38/162 (23.4) 255/1320 (19.3) 115/520 (22.1)	1 (Reference) .834 (.593-1.173) .928 (.643-1.339)	.435 .296 .690		
Family History Positive Negative	115/632 (18.1) 300/1385 (21.6)	.850 (.685-1.054)	.138		
Breast-feeding	229/1192 (19.2)	.869 (.716-1.055)	.156		

doi.org/10.32388/UB8V1E

		Univariate analysis		Multivariate analysis	
Patients Descriptions	**Events/Total (%)**	**Hazard ratio (95% CI)**	**p**	**Hazard ratio (95% CI)**	**p**
Positive Negative	186/825 (22.5)				
Breast site Left Right Bilateral	205/1013 (20.2) 186/935 (19.8) 24/69 (34.7)	1 (Reference) .000 (.000-6.51) <.001 (.000-2.31)	.939 .935		
Location Unilateral Metacron Sencron	391/1948 (20) 17/46 (36.9) 7/23 (30.4)	1 (Reference) 1.590 (.978-2.586) 1.770 (.838-3.739)	.061 .135		
Tumor Quadrant Inner Outer Periareolar Multifokal	80/402 (19.9) 234/1205 (19.4) 54/259 (20.8) 47/150 (31.3)	1 (Reference) .994 (.771-1.281) 1.081 (.766-1.527) 1.819 (1.269-2.608)	.962 .657 **.001**	1 (Reference) .924 (.697-1.225) .713 (.479-1.062) .721 (.466-1.117)	.583 .096 .143
Histopathologic Type Invasive ductal carcinoma Invasive lobular carcinoma Other	344/1652 (20.8) 26/122 (21.3) 45/243(18.5)	1 (Reference) .954 (.640-1.422) .864 (.633-1.179)	.646 .817 .356		
Surgical Type BCS MRM No surgery	122/1016 (12) 226/930 (24.3) 67/71(94.3)	1 (Reference) 1.978 (1.586-2.466) 27.941(20.296-38.465)	**<.001** **<.001**	1 (Reference) .834 (.634-1.090) 1.444 (.676-3.087)	.184 .343
Axillary Surgery Type SLND	37/451(8.2) 304/1477(20.5)	1 (Reference) 2.210 (1.569-3.111)	**<.001** **<.001**	1 (Reference) .645(.427-.975)	**.037** .900

doi.org/10.32388/UB8V1E

		Univariate analysis		Multivariate analysis	
Patients Descriptions	**Events/Total (%)**	**Hazard ratio (95% CI)**	**p**	**Hazard ratio (95% CI)**	**p**
AD No axillary surgery	74/89(83.1)	21.759 (14.573-32.487)		1.056 (.455-2.448)	
Stage I II III IV	23/415 (5.54) 96/881 (10.8) 158/583 (27.1) 138/138 (100)	1 (Reference) 1.960 (1.244-3.090) 5.447 (3.517-8.436) 95.570 (60.478-151.023)	**<.001** **.004** **<.001** **<.001**		
T stage T1 T2 T3 T4	67/670 (10) 220/1048 (20.9) 38/155 (24.5) 89/143 (62.2)	1 (Reference) 2.207 (1.679-2.902) 2.438 (1.637-3.631) 11.090 (8.050-15.279)	**<.001** **<.001** **<.001**	1 (Reference) 1.426 (1.037-1.962) 1.579 (1.012-2.464) 1.794 (1.053-3.057)	**.029** **.044** **.032**
Positive Axillary Node Count 0 1-3 4-9 ≥10	78/861 (9.06) 80/531 (15.07) 145/402 (36.07) 111/222 (50)	1 (Reference) 1.668 (1.221-2.278) 5.000 (3.795-6.587) 7.384 (5.524-9.870)	**.001** **<.001** **<.001**	1 (Reference) .811 (.561-1.171) 1.338 (.928-1.929) 1.644 (1.116-2.423)	.263 .119 **.012**
Metastasis site None Bone Lung Liver Brain Multiple organs	25/1627 (1.53) 142/142 (100) 25/25 (100) 15/15 (100) 21/21 (100) 185/185 (100)	1 (Reference) 156.760 (102.099-240.686) 139.613 (79.878-244.018) 171.002 (89.530-326.613) 173.699 (96.477-312.733)	**<.001** **<.001** **<.001** **<.001**	1 (Reference) 158.568(100.278.250.742) 131.993 (72.208-241.278) 133.403 (64.540-275.738) 129.981 (68.258-247.517) 126.654 (79.530-201.699)	**<.001** **<.001** **<.001** **<.001**

