Impact of Triple Negative Phenotype on Prognosis and Early Onset of Breast Cancer in Iranian Females

Article *in* Asian Pacific journal of cancer prevention: APJCP · January 2011

Source: PubMed

6 authors, including:

Siamak Salami
Shahid Beheshti University of Medical Sciences
49 PUBLICATIONS **571** CITATIONS

SEE PROFILE

RESEARCH COMMUNICATION

Impact of Triple Negative Phenotype on Prognosis and Early Onset of Breast Cancer in Iranian Females

Siamak Salami[1*], Fatemeh Ramezani[1], Toofan Aghazadeh[2], Hossein Afshin-Alavi[3], Behrouz Ilkhanizadeh[4], Davood Maleki[5]

Abstract

Background: Breast cancer in Iranian women occurs about a decade earlier than in Western countries. This study sought to evaluate the impact of triple negative phenotype on early onset of ductal cell breast cancer and its prognosis in Iranian females. Methods: Estrogen and progesterone receptors, Her-2 overexpression and nuclear accumulation of P53 were assessed in sixty surgically resected formalin-fixed paraffin embedded breast invasive ductal carcinomas. They were divided into triple negative and non triple negative phenotypes and impact of the phenotypes were evaluated on prognostic factors of all patients and based on menopausal status. Results: The result showed that the mean age of patients with triple negative breast tumors, especially in postmenopausal group, was significantly lower than with non triple negative phenotypes. Although the latter was significantly associated with higher histological grade, it also showed a significant correlation with smaller size of tumor and a lower rate of axillary lymph node metastasis in young patients. Conclusion: The higher rate of breast cancer with triple negative phenotype in Iranian females is a feasible reason for the reported lower mean age of breast cancers. In premenopausal patients, triple negative phenotype reveals a positive impact on prognostic factors, but it is associated with a poorer prognosis in postmenopausal patients. Hence, a distinct ethnic profile of triple negative phenotype in Iranian females is suggested.

Keywords: Triple negative phenotype - breast cancer - Iranian females - prognostic factors

Asian Pacific J Cancer Prev, **12**, 719-724

Introduction

Breast cancer is still the most common cancer of females in western countries but over the past thirty years, a trend of increasing incidence and mortality from breast cancer in lower resource countries was observed. It has been reported that of the over million newly diagnosed cases of breast cancer in 2009, low- and middle-resource countries will be burdened with 45% of breast cancer cases and 55% of breast cancer-related deaths(Bray and Moller, 2006; Porter, 2009). Higher rate of breast cancer and its related death in developing or underdeveloped Asian countries was also reported (Hirabayashi, 2009).

Although Iran is classified as country with low breast cancer rate, it is the first common cancer in Iranian females (Sadjadi et al., 2005) with 31.4 cancer age-standardized rates (ASR) and 23.16 crude incidence rate per 100,000 women (Mousavi et al., 2007; Mohagheghi et al., 2009).

In spite of lower rate of breast cancer in Iran and beside its increasing rate, breast cancer mainly affects Iranian women about a decade earlier than Western countries (Harirchi et al., 2000; Mousavi et al., 2007; Kolahdoozan et al., 2010). The mean age of the patients at diagnosis was reported as 47.2 (SD = 13.5)- 51.3 (SD=12.5) (Mousavi et al., 2006; Vahdaninia and Montazeri, 2004). It has been shown that more than 36% of the tumors occur in women under 40 years old and breast cancer is a high burden in the community (Mousavi et al., 2006). The overall five year survival rate of breast cancer in Iranian patients was 75 % and lesser one-year breast cancer survival rate was reported than Canadians in British Columbia (Sadjadi et al., 2009). Most of the patients have locally advanced disease (Harirchi et al., 2000).

