Impact of Tumor Biology, Particularly Triple-negative Status, on Response to Pre-operative Sequential, Dose-dense Epirubicin, Cyclophosphamide Followed by Docetaxel in Breast Cancer

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Abstract. *Background: The central objective of this study was to determine the predictive impact of several established tumor biological factors (PgR, ER, HER2 and Ki-67) on response to pre-operative chemotherapy in primary breast cancer. Patients and Methods: 59 primary M0 breast cancer patients received pre-operative sequential dose-dense epirubicin and cyclophosphamide followed by docetaxel (19 patients at dosage 100 mg/m^2, 40 patients at 75 mg/m^2). Results: Pathological complete remission (pCR) occurred in 17 patients (29%) and at least partial remission in 42 (71%). Higher proliferation (Ki-67) and lack of hormone receptors (either or both) were significant predictive factors for pCR; moreover, 8/11 (73%) patients with triple-negative tumors (HER2$^-$/ER$^-$/PgR$^-$) had pCR (p=0.001). Breast conserving surgery was achieved in 46/59 patients (78%). Hand-foot syndrome occurred in 12/19 patients treated at the higher docetaxel dosage but only 1/40 of the remaining patients. Higher docetaxel dosage was associated with improved pCR in the non-triple-negative subgroup. Conclusion: The tumor biology of hormone receptor-negative, especially triple-negative, and highly proliferating breast cancer is associated with strongly positive response to dose-dense, pre-operative epirubicin/cyclophosphamide/docetaxel chemotherapy.*

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Currently, pre-operative therapy strategies in breast cancer are motivated primarily by the aim of down-staging the primary tumor, thus increasing the proportion of patients eligible for breast-conserving surgery. Although randomized trials do not show a general survival advantage for a given chemotherapy regimen administered pre-operatively *vs*. postoperatively (1-6) among those patients receiving primary chemotherapy, the survival advantage of pathological complete remission (pCR) is established (1, 2, 4, 7-11). Hence, improvements in prediction of response to pre-operative chemotherapy regimens could lead to correspondingly refined patient selection for treatment strategies and, ultimately, to a net survival benefit.

In order to address these issues, there has been mounting interest in the impact of clinical and especially tumor biological factors on response to pre-operative chemotherapy. Warm *et al*. (12) found that response (including detailed partial responses) to pre-operative epirubicin and paclitaxel chemotherapy was favorably associated with low levels of progesterone (PgR) or estrogen receptor (ER), high Ki-67 tumor cell percentage, and Her2/neu overexpression, consistent with previous findings (13, 14), particularly the GEPARTRIO study (14).

In a recent large randomized trial, 'triple-negative' (15), high-risk breast cancer patients derived particular survival benefit from dose intensification of adjuvant chemotherapy (epirubicin and cyclophosphamide followed by tandem epirubicin and cyclophosphamide-thiotepa with autologous peripheral blood progenitor support). The term 'triple-negative' refers to tumors lacking HER2 overexpression and also exhibiting negative PgR and ER receptor status; their occurrence is strongly associated with a 'basal' type cancer signature (16, 17). There has been increased focus on optimizing treatment for patients in this category, particularly since these tumors are responsive neither to anti-endocrine nor to trastuzumab therapy.

These and other findings set the stage for the current investigation of the predictive impact of clinical and tumor biological factors (estrogen and progesterone hormone receptors, HER2/neu, and Ki-67) on response to dose-dense sequential epirubicin and cyclophosphamide followed by docetaxel chemotherapy in a representative sample of primary breast cancer patients. This scheme has been developed in the pre-operative setting based on its chemotherapeutic action on the tumor (18-20), and evidence from adjuvant trials (21-23). Efficacy of 4×anthracycline combined with cyclophos-phamide, followed by 4×taxanes in pre-operative breast cancer chemotherapy has been demonstrated in several randomized studies (24-26). The anthracycline-based agent epirubicin is widely used in Germany for the treatment of both primary and metastasized breast cancer, as well as for ovarian cancer and soft-tissue sarcoma (27). Side-effects of epirubicin, while less intensive than those of doxorubicin (28), include hematological and cardiac toxicity, as well as mucositis, nausea and vomiting, reversible hair loss and local skin reactions. Side-effects of cyclophosphamide include fatigue, bone marrow suppression, gastro-intestinal complaints (nausea/vomiting, stomach ache, diarrhea), darkening of the skin/nails and alopecia.

