

RESEARCH ARTICLE

Prevalence of HER-2-Positive Invasive Breast Cancer: A Systematic Review from Iran

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Abstract

Background: The HER-2/neu gene is altered in 15-20% of breast cancer patients. Immunohistochemistry (IHC) is considered to be the most cost-effective method for HER-2 detection in many countries. Approximately 8,000 new cases of breast cancer are observed annually in Iran. The aims of this study were to conduct a systematic review of the literature on the rate of HER-2-positive breast cancer diagnosed by IHC in Iran. **Methods:** A systematic search of the medical literature using the Medline/PubMed, ISI and SID databases revealed articles published in the English and Persian languages evaluating HER-2-positive breast cancer in Iran. **Results:** From 22 studies, 3,033 patients were evaluated, of whom 1,350 were diagnosed as HER-2-positive by IHC HER-2 testing. The mean percentage of HER-2-positive patients was 44.5%, which is higher than that recorded in international statistics. Results of this meta-analysis showed a significant heterogeneity between ratios. There was a statistically significant difference between the results of pre- and post implementation of 2007 American Society of Clinical Oncology/College of American Pathologists (ASCO/CAP) guideline. IHC HER-2 testing has been performed in Iran for over 10 years. Similar to many other countries, before establishment of an infrastructure for IHC diagnostic tests, HER-2 testing was routinely performed in Iran. Our study showed that the statistics reported from Iran varied widely; for instance, the rate of HER-2-positive cases varied from 23.3% to 81.0%. **Conclusions:** Our results demonstrate that the lack of standardization and harmonization of this test have led to marked variations in breast cancer diagnosis in Iran.

Keywords: Breast cancer - HER-2/neu gene - immunohistochemistry - Iranian women

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Introduction

Breast cancer is considered to be one of the most prevalent cancers in many societies, and the incidence of this disease is increasing annually for various reasons. Before the introduction of targeted therapy two decades ago, survival rates had changed little (Mauri et al., 2008). In this latter type of therapy, the signalling pathways of neoplastic cells are blocked by small molecules, inhibiting the rate of proliferation of cancer cells (Chandarlapaty et al., 2011).

The HER-2/neu gene is an epidermal growth factor receptor located on chromosome 17. Statistics show that this gene is altered in 15-20% of breast cancer patients (Vogel CL et al., 2012). This change not only increases the rate of invasiveness and metastasis but also leads to chemoresistance. In 1998, the Food and Drug Administration (FDA) of the United States approved trastuzumab [Herceptin[®], supplied by Genentech, Inc]. for the treatment of HER-2-positive breast cancer. The high cost of this anti HER-2 monoclonal antibody has attracted attention. From the time of its launch, public health has been faced with serious challenges because in addition to its direct costs, trastuzumab-induced

cardiomyopathy incurs indirect costs for public health systems. Therefore, selection of eligible patients for treatment with trastuzumab has always been a challenge for oncologists (Hudis et al., 2007; Dendukuri, 2007). Although in situ hybridization (ISH) diagnostic techniques have been introduced, immunohistochemistry (IHC) is considered to be the most cost-effective method for HER-2 detection in many countries (Moore, 2012). However, this technique has certain limitations in the diagnosis of HER-2-positive breast cancer. Routine IHC HER-2 testing involves biopsy in the operation room. After fixation and tissue processing, IHC staining is performed and results are evaluated by pathologists. Subjective or technical errors in this testing can lead to inaccurate results.

The results of IHC HER-2 testing vary among different countries. There are no comprehensive guidelines for interpretation of results; leading scientific associations have recommended certain guidelines for the harmonization of IHC HER-2 testing. In 2006, the National Comprehensive Cancer Network (NCCN) codified HER-2 testing guidelines. According to this guideline, a score of 3+ is determined as a criterion of reactivity for >10% of tumour cells, which is identical to the corresponding criterion of a previous guideline (Carlson et al., 2006).

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One year later, the American Society of Clinical Oncology/ College of American Pathologists (ASCO/CAP) published a comprehensive guideline for HER-2 testing that has been accepted as the reference guideline worldwide. According to this guideline, to reduce interpretation errors in score 2+, the score 3+ criterion is determined as reactivity of >30% of tumour cells (Antonio et al., 2007).

