Internal Medicine and Medical Investigation Journal
E-ISSN: 2474-7750
Homepage: www.imminv.com

ORIGINAL ARTICLE

Evaluation of the Survival of *HER2*-Positive Breast Cancer Patients Receiving Herceptin Compared With Those Who Did Not Receive It

Ali Akhavan[1], Fariba Binesh[2], Somaye Sadat Hoseini-Dezki[2*], Seyed Mohammad Reza Mortazavizadeh[3]

[1] *Department of Nuclear Medicine, Shahid Sadoughi University of Medical Sciences, Yazd, Iran*

[2] *Department of Pathology, Shahid Sadoughi University of Medical Sciences, Yazd, Iran*

[3] *Department of Internal Medicine, Faculty of Medicine, Islamic Azad University, Yazd, Iran*

Corresponding Author: Somaye Sadat Hoseini-Dezki, E-mail: Somaie_hosseini2959@Yahoo.com

ARTICLE INFO

Article history
Received: Oct 11, 2017
Accepted: Dec 02, 2017
Published: Jun 13, 2018
Volume: 3
Issue: 2

Conflicts of interest: None
Funding: None

Key words
Herceptin,
HER2 Positive,
Breast Cancer

ABSTRACT

Introduction: Breast cancer is the most common type of cancer that affects women. Human epidermal growth factor receptor 2 (*HER2*) is a gene that plays a significant role in the development of breast cancer. The aim of this study was to compare the survival of *HER2*-positive breast cancer patients who received Herceptin with those who did not receive it in Yazd, Iran. **Materials and Methods:** Our study was an analytical cross-sectional study. Sampling was done on all patients with *HER2*-positive invasive ductal carcinoma (*HER2* positive) who were referred to Shahid Sadoughi Hospital or Shahid Ramezanzadeh Hospital from 2003 to 2014. The data were collected using a questionnaire, which included questions on age, type of tumor, tumor grade, recurrence history, tumor size, number of lymph nodes, and whethr Hercepin was received or not received. Data were then entered into SPSS version 18 and analyzed by statistical tests. **Results:** The mean age of the patients was 50.78 ± 10.75 years. The mean survival time was 61.61 ± 2.93 months, and the mean recurrence time was 104.104 ± 41.3 months. The results also showed that there was no statistically significant difference between the 2 groups, despite the higher mean survival time and less recurrence time in patients receiving Herceptin compared with those who did not receive Herceptin (P>0.05). **Conclusions:** According to the results of this study, the efficacy of Herceptin as a neoadjuvant treatment in the survival of *HER2*-positive breast cancer patients has not been established.

INTRODUCTION

Breast cancer is one of the most common cancers in the world today. According to the US Joint Commission on Cancer, 1 in every 8 women is diagnosed with breast cancer (1). Nearly 12% of women in the United States suffer from breast cancer in their lifetime (2, 3). Breast cancer is the second leading cause of cancer death after lung cancer and is the most common cause of cancer death in women aged 55 to 45 years. Treatment for breast cancer includes surgery, radiotherapy, chemotherapy, hormonal therapy, and biological therapy. The treatment strategy is dependent on various factors; one of them is the human epidermal growth factor receptor 2 (HER2). *HER2* is an oncogene that encodes transmembrane glycoprotein with tyrosine kinase activity called P-185, which belongs to the family of epithelial growth factor receptors (4). *HER2* is normal at the lower level in epithelial cells, but it increases in 15% to 30% of cases of breast cancer (5). This gene receives a protein called *HER2* on the cell surface that receives growth signals (6). About 15% to 30% of cancers are *HER2*-positive breast cancer, and the neoadjuvant therapy performed to treat them includes Herceptin (7). Herceptin is a specific monoclonal antibody that identifies and destroys tumor cells by targeting *HER2 (*8). Herceptin is a high-priced drug; for a treatment period of 1 year, it costs about $70,000, which is about 40% of the cost of complete breast cancer treatment (9). Approximately 65% of *HER2*-positive breast cancer patients do not respond to Herceptin, and the disease progresses in approximately 70% of those who have given an early response to Herceptin, 1 year after the onset of treatment (10).

