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ORIGINAL REPORT

Phase II Genomics Study of Ixabepilone as Neoadjuvant Treatment for Breast Cancer

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The Appendix is included in the full-text version of this article, available online at www.jco.org. It is not included in the PDF version (via Adobe® Reader®).

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ABSTRACT

Purpose
This phase II study evaluated the efficacy and safety of ixabepilone as neoadjuvant therapy for invasive breast cancer not amenable to breast conservation surgery. Gene expression studies were undertaken using genes that were identified as potentially associated with sensitivity/resistance to ixabepilone in prior preclinical investigations.

Patients and Methods
Patients with invasive breast cancer ≥ 3 cm were eligible. Ixabepilone 40 mg/m^2 was administered as a 3-hour intravenous infusion on day 1 of a 21-day cycle for four or fewer cycles.

Results
One hundred sixty-one patients were treated. The overall complete pathologic response (pCR) rate was 18% in breast and 29% in estrogen receptor (ER) –negative patients. Gene expression data were available for 134 patients. ER gene expression (*ER1*) was inversely related to pCR in breast and had a positive predictive value (PPV) of 37% and negative predictive value (NPV) of 92%. A 10-gene penalized logistic regression (PLR) model developed from 200 genes predictive of ixabepilone sensitivity in preclinical experiments included ER and *tau* and had higher PPV (45%) and comparable NPV (89%) to *ER1*. Grade 3 to 4 adverse events (AEs) were reported for 32% of patients. Except for neutropenia and leukopenia, all grade 3 to 4 AEs occurred in ≤ 3% of patients. Reversible peripheral neuropathy was experienced by 3% of patients.

Conclusion
ER, microtubule-associated protein *tau*, and a 10-gene PLR model that included ER were identified as predictors of ixabepilone-induced pCR. Results indicate an inverse relation between ER expression levels and ixabepilone sensitivity. Neoadjuvant ixabepilone demonstrated promising activity and a manageable safety profile in patients with invasive breast tumors.

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INTRODUCTION

The development of novel chemotherapeutic agents has significantly improved the prognosis and survival of patients with breast cancer.[1-3] However, breast cancer is a heterogeneous disease; only certain subsets of patients respond to specific chemotherapeutic agents, whereas other patients derive little or no benefit but may be exposed to treatment-associated toxicities. One of the current challenges facing clinicians is how to select patients prospectively who will respond to specific therapeutic regimens, thus minimizing unnecessary toxicities for patients with nonresponsive disease.

Recent pharmacogenomic studies in breast cancer specimens identified gene expression profiles useful for classifying tumor subtypes or predicting response to specific therapeutic agents.[4-6] Despite this progress, many such studies have reported only retrospective correlations with clinical outcomes but failed to produce broadly applicable models for selection of therapy in patients with breast cancer.

To date, prospective analysis of genetic profiles associated with activity and toxicity of novel chemotherapeutic agents has not been performed during the early stages of drug development. For breast cancer, the neoadjuvant setting provides an opportunity to identify predictive biomarkers for novel therapeutic agents because pretreatment biopsies are readily accessible. Moreover, biomarker results obtained with tumor biopsies can be evaluated against pathologic complete response (pCR) in the breast (pCR$_B$), a surrogate end point that demonstrates strong association with disease-free and overall patient survival.[3,7,8]

Ixabepilone is the first in a new class of semi-synthetic antineoplastic agents derived from the natural epothilones and their analogs.[9,10] Ixabepilone promotes tumor cell death by stabilizing microtubules and inducing cell cycle arrest and subsequent apoptosis.[11] Ixabepilone was specifically designed to have low susceptibility to mechanisms of drug resistance present in tumor cells. Phase II clinical trials have demonstrated efficacy of ixabepilone in metastatic breast cancer, including multidrug resistant tumors,[12-15] and in other tumor types, including renal, prostate, pancreatic, and lymphoma.[16-21] A large, randomized, phase III trial comparing ixabepilone plus capecitabine versus capecitabine monotherapy demonstrated improved efficacy for the combination, with significant prolongation of median progression-free survival (5.8 months *v* 4.2 months; $P < .001$) and increased response rate (35% *v* 14%; $P < .001$).[22] A phase II study was designed to evaluate ixabepilone as neoadjuvant monotherapy for breast cancer, incorporating expression analysis of genes identified from prior preclinical investigations as potentially associated with sensitivity/resistance to ixabepilone.

PATIENTS AND METHODS

Study Participants

Women 18 years of age or older with a histologically confirmed diagnosis of T2-4, N0-3, M0 invasive breast adenocarcinoma ≥ 3 cm in diameter not amenable to breast conservation surgery (BCS) were eligible for enrollment. Patients were excluded if they had inflammatory breast cancer, neuropathy > National Cancer Institute Common Toxicity Criteria grade 1, had received prior chemotherapy or radiotherapy to the breast area, or were unfit for breast or axillary surgery. Patients gave written, informed consent before any study-related procedure.

