Local Recurrence and Distant Metastases after Breast Conservation Treatment in Women with Triple Negative Breast Cancer Subtype

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Abstract: Introduction: Recently, gene expression studies using DNA microarrays have identified five common subtypes of breast cancer. The triple negative (TN) phenotype, which includes tumors that do not express ER, PR, or HER2 serves as a proxy for the basal-like subtype. At the present time, there is little clinical data evaluating whether a particular breast cancer subtype is associated with increased rates of local and/or distant recurrence after BCT .**Objective:** to evaluate the outcome after breast conservation therapy for triple-negative early-stage invasive breast carcinoma. **Materials and methods:** Between 2000 and 2010, 421 patients with early stage breast cancer patients who treated with BCT were classified as TN (58) if they were negative for all three receptors (ER, PR, and HER2/*neu)* or as non–TN (363) if they were positive for any of the three markers. These patients were evaluated for isolated local and distant recurrence. **Results:** The local relapse rates among the TN group were nearly equal to those of the non–TN. (5.2% vs. 3.9% *P*= 0.63) five-years overall and disease free survival of the TN group were significantly poorer than the other group(62% vs 85 % *p*=0.002) and (39 % vs 75% *p*= 0.00). The isolated local relapse free survival was 90.3% vs 95.7% between the 2 groups. (*P*= 0.365) while the isolated distant metastases free survival was 52% vs 84%. (*P*= 0.00). **Conclusions:** Although women with TN tumors had a higher rate of local failure and a lower rate of freedom from distant metastases compared with women without TN tumors, the absolute difference was relatively small and is not statistically significant and therefore does not preclude BCT for women with TN early-stage invasive breast carcinoma.

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1. Introduction

Breast cancer encompasses a heterogeneous population of tumors characterized by a variety of clinical, pathological, and molecular features [1-3]. Molecular markers such as estrogen receptor (ER), progesterone receptor (PR), and human epidermal growth factor receptor-2 (HER2) have been used to classify patients into different subtypes. Recently, gene expression studies using DNA microarrays have identified five common subtypes of breast cancer: luminal A (ER or PR positive and HER2 negative), luminal B (ER or PR positive and HER2 positive), HER2 overexpressing (ER and PR negative and HER2 positive), basal-like (ER, PR, and HER2 negative, CK 5/6 positive) and normal breast-like tumor [2,4-6]. HER2 overexpressing and basal-like subtypes are significantly more likely to be grade 3 and are associated with worse recurrence rates and decreased overall survival [3,5-7]. The triple negative (TN) phenotype, which includes tumors that do not express ER, PR, or HER2 on immunohistochemistry (IHC), serves as a proxy for the basal-like subtype with a positive predictive value of approximately 80-97% [8,9]. Although this approach is not complete, several groups have adopted a TN definition for basal-like cancer out of convenience.

Several randomized trials have demonstrated molecular markers were available. The data on ER, PR, and HER2/*neu* were obtained through standard clinical testing, using immunohistochemistry for ER and PR and the HER2/*neu*.

For ER and PR, receptor positivity was based on more than 10% of cells testing positive.

HER2/*neu* scores of 0 and 1 were considered negative, and Positive HER2 was determined using either IHC 3+ staining or amplification on fluorescence *in situ* hybridization (FISH) in patients with IHC 2+.

Patients were classified as triple negative if they were negative for all three receptors or as non–triple negative if they were positive for any of the three markers.

Exclusion criteria for this study included male breast cancer, T3 disease, mastectomy, previous or concurrent malignancy (breast or other site)or patients treated without radiotherapy.

All patients were treated with breast-conserving surgery followed by radiation therapy. The surgical treatment included complete gross excision of the primary tumor. Re-excision of the primary tumor was done if possible if positive margin was proved. Pathologic axillary lymph node staging was performed for all patients. Earlier in the study period, pathologic axillary lymph node staging was generally performed

using a lower axillary lymph node that survival rates following breast-conserving treatment (BCT) were equivalent to those observed with radical mastectomy [10-12]. Given these results, BCT has become an accepted treatment for early stage breast cancer [13]. At the present time, there is little clinical data evaluating whether a particular breast cancer subtype is associated with increased rates of local and/or distant recurrence after BCT [14-17].