Adjuvant taxanes have improved overall survival (OS) in several studies (29, 30), in particular the Geicam study (31, 32). The side-effects include allergic reactions, bone marrow toxicity and cardiac toxicity, as well as possible cumulative liver complications such as neutropenia, leukocytopenia, thrombocytopenia, anemia, and neurological toxicity. Dose-dense application of docetaxel has been shown to improve DFS and OS in the adjuvant setting (33, 34); enhanced efficacy could be due to the minimization of the intercycle tumor cell growth (35-38). In view of the relationship between long-term survival and pathological complete remission (pCR) following pre-operative breast cancer therapy, the main objective of this study was to identify predictive factors under the above therapy scheme for the primary endpoint pCR; additional endpoints involved partial remission, breast conserving surgery, and safety. Ultimately, the goal is to identify the potential for improvement of survival by individualized pre-operative treatment modalities.

Patients and Methods

This prospective, nonrandomized, phase II study was conducted in the Department of OB/GYN at Cologne University on 59 female primary breast cancer patients of median age 50 years (18-65) with histologically proven, advanced (at least cT2), but metastasis-free disease. Patients entered the study between April 2003 and August 2004. Follow-up was performed at the study center. The study was approved by the Institutional Ethics Committee, and all patients gave written informed consent. Exclusion criteria included pregnancy, corticosteroids, motor or sensory neurotoxicity (grade 2 or worse), heart disease, infection, ulcers, diabetes, previous malignancy, previous cytostatic, immune, radiation, or anti-estrogen cancer treatment; adequate liver function and blood levels (neutrophils $\geq 1.5\times10^9$/l, thrombocytes $\geq 100\times10^9$/l, hemoglobin ≥10 g/dl) and ECOG performance status ≤2 were also mandatory.

Breast cancer diagnostics prior to chemotherapy included ultrasound, magnetic resonance imaging (MRI), and 3 or more high-speed core biopsies, which were histologically classified and used for immunohistochemical (IHC) testing, including ER, PgR, Ki-67 tumor cell percentage, HER2/neu overexpression, and tumor grade. ER status was classified positive for immunoreactive score ≥2 or for positive reaction of >10% of tumor cells, and similarly for PgR status. HER2/neu overexpression was considered positive for IHC score 3; amplification status of tumors with IHC score 2 was determined by FISH. Tumor grade 'between 2 and 3' was coded as 2.5.

The protocol consisted of eight cycles of chemotherapy every two weeks, beginning with four cycles of epirubicin (90 mg/m^2) and cyclophosphamide (600 mg/m^2), followed by four dose-dense cycles of docetaxel, of which the first 19 patients were scheduled to receive 100 mg/m^2 ('higher docetaxel dosage arm') and the next 40 patients received 75 mg/m^2 ('lower docetaxel dosage arm'); the arms are defined by intention to treat. The docetaxel reduction was initiated because of grade 3 hand-foot syndrome in 10/19 patients (details see below).

The protocol included supporting measures aimed to reduce fatigue and to maintain tumor oxygenation, the reduction of which is thought to be associated with possible selection pressure toward chemo-resistant tumor cells (39, 40). Two days after the start of anthracycline-containing chemotherapy, patients were to receive pegfilgrastim (Neulasta®) to support granulopoiesis. Erythropoiesis was supported by erythropoetin alfa (Aranesp®) in patients with hemoglobin levels dropping below 12 g/dl.

Clinical and sonographic examinations were performed every second chemotherapy cycle, imaging (MRT and mammography as required) after 8 cycles. According to the protocol, pre-operative therapy was to be continued as long as minimal response occurred (with acceptable toxicity) and was to be discontinued if progression was determined after 2 or 4 cycles.

