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Original Articles

p53 as a Specific Prognostic Factor in Triple-negative Breast Cancer

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Objective: A recent suggestion is that the predictive value of a single biomarker may rely on the genetic background on the tumor and that different breast cancer subgroups may have different predictive markers of response to chemotherapy. The prognostic value of p53 in the outcome of adjuvant anthracycline-containing chemotherapy was evaluated according to molecular subclasses defined using the expression of estrogen receptor, progesterone receptor and human epidermal growth factor receptor 2.
Methods: Subjects were patients ($n = 135$) with invasive ductal carcinoma treated with adjuvant anthracycline-based chemotherapy between 1994 and 2000 in our hospital. Clinicopathological features were reviewed by retrospective examination of medical records.
Results: Overall survival rate was not independently predictive by p53 status ($P = 0.182$). However, in triple-negative cases, there was statistically significant survival difference ($P = 0.034$) and no statistically significant difference ($P = 0.783$) in non-triple-negative cases by p53 status. In the Cox proportional hazard analysis, p53 was also strongly predictive for relapse-free survival ($P = 0.013$) and overall survival ($P = 0.049$) in triple-negative patients.
Conclusions: p53 status could be a specific prognostic factor in triple-negative breast cancer patients treated by adjuvant anthracycline-based regimen. When p53 is positive in triple-negative breast cancer, we could expect poor survival, prompting aggressive or alternative treatment.

Key words: breast cancer – p53 protein – anthracycline – prognosis – biologic marker

INTRODUCTION

Combination regimens that include anthracyclines (epirubicin and doxorubicin) and alkylating agents (cyclophosphamide) administered in an adjuvant setting improve overall survival in patients with early breast cancer (1). p53 status has been one of the most investigated predictive biomarkers for the efficacy of anthracycline-containing chemotherapy (2–8). Despite the many studies, results have been inconsistent, with no association between p53 expression and tumor response to neoadjuvant anthracyclines reported (2,6–9), whereas other reports have associated p53 overexpression with both resistance (3–5,10) and sensitivity (11,12) to preoperative anthracycline-containing chemotherapy. There is no unique explanation to account for these inconsistencies. p53 is involved in regulating cell proliferation and apoptosis, and in promoting chromosomal stability. Disruption of these functions appears to play an important role in carcinogenesis. Mutations in the tumor suppressor gene p53 are present in 18–25% of primary breast carcinomas (13,14). Breast cancer encompasses a spectrum of distinct phenotypes with disparate histopathological, clinical and molecular features. The triple-negative subtype of invasive breast cancers is defined by a lack of expression of estrogen receptor (ER), progesterone receptor (PR) and human epidermal growth factor

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receptor 2 (HER2) (15–17). These account for approximately 10–17% of all breast carcinomas (15,17–22) and this subtype is significantly associated with p53 overexpression (23). Recently, it has been suggested that the predictive value of a single biomarker could rely on the genetic background on the tumor and that different breast cancer subgroups may have different predictive markers of response to chemotherapy (24,25).

Presently, we have evaluated the prognostic value of p53 for the outcome of adjuvant anthracycline-containing chemotherapy according to molecular subclasses defined by the expression of ER, PR and HER2.

PATIENTS AND METHODS

Patients

Patients ($n = 135$) with invasive ductal carcinoma treated with adjuvant anthracycline-based chemotherapy between 1994 and 2000 in KangNam St Mary's Hospital were enrolled. This study was approved by the Institutional Review Board. Clinico-pathological features of the patients were reviewed by the retrospective review of medical records. All patients were received four- or six-cycle anthracycline-containing chemotherapy.

Tissue Microarrays

To construct the tissue microarray block, 3 mm core biopsies obtained from viable morphologically representative areas of paraffin-embedded tumor tissues were assembled on a recipient paraffin block containing 30 biopsies. This was carried out using a precision instrument (Micro Digital, Gunpo-si, Gyeonggi-do, Republic of Korea). After construction, 5 μm sections were cut and the histology was verified by hematoxylin–eosin staining.