The outcome after breast conservation treatment with radiation has not been well described for triple-negative early-stage invasive breast carcinoma. Therefore, the current study was performed to evaluate the outcome after breast conservation treatment with radiation for this subtype of breast carcinoma.

2. Materials and Methods

Between the year 2000 and 2010, 759 patients with American Joint Committee on Cancer stages I-II (18) disease were treated with breast-conserving surgery followed by radiation therapy to the intact breast, with or without systemic therapy, at the Clinical Radiation Oncology Department, Tanta University hospital.

Only those patients in whom ER, PR, and HER2/*neu* status were available were included in the current analysis. This limited the sample to a total of 421 patients in whom all three was defined as clinical evidence of distant disease based on clinical and/or radiographic findings.

All events were calculated from the time of surgery to the time of the event.

For the calculation of overall survival (OS), a death due to any cause was scored as a failure. For the calculation of freedom from distant metastases, a failure was scored at the time of first evidence of distant metastatic disease.

The Kaplan-Meier method was used to calculate OS, freedom from distant metastases, local failure, and distant failure (19) . The time period was defined as the time of surgery of breast carcinoma. The log-rank test was used for statistical comparisons between groups (20) Multivariate analysis was performed using the Cox proportional hazards model (21). Chi - square test was used to compare the characteristics of patients between the 2 groups. All statistical methods was done using SPSS statistical package version 17.

3. Results

In our series, 421 patients were classified according to ER, PR, and HER2 into either TN (58) or non TN (363) patients. TN patients represent about 14 % of the whole study group. Patients, tumor, and treatment characteristics of each group are detailed in table(1). there is a highly significant difference between both groups regarding age with about 45% of triple negative patients less than 40 years compared to 33% of non triple negative patients.

The median age for patients with TN breast carcinoma was 41 years while the median age for patients without a triple negative breast carcinoma was 47 years.

There was no significant difference between patients by T-size, LN status, grade, extensive intraductal component, tumor necrosis, lymphovascular invasion or resection margin. All triple negative patients received chemotherapy compared to 83% of patients in the other group. The most common regimens used were either CMF or FAC or FAC and taxanes . All eligible patients underwent BCS and post operative whole breast irradiation. For the radiation treatment to the whole breast, the median dose was 46 Gy (mean, 45.75 Gy; range, 45-50.4 Gy). The median total dose was 61 Gy (mean, 60.91 Gy; range, 58-66 Gy). Among non triple negative groups, 80,7% of patients received hormone therapy. Few patients with HER 2 overexpression received adjuvant trastuzumab therapy.

During the follow up period, there were 3 (5.2%) and 14 (3.9%) patients in TN and non TN groups respectively who experienced isolated local in breast relapse as the first site of recurrence. the difference was non significant(P= 0.63).

Isolated distant relapse was observed in 19(32.8 %) and 47(12.9 %) patients in TN and non TN groups respectively(p= 0.00).

The 5-years OS and DFS rates of all patients were 82% and 71% respect. Groups respectively (P= 0.00)

By multivariate Cox proportional hazard models, lymph node status, intraductal component, and use of adjuvant systemic treatment were significant prognostic factors affecting OS . Age, tumor size , lymph node status and use of systemic treatment significantly affected DFS (Table2)

4. Discussion

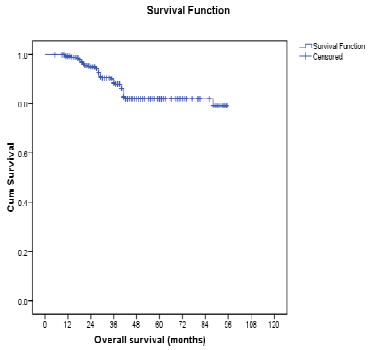
There has been much attention focused on TNBCs since the first identification of basal-like cancers using microarray expression profiling studies in 2005 (22). There is a significant overlap between TNBCs, which are defined by immunohistochemistry, and basaloid like tumors, which are identified from gene expression profiling, such that those two entities are frequently considered synonymous from a clinical perspective.(23-24)