Prognostic factors are diverse tumor or patient related factors which help clinicians to make a clinical decision and to select the appropriate treatment for individual patients needs. They not only allow comparisons between groups of patients at similar risks of recurrence or death, but also provide an understanding of the key mechanisms of metastasis as a central point of breast cancer biology (Bundred, 2001). Tumor size, nodal status and grade are the most important prognostic factors for long term, 5- or 10-year breast cancer survivors (Thorat, 2007; Soerjomataram et al., 2008). Measurement of Estrogen and Progesterone receptors (ER and PgR), has been strongly recommended for every primary invasive breast cancer by American Society of Clinical Oncology (ASCO) and they may also be measured on metastatic lesions if the results would influence treatment planning. Steroid hormone receptor status should be used in both pre- and

*[1]Molecular Research Lab, Department of Clinical Biochemistry and Nutrition, [4]Department of Pathology, [5]Department of Hematology and Oncology, Faculty of Medicine, Urmia University of Medical Sciences, Urmia, [2]Department of Biology, Sciences and Research Campus, Azad Islamic University, [3]Department of Pathology, Day Hospital, Tehran, Iran *For correspondence : salami_si@umsu.ac.ir, salami.si@gmail.com*

postmenopausal patients, to identify patients most likely to benefit from endocrine therapy in both the primary or metastatic breast cancer (Harris et al., 2007).

Amplification of chromosome 17 located proto-oncogene, known as human epidermal growth factor receptor 2 (HER2) or C-erb B2, was reported in approximately 15%-30% of breast cancers breast cancer (Schmitt, 2009). HER-2/neu encodes a trans-membrane glycoprotein with tyrosine kinase activity which plays a role of double edge sword; Over-expression of HER-2/neu is a good predictor of response to trastuzumab (Herceptin), but it is also an independent negative predictor of overall survival and time to relapse in patients with lymph-node-positive breast cancer (Hung and Lau, 1999; Schmitt, 2009). According to the last updated ASCO guideline, Her2 expression and/or amplification should be evaluated in every primary invasive breast cancer either at the time of diagnosis or at the time of recurrence, principally to guide selection of trastuzumab in the adjuvant and/or metastatic setting (Harris et al., 2007).

Based on results of immunohistochemical staining, triple negative breast cancer is a term that has been used to define a biologically diverse group of breast tumors that lack expression of ER, PR, and HER-2 and represents about 15% of all breast cancers (Seal and Chia, ; Viale and Bottiglieri, 2009). Since, they are a particularly difficult to treat and biologically aggressive disease with limited treatment options, optimization of chemotherapy regimens may be key in treating triple negative breast cancer (Conte and Guarneri, 2009).

The current study was done to evaluate the impact of triple negative phenotype on clinicopathological factors of the breast cancer in premenopausal and postmenopausal Iranian females. In view of the fact that infiltrating ductal cell carcinoma (IDCC) was the most common histological type of breast cancer in Iran(Mousavi et al., 2008), the tumors with clear cut diagnosis of IDCC was only included.

Materials and Methods

Tissue samples and study design:

Sixty surgically resected formalin fixed paraffin embedded breast tissues from patients with confirmed invasive ductal carcinoma between 2006 and 2008 were enrolled. This study was approved by the institutional review board and board of medical ethics. At least 4 slides of each tumor were stained using Hematoxylin and Eosin (HE) staining protocol and all tumors with clear cut diagnosis of Invasive Ductal Carcinoma were selected by two expert pathologists independently.

Clinicopathological features of the patients were collected by the retrospective review of medical records. Thirty samples that lack expression of ER, PR, and HER-2 were considered as "Triple Negative" (TN), and remaining samples were named "Non Triple Negative" (NTN). Meanwhile, the samples were divided to premenopausal (>50 years old) and postmenopausal (≥50 years old) groups and patients' and tumors' related prognostic markers of triple negative phenotype were compared to non triple negative ones in each group separately.

Histopathological analysis

Tissue samples from patients were obtained during mastectomy or lumpectomy. Tumor samples were then dissected and block-sized pieces were fixed in 4% paraformaldehyde for 24 h and stored in 70% ethanol at 4oC. Following fixation, samples were trimmed to 3-5 mm in thickness and embedded in paraffin to prepare 5 micron thick sections for haematoxylin and eosin (H & E) staining.