Fluorescence *In Situ* Hybridization (Fish) of *c-erbB2*

FISH was performed using the PathVysion™ HER2/CEN probe (Vysis, Downers Grove, IL, USA). The *c-erbB2* to chromosome 17 centromere ratio was measured in at least 60 nuclei from the tumor cells, and an average score was taken. More than two copies of *c-erbB2* for each chromosome 17 were considered to be a positive sign for *c-erbB2* gene amplification.

Immunohistochemistry

Five-micrometer sections of paraffin-embedded tissue arrays were deparaffinized, rehydrated in a graded series of alcohol solutions and microwave-treated for 10 min in a pH 6.0 citrate buffer. The endogenous peroxidase activity was blocked using 0.3% hydrogen peroxide. The tissue arrays were processed in an automatic immunohistochemistry (IHC) staining machine using standard procedures (Lab Vision autostainer; Lab Vision, Fremont, CA, USA) and a ChemMate™ EnVision™ system (DAKO, Carpinteria, CA, USA). p53 antibody (DO-1; DAKO) was used at a dilution of 1:50. Sections were visualized with 3-3′-diaminobenzidine and counterstained with Mayer's hematoxylin. The p53 expression levels were determined semi-quantitatively based on the positive nuclear staining fraction of tumor cells (score $0 \leq 10\%$; score 1 = 11–25%; score 2 = 26–50%; score $3 \geq 51\%$) and score 0 considered as negative and score 1, 2 and 3 were considered as positive.

Enzyme Immunoassay

ER and PR status were reviewed by medical records. The receptor status had been determined using a commercial enzyme immunoassay according to the instructions of the manufacturer (Abbott Laboratories, Chicago, IL, USA). A result exceeding 15 fmol/mg was considered positive for the presence of the particular receptor.

Statistical Analyses

The duration of survival was defined as the time from operation to death attributed to breast cancer. The overall survival rate and relapse-free survival rate of each subgroup were estimated by the Kaplan–Meier method and the statistical significance of the difference in survival outcomes among subgroups were evaluated by the log-rank test. To evaluate the relationship between each prognostic variable and survival prognosis, the Cox proportional hazard regression analysis was performed. The relative risks were calculated with 95% confidence intervals. A value of $P > 0.05$ was regarded as statistically significant.

RESULTS

Clinico-pathological characteristics of the patients and IHC profiles are summarized in Table 1. There was no statistical difference in distribution for all respective clinico-pathological parameters between the triple-negative and non-triple-negative groups. IHC for p53 was positive for 57 of 135 (42.2%) breast cancer cases. Overexpression of p53 occurred in 13 of 32 (40.6%) patients in the triple-negative group and 44 of 103 (42.7%) patients in the non-triple-negative group. In the triple-negative subgroup, the overall survival rate of p53-positive patients was statistically significantly lower than that of p53-negative patients ($P = 0.034$; Fig. 1a). In the non-triple-negative subgroup and overall patients group, there was no statistical difference (Fig. 1a and c). Only the triple-negative subgroup showed a statistical difference for relapse-free survival ($P = 0.005$; Fig. 2).

Table 2 summarizes the risk factors for overall survival and relapse-free survival in the overall patients group. Univariate analysis revealed that nodal status, disease stage,