In addition, depending on resource levels, certain regional guidelines have been codified. Some of these guidelines have been developed on the basis of the economical status of the concerned countries. The Breast Health Global Initiative and Asian Oncology Summit stratified different countries according to resources into the following four levels: basic, limited, enhanced and maximal. In the basic and limited levels, there are no recommendations for HER-2 diagnosis in breast cancer, but in the other two levels, IHC and ISH tests are recommended (Yip et al., 2008).

Approximately 8000 new cases of breast cancer are observed annually in Iran. According to published reports, the incidence of breast cancer in Iran is 22 cases per 100,000 people, while statistics show that this rate is increasing (Mousavi et al., 2007). The World Bank has ranked Iran in the category of upper middle income countries (World Bank, 2010), and thus Iran is considered as an enhanced-level nation. For this reason, HER-2 testing is considered as one component of the routine diagnostic testing protocol for breast cancer in Iran. From an economic perspective, it is considered cost-effective. However, ISH tests have not been implemented to any extent in Iran; unofficial statistics report a higher rate of HER-2-positive breast cancer diagnosed by IHC. Results of studies published in national and international journals show a relatively large variation in the percentage of HER-2-positive breast cancer in Iranian women.

The aims of this study were to conduct a systematic review of the literature published between 1996-2011 on the rate of HER-2-positive breast cancer diagnosed by IHC in Iran, and to determine any discrepancy between Iranian and world statistics. Our findings should be of interest to policy makers within the Iranian health care system.

Materials and Methods

A systematic search of the medical literature using the Medline/PubMed, ISI and SID databases revealed 38 articles published in the English and Persian languages evaluating HER-2-positive breast cancer in Iran (1996-2012). Key words used were breast cancer, mammary tumour, women, Iran, HER-2 and epidermal growth factor receptor. The following inclusion criteria were used: (a) random sampling must be performed only in Iran and IHC HER-2 testing must be performed in Iranian pathology laboratories; (b) age ranges were not determined; (c) only IHC HER-2 testing for histologically invasive ductal carcinoma and invasive lobular carcinoma was included; (d) whenever HER-2 tests other than IHC were used in the diagnosis of HER-2, only IHC results were included; (e) positivity of HER-2 was defined as score 3+; (f) reactivity >10% or >30% of tumour cells was considered as score 3+. In pre 2007 studies, although authors were unaware

of the >10% method, we considered immunoreactivity as being >10%, but in post2007 studies, reactivity is reported as being >10%, >30% or unknown.

Each article was reviewed twice. If a study had been published only in international congresses or there was no access to the full text of an abstract, we decided to exclude that article from the review. As a result, four studies were excluded. In addition, repeated results or statistically invalid data were excluded, and thus 22 studies were included in the present review. Meta-analysis was used to compare whole ratios. Statistical analysis was performed with Stata software (version 9 supplied by StataCorp LP, Inc). The confidence intervals (CI) of HER-2-positive data are given for both pre- and post 2007 studies.

Results

From 22 studies, 3033 patients (age range, 20-81 years) were evaluated (Table 1), of whom 1350 were diagnosed as HER-2-positive by IHC HER-2 testing. Sample sizes varied from 38-339 patients in each study. The mean percentage of HER-2-positive patients was 44.5%, which is higher than that recorded in international statistics. The rate of HER-2-positive patients was reported to be 23.3-81.0%. These studies were often conducted in public hospitals, with private medical centres and laboratories playing a small role. Studies evaluated HER-2 gene changes in isolation or with other breast cancer biomarkers. The provinces of Iran were represented as follows: Tehran, 11 studies (50.0%); Tabriz, 4 studies (13.6%); Isfahan, 2 studies (9.1%); Mashhad, 2 studies (9.1%) and other areas, 4 studies 18.2% (Table 2). Among the major provinces, Other areas had the greatest percentage of HER-2-positive breast cancer and Isfahan the lowest. There was also a marked variation within provinces; for example, the difference between the results of 2 studies performed in Tabriz was 57.7% and that between the maximum and minimum values in Tehran was 48.2% (Table 2).

Results of this meta-analysis showed a significant heterogeneity between ratios ($p < 0.001$). Because of this, the pooled estimate cannot be reported. This shows that the ratios were not accidental and that one or more factors were involved.