Considering the high prevalence of breast cancer and the use of Herceptin as a neoadjuvant treatment, the high cost, complications, and resistance to this drug, we determined that the survival of patients with *HER2*-positive breast cancer in patients receiving Herceptin compared with those who did not receive it in Yazd, Iran.

MATERIALS AND METHODS

Our study was a cross-sectional, retrospective study. The study population included 272 patients with *HER2*-positive

Published by Mehrabani Publishing LLC.

http://dx.doi.org/10.24200/imminv.v3i2.122

invasive ductal carcinoma (*HER2* positive) who referred to Shahid Sadoughi Hospital or Shahid Ramezanzadeh Hospital, Iran, between 2003 and 2014. A census survey method was conducted, and all patients with breast cancer were enrolled. However, patients with metastatic breast cancer and heart diseases, those receiving Herceptin, those whose records were incomplete, and those who could not afford telephone access were excluded. The records of the patients were collected from Shahid Sadoughi Hospital, Shahid Ramezanzadeh Hospital, and Dr Mortazavi Zadeh's office. Information including patient's age, type of tumor, tumor grade, relapse date, tumor size, and the number of lymph nodes was recorded. Patients' telephone numbers were also recorded. Information about receiving Herceptin as well as their survival was collected through phone calls. The questionnaire was recorded. Survival meant the mortality rate of patients until 2014. In the next stage, patients were staged. In this study, patients who received at least 4 courses of Herceptin were assigned to receive Herceptin. The reason is that considering the cost in the years of the study, which was based on the results of various studies that showed that, on the basis of the prescription of a male patient, 9-week treatment with Herceptin may be sufficient, the Ministry of Health decided to limit the treatment to 9 weeks; therefore, some patients received Herceptin for 1 year while some of them received it for less than 1 year. There was also a need for financial intelligence analysis, because despite planning, 1 year later, Herceptin was discontinued in the middle of the test. Finally, the test results were analyzed using SPSS version 18, chi-square, and Fisher tests. Patient survival time was calculated by reducing the date of diagnosis from the date of the patient's death or the disease recurrence date.

RESULTS

The results of our study showed that the mean age of the participants in the study was 50.78 ± 10.75 years. The minimum age and the maximum age of the participants were 25 and 85 years, respectively. The age group, Herceptin, grade, stage, ER, PR, *P53*, Ki-67, death events, and recurrence events, and their abundance of the study group are presented in Table-1.

The analysis of the results of our study by the chi-square and Fisher tests showed that there was a significant relationship of the death event with the stage of the tumor and that of the incidence of recurrence with the stage and grade of the tumor ($P<0.05$).

The survival of patients was analyzed by the Kaplan-Meier test. The results showed that the average survival time using the Kaplan-Meier test was 119.61 ± 2.93 minutes, the curve of which is shown in Figure-1.

The mean survival time of patients according to the variables of the study was investigated using the log-rank test, the results of which are presented in Table 2. The results of this study showed that the mean survival time in patients receiving Herceptin was 19.11, and in those who did not receive Herceptin, it was 10.99. Despite the higher mean survival time of patients receiving Herceptin, compared with those not receiving it, there was no statistically significant difference between the 2 groups ($P>0.05$).

Table 1. Frequency of patients according to the variables studied

Variable	Subgroup	Frequency	Percent
Age group	<30	6	2/2
	30-39	33	12/1
	40-49	80	29/4
	50-59	105	38/6
	60-69	33	12/1
	>70	15	5/5
Receiving herceptin	Yes	114	41/9
	No	123	45/2
Grade	I	17	6/3
	II	118	43/4
	III	78	28/7
Stage	I	22	8/1
	II	125	46/0
	III	90	33/1
ER	Negative	116	42/6
	Positive	146	53/7
PR	Negative	140	51/5
	Positive	121	44/5
P53	Negative	73	26/8
	Positive	86	31/6
Ki-67	Negative	64	23/5
	Positive	70	25/7
`Death	Yes	50	18/4
	No	194	71/3
Recurrence	Yes	53	19/5
	No	173	63/6