Table 1. Clinico-pathological features of objective patients

	Triple-negative (%)			Non-triple-negative (%)			*P* value
	p53(−)	p53(+)	*P* value	p53(−)	p53(+)	*P* value	
Age							
<40	3 (15.8)	3 (23.0)	0.774	17 (28.8)	12 (27.3)	0.707	0.078
40–50	8 (42.1)	4 (30.8)		30 (50.8)	20 (45.5)		
>50	8 (42.1)	6 (46.2)		12 (20.3)	12 (27.3)		
Menopause							
Pre	10 (52.6)	7 (53.8)	0.449	41 (69.5)	32 (72.7)	0.699	0.151
Post	9 (47.4)	5 (38.5)		17 (28.8)	12 (27.3)		
Unknown	0 (0)	1 (7.7)		1 (1.7)	0 (0)		
T							
T1	6 (31.6)	4 (30.8)	0.919	11 (18.6)	9 (20.5)	0.568	0.233
T2	11 (57.9)	7 (53.8)		32 (54.2)	27 (61.4)		
T3–4	2 (10.5)	2 (15.4)		16 (27.1)	8 (18.2)		
N							
N0	10 (52.6)	5 (38.5)	0.199	19 (32.2)	12 (27.3)	0.283	0.292
N1	2 (10.5)	5 (38.5)		17 (28.8)	8 (18.2)		
N2	5 (26.3)	1 (7.7)		9 (15.3)	13 (29.5)		
N3	2 (10.5)	2 (15.4)		14 (23.7)	11 (25)		
Stage							
I	2 (10.5)	0 (0)	0.277	6 (10.2)	3 (6.8)	0.402	0.078
II	10 (52.6)	10 (76.9)		26 (44.1)	15 (34.1)		
III	7 (36.8)	3 (23.1)		27 (45.8)	26 (59.1)		
Histologic grade							
G1	0 (0)	0 (0)	0.618	2 (3.5)	1 (2.3)	0.499	0.601
G2	9 (47.4)	5 (38.5)		28 (49.1)	17 (38.6)		
G3	10 (52.6)	8 (61.5)		27 (45.8)	26 (59.1)		
Nuclear grade							
G1	2 (10.5)	0 (0)	0.190	7 (12.1)	2 (4.5)	0.286	0.601
G2	12 (63.2)	6 (46.2)		37 (63.8)	27 (61.4)		
G3	5 (26.3)	7 (53.8)		14 (24.1)	15 (34.1)		
Neural invasion							
No	18 (94.7)	12 (92.3)	0.780	52 (88.1)	35 (79.5)	0.234	0.177
Yes	1 (5.3)	1 (7.7)		7 (11.9)	9 (20.5)		
Vein invasion							
No	18 (94.7)	12 (92.3)	0.780	54 (91.5)	40 (90.9)	0.913	0.653
Yes	1 (5.3)	1 (7.7)		5 (8.5)	4 (9.1)		
Lymphatic invasion							
No	8 (42.1)	5 (38.5)	0.837	19 (32.2)	13 (29.5)	0.773	0.316
Yes	11 (57.9)	8 (61.5)		40 (67.8)	31 (70.5)		
Differentiation							
Well	2 (10.5)	0 (0)	0.05	11 (20.4)	3 (8.3)	0.253	0.051
Moderate	13 (68.4)	5 (38.5)		35 (64.8)	25 (69.4)		
Poor	4 (21.1)	8 (61.5)		8 (14.8)	8 (22.2)		

Continued

Table 1. *Continued*

	Triple-negative (%)			Non-triple-negative (%)			*P* value
	p53(−)	p53(+)	*P* value	p53(−)	p53(+)	*P* value	
ER							
Negative	19	13		11 (18.6)	14 (31.8)	0.009	0
Positive	0			47 (79.7)	24 (54.5)		
Unknown	0			1 (1.7)	6 (13.6)		
PR							
Negative	19	13		14 (23.7)	14 (31.8)	0.024	0
Positive	0			44 (74.6)	24 (54.5)		
Unknown	0			1 (1.7)	6 (13.6)		
HER2							
Negative	19	13		44 (74.6)	18 (40.9)	0.001	0
Positive	0			15 (25.4)	26 (59.1)		
p53							
Negative	19 (59.4)			59 (57.3)			0.834
Positive	13 (40.6)			44 (42.7)			
Total	32			103			

ER, estrogen receptor; PR, progesterone receptor; HER2, human epidermal growth factor receptor 2.