Dissociative results pre- and post2007 are shown in Table 3 and Figure 1. 67.6% of sample size was evaluated by IHC HER-2 testing pre2007, of which 49.6% (CI 95%: 47.4%-51.8%) of cases were HER-2-positive. After implementation of the ASCO/CAP 2007 guideline, testing was repeated in 36.4% of patients, of which 33.8% (CI 95%: 30.8%-36.8%) showed a positive result (Figure 1).

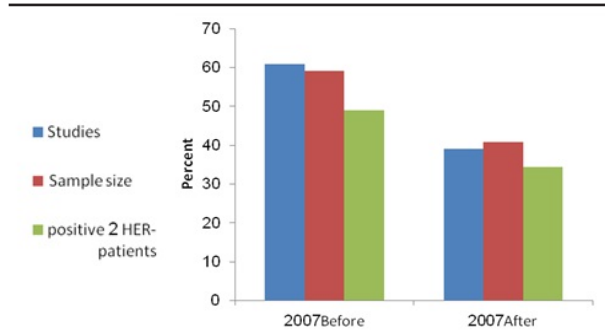
Fourteen studies (63.6%) were performed pre2007 and 8 (36.4%) were performed post2007; 4 (18.2%) were started pre2007 and finished post2007. The criterion for score 3+ in 21 studies (95.4%) was >10% reactivity of tumour cells. In one study (4.6%) performed post2007, the criterion for score 3+ was not established, thus the diagnostic method was considered as unknown. In 8 cases (36.4%) from the studies included, the ASCO/CAP 2007 guideline had not been followed. However, with a 95% CI, the results pre- and post 2007 do not overlap, and

Table 1. Studies Included According to Inclusion Criteria

| Study | Sample size | Number and % of HER-2 + cases | Region of study | Year research |
|--------------------------------|-------------|-------------------------------|-----------------|---------------|
| | | | | |
| Rabiee et al., 2008 | 81 | 31 (38/3%) | Tehran | 1996–2006 |
| Moradi-Marjaneh et al., 2008 | 339 | 176 (51/9%) | Mashhad | 2000–2005 |
| Mirmalek et al., 2005 | 106 | 61 (57/5%) | Tehran | 2001–2003 |
| Mofid et al., 2004 | 102 | 65 (63/7%) | Tehran | 2001–2003 |
| Sirati et al., 2009 | 200 | 143 (71/5%) | Tehran | 2001–2004 |
| Shamsalinia et al., 2010 | 67 | 21 (31/3%) | Ramsar | 2001–2005 |
| Attarian, et al., 2011 | 116 | 27 (23/3%) | Tehran | 2001–2009 |
| Afshar Mighaddam et al., 2008 | 139 | 41 (29/5%) | Isfahan | 2002–2009 |
| Alizadeh et al., 2010 | 105 | 45 (38/0%) | Arak | 2003–2006 |
| Shahriari Ahmadi et al., 2005 | 248 | 161 (64.9%) | Kermanshah | 2003–2005 |
| Moghadaszadeh et al., 2010 | 237 | 88 (37/1%) | Tabriz | 2003–2007 |
| Mirzaei et al., 2010 | 138 | 73 (52/9%) | Tehran | 2004–2005 |
| Kadivar et al., 2010 | 74 | 30 (40/5%) | Tehran | 2004–2007 |
| Mohammadzadeh et al., 2009 | 108 | 29 (26/8%) | Isfahan | 2004–2007 |
| Homaei-Shandiz et al., 2006 | 196 | 67 (34/2%) | Mashhad | 2005–2006 |
| Naghshvar et al., 2007 | 50 | 28 (56/0%) | Sari | 2005–2006 |
| Amouzegar-Hashemi et al., 2010 | 277 | 72 (26/0%) | Tehran | 2005–2009 |
| Ensani et al., 2010 | 38 | 13 (34/2%) | Tehran | 2008–2009 |
| Pourzand et al., 2011 | 105 | 85 (81/0%) | Tabriz | 2008–2010 |
| Kadivar et al., 2010 | 117 | 45 (38/5%) | Tehran | 2009–2010 |
| Halimi et al., 2011 | 116 | 27 (23/3%) | Tabriz | 2010–2011 |
| Salimi et al., 2012 | 74 | 22 (29/7%) | Tehran | 2010–2011 |