		Univariate analysis		Multivariate analysis	
Patients Descriptions	**Events/Total (%)**	**Hazard ratio (95% CI)**	**p**	**Hazard ratio (95% CI)**	**p**
		164.232 (107.535-250.822)			
Skin infiltration Positive Negative	80/152 (52.6) 335/1865 (18)	4.664 (3.642-5.974)	**<.001**	1.249 (.783-1.991)	.351
Surgical margin Positive Negative	69/367 (18.8) 346/1650 (21)	1.004 (.775-1.301)	.975		
Grade 1 2 3	23/304 (7.5) 155/987 (15.7) 237/726 (32.6)	1 (Reference) 2.290 (1.478-3.550) 5.417 (3.528-8.317)	**<.001** **<.001**	1 (Reference) .712 (.424-1.195) 1.017 (.595-1.739)	.198 .950
Mitotic index 1 2 3	97/775 (12.5) 98/637 (15.3) 218/591 (36.8)	1 (Reference) 1.546 (1.164-2.053) 4.303 (3.369-5.497)	**.003** **<.001**	1 (Reference) 1.033 (.734-1.455) .996 (.726-1.367)	.851 .982
ER receptor Positive Negative	300/1598 (18.7) 115/419 (27.4)	.648 (.523-.804)	**<.001**	.838 (.403-1.350)	.324
PR receptor Positive Negative	240/1337 (18) 175/680 (25.7)	.640 (.526-.777)	**<.001**	1.029 (.769-1.376)	.847

		Univariate analysis		Multivariate analysis	
Patients Descriptions	**Events/Total (%)**	**Hazard ratio (95% CI)**	**p**	**Hazard ratio (95% CI)**	**p**
Ki67 <15 ≥15	204/1130 (18) 210/885 (23.7)	1.758 (1.443-2.143)	**<.001**	1.062 (.811-1.389)	.662
HER2 Positive Negative	125/478 (26.1) 290/1539(18.8)	1.646 (1.333-2.032)	**<.001**	1.077 (.730-1.590)	.708
Extensive intraductal component Positive Negative	96/334 (28.7) 319/1683 (18.9)	1.646 (1.310-2.069)	**<.001**	**1.175 (.895-1.542)**	.247
Lymphovascular invasion Positive Negative	211/954 (22.1) 204/1063 (19.1)	1.190 (.981-1.443)	.077	1.077 (.832-1.394)	.574
Perineural invasion Positive Negative	103/437 (23.5) 312/1580(19.7)	1.145 (.917-1.431)	.233		
Chemotherapy None Neoadjuvant Adjuvant	40/324 (12.3) 78/235 (33.1) 297/1458 (20.3)	1 (Reference) 3.202 (2.186-4.690) 1.553 (1.116-2.161)	**<.001** **<.001**	1 (Reference) 1.137 (.699-1.851) .840 (.563-1.252)	.604 .301
Chemotherapy Protocol None FAC AC+TXT Other	40/324 (12.3) 54/166 (32.5) 34/273 (12.4) 279/1235 (22.5)	1 (Reference) 1.858 (1.255-2.750) 1.429 (.569-3.592) 1.694 (1.247-2.301)	**.002** .447 **.001**		

doi.org/10.32388/UB8V1E

		Univariate analysis		Multivariate analysis	
Patients Descriptions	**Events/Total (%)**	**Hazard ratio (95% CI)**	**p**	**Hazard ratio (95% CI)**	**p**
Radiotherapy Positive Negative	295/1757 (16.7) 120/260 (46.1)	.302 (.244-.374)	**<.001**	.470 (.352-.626)	**<.001**
Radiotherapy Type None Breast alone Locoregional	120/260 (46.1) 44/587 (7.5) 51/1170(4.3)	1 (Reference) .127 (.090-.179) .389 (.312-.484)	**<.001** **<.001**		
Tamoxifen period No TMX TMX ≤5 years TMX>5 years	130/652 (19.9) 9/107 (8.4)	1 (Reference) .769 (.623-.949) .283 (.146-.551)	**.014** **<.001**	1 (Reference) 1.022 (.733-1.425) .425 (.196-.922)	.896 **.030**
AI period No AI AI ≤5 years AI >5 years	191/937 (20.3) 32/265 (12)	1 (Reference) .812 (.664-.992) .404 (.278-.589)	**.042** **<.001**	1 (Reference) .861 (.643-1.154) .817 (.505-1.319)	.317 .408
LHRH None LHRH ≤2 years >2 years	8/35 (22.8) 57/343 (16.6)	1 (Reference) 1.242 (.616-2.504) .754 (.570-.998)	.544 .048	1 (Reference) 2.426 (1.057-5.568) 1.225 (.798-1.880)	**.037** .353
Subtyping 2 HER2-enriched TNBC Luminal A Luminal B	51/142 (35.9) 51/236 (21.6) 182/952 (19.1) 131/688 (19.0)	1 (Reference) .479 (.325-.707) .380 (.278-.520) .488 (.353-.675)	**<.001** **<.001** **<.001**	1 (Reference) .794 (.438-1.438) .891 (.336-2.362) 1.157 (.543-2.463)	.447 .817 .706