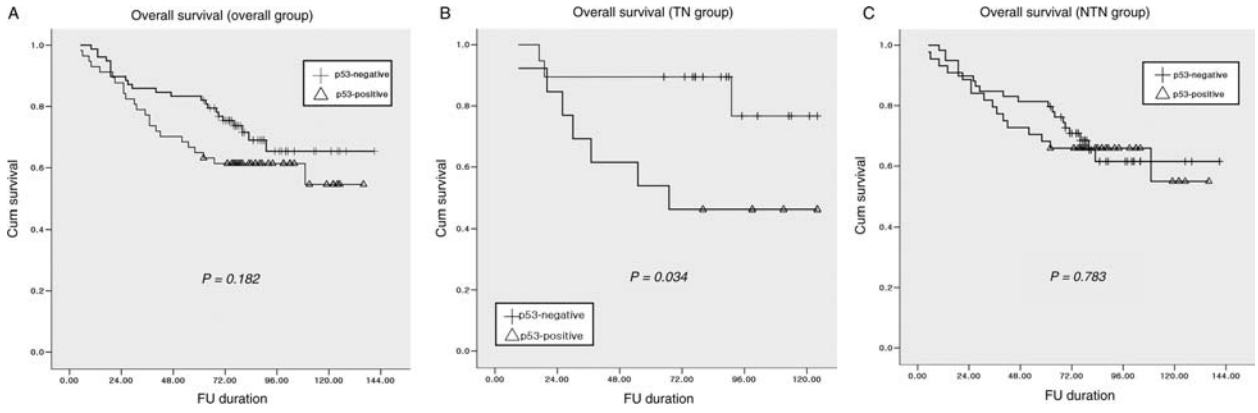

Figure 1. Kaplan–Meier overall survival curve. (a) Overall group, (b) triple-negative group and (c) non-triple-negative group.

nuclear grade, vein invasion, lymphatic invasion, differentiation and HER2 status had prognostic values for overall survival. In multivariate analysis, vein invasion and lymphatic invasion were implicated as independent prognostic factors. For relapse-free survival, similar results were obtained for overall survival in the univariate analysis, and lymphatic invasion and HER2 status were statistically significant in multivariate analysis.

In the triple-negative subgroup, p53 status had statistically significant independent prognostic value for relapse-free survival [$P = 0.013$, RR 5.4 (1.4–20.8)], but, for overall survival, p53 status was significant only in the univariate analysis (Table 3). In the non-triple-negative subgroup, vein invasion was a prognostic factor for overall survival and tumor size, and differentiation was a prognostic factor for relapse-free survival (Table 4).

DISCUSSION

The class discovery expression profile studies pioneered by the Stanford group (26–28) have demonstrated that the well-established morphological and immunohistochemical phenotypic heterogeneity of breast cancer can be confirmed and systematically reclassified into five main groups at the transcription level (26,27). Given that basal-like cancers are preferentially ER-, PR- and HER2-, it has been claimed that the basal-like tumors composed almost entirely of triple-negative