Study Design and Treatment

This was an exploratory, single-arm, phase II, multicenter study of ixabepilone (40 mg/m^2) as neoadjuvant therapy administered as a 3-hour infusion every 21 days for up to four cycles (Fig 1). The number of cycles was selected based on median time to response previously observed with ixabepilone in the metastatic setting.

Patients began ixabepilone therapy after screening, tumor measurement, and collection of core tumor biopsy and blood samples for evaluation of gene expression. Patients with complete responses (CRs), partial responses, or stable disease (SD), assessed between each treatment cycle, continued to the next cycle. Patients with unacceptable toxicity, as determined by study investigator, or progressive disease discontinued ixabepilone. After discontinuation, they received anthracycline-containing combination chemotherapy for up to six additional cycles or local therapy (radiation therapy or surgical intervention) at the investigator's discretion. Use of growth factor for management of cytopenias was at the investigator's discretion. H_1/H_2-blockers with or without corticosteroids were administered before each ixabepilone cycle. Postsurgery, adjuvant anthracycline-based chemotherapy was recommended in addition to adjuvant radiotherapy and hormonal therapy for patients with hormone receptor–positive tumors, and administered at the investigator's discretion. This study was conducted according to International Conference of Harmonization Guidelines of Good Practice, the Declaration of Helsinki, and the institutional review board/independent ethics committee at each study site.

Study Objectives

The primary objective of this study was to analyze pretreatment expression of mRNA from breast tumors to identify potential predictors of response—defined as pCR_B—to ixabepilone in the neoadjuvant setting. Secondary objectives were to assess the rate of pCR in breast and lymph nodes (pCR_{BL}), clinical and radiologic responses, the proportion of patients able to have BCS after treatment, and the safety of ixabepilone.

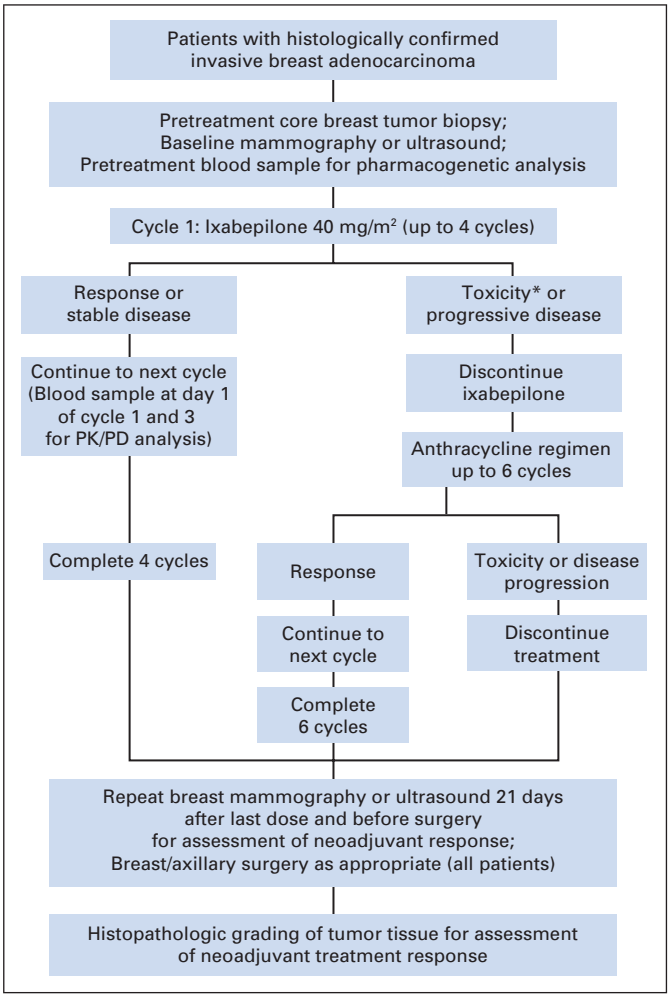

Fig 1. Study schema. (*) Toxicities requiring dose reduction or discontinuation of ixabepilone treatment. PK, pharmacokinetics; PD, pharmacodynamics.

Efficacy Assessment

Efficacy was assessed in relation to pCR_B and best overall response. Pathologic response in the primary tumor site was evaluated by the study site pathologist from the posttherapy surgical specimen using the Sataloff criteria.[23] Independent pathology experts provided central pathologic evaluations for samples from patients achieving pCR. Clinical responses were assessed by physical examination and radiographic measurements. Clinical response in the primary site and axilla was assessed after each treatment cycle. For breast lesions, mammography was the primary radiologic method; for axillary lesions, ultrasound was the primary imaging method. Clinical and radiographic assessments were performed at the end of treatment and approximately 21 days after the last treatment cycle to reassess tumor size and compare with baseline measurements. Best overall responses were determined using physical examination, radiographic examination (mammography or ultrasound), and histopathologic analysis of the primary tumor. After final assessment, patients underwent BCS or mastectomy with axillary lymph node dissection.