		Univariate analysis		Multivariate analysis	
Patients Descriptions	**Events/Total (%)**	**Hazard ratio (95% CI)**	**p**	**Hazard ratio (95% CI)**	**p**
HER2-enriched received Herceptin HER2-enriched did not receive Herceptin	40/120 (33.3) 11/22 (50)	1.515 (.777-2.954)	.223		

Table 8. Univariable and multivariable analysis of Breast Cancer survival using Cox's proportional hazards model within disease-free survival

BMI, body mass index; IDC, invasive ductal carcinoma; ILC, invasive lobular carcinoma; ER, estrogen receptor; PR, progesterone receptor; HER-2, human epidermal growth factor receptor 2, TNM, tumor-node-metastasis staging system based on the system of the American Joint Committee on Cancer

		Univariate analysis		Multivariate analysis	
Patients Descriptions	**Events/Total (%)**	**Hazard ratio (95% CI)**	**p**	**Hazard ratio (95% CI)**	**p**
Age group <35 years 35–50 years > 50 years	21/84 (25) 101/756 (13.3) 241/1177 (20.4)	1 (Reference) .476 (.297-.762) .873 (.559-1.363)	**.002** .550	1 (Reference) .598 (.354-1.012) 1.033(.569-1.876)	.055 .914
BMI <25 ≥25	72/373 (19.3) 291/1644 (17.7)	.919 (.710-1.190)	.524		
Menopause Age (mean) Alive 48.35 years Death 48.14 years	244/2017 (12.1)	1.003 (.977-1.030)	.832		.
Menstruation Age (mean) Alive 13.15 years Death 13.11 years	363/2017(18)	.939 (.870-1.015)	.112		
Menstruation situation Premenopause Postmenopause	114/787 (14.5) 244/1217 (20)	1.582 (1.266-1.976)	**<.001**	.665 (.679-1.403)	.966
Number of births No birth 1-2 birth 3 and more	28/162 (17.3) 209/1320 (15.8) 120/520 (23.1)	1 (Reference) .937 (.631-1.389) 1.298 (.860-1.958)	.745 .214		
Family History Positive Negative	87/632 (13.8) 276/1385 (20)	.709 (.557-.902)	**.005**	.902 (.696-1.168)	.434
Breast-feeding	224/1192 (18.8)	1.156 (.935-1.430)	.180		

		Univariate analysis		Multivariate analysis	
Patients Descriptions	**Events/Total (%)**	**Hazard ratio (95% CI)**	**p**	**Hazard ratio (95% CI)**	**p**
Positive Negative	139/825 (16.8)				
Breast site Left Right Bilateral	185/1013 (18.3) 178/935 (19) 14/69 (20.3)	1 (Reference) .975 (.794-1.198)	.995 .811		
Location Unilateral Metacron Sencron	363/1948 (18.6) 9/46 (19.6) 5/23 (21.7)	1 (Reference) .778(.401-1.509) 1.524 (.630-3.687)	.484 .458 .350		
Tumor Quadrant Inner Outer Periareolar Multifocal	73/402 (18.1) 210/1205 (17.4) 42/259 (16.2) 38/150 (25.3)	1 (Reference) 1.002 (.768-1.308) .951 (.651-1.391) 1.659 (1.121-2.456)	.987 .797 **.011**	1 (Reference) .990 (.734-1.335) .884 (.577-1.353) .713 (.448-1.136)	.948 .569 .155
Histopathologic Type Invasive ductal carcinoma Invasive lobular carcinoma Other	293/1652 (17.7) 22/122 (18) 48/243(19.7)	1 (Reference) .937 (.607-1.445) 1.109 (.817-1.504)	.752 .768 .508		
Surgical Type BCS MRM No surgery	93/1016 (9.15) 221/930 (23.8) 49/71(69)	1 (Reference) 2.344 (1.839-2.987) 19.760 (13.887-28.117)	**<.001** **<.001**	1 (Reference) 1.204 (.873-1.599) 1.154(.561-2.374)	.279 .697
Axillary surgery Sentinel lymph node dissection	21/451(4.7) 286/1477(19.4)	1 (Reference) 3.040 (1.950-4.741)	**<.001** **<.001**	1 (Reference) 1.466 (.896-2.400)	.128 **.004**