Scoring

Allred scoring method and cutoff was used as a clinically validated scoring method for both of ER and PgR and cases were considered positive even if 1% or more of tumor cells showed staining. The HER2-score was based on a 1 to 3 scale: 1 or negative corresponded to tumor cells that were completely negative or faint perceptible staining of less than 10% of the tumor cell membranes; 2+ or suspicious corresponded to weak-moderate staining of the entire tumor cell membranes in more than of 10% of tumor cells; and 3+ indicated strong circumferential staining of the entire tumor cell membranes creating a fishnet pattern in more than of 10% of tumor cells.

For p53 protein, tissue samples were scored on the basis of the intensity of the specific nuclear staining and 10% nuclear staining was assigned as cutoff point. Three slides were stained for each sample and scoring was performed by two different expert observers, independently.

Malignancy grading

Based on equal importance of three tumor features, i.e. tubule formation, nuclear pleomorphism and mitotic count, the malignancy grade of tumor samples were determined by modified version of the Bloom and Richardson grading system and three prognostic categories were assigned as: low risk (I), intermediate risk (II) and high risk (III).

Immunohistochemical analysis (IHC)

Level of Nuclear p53 protein and hormone receptors were evaluated in tissue samples using IHC analysis. After mounting of 5 μm sections on slides, they were deparaffinized in xylene and rehydrated in graded ethanol solutions. Subsequently, they were incubated in citrate buffer (pH=6.0) for 5 min and microwave based antigen retrieval technique (700 W, 2 min and 360 W, 10 min) was applied. For blocking endogenous peroxidase activity, sections were then treated with 3% hydrogen peroxide for 15 min. After pre-incubation in 1% BSA in Tris buffer (TBS; 25 mM Tris-HCl, pH 7.4, 137 mM NaCl, 2.7 mM KCl), sections were incubated for1 hour at room temperature with monoclonal mouse anti-human p53 protein, estrogen receptor and progesterone receptor (DO-7, 1D5 and PgR636 clone; Dako, Denmark) correspondingly, and rinsed three times in TBS. AO485 as an universally approved antibody was used for immunostaining of HER-2. For p53, DO-7 antibody which recognizes an epitope at the N-terminal of the human p53 protein and reacts with wild type as

Table 1. Clinicopathological Characteristics of Patients

Patients	Prognostic Factors		Triple Negative	Non Triple Negative	P value
All patients	Numbers		30	30	
	Age (mean ± SD)		44.9±9.06	50.8±12.07	0.03*
	Side of Involvement %	Right	40.0	40.7	0.95
		Left	60.0	59.3	
	Lymph node involvement %		51.9	75.9	0.06
	Tumor size (mean ± SD)		3.15±1.29	3.72±2.11	0.21
	Tumor grade %	I	0.00	18.5	
		II	16.7	74.1	0.001**
		III	83.3	7.40	
Pre menopausal	Numbers		22	15	
<50 years	Age (mean ± SD)		41.1±7.14	42.47±5.97	0.40
	Side of Involvement %	Right	31.8	33.3	0.92
		Left	68.2	66.7	
	Lymph node involvement %		45.0	85.7	0.03*
	Tumor size (mean ± SD)		3.21±1.42	4.61±2.49	0.03*
	Tumor grade %	I	0.00	20.0	
		II	9.10	80.0	0.001**
		III	90.9	0.00	
Post menopausal	Numbers		8	15	
≥50 years	Age (mean ± SD)		52.2±4.55	61.2±10.7	0.03*
	Side of Involvement %	Right	62.5	50.0	0.58
		Left	37.5	50.0	
	Lymph node involvement%		71.4	66.7	0.82
	Tumor size (mean ± SD)		3.00±0.88	2.83±1.13	0.71
	Tumor grade %	I	0.00	16.7	
		II	37.5	66.70	0.08
		III	62.5	16.7	