Efficacy analyses were conducted on subpopulations of patients with estrogen receptor (ER) –negative, ER/progesterone receptor (PR) –negative, and ER/PR/human epidermal growth factor receptor (HER2)–negative tumors. ER and PR status were defined by immunohistochemistry (IHC) performed locally at each site, using the H score system,[24] with negative cases defined as < 10% tumor cells stained. HER2-negative status was based on fluorescence in situ hybridization (FISH). If FISH results were not available, an IHC test of 0, 1+, or 2+ was classified as HER2 negative; 3+ was considered positive.

Safety Assessments

Safety was assessed throughout the study using number and severity of adverse events (AEs) and clinical laboratory test abnormalities in hematology and chemistry parameters. All were graded using the National Cancer Institute Common Toxicity Criteria version 3.0.

RNA Extraction and Gene Expression Profiling

Core biopsies were taken before treatment and were either snap-frozen or placed in RNAlater solution (Qiagen, Valencia, CA). RNA was extracted at the Karolinska Institute (Stockholm, Sweden) using RNeasy mini kit (Qiagen). Extracted RNA underwent further analysis if the $A_{260/280}$ ratio was 1.8 to 2.1 or 28S/18S ratio determined using a bioanalyzer (Agilent Technologies, Santa Clara, CA) was 0.5 to 2.0 and the total RNA yield ≥ 1 μg. Total RNA (1 μg) was used to prepare targets, as described in the manufacturer's instructions, for hybridization to Affymetrix HU133A2 GeneChip (Affymetrix, Santa Clara, CA).

Statistical Analyses

A sample size of 160 patients was estimated to provide a 90% probability of detecting a gene whose expression was associated with a two-fold increase in odds of a tumor response, assuming 5% of patients would be lost to follow-up and 10% would have insufficient quality or quantity of mRNA. The Appendix (online only) provides a detailed description of methods and statistical tests used to develop single- and multigene predictive models based on expression patterns of individual genes.[25-28] For efficacy analyses, response rate was calculated in all treated patients and exact two-sided 95% Clopper-Pearson CIs were computed.[29]

RESULTS

Patient Disposition and Demographics

Between October 15, 2003, and April 18, 2005, 164 patients were enrolled across 14 study centers in six countries; 161 patients were treated and assessable for response. All 161 treated patients were women with invasive breast adenocarcinoma with a median age of 55 years and a baseline Eastern Cooperative Oncology Group performance status of 0 to 1 (Table 1).

Table 1. Patient Baseline Demographics

Characteristic	Patients Treated (n = 161) No.	%
Age, years		
Median	55	
Range	27-79	
ECOG performance status		
0	154	95.7
1	7	4.3
ER status		
Positive	80	49.7
Negative	72	44.7
Not reported	9	5.6
PR status		
Positive	71	44.1
Negative	76	47.2
Not reported	14	8.7
HER2 status		
Positive	19	11.8
Negative	116	72.0
Not reported	26	16.1
ER/PR/HER2-negative		
Yes	42	26.1
No	119	73.9
Histopathologic subclassification		
Ductal/mammary carcinoma	125	77.6
Lobular	27	16.8
Medullary	2	1.2
Mucinous/colloidal	1	0.6
Other	6	3.7
Histologic grade		
1	20	12.4
2	73	45.3
3	38	23.6
Other	2	1.2
Not reported	28	17.4
Largest indicator/target lesion, cm		
< 2	0	0
2-5	111	68.9
> 5-10	45	28.0
> 10	2	1.2
No indicator/target lesion	3	1.9

Abbreviations: ECOG, Eastern Cooperative Oncology Group; ER, estrogen receptor; PR, progesterone receptor.

Extent of Exposure

One hundred forty-one (88%) of the enrolled patients received four cycles of ixabepilone therapy (median number of cycles, four). Of these patients, 131 (93%) were maintained at 40 mg/m^2 ixabepilone through all four cycles. Eighty-one percent of patients received $\geq$ 95% of planned dose-intensity. Median cumulative dose and median dose-intensity were 160 mg/m^2 (range, 0.4 to 170.7 mg/m^2) and 13 mg/m^2/wk (range, 0.1 to 14.2 mg/m^2/wk), respectively.