Surgery was performed about three weeks after the last chemotherapy cycle; patients who were considered appropriate candidates for breast conserving surgery were offered segmental mastectomy (lumpectomy). Patients who were considered inappropriate for breast-conserving surgery or who did not desire it underwent total mastectomy. Adjuvant therapy was administered according to clinicopathological and tumor biological variables.

Pre-operative tumor size was defined as the maximum dimension obtained from all imaging techniques performed (mammography, breast sonography and MRT). Post-operative tumor size was determined by the pathologist. Responses were initially classified according to standard WHO criteria. In particular, classification as pCR implied absence of any visible tumor tissue (including lymph node metastasis or microinvasive tumor remnants) as assessed by the pathologist); 'pPR' implied at least 50% reduction in all detected malignant lesions (measured in at least 4-week intervals) and no signs of progression in any lesions.

Predictive impacts of factors on response were assessed by univariate (and multivariate) logistic regression and expressed in

Table I. *Characteristics of the patient collective.*

Factor	Status	N	%
ER	Negative	18	31%
	Positive	41	69%
PgR	Negative	24	41%
	Positive	35	59%
HR (PgR or ER positive)	Negative	16	27%
	Positive	43	73%
HER2 overexpression	No	42	71%
Triple negative	Yes	17	29%
	Yes	11	19%
	No	48	81%
Grade	1-2	41	69%
	>2	17	29%
	Unknown	1	2%

Factor	Mean	Standard dev.	N	25th percentile	Median	75th percentile
Ki-67 (% of cells)	25	23	57	10	15	30
Age (years)	50	9	59	43	50	57
Tumor size pre-op (mm)	33	14	59	23	30	38

Primary treatment data	Category	N	%
Chemotherapy (intention to treat)	EC+Doc 100 mg/m^2	19	32%
	EC+Doc 75 mg/m^2	40	68%
	4×EC+4×Doc	53	90%
Cycles	4×EC+3×Doc	1	2%
	4×EC+2×Doc	3	5%
	4×EC+1×Doc	2	3%
Surgery	BCS	24	41%
	MRM	35	59%

terms of unadjusted (and adjusted) odds ratios (OR) as appropriate. The outcome variables for logistic regression were pCR and pPR|pCR (objective remission rate, meaning either pCR or pPR), both coded as binary variables (1: favorable outcome, 0: unfavorable outcome). All status variables (ER, PgR, HER2 overexpression, hormone receptors, triple negativity), as well as docetaxel dosage (100 *vs*. 75 mg/m^2) arms (defined by intention to treat) were coded as binary variables. Grade was entered in models both as a metric variable (including grade 2.5) and as a binary variable (>2 *vs*. ≤2). For the metric variables age, tumor size, and Ki-67 percentage were coded as fractional ranks and entered in logistic regression as continuous variables; hence, no 'optimal cutoffs' were used. To give an intuitive idea of effect size, odds ratios for fractionally ranked continuous variables are reported as a comparison of 75th *vs*. 25th percentile (or *vice versa*).

Distribution-free tests (and/or rank statistics) were also applied as follows: the Mann-Whitney test was used to test for differences in non-normally distributed ordinal or metric variables (such as tumor size or Ki-67) between two sub-groups defined by a binary variable such as pCR, dosage, *etc*. Spearman correlations were computed between factors. Chi-squared or Fisher's exact test were also used for evaluating relationships between categorical variables (*e.g*. dosage *vs*. side-effects). Predictive impact was also characterized in terms of sensitivity, specificity, and positive predictive values where appropriate.