		Univariate analysis		Multivariate analysis	
Patients Descriptions	**Events/Total (%)**	**Hazard ratio (95% CI)**	**p**	**Hazard ratio (95% CI)**	**p**
Axillary dissection No axillary surgery	56/89(62.9)	22.238 (13.458-36.747)		3.251 (1.451-7.283)	
Stage I II III IV	22/415 (5.3) 108/881 (12.3) 150/583 (25.7) 83/138 (60.1)	1 (Reference) 2.199 (1.390-3.477) 5.085 (3.250-7.954) 26.548 (16.530-42.638)	**.001** **<.001** **<.001**		
T Stage T1 T2 T3 T4	57/670 (8.5) 183/1048 (17.5) 37/155 (23.9) 85/143 59.4()	1 (Reference) 2.110 (1.567-2.841) 2.571 (1.699-3.889) 12.764 (9.091-17.920)	**<.001** **<.001** **<.001**	1 (Reference) 1.719 (1.227-2.410) 1.749 (1.099-2.786) 1.843 (1.081-3.143)	**.002** **.018** **.025**
Infiltrated Axillary Node Count 0 1-3 4-9 ≥10	88/861 (10.2) 69/531 (13) 122/402 (30.3) 83/222 (37.4)	1 (Reference) 1.214 (.886-1.664) 3.710 (2.819-4.882) 4.563 (3.379-6.161)	.228 **<.001** **<.001**	1 (Reference) .897 (.624-1.289) 1.390 (.957-2.018) 1.099 (.726-1.662)	.556 .084 .657
Metastasis site None Bone Lung Liver Brain Multiple organs	117/1627(7.19) 73/142 (51.4) 15/25 (60) 10/15 (66.6) 20/21 (95.2) 126/185 (68.1)	1 (Reference) 8.934 (6.667-11.972) 11.240 (6.562-19.252) 14.344 (7.513-27.385) 23.899 (14.826-38.522) 15.101 (11.720-19.458)	**<.001** **<.001** **<.001** **<.001** **<.001**	1 (Reference) 5.123 (3.696-7.100) 4.350 (2.361-8.015) 10.520 (5.270-20.999) 7.798 (4.372-13.909) 5.059 (3.710-6.899)	**<.001** **<.001** **<.001** **<.001** **<.001**

		Univariate analysis		Multivariate analysis	
Patients Descriptions	**Events/Total (%)**	**Hazard ratio (95% CI)**	**p**	**Hazard ratio (95% CI)**	**p**
Skin infiltration Positive Negative	83/152 (54.6) 280/1865 (15)	6.585 (5.127-8.459)	**<.001**	2.093 (1.359-3.223)	**.001**
Surgical margins Positive Negative	73/367 (19.9) 290/1650 (17.6)	1.427 (1.103-1.846)	**.007**	1.236 (.922-1.656)	.156
Grade 1 2 3	28/304 (9.2) 148/987 (15) 187/726 (25.8)	1 (Reference) 1.805 (1.205-2.704) 3.484 (2.341-5.185)	**.004** **<.001**	1 (Reference) .656 (.413-1.042) .535 (.330-.870)	.074 **.012**
Mitotic index 1 2 3	47/775 (6) 57/637 (8.9) 256/591 (43.3)	1 (Reference) 2.157 (1.462-3.182) 12.288 (8.955-16860)	**<.001** **<.001**	1 (Reference) 1.819 (1.182-2.799) 5.904 (4.086-8.532)	**.006** **<.001**
ER receptor Positive Negative	254/1598 (15.9) 109/419 (26)	.578 (.462-.723)	**<.001**	.758 (.410-1.404)	.379
PR receptor Positive Negative	213/1337 (15.9) 150/680 (22)	.641 (.520-.790)	**<.001**	.990 (.711-1.378)	.950
Ki67 <15 ≥15	183/1130 (16.2) 179/885 (20.2)	2.025 (1.636-2.507)	**<.001**	2.627 (1.478-4.670)	**.001**