ER: Esterogen receptor, ; PR: Progestron receptor

The results of our study on the mean survival time of patients in terms of the grade of the disease and receiving Herceptin showed that the average survival rate of grade I patients receiving Herceptin and those not receiving it, was not measurable, because all patients in both the groups survived until the end of the study. The mean survival times of grade II patients receiving Herceptin and those who did not receive were 11.07 and 102.58 months, respectively, with a statistically significant difference ($P=0.061$). There was no statistically significant difference between the mean survival time of grade III patients receiving Herceptin and those who were not ($P=0.962$).

Also, the results of the study on the mean survival time in terms of the stage of the disease and receiving Herceptin showed that none of the 11 patients who received Herceptin and was in stage I died, so the average survival rate for this group could not be obtained. The mean survival times of stage II patients who received Herceptin, and those who did not, were 11.16 and 131.63 months, respectively ($P=0.220$). The mean survival times of stage III patients who received Herceptin and those who did not were 10.088 and 87.95 months, respectively. There was no statistically significant difference between them ($P=0.456$).

Table 2. Frequency and descriptive index of survival time in terms of the variables studied

Variable	Subgroup	Total number	Death	Alive number	Survival mean	*P* value
Receiving herceptin	Yes	110	11	99 (90.0%)	117.19	0.054
	No	115	28	87 (75.7%)	190.79	
Age group	<30	4	0	4 (100%)	---	0.122
	39-30	29	3	26 (89.7%)	108.05	
	49-40	64	8	56 (87.5%)	115.66	
	59-50	91	16	75 (82.4%)	112.97	
	69-60	27	9	18 (66.7%)	95.71	
	>70	10	3	7 (70%)	99.85	
Grade	I	12	0	12 (100%)	---	0.145
	II	94	12	82 (87.2%)	108.60	
	III	69	14	55 (79.7%)	98.54	
Stage	I	18	1	17 (94.4%)	115.92	<0.001
	II	105	9	96 (91.4%)	122.43	
	III	74	21	53 (71.6%)	97.82	
ER	Negative	102	20	82 (80.4%)	108.35	0/322
	Positive	117	18	99 (84.6%)	117.76	
PR	Negative	121	23	98 (81.0%)	111.42	0/313
	Positive	97	14	83 (85.6%)	114.12	
P53	Negative	58	11	47 (81.0%)	114.99	0/665
	Positive	83	11	72 (86.7%)	116.13	
Ki-67	Negative	56	4	52 (92.9%)	118.07	0/065
	Positive	62	10	52 (83.9%)	92.26	