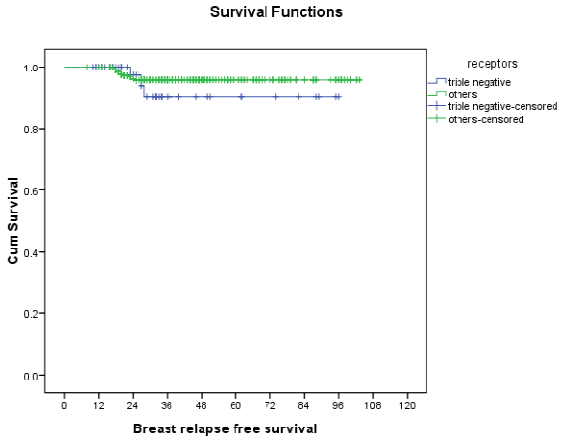
Table (1): Patients characteristics.

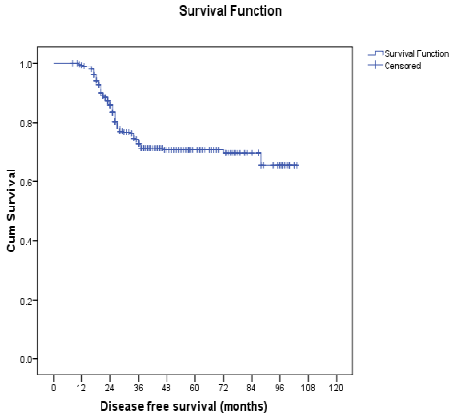
		Receptors							
		Triple negative N=58		Non -triple negative N=363		Total		Chi-square	
		N	%	N	%	N	%	X^2	*P*-value
AGE	<40 y	26	45	121	33.3	155	36.8	13.747	0.000
	>40 y	32	55	242	66. 7	266	63.2		
Tumor size	T1	12	20.69	103	28.37	115	27.32	1. 67	0.43
	T2	46	79.31	260	71.63	306	72.68		
L.N.	N0	12	20.69	95	26.17	107	25.42	1.663	0.435
	N1	24	41.38	159	43.80	183	43.47		
	N2-3	22	37.93	109	30.03	131	31.12		
GRADE	G1	8	13.79	45	12.40	53	12.59	0.111	0.946
	G2	39	67.24	251	69.15	290	68.88		
	G3	11	18.97	67	18.46	78	18.53		
EIC	Positive	18	31	93	25.6	111	26.4	1.295	0.523
	Negative	28	48.3	204	56.2	232	55.1		
	unknown	12	20.7	66	18.2	78	18.5		
Necrosis	yes	6	10.3	33	9.1	39	9.3	0.125	0.939
	no	42	72.4	263	72.5	305	72.4		
	unknown	10	17.2	67	18.5	77	18.3		
LVI	yes	20	34.5	115	31.7	135	34.53	0.503	0.778
	no	33	56.9	223	61.4	256	65.47		
	unknown	5	8.6	25	6.9	30	7.1		
RM	negative	49	84.48	312	85.95	361	85.75	0.341	0.843
	positive	2	3.45	8	2.20	10	2.38		
	close	7	12.07	43	11.85	50	11.88		
Systemic treatment	Chemotherapy alone	58	100.00	70	19.28	128	30.40	153.979	0.000
	Hormonal alone	0	0.00	60	16.53	60	14.25		
	both	0	0.00	233	64.19	233	55.34		
Isolated local recurrence	present	3	5.2	14	3.9	17	4	0.223	0.639
	absent	55	94.8	349	96.1	404	96		
Isolated distant recurrence	present	19	32.8	47	12.9	66	15.7	14.848	0.000
	absent	39	67.2	316	32.8	355	84.3		

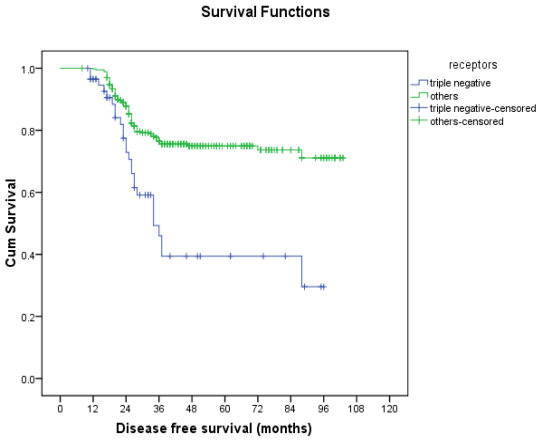
Table (2): Cox proportional hazards multivariate model for overall (OS) and disease-free survival (DFS)