doi.org/10.32388/UB8V1E

		Univariate analysis		Multivariate analysis	
Patients Descriptions	**Events/Total (%)**	**Hazard ratio (95% CI)**	**p**	**Hazard ratio (95% CI)**	**p**
HER2 Positive Negative	98/478 (20.5) 265/1539(17.2)	1.500 (1.188-1.894)	**.001**	1.154 (.729-1.827)	.541
Extensive intraductal component Positive Negative	90/334 (27) 273/1683 (16.2)	1.815 (1.430-2.304)	**<.001**	1.193 (.879-1.621)	.258
Lymphovascular invasion Positive Negative	187/954 (19.6) 176/1063 (16.5)	1.242(1.011-1.527)	**.039**	1.099 (.844-1.431)	.484
Perineural invasion Positive Negative	97/437 (22.1) 266/1580(16.8)	1.215 (.963-1.533)	.101		
Chemotherapy None Neoadjuvant Adjuvant	42/324 (13) 57/235 (24.3) 264/1458 (57.6)	1 (Reference) .816 (.483-1.379) .648 (.437-.959)	.447 **.03**	1 (Reference) .774 (.458-1.309) .628 (.424-.930)	.340 **.02**
Chemotherapy Protocol None FAC AC+TXT Other	42/324 (13) 55/166 (33.1) 44/273 (16.1) 216/1235 (17.4)	1 (Reference) 1.483 (1.004-2.192) 2.269 (1.113-4.627) 1.230 (.900-1.682)	**.048** **.024** .194		
Radiotherapy Positive Negative	276/1757 (15.7) 87/260 (33.4)	.427 (.335-.543)	**<.001**	.885 (.637-1.230)	.467
Radiotherapy Type	87/1757 (15.7)	1 (Reference)	**<.001**		

		Univariate analysis		Multivariate analysis	
Patients Descriptions	**Events/Total (%)**	**Hazard ratio (95% CI)**	**p**	**Hazard ratio (95% CI)**	**p**
No Breast alone Locoregional	48/587 (8.2) 228/1170(19.5)	.220 (.155-.313) .524 (.409-.672)	**<.001**		
Tamoxifen period No TMX TMX ≤5 years TMX>5 years	262/1258 (20.8) 96/652 (14.7) 5/107 (4.6)	1 (Reference) .539 (.426-.683) .146 (.060-.354)	**<.001** **<.001**	1 (Reference) .540 (.376-775) .141 (.075-.367)	**.001** **<.001**
AI period No AI AI ≤5 years AI >5 years	169/815 (20.7) 178/937 (19) 16/265 (6)	1 (Reference) .828 (.671-1.022) .193 (.116-.323)	.079 **<.001**	1 (Reference) .612 (.442-.848) .140 (.092-.259)	**.003** **<.001**
LHRH No LHRH ≤2 years >2 years	324/1634 (19.8) 7/35 (20) 31/343 (9)	1 (Reference) 1.121 (.530-2.370) .430 (.298-.622)	.765 **<.001**	1 (Reference) 1.402 (.587-3.345) 1.004 (.613-1.644)	.447 .987
Subtyping2 HER2-enriched TNBC Luminal A Luminal B	45/142 (32) 49/236 (20.7) 178/952 (18.7) 91/688 (13.2)	1 (Reference) .493 (.330-.737) .368 (.266-.510) .391 (.275-.557)	**.001** **<.001** **<.001**	1 (Reference) .900 (.471-1.722) 10.551 (2.956-37.668) 1.268 (.584-2.755)	.751 **<.001** .548
HER2-enriched received Herceptin HER2-enriched did not receive Herceptin	33/120 (27.5) 14/22 (63.6)	2.109 (1.121-3.965)	**.021**		

Table 9. Univariable and multivariable analysis of Breast Cancer survival using Cox's proportional hazards

doi.org/10.32388/UB8V1E

model within overall survival.

BMI, body mass index; IDC, invasive ductal carcinoma; ILC, invasive lobular carcinoma; ER, estrogen receptor; PR, progesterone receptor; HER-2, human epidermal growth factor receptor 2, TNM, tumor-node-metastasis staging system based on the system of the American Joint Committee on Cancer