Abbreviations: ER: Esterogen receptor, ; PR: Progestron receptor

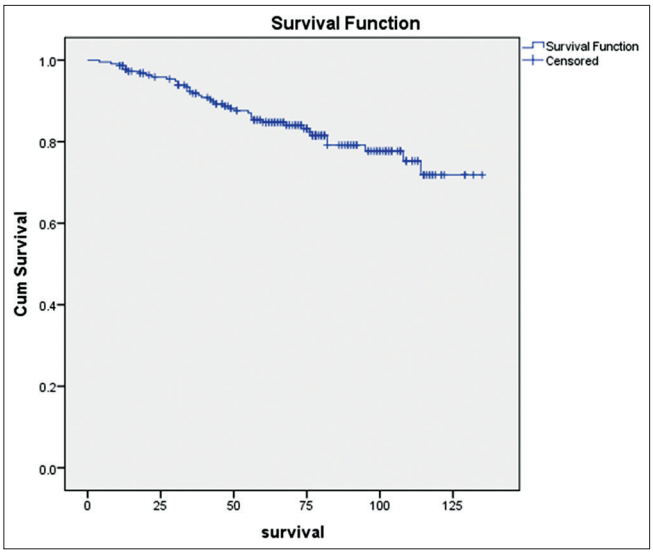

Figure 1. Survival Curve of Patients Studied by the Month

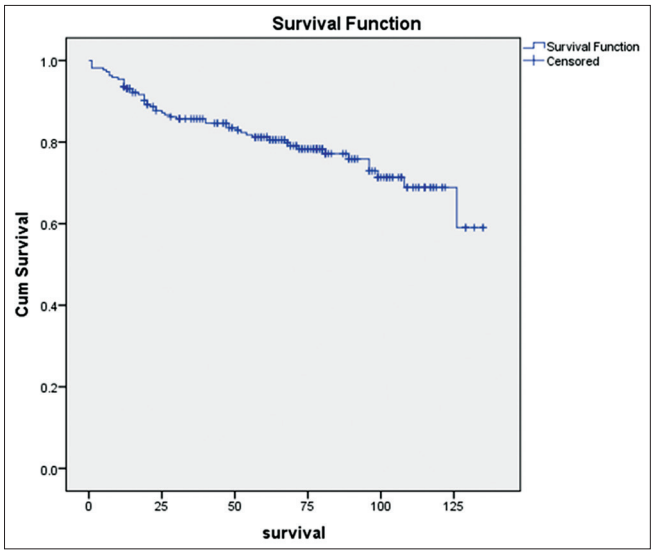

Figure 2. Recurrence Curve in Patients Evaluated by the Month

The recurrence of cancer in patients was analyzed by the Kaplan-Meier test. The results showed that the mean recurrence time using the Kaplan-Meier test was 104/104 ± 41.3 months, the curve of which has been shown in Figure 2.

The mean recurrence time of patients according to the variables of the study was investigated using the log-rank test, the results of which are presented in Table 3. The results of this study showed that the mean recurrence time of patients receiving Herceptin was 10.47% and of those who did not receive was 21.25%. However, the mean recurrence time of patients receiving Herceptin was significantly lower than that of those who received Herceptin. There was no statistically significant difference between these 2 groups (P>.05).

The results of our study on the mean recurrence time of patients in terms of the grade of the disease and receiving Herceptin showed that the mean recurrence time of grade I patients who received Herceptin and or who did not was not measurable because all patients from these 2 groups were alive

Table 3. Frequency and descriptive indices of recurrence time according to the variables studied

Variable	Subgroup	Total number	Recurrent number	No recurrent number	Survival mean	*P* value
Receiving herceptin	Yes	111	21	90 (81.1%)	104.37	0.569
	No	107	28	79 (73.8%)	105.21	
Age group	30 >	2	0	2 (100%)	---	0.217
	30-39	31	10	21 (67.7%)	86.31	
	40-49	62	11	51 (82.3%)	102.54	
	50-59	86	17	69 (80.2%)	111.57	
	60-69	26	6	20 (76.9%)	106.00	
	70 <	11	5	6 (54.5%)	90.94	
Stage	I	18	2	16 (88.9%)	108.83	0.005*
	II	105	15	90 (85.7%)	115.01	
	III	67	23	44 (65.7%)	90.11	
Grade	I	12	0	12 (100%)	---	0.004*
	II	89	13	76 (85.4%)	105.72	
	III	67	22	45 (67.2%)	84.28	
ER	Negative	97	22	75 (77.3%)	106.57	0.847
	Positive	116	25	91 (78.4%)	108.93	
PR	Negative	117	29	88 (75.2%)	103.72	0.249
	Positive	96	18	78 (81.3%)	108.61	
P53	Negative	64	17	47 (73.4%)	106.47	0.862
	Positive	78	17	61 (78.2%)	105.54	
Ki-67	Negative	57	12	45 (78.9%)	97.89	0.409
	Positive	62	16	46 (74.2%)	82.66	

ER: Esterogen receptor, ; PR: Progestron receptor

at the end of the study. The mean relapse times of grade II patients who received Herceptin and those who did not were 92.34 and 10.58 months, respectively, with a significant difference (*P*=0.491). In other words, cancer recurred sooner in patients who received Herceptin than those who did not. There was no significant difference between the mean relapse time of grade III patients receiving Herceptin and not (*P*=0.409).