Variable	OS			DFS		
	HR	95% CI	*p*-value	HR	95% CI	*p*-value
AGE	.896	.446-1.797	.756	.456	.286-.729	.001
TSIZE	1.927	.527-7.044	.321	2.443	1.138-5.246	.022
LN	2.603	1.263-5.362	.009	2.150	1.406-3.285	.000
grade	.864	.444-1.683	.668	.877	.582-1.322	.531
EIC	.349	.191-.637	.001	.730	.512-1.041	.082
necrosis	.760	.419-1.377	.365	.971	.682-1.382	.869
LVI	.914	.506-1.652	.767	.796	.546-1.162	.238
Tumor subtype	.844	.379-1.876	.677	.935	.552-1.583	.801
Systemic treatment	.681	.470-.987	.043	.533	.419-.678	.000


Survival Function
Survival Function
Censored
Cum Survival
Overall survival (months)


Survival Functions
receptors
triple negative
others
triple negative-censored
others-censored
Cum Survival
Breast relapse free survival


Survival Function
Survival Function
Censored
Cum Survival
Disease free survival (months)


Survival Functions
receptors
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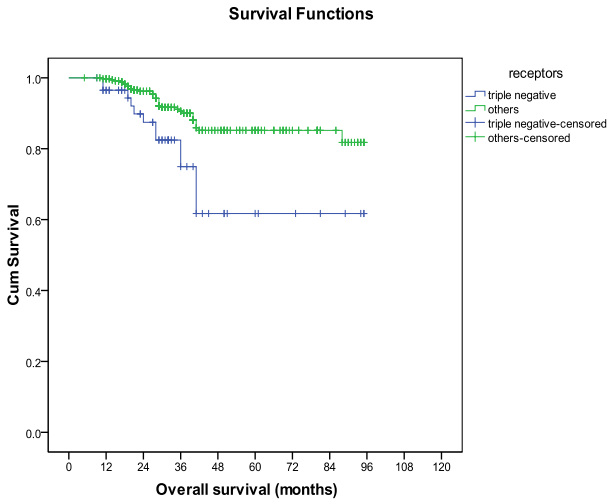
others
triple negative-censored
others-censored
Cum Survival
Disease free survival (months)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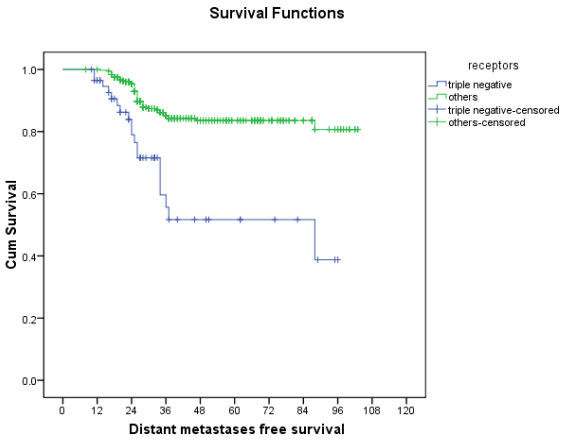
Overall survival (months)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Cum Survival
Distant metastases free survival