Table II. *Significant Spearman correlations between factors. Age and HER2 had no significant correlations with PgR, ER, Ki-67, grade, or tumor size. The associations of higher Ki-67 with negative ER, PgR, or HR receptor status were also significant by the Mann-Whitney test with p<0.001.*

Spearman correlation	ER	Ki-67	Grade	Tumor size
PgR	0.613	–.445	–0.288	*n.s.*
(*p*-value)	<0.001	.001	0.029	
ER		–.509	–.257	0.310
(*p*-value)		<0.001	0.051	0.022
Ki-67	–0.509		0.432	*n.s.*
(*p*-value)	<0.001		0.001	

Results

Characteristics of the patient collective. Frequency distributions of the explanatory clinical and tumor biological factors of the patient collective and of treatment variables are summarized in Table I. Although the study was not designed as a randomized trial of docetaxel dosage, there were no statistically significant imbalances in any of these factors between the docetaxel dosage arms (defined by intention to treat).

A look at the joint distribution of factors will be useful for understanding their impact on endpoints: Table II summarizes Spearman correlations between the predictive factors in our collective, which were typical for primary breast cancer. Age and HER2 overexpression were not significantly correlated with any of the other factors. PgR and ER were (as usual) strongly correlated with each other, and both were rather strongly negatively correlated with proliferation marker Ki 67 (*i.e*. negative hormone receptor status was associated with higher proliferation); PgR and ER were weakly negatively correlated with tumor grade (viewed as a metric variable). Tumor size was weakly correlated with positive ER. Grade and Ki-67 exhibited a moderately positive correlation.

Safety and side-effects. There were no severe adverse events, such as grade 3 or 4 cardiotoxicity or neutropenia. Reversible alopecia occurred in nearly all patients in both dosage arms. Nausea affected 12 patients (2 severely) and did not significantly depend on docetaxel dosage; third-degree hand-foot syndrome occurred in 12/19 patients receiving 100 mg/m^2 docetaxel, but only 1/40 of the remaining patients

Table III. *a: Univariate impact of factors on endpoints pCR and pPR/pCR (meaning 'at least pPR') expressed as odds ratios (OR) as calculated by univariate logistic regression; 95% lower/upper confidence levels (LCL/UCL) are reported for the log odds ratios (i.e., conversion to OR confidence intervals is obtained by exponentiation). The odds ratios refer to the indicated level or category vs. the indicated reference; e.g., patients with triple negative tumors had OR=11.56 for pCR compared to the rest; patients with tumor size at 25th percentile (23 mm) had OR=3.25 for pCR compared to those with tumor size at 75th percentile (38 mm). In the table, the odds ratios reported for grade were obtained by entering grade as a metric variable with values (2, 2.5, 3). b: Multivariate model for pCR (only univariate models for pPR/pCR were obtained).*

a

Factor	Univariate							
	pCR				pCR/pPR			
	Log OR	LCL UCL	*P*-Value	OR	Log OR	LCL UCL	*P*-Value	OR
Docetaxel dosage (100 *vs.* 75 mg/m^2)	-	-	-	-	-	-	-	-
Triple negative status	2.45	0.94 3.96	0.002	11.56	-	-	-	-
Negative ER (*vs.* positive)	1.80	0.57 3.04	0.004	6.07	1.53	0.29 2.76	0.06*	4.62
Negative PgR (*vs.* positive)	1.79	0.55 3.03	0.005	6.00	2.11	0.87 3.35	0.01	8.25
Negative HR (*vs.* positive)	1.73	0.48 2.98	0.007	5.62	2.18	0.93 3.44	0.04	8.89
High Ki-67 (75th *vs.* 25th percentile)	1.77	0.50 3.04	0.006	5.87	1.34	0.07 2.61	0.02	3.82
Small tumor (25th *vs.* 75th percentile)	1.18	0.09 2.27	0.033	3.25	-	-	-	-
HER2 overexpression	-	-	-	-	-	-	-	-
Grade* (metric)	1.95	0.47 3.43	0.01	7.01	2.19	0.12 4.25	0.038	8.91
Age	-	-	-	-				

*Grade coded as a binary variable >2 *vs.* ≤2 was also univariately significant for pCR with log OR=1.70 (0.45-2.95, p=0.008).

b

Factor	Multivariate			
	pCR			
	Log OR	LCL UCL	*p*-Value	OR
Triple negative status	2.38	0.79 3.98	0.003	10.84
Grade (metric)	1.72	0.07 3.36	0.041	5.56