well as the mutant type of the p53 protein, was used. For the HRP method, the sections were incubated with the HRP-labeled secondary antibodies (DAKO, Denmark; dilution 1:100) for 30 min at RT. Slides were incubated with 3,3'-diaminobenzidine (DAB) (Dako DAB tablet dissolved in deionized water; Dako, Denmark) for 5 min at room temperature after three times washing steps with TBS. Positive control slides for p53 protein (Human Squamous Cell Carcinoma) were obtained from Dako (T1076; Dako-Denmark) and used to show the quality and specificity of the DO-7 antibody. In each series, a non primary antibody incubated slide was also used as negative control.

Statistical methods

The impact of triple negative phenotype on prognostic factors like as malignancy grade, lymph node involvement, tumor size and side of involvement in premenopausal and post menopausal patients were tested by means of valid statistical tests and P<0.05 was considered as statistically significant difference.

Results

Age

The age of all patients whose tumor samples were included in this study was ranged from 28-82 with mean age of 47.9 years (± 11.0). The median age was 48 years. A significant difference was found between mean age of patients with triple negative and non triple negative tumors (p=0.03). While, the age of patients with triple negative tumors was ranged from 28-64 years with mean age of 44.9 years (± 9.06), it was ranged from 30-82 years with mean age of 50.8 (± 12.1) (Table 1, Figure 1).

The mean age of premenopausal patients with triple negative tumors (41.2±7.14) and non triple negative tumors (42.5±5.97) was not significantly different (p=0.4). However, the mean age of postmenopausal patients with

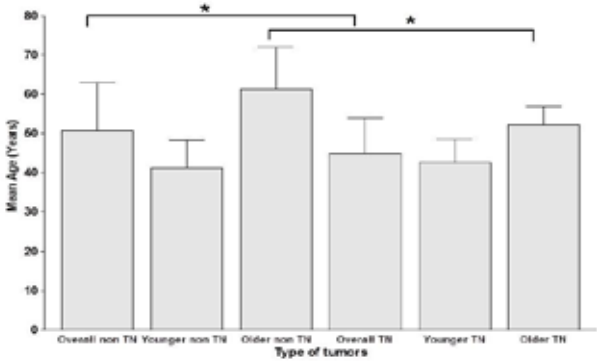

Figure 1. Comparison of Mean Age of Patients with Triple Negative with Non Triple Negative Tumors

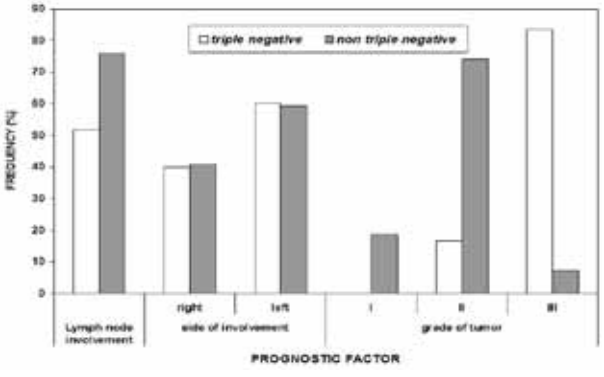

Figure 2. Status of Prognostic Factors in Patients with Triple Negative and Non Triple Negative Tumors

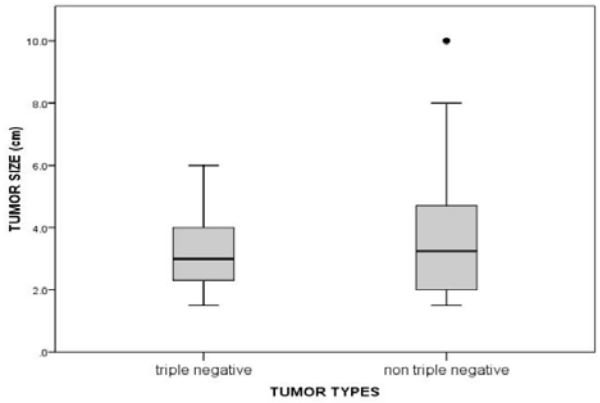

Figure 3. Comparison of Tumor Size in Patients with Triple Negative and Non Triple Negative Tumors

triple negative tumors (52.3±4.55) was significantly lower than with non triple negative tumors (61.2±10.8) (p=0.03).