Also, the results of the study on the mean survival time in terms of stage I patients receiving Herceptin showed that those who received Herceptin had no recurrent events (11); therefore, the mean recurrence time could not be obtained for this group. However, the mean recurrence time for the group of stage I patients who did not receive Herceptin was 50.92 months. The median recurrence times of stage II patients who received Herceptin and those who did not were 103.39 and 17/112 months, respectively, meaning that breast cancer soon recurred in stage II patients who received Herceptin. There was, however, statistically no significant difference between them (*P*=0.515). The median recurrence times of stage III patients who received Herceptin and those did not were 88.81 and 82.49 months, respectively, showing that there was no statistically significant difference between them (*P*=0.896).

DISCUSSION

The aim of this study was to evaluate the survival of *HER2*-positive breast cancer patients who received Herceptin compared with those who did not receive it.

The mean age of 272 patients studied at the time of diagnosis was 50.78 ± 10.75 years, and most patients were in the age group of 50 to 59 years. In a study conducted in Tehran (11), the mean age of patients was 51.3 years, which is consistent with our study results. Other studies have also reported that an average age of breast cancer patients is between 45 and 50 years (12), which is consistent with our study. The age distribution of women with breast cancer in the country indicates that the diagnostic age of the disease in the country is lower than that in Western Europe and North America, and also that women in Iran are more likely to develop breast cancer (13).

Of the 272 patients studied in our research, 50 (18.4%) died. In a study conducted in 2011 (11), the death rate of patients suffering from breast cancer was 18.8%, which is similar to the results of our study. In our study, the mean survival time was 2.93; however, there was no statistically significant relationship between the age and survival time. The relationship between the age and survival time of patients in different studies had different results. The results of a study conducted in 2008 (14) and that of another study conducted in 2009 (15) showed an inverse relationship between the patient's age and the survival time of the patients; however, no significant correlation was found between the age and the survival time of the patients, which is consistent with the results of our study (16, 17). It seems that the most important factor affecting the difference between the

age and survival time of patients is the number of patients in the study. Therefore, if the sample size is bigger, a meaningful relationship between the age and survival time can be established. In our study, the average survival time of patients receiving Herceptin was 19/117 months and that of those who did not receive Herceptin was 10.98 months. This difference was not statistically significant. Many studies have been conducted on the effect of Herceptin on the survival time of patients with breast cancer, yielding controversial results. In some of these studies, Herceptin was shown to increase the survival time of patients, whereas in other studies, it had no effect on the patients suffering from the breast cancer. In a study conducted by Smith, the survival rate of patients treated with Herceptin was higher than for those who did not receive Herceptin treatment, and this relationship was statistically significant, which is consistent with our study outcomes (18). The reason for this difference can be attributed to the difference in the type of study—the study by Smith was performed as a randomized controlled trial, but ours was a cross-sectional and retrospective study. In one of the studies conducted in 2013, a group of patients was treated with doxorubicin, cyclophosphamide, and paclitaxel, whereas another group was treated with the combination of the above drugs and Herceptin. The study results showed an additional 10-year survival time in the latter group; in addition, the relationship between the age and the survival time was significant (19). In another study, the survival rate of 1 year in patients receiving Herceptin was 26%, whereas it was 12% in the group not receiving Herceptin (20). In another study, conducted in France, the patients were divided into 2 groups. The first group received 12-month treatment with Herceptin and the second group received 6-month treatment with it. The results of this study showed that the survival time in the first group was 2 years more than that in the second group (21). Our study showed a statistically significant difference between the mean survival rate of patients who had positive or negative ER and Herceptin receptors. In several studies, the effects of estrogen receptors on survival rates were significant (11, 21, 22), which did not match the results of our study. The results of a study conducted in 1999 (23) and another conducted in 2001 (24) showed that positive or negative estrogen receptors do not play a role in the survival of patients (23, 24), which was consistent with our study results. The average survival time of patients in terms of positive or negative PR in our study, contrary to some other studies, was not significant (25, 26), which was not consistent with our study outcomes. However, in a 2008 study, a significant relationship was found between the mean survival time and positive or negative *PR* (27, 28), which was consistent with the results of our study. The stage of the disease is thought to be an effective factor in the survival of patients. In our study, in the groups of patients who did not receive Herceptin and were at the first, second, and third stages, the survival times were 110/1, 118/89, and 87/95 months, respectively. The difference was statistically significant. In a study of 2011 (11), patients diagnosed with the first stage of the disease showed a higher survival than those who were at the second and third stages, and they had a significant relationship (11).