(p<0.001). Due to the side-effects, three of the patients scheduled for 100 mg/m^2 docetaxel discontinued pre-operative therapy with docetaxel (one following the first cycle and two following the third cycle); docetaxel was reduced from 100 mg/m^2 to 75 mg/m^2 after one cycle in 8/19 patients, after 2 cycles in 3/19, and after 3 cycles in one patient. One patient underwent surgery for a perforated duodenal ulcer, but elected to continue pre-operative therapy (with reduced dexamethasone). As a consequence of the observed side effects, which amount to a dose-limiting toxicity of the taxane, an amendment to the study protocol was approved by the Ethics Committee for the remaining patients in the study: i) docetaxel 75 mg/m^2; ii) growth factor (pegfilgrastim administration) was then left to physician discretion. No other protocol details were affected.

Of the 40 patients scheduled to receive docetaxel at 75 mg/m^2 (intention to treat), 36 completed all 8 chemotherapy cycles (including 4×docetaxel); 3 completed 6 cycles (2×docetaxel), and one completed 5 cycles (1×docetaxel); however, none of these 4 discontinuations were necessitated by medical indication.

Univariate predictors of response. Of the 59 patients, there were 17 pCR (29%), 25 pPR (42%), 9 minimal responses (15%), 7 stable disease responses (12%) and only one progression (2%). Breast conserving surgery was achieved in 24 patients (41%).

Table IIIa describes the impact of factors on endpoints pCR and pPR/pCR in terms of an unadjusted OR obtained by univariate logistic logistic regression. The table lists 95% lower/upper confidence levels (LCL/UCL) for log OR rather than for the OR itself; confidence intervals on the OR are easily computed by exponentiation of the LCL and UCL. Triple-negative status (HER2$^-$/ER$^-$/PgR$^-$) was the strongest predictor of pCR, with an OR of almost twelve (compared to patients who were positive for either HER2, ER, or PR); 8/11 (73%) of the patients with triple-negative tumors had pCR, compared to 9/48 (19%) of others (p=0.001); all but one patient with triple-negative tumor had at least pPR. Negative ER, PgR, or HR (*i.e.* ER and PgR both negative) were all predictive of pCR, with odds ratios of about 6; 12/24 PgR$^-$ compared to 5/35 ER$^+$ (p=0.003), 10/18 ER$^-$ compared to 7/41 ER$^+$ (p=0.003), and 9/16 HR$^-$ compared to 8/43 HR$^+$ (p=0.005) had pCR. There were 4/10 (40%) pCR among patients with 'mixed' hormone receptor status.

High proliferation as characterized by Ki-67 percentage was favorably associated with pCR: *e.g.*, a patient with Ki-67 at the 75th percentile (30%) had an OR of nearly 6 for pCR compared to a patient at the 25th percentile (10%). Poor tumor grade was also significantly associated with better pCR, whether coded as a 'metric' variable or as a binary variable. Recall that poor grade was associated with higher Ki-67 (proliferation) and negative receptor status.

Small tumor size also favorably affected the odds of pCR: A hypothetical patient with tumor size at the 25th percentile (23 mm) would have OR=3.25 compared to a patient with tumor size at the 75th percentile (38 mm). HER2 expression (alone) and age were not significant predictors of pCR. Negative ER status (16/18), negative PgR status (22/24), and negative HR status (15/16) were all significantly associated with pPR/pCR (partial or complete remission) with OR ranging from 4 to almost 9. Tumor grade was also a significant univariate factor for pPR/pCR. Breast-conserving surgery was achieved in 46/59 patients (78%). None of the predictive factors had a significant impact on breast-conserving surgery, nor did response itself (pCR, pPR/pCR).