Table 2. HER-2 Statistics in Different Provinces of Iran

| Number of studies | Sample size and percentage of total number of casespos. | Number and percentage of HER-2 Cases | Difference between the highest and lowest results |
|-------------------|---|--------------------------------------|---|
| 11 (50/0%) | 1323 (43/6%) | 582 (44/0%) | 48/2% |
| 3 (13/6%) | 458 (15/1%) | 200 (43/7%) | 57/7% |
| 2 (9/1%) | 247 (8/1%) | 70 (28/3%) | 2/7% |
| 2 (9/1%) | 535 (17/7%) | 243 (45/2%) | 17/7% |
| 4 (18/2%) | 470 (15/5%) | 255 (54/2%) | 33/6% |

**Figure 1. Dissociation of Studies Performed Pre- and After 2007**

therefore these results are statistically significant. This indicates implementation of the ASCO/CAP guideline by researchers.

Discussion

In order to increase the survival rate, trastuzumab, a monoclonal antibody, is administered to patients in whom

the HER-2/neu gene is overexpressed or abnormally amplified. In addition to the direct costs associated with this drug, indirect costs accrue through the triggering of acute cardiomyopathy. The role of the HER-2 receptor has been acknowledged by pathobiologists since the late 1970s. In the early 1990s, Slamon and colleagues conducted some studies on this receptor. They discovered that alterations of this receptor in breast cancer indicated poor prognosis. On the other hand, the use of trastuzumab leads to improved disease-free survival and overall survival rates (Smith et al., 2007).

For evaluating HER-2 receptor alterations in breast cancer, diagnostic tests such as IHC, ISH and q-PCR have been proposed. The FDA has granted approval for IHC and FISH testing. Although FISH provides greater precision than IHC, it is not considered as the standard testing method (Barberis et al., 2008; Jeremy et al., 2009). IHC HER-2 testing is regarded as being the most cost-effective because it is not only feasible in most pathology laboratories but its reproducibility is also excellent. Although this test is easy and inexpensive, false-positive and false-negative results are considered its main disadvantages (Elkin et al., 2004; Gown et al., 2008).

The direct cost of trastuzumab therapy for one patient in the United States was estimated to be approximately \$70,000–110,000 annually in 2006 (Antonio et al., 2007). It should be emphasized that 15%–20% of diagnoses were considered as false-positives in that year. Thus in 2007, ASCO/CAP developed a guideline for the diagnosis of HER-2 by reviewing articles published over the previous 19 years. The aim of this guideline was to minimize undertreatment and overtreatment in HER-2-positive breast cancer (Antonio et al., 2007).

Similar to many other countries, before establishment of an infrastructure for IHC diagnostic tests, HER-2 testing was routinely performed in Iran. This led to fluctuations in results from different laboratories. Our study showed that the statistics reported from Iran varied widely: for instance, the rate of HER-2-positive cases varied from 23.3% to 81.0%. From the 22 studies included in this systematic review, the average rate of HER-2-positive breast cancer was 44.5%, significantly higher than the reported international values of 20%–30%.

The use of ISH in Iran has been limited due to economical and technical reasons and the shortage of personnel with required expertise, as validated by the small number of studies involving ISH. Ghaffari and colleagues reported the concordance in Iran between IHC and FISH testing for scores 2+ and 3+ as 27.9% and 61.5%, respectively (Ghaffari et al., 2011). This study was the only comparison performed between IHC and FISH in Iran. The sample size for scores 2+ and 3+ was 86 and 13 patients, respectively. According to the ASCO/CAP 2007 guideline, if the process of IHC testing from the time of biopsy to microscopic evaluation meets appropriate standards, concordance between FISH and IHC will be at least 95% (Gown et al., 2008). In our study, despite the small sample size, there was a significant discordance between IHC and FISH in contrast to findings from international statistics.

Since the explanation of IHC results is subjective

in nature, inter- and intra-observer variations exist worldwide. Kadivar and colleagues evaluated agreement between different observers for varying HER-2 scores in Iran and showed that, between positive and negative cases, the score was acceptable, but it was at an intermediate level between positive and weakly positive cases (Kadivar et al., 2009). In a study by Sa'ati and colleagues on 580 Iranian patients between 2000 and 2003, 38% of breast cancer cases were HER-2-positive. The authors of that research introduced their article with the following question: 'Is the high rate of HER-2-positive breast cancer in Iran true or false?' They discussed the proposal that, since ISH diagnostic methods had not been developed appropriately, weakly positive cases had been reported as HER-2-positive (Saatee et al., 2006). The studies of Ghaffari and Kadivar and their colleagues supported this theory. Therefore, the high rate of HER-2-positive cases in Iran may be the result of inadvertent error when grading weakly positive or equivocal cases as positive.