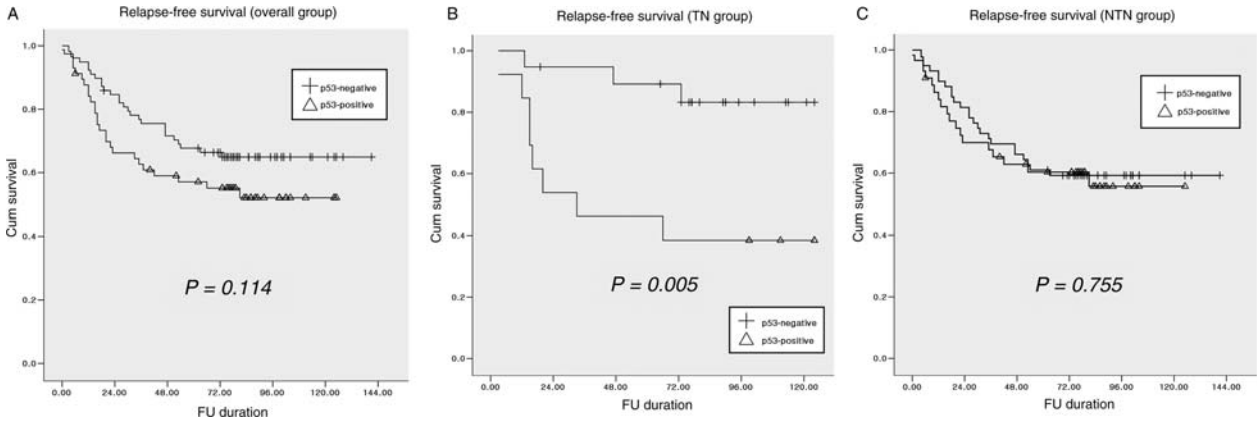

Figure 2. Kaplan–Meier relapse-free survival curve. (a) Overall group, (b) triple-negative group and (c) non-triple-negative group.

Table 2. Cox proportional hazard analysis results for the overall patients group

	Overall survival (overall group)			Relapse-free survival (overall group)		
	Univariate	Multivariate	RR (95% CI)	Univariate	Multivariate	RR (95% CI)
Age	0.447			0.357		
Menopause	0.557			0.627		
T	0.296			0.029	0.753	
N	<0.001	0.091		<0.001	0.255	
Stage	<0.001	0.933		<0.001	0.629	
H-grade	0.331			0.080		
N-grade	0.020	0.621		0.017	0.573	
N invasion	0.246			0.065		
V invasion	<0.001	0.010	3.0 (1.3–7.1)	0.001	0.207	
L invasion	0.001	0.002	46.7 (4.1–529.0)	<0.001	0.001	27.7 (3.8–200.3)
Differentiation	0.032	0.919		0.007	0.295	
ER	0.446			0.483		
PR	0.443			0.253		
HER2	0.023	0.544		0.006	0.011	2.4 (1.2–4.7)
TN	0.635			0.509		
p53	0.185			0.118		

RR, relative risk; CI, confidence interval; T, tumor stage; N, nodal stage; H-grade, histologic grade; N-grade, nuclear grade; N invasion, neural invasion; V invasion, vein invasion; L invasion, lymphatic invasion; TN, triple negativity.

phenotype could reliably be used as a surrogate for basal-like breast cancer (18). Despite the controversy regarding the similarities between basal-like and triple-negative cancers, the latter group is of clinical relevance, as chemotherapy is currently the only modality of systemic therapy available for patients with triple-negative cancers. Triple-negative cancers display more aggressive clinical behavior, distinctive metastatic patterns and poorer prognosis when compared with other breast cancer subtypes (22). Thus, it is of the highest importance to elucidate prognostic factors and key biomarkers of triple-negative cancers. To this end, we presently sought to evaluate the relevance of p53 in triple-negative breast cancers in comparison to non-triple-negative cancers.

Heterogeneity of breast cancer molecular subclasses among studies could account for the heterogeneity of results when the predictive value of a single biomarker is investigated (25,29). In addition, p53+/triple-negative tumors exhibit a higher rate of pCR (22%) when compared with both p53−/triple-negative (10%) and non-triple-negative tumors in neoadjuvant settings (25,29). Likewise, the present results implicate p53 status as having prognostic value in the treatment of triple-negative breast cancer using adjuvant

Table 3. Cox proportional hazard analysis results for the triple-negative subgroup