Clinical and Radiologic Responses

Clinical assessment revealed that CR, partial response, and SD were achieved in 21.1%, 55.9%, and 16.8% of patients, respectively. Radiologic assessment showed that 11.8%, 37.9%, and 38.5% of patients achieved CR, partial response, and SD, respectively. A best overall response rate of 61% was observed for this study (CR = 17%; partial response = 44%), and SD was observed in an additional 31% of patients (Table 2).

pCR

The investigator-assessed pCR_B and pCR_{BL} rates were 18% (95% CI, 12% to 25%) and 11% (95% CI, 6% to 16%), respectively (Table 2). Independent assessment of patient responses yielded pCR_B and pCR_{BL} rates of 14% (95% CI, 9% to 20%) and 9% (95% CI, 5% to 15%), respectively. In patients with ER-negative, ER/PR-negative, and ER/PR/HER2-negative tumors (as determined by locally read IHC and FISH), the investigator-assessed pCR_B rates were 29% (95% CI, 19% to 41%), 33% (95% CI, 21% to 46%), and 26% (95% CI, 14% to 42%), respectively (Table 2). Corresponding pCR_{BL} rates were 19% (95% CI, 11% to 30%), 23% (95% CI, 13% to 35%), and 19% (95% CI, 9% to 34%). Responses were also examined by ER and HER2 status. A higher pCR_B rate (46.1%) was observed in patients with ER-negative/HER2-positive tumors (Table 2).

BCS

After completing four cycles of ixabepilone, 154 of 161 patients underwent surgery. Of these, 50 patients (32%) underwent nonmastectomy BCS, and 104 patients (68%) underwent mastectomy (Table 2).

Table 2. Patient Response Rates and Surgical Outcomes (n = 161)

Response Category	Patients With Response	Total Patients	%	95% CI
pCR_B, breast				
All treated patients	29	161	18	12 to 25
ER-negative tumors*	21	72	29	19 to 41
ER/PR-negative tumors*	20	61	33	21 to 46
ER/PR/HER2-negative tumors†	11	42	26	14 to 42
pCR, breast + lymph nodes				
All treated patients	17	161	11	6 to 16
ER-negative tumors*	14	72	19	11 to 30
ER/PR-negative tumors*	14	61	23	13 to 35
ER/PR/HER2-negative tumors†	8	42	19	9 to 34
Best overall response‡				
All patients	98	161	61	
CR	28	161	17	
Partial response	70	161	44	
End point surgery (n = 154)				
BCS	50	154	32	
Mastectomy	104	154	68	

Patient Responses (pCR_B) by ER/HER2 Status§	ER-Negative/HER2-Negative	ER-Negative/HER2-Positive	ER-Positive/HER2-Negative	ER-Positive/HER2-Positive
Responders, No.	11	6	7	1
Nonresponders, No.	39	7	59	4
Response rate, %	22.0	46.1	10.6	20.0

Abbreviations: pCR_B, pathologic complete response in the breast; ER, estrogen receptor; PR, progesterone receptor; pCR, pathologic complete response; CR, complete response; BCS, breast conservation surgery.
*ER and PR status were determined by immunohistochemistry (IHC).
†HER2 status was based on fluorescence in situ hybridization (FISH). IHC was used if FISH results were not available.
‡Best overall response was defined based on review of clinical (physical examination), radiographic (mammography or ultrasound), and histopathologic evidence at the primary tumor site.
§Data based on 134 patients for whom data for ER and HER2 expression status were available.

Gene Expression Analysis

To guide clinical investigations, markers identified through preclinical analysis were used to evaluate genes associated with sensitivity to ixabepilone. Twenty-three breast cancer cell lines were classified as sensitive (half-maximal inhibitory concentration [IC_{50}] < mean of the log IC_{50}) or resistant (IC_{50} > mean of the log IC_{50}) to ixabepilone in in vitro cell proliferation assays. Genes correlating with ixabepilone-sensitive or ixabepilone-resistant phenotypes were identified. The top 200 genes with expression levels highly correlated with the sensitivity-resistance class distinction were identified using the GeneCluster (Broad Institute, Cambridge, MA) package (Appendix Table A1, online only). Expression levels of the top 200 of these gene candidates are illustrated in Figure 2A. Subsequently, the top preclinical 200 genes linked with ixabepilone sensitivity were further analyzed using a pathway analysis tool. This analysis revealed that the ER network, including ER and *tau*, was most significantly correlated with the sensitivity to ixabepilone. The ER and microtubule-associated protein *tau* were among these top 200 genes. The expression patterns of ER and *tau* were highly correlated with resistance to ixabepilone (Fig 2B and 2C) and to each other (Pearson correlation = 0.78; $P < .001$) in the cell line study.

Of 161 treated patients, 134 patients met the inclusion criteria for the gene expression data set (mRNA samples of sufficient quantity [≥ 1 μg] and quality ([$A_{260/280}$ ratio of 1.8 to 2.1] or Agilent 28S/18S of 0.5 to 2.0). Among these patients, 23 patients (17%) achieved a pCR_B. These 134 patients then were randomly assigned to training (n = 67) or test sets (n = 67), stratified by response. Data from the training and test sets were used to identify and independently validate, respectively, the gene expression profile that predicts response. In addition, ER and *tau* were also examined as predictors of ixabepilone response.