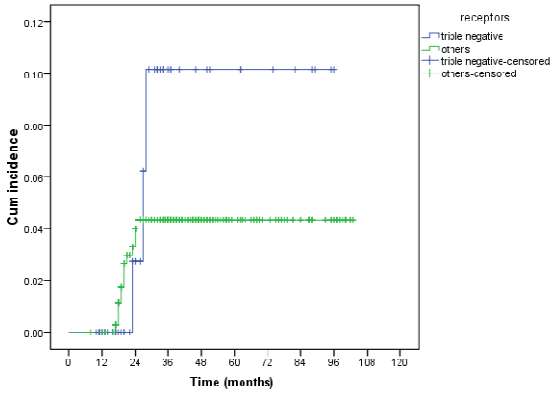

Figure 1. Cumulative incidence of local recurrence as the first event

TNBCs have a poor prognosis and do not respond to therapy directed at known breast cancer growth factor targets, including hormonal therapy and trastuzumab. TNBCs are fast-growing tumors, usually have a high histologic grade, and tend to metastasize earlier compared with breast cancers that express hormone receptors (16,25) . The predominant sites of metastases for TNBCs differ from the luminal subtype. They are more frequently node negative and spread, in70%of patients, to the lung and brain, whereas luminal cancers tend to metastasize, in 70% of patients, to the bone and liver. (16,22.)

TNBC tumors seem to be more sensitive to chemotherapy, and although there is a sharp decrease in survival early after diagnosis, survival plateaus after 8 to 10 years (16) .

In the current study, we evaluated 421 patients with conservatively treated breast cancer in whom all three markers were available to validate the prognostic utility of this classification scheme and to determine whether triple negative breast cancers have a more aggressive locoregional and distant relapse rate.

Among all patients included, the triple negative cohort still had a poorer prognosis than the non–triple negative cohort, but the two cohorts had a similar local relapse-free survival (90.3 vs 95.7 in TN and non TN groups respectively, *P*= 0.365).

In fact, the local relapse rates among the triple negative cohort were nearly equal to those of the non–triple negative cohort (5.2% and 3.9% of patients in TN and non TN groups *P*= 0.63).

This is of particular importance given the fact that triple negative patients are significantly younger, and younger women have been shown to have a higher rate of local relapse compared with older women(26-31), therefore, those patients should not be considered poor candidates for breast-conserving therapy.

The fact that these women with triple negative tumors had similar local relapse rates indicates that these tumors are not radiation resistant. This is consistent with the fact that these basal-like tumors are theoretically responsive to DNA-damaging agents and should, therefore, be relatively radiation sensitive. Larger patient cohorts with a longer follow-up will be required to further validate these data.

Only limited data are available for local control after breast conservation treatment with radiation relative to biologic subtype.

Haffty et al reported that there was no significant difference in local failure at 5 years for the triple-negative subgroup versus the non–triple-negative subgroup (17% vs. 17%, respectively; *P* = .82) (14) .

Nguyen *et al.,* reported the outcome for patients grouped according to ER status, PgR status, and HER2 status as a surrogate for biologic subtype of disease (15). The 5-year rate of local recurrence was 7.1% for the basal-like (triple-negative) subgroup, 8.4% for the HER2 subgroup, 1.5% for the luminal B subgroup, and 0.8% for the luminal A subgroup. In comparison with the luminal A subgroup, there was an increased rate of local failure for the basal-like (triple-negative) subgroup (*P* = .0099) and the HER2 subgroup (*P* = .012).

In a study evaluating HER2 as a single factor, **Harris *et al.,*** reported no difference in local failure at 5 years for the HER2-positive group compared with the HER2-negativegroup (0% vs. 2%, respectively; *P* = .15)[32]. **Parikh *et al.,*** reported an increased rate of local recurrence for premenopausal patients with CK19-negative tumors compared with CK19-positive tumors (relative risk, 3.54; *P* < .01) [33] .

For 3038 patients treated with either breast conservation treatment or mastectomy, **Cheang *et al.,*** used a panel of tumor markers to define biologic subtype of disease [34] . The 10-year rate of local relapse was 7% for the luminal A subgroup, 11% for the luminal B subgroup, 15% for the luminal/HER2-positive subgroup, 15% for the basal-like subgroup, and 18% for the HER2-positive subgroup.

For 1601 patients treated with either mastectomy or breast conservation, **Dent *et al.,*** reported no significant difference in local failure for the triple-negative group compared with the non–triple-negative group (HR, 0.8; *P* = .38) [16] .