	Overall survival (TN group)			Relapse-free survival (TN group)		
	Univariate	Multivariate	RR (95% CI)	Univariate	Multivariate	RR (95% CI)
Age	0.940			0.631		
Menopause	0.444			0.434		
T	0.898			0.642		
N	0.039	0.359		0.481		
Stage	0.835			0.945		
H-grade	0.585			0.849		
N-grade	0.331			0.144		
N invasion	0.241			0.299		
V invasion	<0.006	0.069		0.354		
L invasion	0.048	0.039	11.5 (1.1–116.6)	0.087		
Differentiation	0.998			0.895		
p53	0.049	0.113		0.013	0.013	5.4 (1.4–20.8)

Table 4. Cox proportional hazard analysis results for the non-triple-negative subgroup

	Overall survival (NTN group)			Relapse-free survival (NTN group)		
	Univariate	Multivariate	RR (95% CI)	Univariate	Multivariate	RR (95% CI)
Age	0.479			0.497		
Menopause	0.569			0.720		
T	0.167			0.020	0.005	23.3 (2.9–189)
						2.1 (0.5–8.7)
N	0.013	0.691		0.004	0.716	
Stage	<0.001	0.527		<0.001	0.181	
H-grade	0.116			0.028	0.639	
N-grade	0.073			0.116		
N invasion	0.452			0.121		
V invasion	<0.001	0.044	2.6 (1.0–6.6)	0.002	0.067	
L invasion	0.008	0.919		0.003	0.942	
Differentiation	0.003	0.307		0.001	0.015	1.4 (0.2–9.1)
						1.5 (0.5–4.9)
p53	0.784			0.756		

NTN, non triple negative.

anthracycline-containing chemotherapy. However, the present study differs from previous observations in several ways. First, our study was done in the adjuvant setting, so an immediate response could not be discerned. Second, the outcome of anthracycline-containing chemotherapy differs from that reported previously (29). Our results indicate that overall survival and relapse-free survival rate of patients overexpressing p53 are worse than patients harboring triple-negative breast cancer cells. In other words, p53 in the triple-negative breast cancer was a poor prognostic factor in our study. In thinking about the past inconsistencies of study results, we are prompted by our present observations to suggest that p53-positive, triple-negative breast cancer carries a poor long-term outcome, even if the cancer displays an initially higher response rate for anthracycline-containing regimens. This paradox is consistent with the data suggesting

that the result of higher sensitivity to neoadjuvant anthracycline in subtypes known to have a poor prognosis is explained by the high relapse among those with residual disease (30), and that triple-negative phenotype is associated with shorter survival despite being associated with a higher response rate to neoadjuvant chemotherapy (31).

There have been many studies concerning the predictive role of p53 for anthracyclines (2–8). However, most of these were preclinical studies or were in a neoadjuvant setting. The present study is the first to evaluate the subclass-specific prognostic value of p53 for the outcome of adjuvant anthracycline-containing chemotherapy. In contrast to previous observations (23), our results demonstrate that p53 expression rate of triple-negative breast cancer is similar to non-triple-negative cancer (40.6% versus 42.7%). A principle explanation for the discrepancies reported to date concerns the various methods used to assess p53 status. Other explanations (and limitations) for our findings are that our sample size was small and involved a retrospective examination. Nevertheless, the present and previous studies agree that the predictive role of p53 involves a complex interplay between the genetic background and molecular classification.

In conclusion, we have found that p53 status is a strong prognostic factor for relapse-free survival and overall survival only for the triple-negative group in patients treated with adjuvant anthracycline-containing chemotherapy. Under these treatment conditions, expression of p53 could provide information concerning a poor outcome in triple-negative breast cancer. In such cases, consideration might well be given to more aggressive or alternative treatment such as Bevacizumab or dasatinib. There is no definite answer to optimal management of triple-negative tumors at this moment. However, this is a field that is rapidly evolving and evidence-based answers may emerge in the near future.

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Conflict of interest statement

None declared.