Some researchers, whose works have been included in this systematic study, acknowledge genetic differences in Iranian patients and the effect of environmental factors on the HER-2/neu gene as reasons for statistical differences between Iran and other parts of the world (Sirati et al., 2009; Shamsalinia et al., 2010). The effect of genetic factors on breast cancer is obvious. Mousavi and colleagues conducted a large epidemiological study on breast cancer in Iran and demonstrated that the incidence age of breast cancer in Iranian women was 40-49 years (30% in women <30 years) (Mousavi et al., 2007). Results from other studies indicate that the incidence age of breast cancer in Iran is lower than that in Western countries, and that the disease is more invasive (Harirchi et al., 2011). Results from India and Pakistan were similar to those from Iran; women in these countries showed an incidence pattern of breast cancer different to that in women from USA (Naeem et al., 2008; Kakarala., 2010). Data on the inheritance of the HER-2 gene are inconsistent, and it is not yet clear whether mutation of this gene can occur in germinal cells (Espinosa et al., 2003; Montgomery et al., 2003). However, the different pattern of breast cancer in Iran in comparison with Western countries suggests genetic differences (Fattahi et al., 2009).

Despite discussions, some Iranian researchers who found the rate of HER-2-positive breast cancer higher in Iran than that from international statistics have proposed the probable role of genetic factors (Sirati et al., 2009; Shamsalinia et al., 2010). Nevertheless, these claims must be evidence-based and complementary tests should be performed to corroborate them. Variation in the HER-2-positive rate among Iranian provinces is striking. As shown in Table 2, the difference in one area was 57.7% and in another it was 48.2%. Nonetheless, because of low sample size and reproducibility, it is impossible to evaluate the role of genetic and environmental factors, but marked variation in results within a region raises questions about the validity of IHC as opposed to genetic variation. We can begin to discuss the role of genetic and environmental factors once the validity and reliability of IHC testing have been proved.

This systematic review reveals that the criterion for score 3+ of >10% reactivity in studies performed post 2007 can lead to an increased incidence of false-positive results.

There is no doubt that evaluation of the IHC HER-2 testing is an international issue. Several guidelines have been developed at both national and regional levels since the 1990s, the aim of which is the development of good laboratory practice. At present, the ASCO/CAP 2007 guideline is considered the main worldwide reference, and various countries are aiming to enhance the validity of results by improvement in laboratory techniques. For example, because of frequent errors by Spanish pathologists in the evaluation of HER-2 testing results, in 2009 the Spanish Society of Pathologists (SEAP) and Spanish Society of Medical Oncologists (SEOM) developed a consensus-based guideline (the 'SEAP/SEOM' guideline) to improve the quality of HER-2 testing (Albanell et al., 2009). In Malaysia, a clinical practice guideline group recommended that all 2+ and 3+ scores be approved by FISH (Academy of Medical of Malaysia., 2010). Because of the high rate of HER-2-positive tests in Iran, in order to decrease the rate of false-positive and false-negative results, standardization of the HER-2 IHC testing in association with the development of ISH techniques appears essential.

Countries with the same level of resources as Iran, such as Saudi Arabia, Egypt and Taiwan, have shown the same variations in the HER-2-positive breast cancer rate (El-A Helal et al., 2000; Kuo et al., 2007; Arafah., 2012). Taken together, we maintain that standardization is the first step toward improvement in IHC diagnosis by the HER-2 biomarker in breast cancer in Iran. In order to meet these standards, harmonization of all IHC stages, establishment of controls, elevation of laboratory quality, development of the ISH technique and training of staff must be taken into consideration.

As of January 2011, the Health Ministry of Iran in collaboration with Iranian pathology, medical oncology and cancer societies have made great strides in improving this diagnostic technique and have established an electronic breast cancer diagnostic system in drugstores and laboratories (Mechanized system of breast cancer management Iran ., 2011). Since it is possible that this test may also be used in other forms of cancer such as gastric adenocarcinoma, standardization of the IHC HER-2 testing appears essential. This will also help in the identification of patients eligible for trastuzumab therapy and will reduce both the direct and indirect costs of trastuzumab to public health systems.