Consistent with preclinical observations, low expression of ER and many ER-coregulated genes were found in responders (compared with nonresponders). ER expression was identified as a predictive marker for response to ixabepilone using the logistic regression model; ER expression was inversely related to pCR_B. It had an area under the curve of 0.745 (Fig 3), a positive predictive value (PPV) of 37%, and a negative predictive value of 92% for pCR_B (Table 3). When ER protein expression was assessed by IHC, the response prediction was not as strong (PPV = 29%) as that observed with gene expression levels (Table 3). *Tau* was also identified as a marker of response, but its predictive power (PPV = 29%) was not as strong as ER (Fig 3 and Table 3).

The response probability was further assessed in the test set using a 10-gene penalized logistic regression model. The 10-gene model had a higher PPV (45%), but was comparable in CIs to the single-gene logistic regression model for ER, and also had a similar negative predictive value (89%; Table 3). Interestingly, ER was independently included in the 10-gene model as part of the penalized logistic regression model discovery process. Genes in the 10-gene model, correlation coefficients, and predicted probability of response in the patient test set are shown in Figure 4 and Table 4.

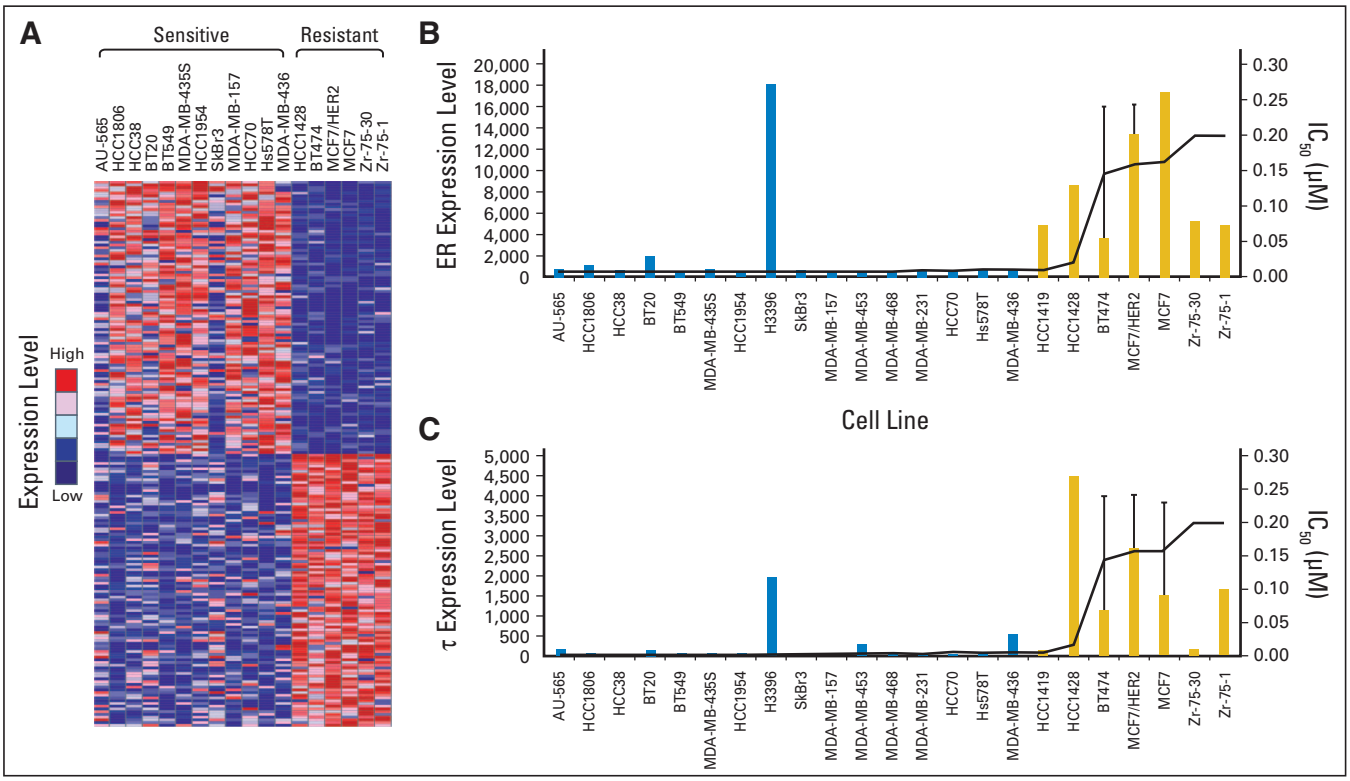

Fig 2. Correlation between gene expression patterns and sensitivity to ixabepilone in breast cancer cell lines. (A) Heat map depicting the top 200 genes correlating with ixabepilone sensitivity. The matrix represents normalized expression patterns for each gene (red indicates high expression, blue indicates low expression). Sensitive and resistant cell lines are indicated. (B) Correlation between estrogen receptor (ER) expression levels and ixabepilone sensitivity in human breast cancer cell lines. (C) Correlation between *tau* expression levels and ixabepilone sensitivity in human breast cancer cell lines.