Multivariate and subgroup results. Both triple-negative status and tumor grade were independent factors in multivariate logistic regression for the outcome pCR (Table IIIb). This independence appears remarkable at first glance, in that tumor grade is correlated with negative receptors, but is less so upon closer evaluation: the multivariate independence of tumor grade as a predictor of pCR is attributable to the patients who were not triple negative. It is worth mentioning that that if one combines these two factors into a simple binary variable (=1 if patient is either triple negative or grade=3, or both; 0 otherwise), one obtains in-sample sensitivity of 71% (12/17), specificity of 88% (37/42), and positive predictive value of 71% (12/17) for prediction of pCR (p<0.001). Ki-67 was not significant for pCR in the subgroup of HR$^-$ or non-triple negative patients (nor in multivariate analysis including any of the hormone receptors, data not shown).

Although not significant for pCR among all patients, higher dosage of docetaxel was associated with somewhat improved pCR in the subgroup of non-triple-negative patients, with 6/17 pCR at 100 mg/m^2 *vs*. 3/31 pCR at 75 mg/m^2 docetaxel (p=0.03) (OR=5.1, CI 1.1-24.0).

Discussion

Pre-operative sequential, dose-dense epirubicin and cyclophosphamide followed by docetaxel as administered in this study led to 29% PCR and 71% complete or partial remission together with 78% breast-conserving surgery. These relatively favorable response rates in our (smaller) collective as a whole are consistent with the results of the NSABP B-27 study for the same sequence of chemotherapeutic agents (26). Improvement of remission in pre-operative chemotherapy by augmenting anthracycline-containing regimens with a taxane has been confirmed in several large trials (5, 14, 24, 25).

Prospective randomized pre-operative chemotherapy studies (5, 25, 41) have reported pCR rates about 3-4 times higher in HR-negative tumors than in HR-positive tumors). The corresponding ratio of about 3 (OR=about 6) of the present study confirms the predictive impact of HR$^-$ status for this regimen. Thus, in view of the possible survival advantage for patients achieving pCR, pre-operative sequential, dose-dense epirubicin, cyclophosphamid followed by docetaxel chemotherapy would appear to be a strong alternative in metastasis-free patients with tumors classified cT2 or worse and negative ER and PgR status. The predictive impact of a negative ER status and a negative PgR status on their own was comparable to that of negative HR in terms of the odds ratio; this would suggest that pre-operative chemotherapy might also be an option for patients with 'mixed' hormone receptor status (4/10 had pCR). The evidence (42, 43) points to a favorable response to either pre-operative endocrine therapy or pre-operative chemotherapy for patients with negative PgR status and positive ER status. However, possible benefits of a combination of both therapy regimens in the pre-operative setting for this patient group are not yet established; a definitive statement on pre-operative therapy for mixed receptor status should await larger trials, particularly in view of possible endocrine therapy options.

A significant predictive impact of proliferation as measured by Ki-67 on pCR has been reported in previous pre-operative chemotherapy studies (44, 45). A predictive role of Ki-67 was also found by Warm *et al*. (12). In the present study, the predictive impact of Ki-67 on pCR was strong, corresponding to an OR of about 6 for a tumor with Ki-67 at the 75th percentile compared to one with Ki-67 at the 25th percentile. Our results for pre-operative sequential, dose-dense epirubicin and cyclophosphamide followed by docetaxel confirm the generally observed positive association between high cell proliferation and 'chemosensitivity', particularly in response to pre-operative chemotherapy (46, 47).

Ideally, specific predictive relationships between tumor biological factors and probability of response to each therapy option are needed for optimal individualization of care. Considering the combinatorics of different therapy regimens and of pre-operative *vs*. adjuvant administration of each regimen in breast cancer, a more systematic understanding of why particular patient groups respond to certain pre-operative chemotherapy regimens is necessary. Different tumor biological processes important for breast cancer metastasis are known to act on different time scales, as reflected in time-varying effects of factors such as uPA/PAI-1 (48) on prognosis. The latency time to breast cancer after known exposure to ionizing radiation is inversely related to proliferation rate (49). Since some tumors proliferate quickly but are less dangerous in terms of invasion, angiogenesis, and other metastatic processes, one could hypothesize that such tumors might be excellent candidates for pre-operative therapy. This type of therapy would slow down the dominant

metastatic process in these tumors (proliferation) and help confine disease at an early stage. Ultimately, disease confinement at an early stage might be expected to contribute to a survival benefit.