This systematic review was unable to cover all aspects of IHC HER-2 testing in Iran, because such a study would require a larger sample size and IHC results must be controlled by FISH. The lack of access to such data was the major limitation of this study. In addition, in some studies performed in developed countries, IHC HER-2 testing results have been evaluated both within and between laboratories, and the fact that such data are lacking for Iran can be readily considered as another limitation of our study

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References

- Academy of Medical of Malaysia (2010). Clinical Practice Guidelines (CPGs). *Management Breast Cancer*. <http://www.moh.gov.my/cpgs>.
- Afshar Moghaddam N, Mahsoni P, Rajabi P, Pooyan Tabibi A (2008). Correlation of Her-2/Neu and Tp53 Expressions with Clinicopathologic Characteristics in Infiltrative Ductal Breast Carcinomas. *Iranian J Pathology*, **3**, 75-80
- Albanell J, Andreu X, et al (2009). Guidelines for HER2 testing in breast cancer: a national consensus of the Spanish Society of Pathology (SEAP) and the Spanish Society of Medical Oncology (SEOM). *Clin Transl Oncol*, **11**, 363-75
- Alizadeh SA, Moshfeghi K, Kalantari M, Ebrahimi Kh (2010). Lymph-node involvement and tumor markers in patients with breast Cancer. *Arak Med University J*, **12**, 44-50
- Amouzegar Hashemi F, Alaleh A, Kazemian A, Haddad P (2010). Local recurrence after breast conserving surgery in breast cancer: a study in Cancer Institute, Tehran. *Tehran University Med J*, **67**, 863-9
- Antonio C Wolff, M Elizabeth H Hammond, et al (2007). American society of clinical oncology/college of american pathologists guideline recommendations for human epidermal growth factor receptor 2 testing in breast Cancer. *J Clin Oncol*, **25**, 118-45
- Arafah M (2012). Correlation of hormone receptors with her 2/neu protein expression and the histological grade in invasive breast cancers in a cohort of Saudi Arabia. *Turk Patoloji Derg*, **28**, 38-43
- Attarian H, Pirzadeh A, Rezvani H, et al (2011). Clinicopathologic manifestations of Iranian patients with breast cancer. *Pak J Med Sci*, **27**, 182-5
- Barberis M, Pellegrini C, Cannone M, et al (2008). Quantitative PCR and HER2 testing in breast cancer a technical and cost-effectiveness analysis. *Am J Clin Pathol*, **129**, 563-70
- Carlson RW, Moench SJ, **One author** et al (2006). HER2 testing in breast cancer: NCCN task force report and recommendations. *J Natl Compr Canc Netw*, **4**, 1-22
- Chandarlapaty S, Modi Sh, Sloan-Kettering M (2011). Targeted therapy for human epidermal growth factor receptor 2-positive breast cancer: can there be too many active drugs? *J Clin Oncol*, **10**;29(23):3111-Ahn ER,
- Dendukuri N, Khetani K, McIsaac M, Brophy J (2007). Testing for HER2- positive breast cancer: a systematic review and cost-effectiveness analysis. *CMAJ*, **176**, 1429-33
- El-A Helal T, Khalifa A, Kamel AS (2000). Immunohistochemical expression of p53 and c-erbB2 proteins in breast cancer in Egypt. *Anticancer Res*, **20**, 2145-50
- Elkin EB, Weinstein MC, Winer EP, et al (2004). HER-2 testing and Trastuzumab therapy for metastatic breast cancer: A cost-effectiveness analysis. *J Clin Oncol*, **22**, 854-63
- Ensani F, Kargar M, Davanloo M, Irvanloo G, Khazaiepour Z (2010). Comparison between Immunohistochemistry and Chromogenic in Situ Hybridization Method in Assessment of HER-2 Status in Breast Cancer. *Iranian J Surgery*, **7**, 8-16
- Espinosa AB, Tabernero MD, García-Macías MC, et al (2003). Her-2/neu Gene Amplification in Familial vs Sporadic Breast Cancer. *Am J Clin Pathol*, **120**, 917-27