Tolerability

The majority of AEs were mild to moderate and manageable. Seventy-two percent of patients experienced AEs of grade 1 or 2 (Table 5). Severe AEs occurred in 32% of patients (grade 3 in 27%; grade 4 in 5%). A total of 68 patients reported mild to moderate treatment-related neuropathy, most of which were grade 1 or 2 (grade 1, 26%; grade 2, 14%). The most common treatment-related neuropathy was sensory (grade 1, 15%; grade 2, 4%; grade 3, 1%). Neuropathy was reversible and resolved within 6 to 12 weeks after the last dose of ixabepilone.

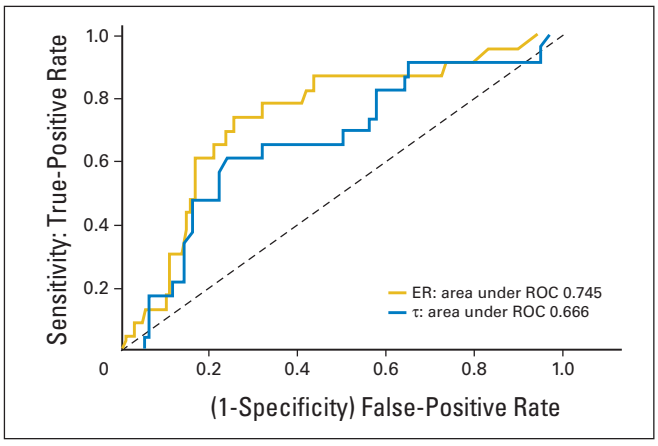

Fig 3. Receiver operating characteristic (ROC) curves for estrogen receptor (ER) and *tau* expression.

Hematologic AEs, primarily neutropenia and leukopenia, were manageable and did not result in notable dose reductions or discontinuations. Grade 3 to 4 neutropenia and leukopenia were observed in 14% and 8% of patients, respectively. Febrile neutropenia was reported in only two patients (3%). Grade 3 to 4 hypersensitivity reactions were reported in 1% of patients. Eleven patients (7%) discontinued therapy because of treatment-related AEs.

DISCUSSION

This study demonstrates that pathways involving ER and *tau* (an ER-regulated gene) are integral to ixabepilone sensitivity. Analyses confirmed ER expression as a predictive marker for response to ixabepilone, which had already been shown with anthracyclines and paclitaxel.[30] ER gene expression—a continuous variable, centrally assessed—was a better predictor of response than ER levels assessed locally by IHC, which is a categoric variable and might be subject to sampling errors. There was, however, a higher pCR_B in the ER-negative group using IHC as compared with the overall population. *Tau* was also identified as a marker of response, indicating an inverse correlation between ixabepilone sensitivity and *tau* levels. Recent studies have suggested that low *tau* expression makes microtubules more vulnerable to paclitaxel and also hypersensitizes breast cancer cells to the drug. Preincubation of tubulin with *tau* decreases paclitaxel binding and paclitaxel-induced microtubule polymerization.[30] Interestingly, *tau* was identified as a marker of response to the

Table 3. Statistical Power of Regression Models Predictive of Ixabepilone-Induced pCR_B

Model	PPV %	PPV 95% CI	NPV %	NPV 95% CI	Sensitivity %	Sensitivity 95% CI	Specificity %	Specificity 95% CI	Error (%)
10-gene regression model	45	21 to 72	89	79 to 95	45	21 to 72	89	79 to 95	33
ER gene expression	37	19 to 59	92	80 to 97	64	35 to 85	79	66 to 87	29
ER status, IHC	29	20 to 41	90	81 to 95	72	54 to 85	59	50 to 67	34
tau gene expression	29	14 to 50	89	77 to 95	55	28 to 79	73	60 to 83	36

Abbreviations: pCR_B, pathologic complete response in the breast; PPV, positive predictive value; NPV, negative predictive value; ER, estrogen receptor; IHC, immunohistochemistry.

paclitaxel plus fluorouracil, doxorubicin, and cyclophosphamide regimen in a separate clinical study.[4]

A 10-gene model that predicts pCR_B and objectively included ER as part of the model was discovered using the top 200 preclinical markers identified in this study. A better understanding of ER and *tau* pathways and other signaling cascades will help define gene expression patterns unique to ixabepilone and thus facilitate selection of patients most likely to derive the greatest long-term clinical benefit from neoadjuvant ixabepilone therapy.