CONCLUSION

In our study, the difference in mean survival time in the group receiving Herceptin was statistically significant at the second stage. In the main study that led to the confirmation of Herceptin by the Food and Drug Administration, all tests for HER2 positive status were rechecked in a reference laboratory, something which is virtually impossible in our country because there is no reference laboratory in Iran for rechecking.

Our study failed to demonstrate the efficacy of Herceptin adjuvant therapy in patients suffering from breast cancer. According to this study, as well as the results of other studies, a general screening for the use of Herceptin in Iran as an adjuvant treatment therefore seems necessary.

ACKNOWLEDGMENTS

The authors thank all staff members of Dr Mortazavi Zadeh's office, especially Mrs. Rajaei (Secretary of the office) and Mr. Karimi (Secretary of the office), for participating in this study.

AUTHER CONTRIBUTIOB

All authors contributed equally to this study.

CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest.

COMPLIANCE WITH ETHICAL STANDARDS

The nature of confidentiality was observed in this study and observations reported without any name of participants.

REFERENCES

1. WHO (October 2010). "Cancer". World Health Organization. Retrieved 5 January 2011.
2. Benson JR, Jatoi I. The global breast cancer burden. Future Oncol. 2012;8:697–702.
3. Coughlin S, Ekwueme D. Breast cancer as a global health concern. Cancer Epidemiol. 2009;33:315–8.
4. Maximiano S, Magalhaes P, Guerreiro MP, Morgado M. Trastuzumab in the Treatment of Breast Cancer. BioDrugs: clinical immunotherapeutics, biopharmaceuticals and gene therapy. 2016;30(2):75-86.
5. Nitta H, Kelly BD, Allred C, Jewell S, Banks P, Dennis E, et al. The assessment of HER2 status in breast cancer: the past, the present, and the future. Pathology international. 2016 Jun;66(6):313-24. PubMed PMID: 27061008.
6. Garnock-Jones KP, Keating GM, Scott LJ. Spotlight on trastuzumab as adjuvant treatment in human epidermal growth factor receptor 2 (HER2)-positive early breast cancer. BioDrugs. 2010;24:207–9.
7. McKeage K, Lyseng-Williamson KA. Trastuzumab: a pharmacoeconomic review of its use in early breast cancer. Pharmacoeconomics. 2008;26:699–719.