Side of involvement:

Compared to right breast cancer, 59.6% of patients had left side involvement. Not statistically significant difference was found in side of involvement between patients with triple negative tumors and non triple negative ones (p=0.95) (Table 1).

Although the involvement of left breast in the premenopausal patients with triple negative or non triple negative tumors was higher than right side, a positive but not significant trend of right breast cancer, particularly with triple negative phenotype, was found in postmenopausal patient (p=0.12).

Involvement of Axillary lymph nodes:

Axillary lymph nodes were involved in 64.3% of patients. Frequency of lymph node involvement in patients with non triple negative tumors was higher than patients with non triple negative tumors (51.9% vs. 75.9%) (p=0.06). The difference between prevalence of Axillary lymph nodes involvement in premenopausal patients with triple negative phenotype, is significantly lower than non triple negative phenotypes (45% vs. 85.7%) (p=0.03)(Table 1, Figure 2). The frequency of lymph node involvement in postmenopausal patients with triple negative tumors was higher than non triple negative tumors but it was not statistically significant (71.4% vs. 66.7%) (p=0.82).

Tumor size

Size of tumors were measured based on their longest length and they were divided to small (≤2 cm), medium (>2cm and ≤4cm) and large (>4cm) tumors. 49.2 % of tumors were medium size. The mean size of tumors was 3.43±1.75 centimeter. Not significant difference was found between size of triple negative and non triple negative tumors (3.15±1.29, 3.72±2.11 respectively) (p=0.21).

Although the mean size of tumor in premenopausal patients with non triple negative tumors was significantly larger than postmenopausal patients (4.61±2.49 vs. 2.83±1.13) (p=0.02), no significant differences was found in mean tumor size in premenopausal and postmenopausal patients with triple negative phenotype (3.21±1.42 vs. 3.00±0.88) (p=0.7) (Table 1, Figure 3).

Grade

Using modified version of the Bloom and Richardson grading system, the malignancy grade of tumors were defined as a low grade (I), intermediate grade (II) and high grade (III). Their prevalence was 8.8%, 43.9% and 47.4%, respectively, with a difference was found between triple negative and non triple negative tumors (p=0.001).

In contrast to higher frequency of grade II tumors in patients with non triple negative tumors, grade III tumors were the most common type in the patients with triple negative phenotype. A similar pattern was found in premenopausal patients (p=0.001) but in spite of similar trend, not significant difference was found for postmenopausal patients (p=0.08).

Immunohistochemistry:

Slides with more than 80% malignant cells were used for immunohistochemical assessment. Inter- and intra-observer reproducibility in the evaluation of all immunostaining methods were at least 95 percent, indicating reliability.

Hormone receptors:

Specific receptors were detected in 31 and 41.4% of tumors for estrogen and progesterone, respectively. An intimate association were found between estrogen and progesterone receptors (p=0.01). ER positive tumors in right breast involvement and PR positive tumors in left breast involvement were more frequent, but the differences are not significant (p>0.5).

Hormone receptor negative tumors (ER-, PR-) in left breast involvement were significantly higher than right breast involvement (p=0.042) and side of involvement strongly predicts hormone receptor status (R2=0.934, p=0.009).

Involvement of lymph node and larger tumor size were also found as strong predictors of negative ER-PR status of breast tumors (R2=0.934, p=0.002). Level of hormone receptors in tumors with higher grade was lower than well differentiated low grade tumors but significant correlation or prediction was not found between ER status with grade (p>0.5). Meanwhile, a significant difference was found for progesterone receptor only (p=0.045). No significant difference was found between hormone receptor status in the patients younger or older than 50 years old. Assessment of hormone receptors in tumors with different size was not revealed a significant correlation but less hormone receptors positivity was found in larger tumors. With regard to hormone receptor status and grade of tumors, higher grade tumors are tend to be hormone receptor negative but the difference was not statistically significant.