A recent Cancer and Leukemia Group B clinical study has demonstrated that expression and/or amplification of HER2 is associated with greater clinical benefit in patients with breast cancer treated with adjuvant paclitaxel.[31] As part of our analysis, patient responses to ixabepilone were analyzed by ER and HER2 status (determined by IHC). The overall response rate (pCR_B) was 18% on the basis of 134 patients for whom data for ER and HER2 status were available. We observed the highest response rate, 46.1%, in patients with ER-negative/HER2-positive tumors, suggesting that ixabepilone is

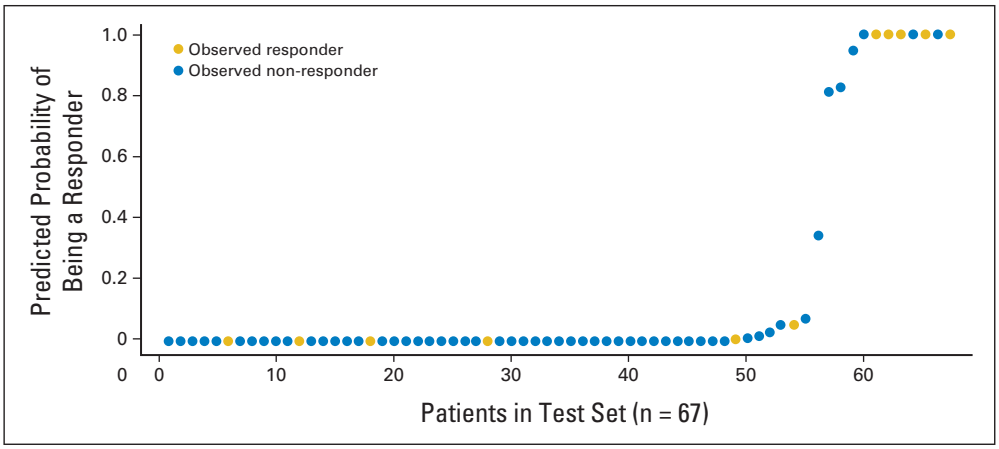

Fig 4. Predicted probability of response in the patient test set using the 10-gene penalized logistic regression model. The predicted probability of response is close to 1 or 0 because estimated coefficients in the 10-gene model were relatively large. This may be explained, at least in part, by amplification of important coefficients and shrinking of less important coefficients resulting from the penalized logistic regression analysis.

Table 4. Composition of 10-Gene Penalized Logistic Regression Model and Coefficients

Gene No.	Coefficient in the PLR Model	Affymetrix Identifier	Gene Name	Gene Ontology
1	−1.40	205225_at	Estrogen receptor 1	Nuclear hormone receptor, cellular proliferation and differentiation
2	−1.92	203637_s_at	Midline 1 (Opitz/BBB syndrome)	Microtubule cytoskeleton organization and biogenesis
3	1.15	200600_at	Moesin	Cell motility
4	1.82	213400_s_at	Transducin (β) -like 1	Signal transduction
5	−2.54	210042_s_at	Cathepsin Z	Proteolysis and peptidolysis
6	14.10	204567_s_at	ATP-binding cassette, subfamily G (WHITE), member 1	Small molecule transport
7	−5.73	210239_at	Iroquois homeobox protein 5	Regulation of transcription, DNA dependent
8	−3.73	201484_at	Suppressor of Ty (*S. cerevisiae*) 4 homolog 1	Chromatin modeling
9	6.67	209040_s_at	Proteasome (prosome, macropain) subunit, β type, 8 (large multifunctional protease 7)	Proteolysis and peptidolysis, ubiquitin-dependent protein catabolism
10	3.42	211392_s_at	Zinc finger protein 278	

Abbreviations: PLR, penalized logistic regression; ATP, adenosine triphosphate.

Table 5. Overall Treatment-Related Adverse Events Occurring in ≥ 10% of Patients (n = 161)

Adverse Event	Grade 1 (%)	Grade 2 (%)	Grade 3 (%)	Grade 4 (%)
Nonhematologic				
Alopecia	29	57	0	0
Myalgia	26	22	2	0
Peripheral neuropathy	26	14	3	0
Peripheral sensory neuropathy	15	4	1	0
Arthralgia	17	12	1	0
Asthenia	18	8	1	0
Fatigue	15	4	1	0
Nausea	16	3	0	0
Diarrhea	8	5	2	0
Bone pain	11	4	0	0
Hypersensitivity	6	4	1	0
Stomatitis	6	4	1	0
Hematologic*				
Neutropenia	2	5	10	4
Leukopenia	1	5	7	1

NOTE. The following adverse events occurred in < 10% of patients: muscular weakness (9%), vomiting (8%), constipation (7%), anorexia (7%), mucosal inflammation (6%), motor neuropathy (1%).
*Clinical laboratory evaluation.

most efficacious in this population, although the result is not statistically significant given the small sample size (Fisher's exact test, $P = .157$). This is consistent with the recent results indicating chemotherapeutic benefit in the adjuvant setting in ER-negative/HER2-positive breast cancer.[31]

In this large, phase II study, ixabepilone (40 mg/m^2) demonstrated substantial antitumor activity, as well as an acceptable safety profile when administered for four cycles as neoadjuvant therapy to patients with invasive breast adenocarcinoma not amenable to primary BCS.