There are emerging data for using genomic factors to predict the risk of local recurrence after breast conservation treatment. Using a 21-gene recurrence score assay, Mamounas et al evaluated the risk of local-regional recurrence for patients treated in the National Surgical Adjuvant Breast and Bowel Project B-14 and B-20 trials for patients with node-negative, ER-positive breast cancer[35] . After breast

conservation treatment with radiation for 390 patients, there was a strong interaction of local-regional failure with age. For patients aged < 50 years, the 10-year rate of local-regional failure was 12.5% for tumors with a low-risk recurrence score, 27.7% for tumors with an intermediate-risk recurrence score, and 26.5% for tumors with a high-risk recurrence score (P = .057). However, the 10-year rates of local-regional recurrence were relatively low for patients aged ≥ 50 years, regardless of the results of the recurrence score assay (3.6% vs. 3.7% vs. 4.8%, respectively; P = .663).

Nuyten *et al.,* reported that the wound response signature obtained by gene expression profiling was able to segregate the patients after breast conservation treatment into a high risk versus a low risk of local recurrence at 10 years (29% vs. 5%, respectively; P = .0008) [36] . The wound signature profile remained a significant predictor for local recurrence on multivariate analysis (P = .01)

Freedman *et al.,* [17] defined three groups: ER or PR (+), HER2, and TN. Patients in the TN and HER2 groups were more likely to have T2 and grade 3 diseases. The median age of the TN and HER2 groups was both 54 years, which was also older than that of the present study. They also reported that there was no overall difference in total locoregional recurrence rates between the three groups (p=0.13). Additionally, a higher rate of distant metastases in the HER2 group was observed (11.9%, p=0.009) which translated into a lower recurrence-free survival rate (84%, p=0.005).

It is also notable that, despite the use of adjuvant systemic chemotherapy, triple negative patients seemed to have a poor prognosis, with a distant metastasis-free survival rate of only 52% at 5 years. The interpretation of these findings remains debatable given the relatively small number of patients and the retrospective nature of this study, and further studies evaluating the impact of adjuvant systemic therapy in triple negative patients is warranted.

By definition of their lack of receptors for ER, PR, and HER2/ *neu*, patients with triple negative tumors are not candidates for adjuvant hormonal therapy or trastuzumab[37]. Basal-like tumors have been shown to overexpress HER1 (epidermal growth factor), and patients with these tumors may be candidates for prospective studies evaluating targeted antibodies against epidermal growth factor that are already in clinical use [5,38] .

Clearly, additional prospective and retrospective studies are warranted to further refine prognosis and optimize treatment in patients with triple negative breast cancers. We have demonstrated here, using the simple commonly available markers of ER, PR, and HER2/*neu*, that patients with triple negative breast cancers have a relatively poor prognosis, with a poorer distant metastasis-free, disease-free, and Overall survival.

The current study has several potential limitations. Firstly, because HER2 testing has evolved over time, there was not uniform testing for all of the breast carcinomas in the current study.

Second, the relatively small number of patients in the triple-negative subset limited the power to detect small, but potentially statistically significant, differences. Whether alternative forms of chemotherapy for these breast cancer patients, with or without biologic modifiers, will prove superior can only be addressed by well-designed prospective studies. Third, the groups were differentiated based on tumor markers, not gene profiling. Nonetheless, segregating patients into prognostic groups on the basis of tumor markers is clinically relevant, particularly in view of targeted therapies currently available.

Another potential weakness of the study is unavoidable selection biases in a retrospective series such as this. For the current study, only patients who had available ER, PR, and HER2/*neu* data were included.

Finally, the patients were treated largely before the routine use of trastuzumab and lapatinib. Thus, the results in the current study, particularly for the HER2-positive patients, could be substantially different for patients treated using newer targeted therapies.

Conclusion

Although women with triple-negative tumors had a higher rate of local failure and a lower rate of freedom from distant metastases compared with women without triple-negative tumors, the absolute difference was relatively small and not statistically significant and therefore does not preclude breast conservation treatment with radiation for women with triple-negative early-stage invasive breast carcinoma.