- Fattahi MG, Mojtahedi Z, Karimaghaee N, et al (2009). Analysis of BRCA1 and BRCA2 mutations in Southern Iranian breast cancer patients. *Arch Iran Med*, **12**, 584-7
- Ghaffari SR, Sabokbar T, Dastan J, Rafati M, Moossavi Sh (2011). Her 2 Amplification Status in Iranian Breast Cancer Patients: Comparison of Immunohistochemistry (IHC) and Fluorescence in situ Hybridisation (FISH). *Asian Pac J Prev*, **12**, 1031-4.
- Gown AM, Goldstein LC, Barry TS, et al (2008). High concordance between immunohistochemistry and fluorescence in situ hybridization testing for HER2 status in breast cancer requires a normalized IHC scoring system. *Mod Pathol*, **21**, 1271-7
- Gown AM (2008). Current issues in ER and HER2 testing by IHC in Breast Cancer. *Mod Pathol*, **21**, 8-15
- Halimi M, Aghbali A, Tabrizi D, Olad Sahebmadarek E (2011). Expression of epidermal Growth Factor Receptor Tyrosine Kinase Family in Fine Needle Aspiration and permanent Specimens of Invasive Lobular and Ductal Breast cancer. *Pak J Biol Sci*, **14**, 584-9
- Harirchi I, Kolahdoozan S, Karbakhsh M, et al (2011). Twenty years of breast cancer in Iran: downstaging without a formal screening program. *Ann Oncology*, **22**, 93-7
- Homaei-shandiz F, Ghavam naseri MR, Sharifi N, et al (2006). Evaluation of the Relationship Between Human Epidermal Growth factor receptore-2/neu (c-erbB-2) Amplification and pathologic Grading in Patterns with Breast Cancer. *Saudi Med J*, **27**, 1810-4
- Hudis CA, MD (2007). Trastuzumab - Mechanism of Action and Use in Clinical Practice. *N engl J med*, **357**, 39-51
- Jeremy St. J. Thomas (2009). Quality Assurance in Her-2 Testing-Redefining the Gold Standard. *Clin Chem*, **55**, 1265-7
- Kadivar M, Askarzadeh P, Shayanfar N, Sadeghipour AR, Bahraminejad M (2009). Interpretation of immunohistochemistry results of HER2/neu expression in invasive breast cancer: evaluation of Interobserver and Intraobserver Agreement. *Razi J Medical Science*, **16**, 147-55
- Kadivar M, Esmaili A, Joolae A (2010). evaluation of microvessel density and vascular endothelial growth factor in invasive breast carcinoma via immunohistochemistry and their correlation with clinicopathologic parameters. *Razi J Medical Sci*, **16**, 46-56
- Kadivar M, Rezaee M, Jadidfard R, Joolae A (2010). Evaluation of histopathology and biologic markers in premenopausal (under 40 years) and postmenopausal (over 60 years) women with breast cancer in hazrat-e-rasoul and atieh hospitals. *Razi J Medical Sci*, **17**, 49-57
- Kakarala M, Rozek L, Cote M, Liyanage S, Brenner DE (2010). Breast cancer histology and receptor status characterization in Asian Indian and Pakistani women in the U.S. a SEER analysis. Kakarala et al. *BMC Cancer*, **10**, 2-8
- Kuo SJ, Wang BB, Chang CS et al (2007). Comparison of immunohistochemical and fluorescence in situ hybridization assessment for HER-2/NEU status in Taiwanese breast cancer patients. *Taiwan J Obstet Gynecol*, **46**, 146-53
- Mauri D, Polyzos NP, Salanti G, Pavlidis N, Ioannidis JP (2008). Multiple-treatments meta-analysis of chemotherapy and targeted therapies in advanced breast cancer. *J Natl Cancer Inst*, **100**, 1780 -91
- Mechanized system of breast cancer management Iran (2011). <http://bcancer.ir>. (Persian)
- Mirmalek S.A, Tirgary F, Alizadeh HR (2005). Study and comparison between malignant breast tumors and related surrounding benign tissue about HER-2 Receptor Persistence. *Iranian J surgery*, **13**
- Mirzaei HR, Sabahat A, Nasrollahi F, Mohammadi-Yeganeh L (2010). Correlation between ER, PR, HER2 receptors and prognostic factors in breast cancer. *Pejouhandeh*, **15**, 152-6
- Mofid B, Jalali NMR, Kian L, Zayeri F (2004). Association of