The study end point of pCR_B, which correlates with overall survival, was comparable to values reported in trials of other single agents in the neoadjuvant setting. The 18% pCR_B rate reported after four cycles of ixabepilone compared favorably with pCR_B rates in studies of single-agent taxanes also administered for four cycles as neoadjuvant therapy. Docetaxel achieved a pCR_B rate of 10% and pCR_{BL} a rate of 7%.[32] Studies of single-agent paclitaxel 250 mg/m^2 and 200 mg/m^2 yielded pCR_B rates of 8% and 4%,[33,34] respectively. In a large trial, the pCR achieved with doxorubicin and cyclophosphamide was 13%.[35] Higher pCR rates than those mentioned here have been achieved with the taxanes, but they were the result of an increased dose, a higher number of treatment cycles, and/or combination regimens.[32,36,37]

Ixabepilone had an acceptable safety profile and a low discontinuation rate (7%) due to treatment-related AEs. It compared favorably with other chemotherapeutic agents in the neoadjuvant setting and with single-agent ixabepilone in the metastatic setting. The more favorable safety profile for ixabepilone as neoadjuvant therapy could reflect fewer treatment cycles as compared with studies in metastatic disease. It could also be attributed to a lower rate of subclinical neuropathy at baseline in this chemotherapy-naive population. In this study, neuropathy was primarily sensory and reversible. Incidence of grade 3 peripheral neuropathy (3%) was substantially lower than reported in previously pretreated patients with metastatic disease (eg, 20% in anthracycline-pretreated,[13] 12% in taxane-resistant,[14] and 13% in anthracycline-, taxane-, and capecitabine-resistant populations[15]). Grade 3 to 4 neutropenia related to treatment with ixabepilone in the neoadjuvant setting (14%) was also lower than with other chemotherapeutic agents: 93% grade 3 to 4 neutropenia with four cycles of neoadjuvant docetaxel,[32] and 70.5% grade 3 to 4 neutropenia with six cycles of neoadjuvant docetaxel.[38]

Using gene expression assays, this study successfully identified patients who were most likely to benefit from neoadjuvant ixabepilone treatment. In addition, the study validated preclinical markers (ie, ER and *tau*) in the clinical setting, indicating the value of preclinical studies early in drug development to identify candidate efficacy biomarkers that can be used in decision making. The findings of this study will be used in a large-scale prospective neoadjuvant study. Patients will be selected to explore the predictive value of markers for identifying patients who may benefit most from ixabepilone-based therapy. Given the efficacy demonstrated by ixabepilone in metastatic breast cancer, including in heavily pretreated patients and those with multiresistant disease[12-15,22] and in the neoadjuvant setting,[39-42] further studies of ixabepilone in earlier breast cancer settings are warranted.

AUTHORS' DISCLOSURES OF POTENTIAL CONFLICTS OF INTEREST

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Glossary Terms

Gene expression analysis: Technique for the simultaneous quantification of the mRNA expression level of thousands of genes. Can be performed using microarrays, RT-PCR, or other technologies for measuring gene expression.

Gene expression profile: The expression of a set of genes in a biologic sample (eg, blood, tissue) using microarray, RT-PCR, or other technology capable of measuring gene expression.

Tubulin: Component of microtubules, which are essential components of the eukaryotic cytoskeleton. The three tubulin families are alpha, beta, and gamma. Alpha and beta tubulins are components of microtubules, and gamma tubulins play a critical role in microtubule assembly.

Neoadjuvant therapy: The administration of chemotherapy prior to surgery. Induction chemotherapy is generally designed to decrease the size of the tumor prior to resection and to increase the rate of complete (R0) resections.

Pharmacogenomic: The study of how a person's genome can affect their reaction to medications.

Biomarker (biologic marker): A characteristic that is objectively measured and evaluated as an indicator of normal biologic processes, pathogenic processes, or pharmacologic responses to a therapeutic intervention.

ER (estrogen receptor): Belonging to the class of nuclear receptors, estrogen receptors are ligand-activated nuclear proteins present in many breast cancer cells that are important in the progression of hormone-dependent cancers. After binding, the receptor-ligand complex activates gene transcription. There are two types of estrogen receptors α and β. ERα is one of the most important proteins controlling breast cancer function. ERβ is present in much lower levels in breast cancer and its function is uncertain. Estrogen-receptor status guides therapeutic decisions in breast cancer.

Pathologic complete response (pCR): The absence of any residual tumor cells in a histologic evaluation of a tumor specimen is defined as a complete pathologic response.