Figures

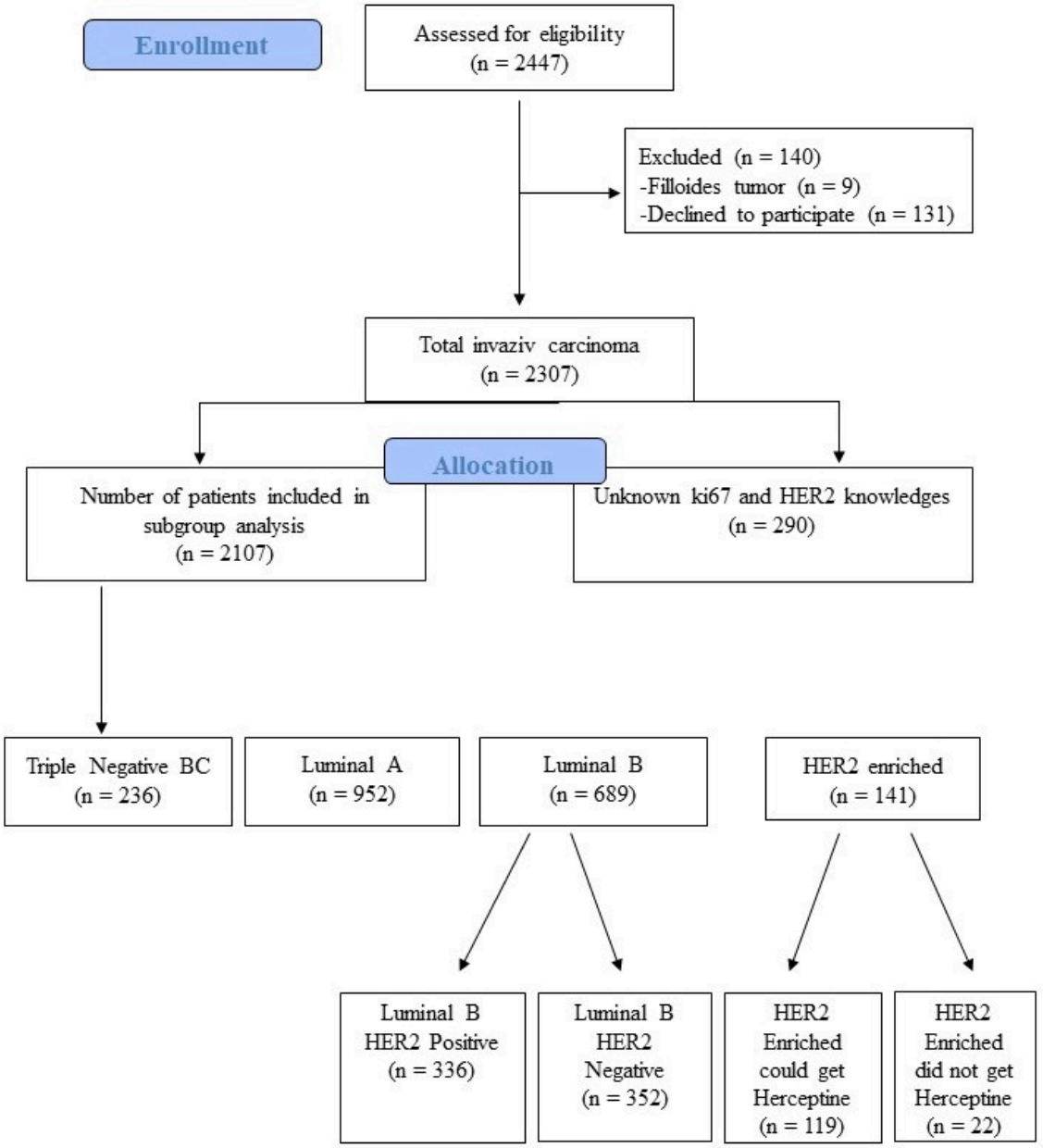

Figure 1. Distribution of BC patients in our series by subtyping

doi.org/10.32388/UB8V1E

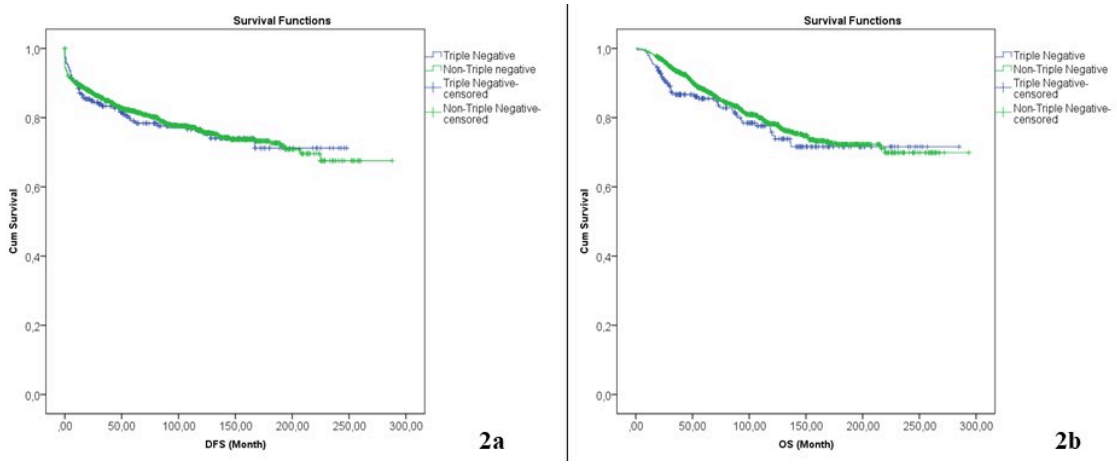

Figure 2. Survival curve of DFS **(a)** and OS **(b)** for the TNBC and NTNBC subgroups producing subtype 1 using the Kaplan–Meier method.

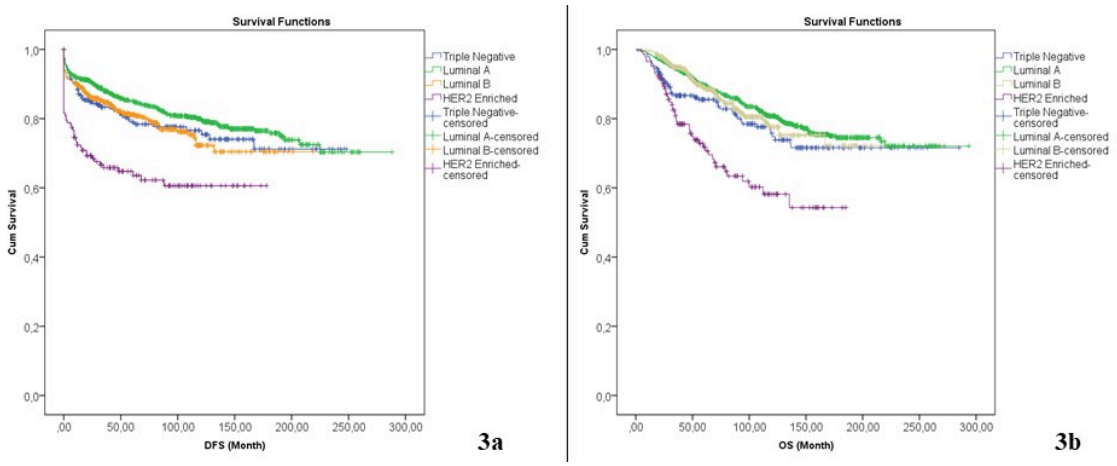