References

1. Early Breast Cancer Trialists' Collaborative Group (EBCTCG). Effects of chemotherapy and hormonal therapy for early breast cancer on recurrence and 15-year survival: an overview of the randomised trials. *Lancet* 2005;365:1687–717.
2. Makris A, Powles TJ, Dowsett M, Osborne CK, Trott PA, Fernando IN, et al. Prediction of response to neoadjuvant chemoendocrine therapy in primary breast carcinomas. *Clin Cancer Res* 1997;3:593–600.
3. Berns EM, Foekens JA, Vossen R, Look MP, Devilee P, Henzen-Logmans SC, et al. Complete sequencing of TP53 predicts poor response to systemic therapy of advanced breast cancer. *Cancer Res* 2000;60:2155–62.
4. Geisler S, Lonning PE, Aas T, Johnsen H, Fluge O, Haugen DF, et al. Influence of TP53 gene alterations and c-erbB-2 expression on the response to treatment with doxorubicin in locally advanced breast cancer. *Cancer Res* 2001;61:2505–12.
5. Kandioler-Eckersberger D, Ludwig C, Rudas M, Kappel S, Janschek E, Wenzel C, et al. TP53 mutation and p53 overexpression for prediction of response to neoadjuvant treatment in breast cancer patients. *Clin Cancer Res* 2000;6:50–6.
6. MacGrogan G, Mauriac L, Durand M, Bonichon F, Trojani M, de Mascarel I, et al. Primary chemotherapy in breast invasive carcinoma: predictive value of the immunohistochemical detection of hormonal receptors, p53, c-erbB-2, MiB1, pS2 and GST pi. *Br J Cancer* 1996;74:1458–65.
7. Niskanen E, Blomqvist C, Franssila K, Hietanen P, Wasenius VM. Predictive value of c-erbB-2, p53, cathepsin-D and histology of the primary tumour in metastatic breast cancer. *Br J Cancer* 1997;76:917–22.
8. Rozan S, Vincent-Salomon A, Zafrani B, Validire P, De Cremoux P, Bernoux A, et al. No significant predictive value of c-erbB-2 or p53 expression regarding sensitivity to primary chemotherapy or radiotherapy in breast cancer. *Int J Cancer* 1998;79:27–33.
9. Mathieu MC, Koscielny S, Le Bihan ML, Spielmann M, Arriagada R. p53 protein overexpression and chemosensitivity in breast cancer. Institut Gustave-Roussy Breast Cancer Group. *Lancet* 1995;345:1182.
10. Clahsen PC, van de Velde CJ, Duval C, Pallud C, Mandard AM, Delobelle-Deroide A, et al. p53 protein accumulation and response to adjuvant chemotherapy in premenopausal women with node-negative early breast cancer. *J Clin Oncol* 1998;16:470–9.
11. Colleoni M, Orvieto E, Nola F, Orlando L, Minchella I, Viale G, et al. Prediction of response to primary chemotherapy for operable breast cancer. *Eur J Cancer* 1999;35:574–9.
12. Faneyte IF, Schrama JG, Peterse JL, Remijnse PL, Rodenhuis S, van de Vijver MJ. Breast cancer response to neoadjuvant chemotherapy: predictive markers and relation with outcome. *Br J Cancer* 2003;88:406–12.
13. Alsner J, Yilmaz M, Guldberg P, Hansen LL, Overgaard J. Heterogeneity in the clinical phenotype of TP53 mutations in breast cancer patients. *Clin Cancer Res* 2000;6:3923–31.
14. Pharoah PD, Day NE, Caldas C. Somatic mutations in the p53 gene and prognosis in breast cancer: a meta-analysis. *Br J Cancer* 1999;80:1968–73.
15. Bauer KR, Brown M, Cress RD, Parise CA, Caggiano V. Descriptive analysis of estrogen receptor (ER)-negative, progesterone receptor (PR)-negative, and HER2-negative invasive breast cancer, the so-called triple-negative phenotype: a population-based study from the California cancer Registry. *Cancer* 2007;109:1721–8.
16. Carey LA, Dees EC, Sawyer L, Gatti L, Moore DT, Collichio F, et al. The triple negative paradox: primary tumor chemosensitivity of breast cancer subtypes. *Clin Cancer Res* 2007;13:2329–34.
17. Haffty BG, Yang Q, Reiss M, Kearney T, Higgins SA, Weidhaas J, et al. Locoregional relapse and distant metastasis in conservatively managed triple negative early-stage breast cancer. *J Clin Oncol* 2006;24:5652–7.
18. Dent R, Trudeau M, Pritchard KI, Hanna WM, Kahn HK, Sawka CA, et al. Triple-negative breast cancer: clinical features and patterns of recurrence. *Clin Cancer Res* 2007;13:4429–34.
19. Foulkes WD, Metcalfe K, Hanna W, Lynch HT, Ghadirian P, Tung N, et al. Disruption of the expected positive correlation between breast tumor size and lymph node status in BRCA1-related breast carcinoma. *Cancer* 2003;98:1569–77.
20. Harris LN, Broadwater G, Lin NU, Miron A, Schnitt SJ, Cowan D, et al. Molecular subtypes of breast cancer in relation to paclitaxel response and outcomes in women with metastatic disease: results from CALGB 9342. *Breast Cancer Res* 2006;8:R66.
21. Morris GJ, Naidu S, Topham AK, Guiles F, Xu Y, McCue P, et al. Differences in breast carcinoma characteristics in newly diagnosed African-American and Caucasian patients: a single-institution compilation compared with the National Cancer Institute's Surveillance, Epidemiology, and End Results database. *Cancer* 2007;110:876–84.
22. Rakha EA, El-Sayed ME, Green AR, Lee AH, Robertson JF, Ellis IO. Prognostic markers in triple-negative breast cancer. *Cancer* 2007;109:25–32.
23. Tan DS, Marchia C, Jones RL, Savage K, Smith IE, Dowsett M, et al. Triple negative breast cancer: molecular profiling and prognostic impact