The evidence from this and other studies points toward a predictive effect of proliferation markers for favorable response to pre-operative chemotherapy. This response might be strong enough to provide a long-term survival advantage to this group of patients compared to those receiving the same chemotherapy regimen in the adjuvant setting. A trial of breast cancer patients with positive proliferation markers (S-Phase fraction or Ki-67), randomized between pre-operative and adjuvant chemotherapy treatment arms would be useful.

On the other hand, since proliferation is correlated with negative receptors, the impact of Ki 67 on pCR could have been influenced by multicollinearity, since negative receptor status is also a strong pCR predictor. Indeed, Ki-67 was not a significant predictor of pCR in HR^+ or non-triple-negative subgroups, nor was it significant in multivariate logistic regression including any receptor variable or triple negativity. Hence, an intriguing issue for the future is whether proliferation is causally relevant to the tumor biological mechanism leading to favorable response in tumors with high Ki-67, or whether other characteristics associated with a receptor-negative or triple-negative phenotype are biologically more relevant.

The factors triple negativity and tumor grade were independent predictors of pCR, and the combination (triple negativity or tumor grade=3) had in-sample sensitivity of 71%, positive predictive value of 71%, and specificity of 88%. A strong predictive impact of these factors for the current dose-dense regimen (sequential epirubicin and cyclophosphamide followed by docetaxel) should be investigated in a larger prospective study and would be of considerable clinical interest if confirmed. In the current study, docetaxel dosage had impact on remission within the group of non-triple negative patients. However, the toxicities of 100 mg/m^2 docetaxel may require dose reduction in some patients.

The case for improved long-term survival from preoperative chemotherapy (compared to an equivalent adjuvant regimen) has yet to be made, with most studies showing either no significant survival difference (11) or only an inconclusive advantage for pre-operative treatment (1, 2, 4, 7-10). However, predictive factors for pathological complete response to particular therapy strategies identified in the pre-operative setting could be utilized in the design of trials with long-term survival as the primary endpoint to improve individualized breast cancer treatment and ultimately lengthen long-term survival.

In the current study, 8/11 triple-negative tumors had pCR in response to the present pre-operative chemotherapy regimen of sequential, dose-dense epirubicin and cyclophosphamide followed by docetaxel. In (50) a 40% pCR rate (86% at least pPR) was observed in triple-negative patients with T2-T3, N0-3 tumors (similar to our triple-negative subgroup) who received pre-operative chemotherapy consisting of four courses of cisplatin-containing, ECF followed by three courses of weekly paclitaxel. In the GEPARTRIO trial (14), pCR exceeding 40% was found among triple negative tumors following pre-operative TAC chemotherapy.

Conclusion

The present study highlights the predictive role of triple-negativity for response to pre-operative chemotherapy in primary breast cancer, at least for the present dose-dense regimen, and has potential implications for clinical management of this patient group: If the general correlation between pCR and long-term survival holds in this group, then the present results would support pre-operative chemotherapy in triple-negative patients, particularly since their targeted options (such as endocrine or trastuzumab therapy) are currently still limited. Moreover, given the known, very aggressive tumor biology (50) of triple-negative tumors, it may be of advantage to patients to treat them in the pre-operative setting, where their response to a particular chemotherapy regimen can be closely monitored.

Finally, if pre-operative chemotherapy of triple-negative breast cancer patients becomes an established standard, it will be important to address the issue of clinical management of those (few) triple-negative patients who fail to respond to state-of-the-art pre-operative chemotherapy. Novel targeted agents such as bevacizumab may be useful in this setting, as currently explored by the GeparQuinto protocol (51).

Competing Interests

The Authors declare that they have no competing interests

Author's Contributions

MW, EG, MS, MH, AT and PM conceived the study, and participated in its design and coordination. RK performed statistical analysis. MW, NH, and RK drafted the manuscript. All Authors read and approved the final manuscript.

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