Over expression of C-erb B2 (Her 2)

Strong circumferential staining of the entire tumor cell membranes creating a fishnet pattern was found in 44.6% of tumors. No significant correlation were found between Her 2 over expression and side of involvement, lymph node metastasis or tumor grade.

Nuclear accumulation of P53

Nuclear accumulation of p53 was found in 31% of cases. In comparison to right breast involvement, more P53 positive tumors was found in the left breast but the difference in this case did not reach statistical significance (p>0.1).

Correlation among clinicopathological factors:

Significant correlation was found between tumor size with age at diagnosis and menstruation status (p=0.016, p=0.018). Breast tumors in younger and premenopausal patients are smaller than elder postmenopausal females.

Significant correlation was also found between PgR with patient age, menstruation status and grade of tumor (p=0.05 and p=0.01, respectively). It means that breast tumors in elder and postmenopausal females are tending to be steroid receptors positive.

Accumulation of p53 protein:

An inverse significant correlation was also found between p53 mutation and steroid hormone receptors, ER and PgR (p=0.02and p=0.04, respectively), hence risk of poor prognosis in the patient with coincident nuclear accumulation of p53 and lack of steroid hormone receptors are evidently greater than the patients with either (OR=1.75, 95% CI=1.11- 2.75)

Discussion

As it aforementioned, breast cancer in Iranian women occurs about a decade earlier than Western countries (Harirchi et al., 2000; Kolahdoozan et al., 2010; Mousavi et al., 2007). The mean age of the patients at diagnosis was reported as 47.2 ±13.5 - 51.3±12.5 (Vahdaninia and Montazeri, 2004; Mousavi et al., 2006).

In the current study, the mean age of all patients was 47.9±10.98 years which is consistent with the reported results and proves the early onset of breast cancer in Iranian females. Meanwhile, the mean age of the patients with triple negative phenotype was significantly lower than non triple negative ones. While the mean age of the premenopausal patient (>50 years old) was not revealed a significant difference between triple and non triple negative tumors, it was significantly lower in the postmenopausal patients (≥50 years old) with triple negative phenotype than non triple negative cases.

Higher rate of triple negative breast cancer in premenopausal females was reported in china (Zhou and Liu, 2010). In contrast to high premenopausal population of triple negative ductal cell carcinoma, a high proportion of postmenopausal patients with triple negative breast squamous cell carcinoma, spindle cell carcinoma, and metaplastic carcinoma with bone/cartilage metaplasia was reported(Iwase et al., 2010). We found that the mean size of tumor in premenopausal patients with non triple negative tumors are significantly bigger than postmenopausal patients (4.61±2.49 vs. 2.83±1.13) but no significant differences was found in premenopausal and postmenopausal patients with triple negative phenotype (3.21±1.42 vs. 3.00±0.88). Although not significant differences was found between triple negative and non triple negative breast tumors but triple negative tumors in postmenopausal patients were larger than non triple negative tumors and less hormone receptors positivity was found in larger tumors. A positive relation between tumor size and triple negative phenotype was reported (Dent et al., 2007; Rakha et al., 2007; Zhou and Liu, 2010).

Results of current study revealed that the grade III tumors were the most common type in the Iranian patients with triple negative phenotype which is significantly different from non triple negative tumors. Similar resulted were also reported in other countries (Dent et al., 2007; Rakha et al., 2007).

It has been reported that the axillary nodes metastasis rate in triple negative breast tumors are lower than non triple negative tumors (Zhou and Liu, 2010). A Similar and significantly lower rate of lymph node involvement in patients with triple negative breast tumors was found in the current study. Whereas the lack of lymph node involvement is considered as a marker of good prognosis in non triple negative breast cancers (Kwon et al., 2010), in lymph node-negative patients, triple negative disease was associated with poorer five-year breast cancer-specific survival (Lai et al., 2010).