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5. References

1. **Jae Myoung Noh, Doo Ho Choi, Seung Jae Huh, Won Park *et al.*** (2011): Patterns of Recurrence after Breast-Conserving
2. **Sørlie T**. (2004): Molecular portraits of breast cancer: tumour subtypes as distinct disease entities. Eur J Cancer;40:2667-75.
3. **Sørlie T, Perou CM, Tibshirani R, Aas T, Geisler S, Johnsen H, *et al.*** (2001): Gene expression patterns of breast carcinomas distinguish tumor

subclasses with clinical implications. Proc Natl Acad Sci U S A;98:10869-74.

4. **Brenton JD, Carey LA, Ahmed AA, Caldas C.** Molecular classification and molecular forecasting of breast cancer: ready for clinical application? J Clin Oncol 2005;23:7350-60.
5. **Sørlie T, Tibshirani R, Parker J, Hastie T, Marron JS, Nobel A, *et al.*** (2003): Repeated observation of breast tumor subtypes in independent gene expression data sets. Proc Natl Acad Sci U S A;100:8418-23.
6. **Sotiriou C, Neo SY, McShane LM, Korn EL, Long PM, Jazaeri A, *et al.* (**2003): Breast cancer classification and prognosis based on gene expression profiles from a population-based study. Proc Natl Acad Sci U S A; 100:10393-8.
7. **Carey LA, Perou CM, Livasy CA, Dressler LG, Cowan D, Conway K, *et al.*** (2006): Race, breast cancer subtypes, and survival in the Carolina Breast Cancer Study. JAMA;295:2492-502.
8. **Nielsen TO, Hsu FD, Jensen K, Cheang M, Karaca G, Hu Z, *et al.*** (2000); Immunohistochemical and clinical characterization of the basal-like subtype of invasive breast carcinoma. Clin Cancer Res 2004;10:5367-74.
9. **Rakha EA, Reis-Filho JS, Ellis IO.** (2008): Basal-like breast cancer: a critical review. J Clin Oncol;26:2568-81.
10. **van Dongen JA, Voogd AC, Fentiman IS, Legrand C, Sylvester RJ, Tong D, et al.** Long-term results of a randomized trial comparing breast-conserving therapy with mastectomy: European Organization for Research and Treatment of Cancer 10801 trial. J Natl Cancer Inst 2000;92:1143-50.
11. **Veronesi U, Cascinelli N, Mariani L, Greco M, Saccozzi R, Luini A, *et al.*** (2002):Twenty-year follow-up of a randomized study comparing breast-conserving surgery with radical mastectomy for early breast cancer. N Engl J Med;347:1227-32.
12. **Poggi MM, Danforth DN, Sciuto LC, Smith SL, Steinberg SM, Liewehr DJ, *et al.*(**2003): Eighteen-year results in the treatment of early breast carcinoma with mastectomy versus breast conservation therapy: the National Cancer Institute Randomized Trial. Cancer; 98:697-702.
13. **Lazovich D, Solomon CC, Thomas DB, Moe RE, White E.** (1999): Breast conservation therapy in the United States following the 1990 National Institutes of Health Consensus Development Conference on the treatment of patients with early stage invasive breast carcinoma. Cancer;86: 628-37.
14. **Haffty BG, Yang Q, Reiss M, Kearney T, Higgins SA, Weidhaas J, *et al.*** (2006): Locoregional relapse and distant metastasis in conservatively managed triple negative early-stage breast cancer. J Clin Oncol;24:5652-7.
15. **Nguyen PL, Taghian AG, Katz MS, Niemierko A, Abi Raad RF, Boon WL, *et al.*** (2008): Breast cancer subtype approximated by estrogen receptor, progesterone receptor, and HER-2 is associated with local and distant recurrence after breast-conserving therapy. J Clin Oncol;26:2373-8.
16. **Dent R, Trudeau M, Pritchard KI, Hanna WM, Kahn HK, Sawka CA, *et al.*** (2007): Triple-negative breast cancer: clinical features and patterns of recurrence. Clin Cancer Res.;13(15 Pt 1):4429-34.
17. **Freedman GM, Anderson PR, Li T, Nicolaou N.** (2009): Locoregional recurrence of triple-negative breast cancer after breast-conserving surgery and radiation. Cancer;115:946-51.
18. **Greene FL, Page DL, Fleming ID, *et al.* (**2002): **eds.** American Joint Committee on Cancer. AJCC Cancer Staging Manual. 6th ed. New York: Springer-Verlag; **Kaplan EL, Meier P. (**1958): Nonparametric estimation from incomplete observations. *J Am Stat Assoc*; 53:457-81.