Figure 3. Survival curve of DFS **(a)** and OS **(b)** for TNBC, Luminal A, Luminal B, and HER2-enriched subgroups producing subtype 2 using the Kaplan–Meier method.

doi.org/10.32388/UB8V1E

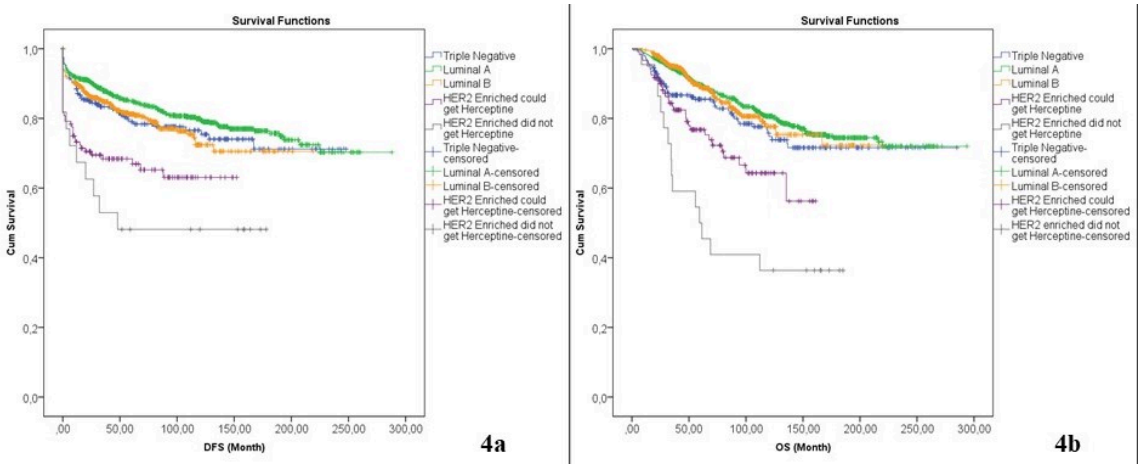

Figure 4. Survival curve of DFS **(a)** and OS **(b)** for TNBC, Luminal A, Luminal B, and HER2 -enriched subgroups that received Herceptin and the HER2-enriched subgroups that did not receive Herceptin producing subtype 3 using the Kaplan–Meier method.