- HER2 With prognostic factors of breast cancer. *J the Faculty of Med*, **28**, 29-32
- Moghadaszadeh M, Nikanfar AR, Hemati MR, Gharibi B, Sardashti S (2010). Interaction between Her-2 and Hormone Receptor in Breast Cancer. *International J Hematology Oncology and Stem cells Res*, **4**, 14-16
- Mohammadzadeh F, Ghasemibasir HR, Rajabi P, et al (2009). Correlation of E-cadherin expression and routine immunohistochemistry panel in breast invasive ductal carcinoma. *Cancer Biomark*, **5**, 1-8
- Montgomery KG, Gertig DM, Baxter SW, et al (2003). The HER2 I655V Polymorphism and Risk of Breast Cancer in Women _ Age 40 Years. *Cancer Epidemiology, Biomarkers and Prevention*, **12**, 1109-11
- Moore S (2012). Heart failure in a breast cancer survivor. *Oncol Nurs Forum*, **39**, 147-50 .
- Moradi-Marjaneh M, Homaei-Shandiz F, Shamsian SAA, Eftekhar-Zadeh Mashhadi I, Hedayati-Moghadam MR (2008). Correlation of HER2/neu Over Expression, p53 Protein Accumulation and Steroid Receptor Status with Tumor Characteristics: An Iranian Study of Breast Cancer Patients. *Iranian J Publ Hlth*, **37**, 19-28
- Mousavi SM, Montazeri A, Mohagheghi MA et al (2007). Breast cancer in iran: an epidemiological review. *Breast J*, **13**, 383-91
- Naeem M, Nadim Khan, Aman Z et al (2008). Pattern of breast cancer: experience at lady reading hospital, peshawar. *J Ayub Med Coll Abbottabad*. **20**, 22-5
- Naghshvar F, Torabizade ZH, Emadian O, Zareh A, Ghahramani M (2007). Status of estrogen, progesterone receptors and HER-2/neu expression in invasive breast cancer. *J Gorgan university Medical Sci*, **8**, 65-68
- Pourzand A, Fakhree MBA, Hashemzadeh Sh, Halimi M, Daryani A (2011). Hormone Receptor Status in Breast Cancer and its Relation to Age and Other Prognostic Factors. *Breast Cancer*, **5**, 87-92
- Rabiee Hashemi SM, Rabiee Hashemi S (2008). HER-2, P53 and hormonal receptors protein expression as predictive factors in breast cancer prognosis. *Chin J Clin Oncol*, **5**, 413-7
- Saatei S, Afrakhteh M, Sadrolhefazi B (2006). High Prevalence of HER-2/ neu overexpression among Iranian women with breast cancer: True or False? *Breast J*, **12**, 499-501
- Salimi M, Mozdarani H, Majidzadeh K (2012). Expression pattern of ATM and cyclin D1 in ductal carcinoma, normal adjacent and normal breast tissues of Iranian breast cancer patients. *Med Oncol*, **29**, 1502-9
- Shahriari Ahmadi A, Ghavamzadeh A, Amiri N et al (2005). Clinical, biological and pathological characteristics of breast cancer patients at the taleghani university hospital in kermanshah, Iran. *IJHOBMT*, **2**, 6-11
- Shamsalinia A, Keihanian Sh, Ghaffari F, Mansore Saravi M, Najafi Ashtian M (2010). Association between expression of her-2/neu gene and estrogen receptors, progesterone receptors and p53 gene among female with primary breast cancer. *The Iranian J Obstetrics, Gynecology and Infertility*, **13**, 43-48
- Sirati F, Ghahhari.A, Alavi N (2009). Determination of the Prevalence of Prognostic factors(HER-2/neu, P53, PR, ER) in breast cancer and assessment of their correlation with each others and age and menopause state of patients. *Iranian J of breast disease*, **1**, 24-31
- Smith I, Procter M, et al (2007). 2-year follow-up of trastuzumab after adjuvant chemotherapy in HER2-positive breast cancer: a randomised controlled trial. *Lancet*, **396**, 29-36
- Vogel CL (2012). Dual HER2-targeted approaches in HER2-positive breast cancer. *Breast Cancer Res Treat*, **131**, 371-83
- World Bank (2010). Gross domestic product World Development Indicators database. <http://www.worldbank.org>.; Accessed 1 July 2011
- Yip CH, Smith RA, et al (2008). Guideline implementation for breast healthcare in low- and middle-income countries. *Cancer*, **113**, 2221-43