20-Mantel N. (1966): Evaluation of survival data and two new rank order statistics arising in its consideration. *Cancer Chemother Rep*; 50:163-70.

21. **Cox DR.** (1972): Regression models and life tables. *J Roy Statist Soc Ser B*; 34:187-220.

22-Perou CM, Sørlie T, Eisen MB, *et al.* (2000**):** Molecular portraits of human breast tumours. Nature 406:747-752,

23-Foulkes WD, Smith IE, Reis-Filho JS(2010**):** Triple-negative breast cancer. N Engl J Med., 363:1938-1948,

24-Brenton JD, Carey LA, Ahmed AA, *et al.* (2005**):** Molecular classification and molecular forecasting of breast cancer: Ready for clinical application? J Clin Oncol 23:7350-7360,

25- Collett K, Stefansson IM, Eide J, *et al.* (2005**):** A basal epithelial phenotype is more frequent in interval breast cancers compared with screen detected tumors. Cancer Epidemiol Biomarkers Prev 14:1108-1112,

26. **Bartelink H, Horiot JC, Poortmans P, *et al.*** (2001**):** Recurrence rates after treatment of breast cancer with standard radiotherapy with or without additional radiation. N Engl J Med., 345:1378-1387,
27. **de la Rochefordiere A, Asselain B, Campana F, *et al.*** (1993**):** Age as prognostic factor in premenopausal breast carcinoma. Lancet, 341:1039-1043.
28. **Fowble BL, Schultz DJ, Overmoyer B, *et al.*** (1994**):** The influence of young age on outcome in early stage breast cancer. Int J Radiat Oncol Biol Phys., 30:23-33,
29. **Harrold EV, Turner BC, Matloff ET, *et al.*** (1998**):** Local recurrence in the conservatively

treated breast cancer patient: A correlation with age and family history. Cancer J Sci Am 4:302-307,

30. Recht A, Connolly JL, Schnitt SJ, *et al.* (1988)**:** The effect of young age on tumor recurrence in the treated breast after conservativesurgery and radiotherapy.Int J Radiat Oncol Biol Phys 14:3-10,

31. Vicini FA, Recht (2002 **A): A**ge at diagnosis and outcome for women with ductal carcinoma-in-situ of the breast: A critical review of the literature. J Clin Oncol., 20:2736-2744,

32-Harris EE, Hwang WT, Lee EA, *et al.* (2006): The impact of HER-2 status on local recurrence in women with stage I-II breast cancer treated with breast-conserving therapy. *Breast J*; 12:431-6.

33-Parikh RR, Yang Q, Higgins SA, *et al.* (2008): Outcomes in young women with breastcancer of triple-negative phenotype: the prognostic significance of CK19 expression.*Int J Radiat Oncol Biol Phys*; 70:35-42.

34-Cheang MC, Voduc D, Tyldesley S, *et al.* (2008): Breast cancer molecular subtypes and locoregional recurrence. *J Clin Oncol*; 26(15 suppl):9s (Abstract 510).

35-Mamounas E, Tang G, Bryant J, *et al.* Association between the 21-gene recurrence score assay (RS) and risk of locoregional failure in node-negative ER-positive. breast cancer: results from the NSABP B-14 and NSABP B-20. Proceedings of the San Antonio Breast Cancer Symposium; December 8-11, 2005. Abstract 29.

36-Nuyten DS, Kreike B, Hart AA, *et al.* (2006): Predicting a local recurrence after breast conserving therapy by gene expression profiling. *Breast Cancer Res*; 8:R62.

37-Romond EH, Perez EA, Bryant J, *et al.* (2005)**:** Trastuzumab plus adjuvant chemotherapy for operable HER2-positive breast cancer. N Engl J Med., 353: 1673-1684,.

38-Matos I, Dufloth R, Alvarenga M, *et al.* (2005)**:** P63, cytokeratin 5, and P-cadherin: Three molecular markers to distinguish basal phenotype in breast carcinomas. Virchows Arch 447:688-694.

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