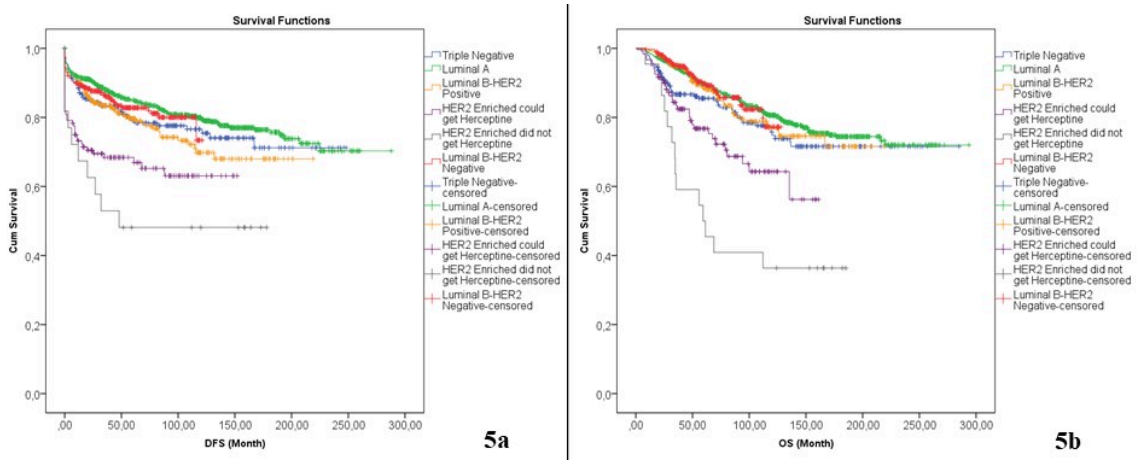

Figure 5. Survival curve of DFS **(a)** and OS **(b)** for the TNBC, Luminal A, and Luminal B subgroups that received Herceptin, the Luminal B subgroups that did not receive Herceptin, the HER2-enriched subgroup that received Herceptin, and the HER2-enriched subgroup that did not receive Herceptin, producing subtype 4 using the Kaplan–Meier method.

doi.org/10.32388/UB8V1E

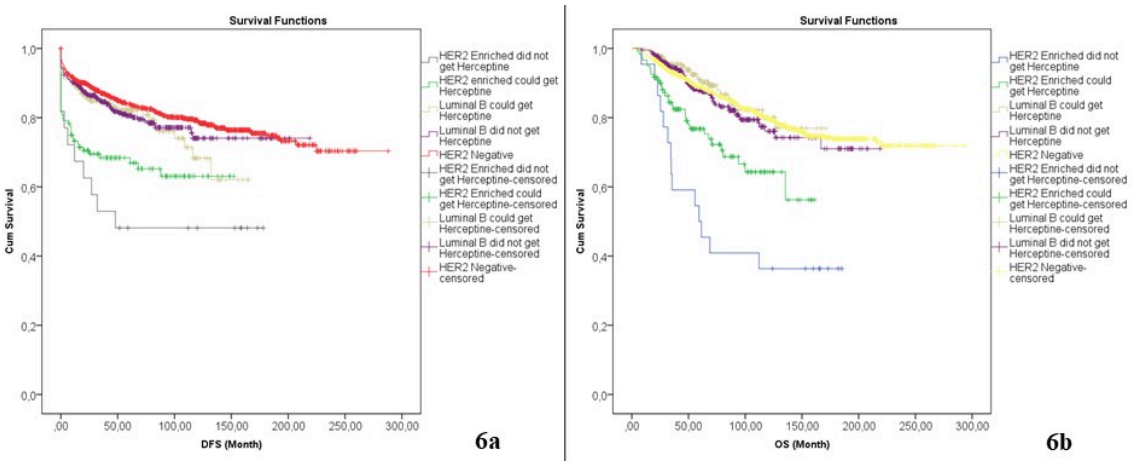

Figure 6. Survival curves of DFS **(a)** and OS **(b)** for the HER2-negative, Luminal B subgroup receiving Herceptin, the Luminal B subgroup that did not receive Herceptin, the HER2-enriched subgroup that received Herceptin, and the HER2-enriched subgroup that did not receive Herceptin, producing subtype 5 using the Kaplan–Meier method.

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Declarations

Funding: The author(s) received no specific funding for this work.

Potential competing interests: The author(s) declared that no potential competing interests exist.