We found that triple negative breast cancers in Iranian females occur in narrower range of age with lower mean and median than non triple negative breast cancers and consequently higher frequency of breast cancer with triple negative phenotype in Iranian females might be one of the possible explanations for lower mean age of breast cancer in Iran. Although earlier reports suggested that the reported differences in survival among ethnic groups are not due to biologic differences among them (Weiss and Amberson, 1995), unique characteristics of distinct breast cancer subtypes has been reported newly which suggests possibly different pathways for their development (Menashe et al., 2009; Trivers et al., 2009). Therefore, the possibility of race effect on frequency of triple negative breast cancer in Iranian females should be evaluated.

The impact of triple negative phenotype on prognosis of breast cancer in premenopausal patients was more prominent then postmenopausal ones. Very strong positive relationship was found between triple negative phenotype and higher grade of tumors. Meanwhile, smaller mean size of tumors with lower rate of axillary lymph node in premenopausal patients with triple negative phenotype implicates a better prognosis than postmenopausal patients and reveals a similar impact of triple negativity on Iranian patients as it reported in Chinese patients (Yin et al., 2009), but it is different from Japanese patients (Osako et al., 2008). The reported overall 5-year survival rate (Vahdaninia and Montazeri, 2004) could be discussed, at least in part, by this findings.

In conclusion, the result of current study, as the first one, pointed the finger at curtail contribution of triple negative phenotype on early onset of breast cancer rate in Iran. In contrast to poor prognosis of triple negative phenotype in postmenopausal patients, premenopausal patients are more vulnerable to a positive influence of triple negativity. As it suggested recently (Cleator et al., 2008), a unique ethnic profile of triple negative breast cancer in Iranian patients seems possible and a larger

project, a population study, is strongly suggested.

References

Bray F, Moller B (2006). Predicting the future burden of cancer. *Nat Rev Cancer*, **6**, 63-74.

Bundred NJ (2001). Prognostic and predictive factors in breast cancer. *Cancer Treat Rev*, **27**, 137-42.

Cleator SJ, Palmieri C, Coombes CR (2008). The ethnic profile of triple-negative breast cancer. *Onkologie*, **31**, 580-2.

Conte P, Guarneri V (2009) Triple-negative breast cancer: current management and future options. *Ejc Supplements*, **7**, 14-8.

Dent R, Trudeau M, Pritchard KI, et al (2007). Triple-negative breast cancer: clinical features and patterns of recurrence. *Clin Cancer Res*, **13**, 4429-34.

Harirchi I, Ebrahimi M, Zamani N, et al (2000). Breast cancer in Iran: a review of 903 case records. *Public Hlth*, **114**, 143-5.

Harris L, Fritsche H, Mennel R, et al (2007). American Society of Clinical Oncology 2007 update of recommendations for the use of tumor markers in breast cancer. *J Clin Oncol*, **25**, 5287-312.

Hirabayashi YZM (2009). Comparison of time trends in breast cancer incidence (1973-2002) in Asia, from cancer incidence in five continents, Vols IV-IX. *Japanese J Clin Oncol*, **39**, 411-2.

Hung MC, Lau YK (1999). Basic science of HER-2/neu: a review. *Semin Oncol*, **26**, 51-9.

Iwase H, Kurebayashi J, Tsuda H, et al (2010). Clinicopathological analyses of triple negative breast cancer using surveillance data from the registration committee of the Japanese breast cancer society. *Breast Cancer*, **17**, 118-24.

Kolahdoozan S, Sadjadi A, Radmard AR, et al (2010). Five common cancers in Iran. *Arch Iran Med*, **13**, 143-6.

Kwon JH, Kim YJ, Lee KW, et al (2010). Triple negativity and young age as prognostic factors in lymph node-negative invasive ductal carcinoma of 1 cm or less. *BMC Cancer*, **10**, 557.