in adjuvant anthracycline-treated patients. *Breast Cancer Res Treat* 2008;111:27–44.
24. Andre F, Pusztai L. Heterogeneity of breast cancer among patients and implications for patient selection for adjuvant chemotherapy. *Pharm Res* 2006;23:1951–8.
25. Andre F, Pusztai L. Molecular classification of breast cancer: implications for selection of adjuvant chemotherapy. *Nat Clin Pract Oncol* 2006;3:621–32.
26. Perou CM, Sorlie T, Eisen MB, van de Rijn M, Jeffrey SS, Rees CA, et al. Molecular portraits of human breast tumours. *Nature* 2000;406:747–52.
27. Sorlie T, Perou CM, Tibshirani R, Aas T, Geisler S, Johnsen H, et al. Gene expression patterns of breast carcinomas distinguish tumor subclasses with clinical implications. *Proc Natl Acad Sci USA* 2001;98:10869–74.
28. Sorlie T, Tibshirani R, Parker J, Hastie T, Marron JS, Nobel A, et al. Repeated observation of breast tumor subtypes in independent gene expression data sets. *Proc Natl Acad Sci USA* 2003;100:8418–23.
29. Bidard FC, Matthieu MC, Chollet P, Raoefils I, Abrial C, Domont J, et al. p53 status and efficacy of primary anthracyclines/alkylating agent-based regimen according to breast cancer molecular classes. *Ann Oncol* 2008;19:1261–5.
30. Carey LA, Dees EC, Sawyer L, Gatti L, Moore DT, Collichio F, et al. The triple negative paradox: primary tumor chemosensitivity of breast cancer subtypes. *Clin Cancer Res* 2007;13:2329–34.
31. Keam B, Im SA, Kim HJ, Oh DY, Kim JH, Lee SH, et al. Prognostic impact of clinicopathologic parameters in stage II/III breast cancer treated with neoadjuvant docetaxel and doxorubicin chemotherapy: paradoxical features of the triple negative breast cancer. *BMC Cancer* 2007;7:203.