8. Patani N, Mokbel K. Herceptin and breast cancer: an overview for surgeons. Surg Oncol. 2010;19:11–21.
9. Pinto AC, Ades F, de Azambuja E, Piccart-Gebhart M. Trastuzumab for patients with HER2 positive breast cancer: delivery, duration and combination therapies. Breast. 2013;22:S152–5.
10. Slamon D, Eiermann W, Robert N, Pienkowski T, Martin M, Press M, et al. Adjuvant trastuzumab in HER2-positive breast cancer. N Engl J Med. 2011;365:1273–83.
11. Khodabakhshi R, Gohari MR, Moghadamifard Z. Disease-Free Survival of Breast Cancer Patients and Identification of Related Factors. Razi Journal of Medical Sciences.2011;18(89):27-39.
12. Gohari MR, Mahmoudi M, Kazem M, Pasha E, Khodabakhshi R. Recurrence in breast cancer analysis with frailty model. Saudi Med J; 2006. 27(8):1187-93.
13. Vahdaninia M, Montazeri A. Breast cancer in Iran: a survival analysis. Asian Pac J Cancer Prev; 2004. 5(2):223-25.
14. Mousavi SM, Mohagheghi MA, Mousavi Jerrahi A, Nahvijou A, Seddighi Z. Outcome of breast cancer in Iran: a study of Tehran center registry data. Asian Pac J Cancer Prev; 2008.9(2):275-78.
15. Sajadi A, Gregory H, Bajdik CH, Bashash M, Ghorbani A, Nouraie M, et al. Comparison of breast cancer survival in two populations: Ardabil, Iran and British Columbia, Canada. BMC Cancer; 2009. 9: 381-86.
16. Heydari ST, Mehrabani D, Tabei SZ, Azarpira N, Vakili MA. Survival of breast cancer in southern Iran. IJCP; 2009. 1: 51-54.
17. Akbari ME, Khayamzadeh M, Khoshnevis SJ, Nafisi N, Akbari A. Five and ten years survival in breast cancer patients mastectomies vs. breast conserving surgeries personal experience. IJBC;2008. 1(2): 53-56.
18. Ian Smith, Marion Procter, Richard D Gelber, Sébastien Guillaume, Andrea Feyereislova, Mitch Dowsett, Aron Goldhirsch, Michael Untch,et all. 2-year follow-up of trastuzumab after adjuvant chemotherapy in HER2-positive breast cancer: a randomized controlled trial. Lancet 2007; 369: 29–36.
19. Romond EH, Suman VJ, Jeong J-H, et al: Trastuzumab plus adjuvant chemotherapy for HER2-positive breast cancer: Final planned joint analysis of overall survival (OS) from NSABP B-31 and NCCTG N9831. 2012 San Antonio Breast Cancer Symposium. Abstract S5-5. Presented December 7, 2012.
20. Karam I, Hamilton S, Nichol A, Woods A, Speers C, Kennecke H. (2013). Population-based outcomes after brain radiotherapy in patients with brain metastases from breast cancer in the Pre-Trastuzumab and Trastuzumab eras. Radiation Oncology, 8 (1), 12.
21. Koizumi M, Yoshimoto M, Kasumi F, Iwase T. An open cohort study of bone metastasis incidence following surgery in breast cancer patients. BMC Cancer; 2010. 10:381-85.
22. Alexieva FJ, Van putten WLJ, Blankenstein A, Blonk vander J, Klijn JGM. The prognostic value and relationships of patient characteristics, estrogen and progestin receptors, and site of relapse in primary breast cancer. Cancer; 1988. 4:758–68.
23. Horita K, Yamaguchi A, Hirose K, Ishida M, Noriki S, Imamura Y, et al. Prognostic factors affecting disease-free survival rate following surgical resection of primary breast cancer. Eur J Histochem; 2001. 45(1):73-84.
24. Wolberg WH, Street WN, Mangasarian OL. Importance of nuclear morphology in breast cancer prognosis; 1999. 5:3542–48.
25. Heitz F, Rochon J, Harter P, Lueck HJ, Fisseler-Eckhoff A, Barinoff J, et al. Cerebral metastases in metastatic breast cancer: diseasespecific risk factors and survival. Ann Ancol. 2011. 22(7):1571-81.
26. Louis P, Pertschuk DO, Joseph G, Feldman DPH, Karen RN, Anne C. et al. lmmuonocytochermical detection of progesterone receptor in breast cancer with monoclonal antibody. Relation to biochemical assay, disease-free survival and clinical endocrine response. Cancer; 2006. 62(2):342-49.
27. Dawood S, Broglio K, Esteva FJ, Ibrahim NK, Kau SW, Islam R, et al. Defining prognosis for women with breast cancer and CNS metastases by HER2 status. Ann Ancol. 2008. 19:1242–48.
28. Mehranfar S, Zeinali S, Hosseini R, Mohammadian M, Akbarzadeh A, Feizi AH. History of Leukemia: Diagnosis and Treatment from Beginning to Now. Galen Medical Journal. 2017 Apr 1;6(1):12-22.