Lai HW, Kuo SJ, Chen LS, et al (2010). Prognostic significance of triple negative breast cancer at tumor size 1 cm and smaller. *Eur J Surg Oncol*, **37**, 18-24.

Menashe I, Anderson WF, Jatoi I, et al (2009). Underlying causes of the black-white racial disparity in breast cancer mortality: a population-based analysis. *J Natl Cancer Inst*, **101**, 993-1000.

Mohagheghi MA, Mosavi-Jarrahi A, Malekzadeh R, et al (2009). Cancer incidence in Tehran metropolis: the first report from the Tehran Population-based Cancer Registry, 1998-2001. *Arch Iran Med*, **12**, 15-23.

Mousavi SM, Mohaghegghi MA, Mousavi-Jerrahi A, et al (2006). Burden of breast cancer in Iran: a study of the Tehran population based cancer registry. *Asian Pac J Cancer Prev*, **7**, 571-4.

Mousavi SM, Mohagheghi MA, Mousavi-Jerrahi A, et al (2008). Outcome of breast cancer in Iran: a study of Tehran cancer registry data. *Asian Pac J Cancer Prev*, **9**, 275-8.

Mousavi SM, Montazeri A, Mohagheghi MA, et al (2007). Breast cancer in Iran: an epidemiological review. *Breast J*, **13**, 383-91.

Osako T, Nishimura R, Okumura Y, et al (2008). Premenopausal status reflects an unfavorable prognosis in triple-negative breast cancer. *J Clin Oncol (Meeting Abstracts),* **26**, 22208-.

Porter PL (2009). Global trends in breast cancer incidence and mortality. *Salud publica de Mexico*, **51**, S141-6.

Rakha EA, El-Sayed ME, Green AR, et al (2007). Prognostic markers in triple-negative breast cancer. *Cancer*, **109**, 25-32.

Sadjadi A, Hislop TG, Bajdik C, et al (2009). Comparison of breast cancer survival in two populations: Ardabil, Iran and British Columbia, Canada. *BMC Cancer*, **9**, 381.

Sadjadi A, Nouraie M, Mohagheghi MA, et al (2005). Cancer occurrence in Iran in 2002, an international perspective. *Asian Pac J Cancer Prev*, **6**, 359-63.

Schmitt F (2009). HER2+ breast cancer: how to evaluate? *Adv Ther*, **26**, S1-8.

Seal MD, Chia SK (2010). What is the difference between triple-negative and basal breast cancers? *Cancer J*, **16**, 12-6.

Soerjomataram I, Louwman MW, Ribot JG, et al (2008). An overview of prognostic factors for long-term survivors of breast cancer. *Breast Cancer Res Treat*, **107**, 309-30.

Thorat MA, Badve S (2007). Prognostic factors in invasive breast carcinoma: Do new molecular techniques/profiling add significantly to traditional histological factors? *Current Diagnostic Pathology*, **13**, 116-25.

Trivers KF, Lund MJ, Porter PL, et al (2009). The epidemiology of triple-negative breast cancer, including race. *Cancer Causes Control*, **20**, 1071-82.

Vahdaninia M, Montazeri A (2004). Breast cancer in Iran: a survival analysis. *Asian Pac J Cancer Prev*, **5**, 223-5.

Viale G, Bottiglieri L (2009). Pathological definition of triple negative breast cancer. *Eur J Cancer*, **45**, 5-10.

Weiss SE, Tartter PI, Ahmed S, et al(1995). Ethnic differences in risk and prognostic factors for breast cancer. *Cancer*, **76**, 268-74.

Yin WJ, Lu JS, Di GH, et al (2009). Clinicopathological features of the triple-negative tumors in Chinese breast cancer patients. *Breast Cancer Res Treat*, **115**, 325-33.

Zhou X, Liu Q (2010). Clinicopathological characters of triple negative breast cancer. *Chinese J Cancer Res*, **22**, 17-20.