A Phase II Trial of Erlotinib in Combination with Bevacizumab in Patients with Metastatic Breast Cancer

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Abstract

Purpose: To evaluate the efficacy and toxicity of erlotinib plus bevacizumab in patients with metastatic breast cancer (MBC), targeting the epidermal growth factor receptor (EGFR/HER1) and the vascular endothelial growth factor (VEGF) pathway.

Experimental Design: Thirty-eight patients with MBC were enrolled and treated at two institutions with erlotinib, a small molecule EGFR tyrosine kinase inhibitor (150 mg p.o. daily) plus bevacizumab, an anti-VEGF antibody (15 mg/kg i.v. every 3 weeks). Patients had one to two prior chemotherapy regimens for metastatic disease. The primary end point was response rate by Response Evaluation Criteria in Solid Tumors criteria using a Simon 2-stage design. Secondary end points included toxicity, time to progression, response duration, and stabilization of disease of $\geq$26 weeks. Correlative studies were done on tumor tissue, including EGFR expression and mutation analysis.

Results: One patient achieved a partial response for 52+ months. Fifteen patients had stable disease at first evaluation at 9 weeks; 4 of these patients had stable disease beyond 26 weeks. Median time to progression was 11 weeks (95% confidence interval, 8-18 weeks). Diarrhea of any grade was observed in 84% of patients (grade 3 in 3%); 76% experienced grade 1 or 2 skin rash, and 18% developed hypertension (grade 3 in 11%). The level of EGFR expression was not predictive of response to therapy.

Conclusions: The combination of erlotinib and bevacizumab was well-tolerated but had limited activity in unselected patients with previously treated MBC. Biomarkers are needed to identify those MBC patients likely to respond to anti-EGFR/HER1 plus anti-VEGF therapy.

Breast cancer is the second leading cause of cancer-related mortality among women in the United States. Although a number of agents have activity in breast cancer, metastatic disease remains incurable. New targeted treatments that delay disease progression while reducing toxicity would therefore represent a significant advance in the care of women with breast cancer.

Vascular endothelial growth factor (VEGF) is a central regulator of both normal and pathologic angiogenesis, which is essential for the growth and metastasis of solid tumors (1). VEGF therefore serves as a therapeutic target for inhibiting tumor growth. As proof of this concept, bevacizumab (Avastin; Genentech), a humanized monoclonal antibody that binds the VEGF-A ligand, improved overall survival when added to chemotherapy in patients with metastatic colorectal cancer (2) and non–small cell lung cancer (NSCLC; ref. 3). As a single agent, bevacizumab is also active in metastatic renal cell carcinoma (4) and ovary cancer (5). In metastatic breast cancer (MBC), single-agent bevacizumab produced objective responses in 9.3% of patients in a phase I/II trial (6). In combination with weekly paclitaxel, bevacizumab doubled response rate and significantly prolonged progression-free survival compared with chemotherapy alone as first-line treatment of MBC (progression-free survival, 11.8 versus 5.9 months; hazard ratio, 0.60, $P < 0.001$; ref. 7).

Members of the human epidermal growth factor receptor (EGFR) family (ErbB family) are also proven therapeutic targets for cancer therapy (8). In breast cancer, targeting the human epidermal growth factor receptor 2 (HER2) with trastuzumab (Herceptin; Genentech) improves survival in patients with HER2-positive breast cancer in both the adjuvant (9) and metastatic setting (10). The EGFR or HER1 is another

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Translational Relevance

This phase 2 trial adds to the growing body of evidence for minimal activity of anti-endothelial growth factor receptor (EGFR) therapy in breast cancer, and the uncertain benefits of anti-EGFR plus anti – vascular endothelial growth factor therapy in solid tumors. Although based on preclinical data suggesting increased synergy for this combination, this study did not meet predetermined goals of efficacy in unselected patients. This negative result is clinically relevant and important to share with the research community. Several single-arm phase 2 studies have shown promise for this regimen in tumors including non – small cell lung cancer, renal cell carcinoma, and carcinoma of unknown primary; however, this combination has not been validated by phase 3 trials. In addition, this study includes tissue-based correlative work. The level of EGFR expression in breast cancer tumor tissue was not predictive of response to therapy, and EGFR tyrosine-kinase domain mutations were not detected in the 25 patients with sufficient tumor tissue available for this analysis.

member of the ErbB/HER family. EGFR/HER1 is expressed and abnormally activated in several epithelial tumors (11). Binding of the epidermal growth factor or transforming growth factor α ligand to EGFR triggers downstream signaling pathways that mediate a variety of cellular responses, including cellular proliferation, angiogenesis, and apoptosis (8). Anti-EGFR therapy with erlotinib (Tarceva; OSI Pharmaceuticals), an orally active EGFR tyrosine kinase inhibitor, improves survival in patients with NSCLC (12), and activating mutations in the EGFR gene are predictive of response to therapy in this disease (13, 14). In breast cancer, EGFR expression has ranged widely from 8.3% to 91% in the reported literature (15) and has been associated with a decrease in relapse-free and overall survival (16). However, recent studies using the anti-EGFR tyrosine kinase inhibitors erlotinib and gefitinib (Iressa; Astra-Zeneca) as monotherapy in patients with breast cancer have reported limited activity, with response rates of <5% (17, 18).

Preclinical data suggest that the EGFR signaling pathway plays a role in the regulation of angiogenesis (19 – 22). Anti-EGFR therapy with a monoclonal antibody decreases production of angiogenic factors including VEGF, basic fibroblast growth factor, and interleukin-8 (23). In xenograft models, anti-EGFR plus anti-VEGF therapy has increased activity compared with either agent alone (24, 25). We therefore hypothesized that targeting both the EGFR and VEGF pathways may suppress common downstream signaling pathways and increase and/or prolong antitumor activity. Phase II trials of bevacizumab plus erlotinib have reported promising results for patients with renal cell carcinoma (26), NSCLC (27), and carcinomas of unknown primary site (28); and phase I data in NSCLC showed no evidence of a negative pharmacokinetic interaction between these 2 agents (27). Our study sought to evaluate the combination of erlotinib and bevacizumab as targeted therapy in patients with MBC.

Patients and Methods

Patient eligibility. A total of 38 patients with MBC were enrolled at Memorial Sloan-Kettering Cancer Center, New York, NY, and the University of California San Francisco Comprehensive Cancer Center, San Francisco, CA, from February 2003 to December 2004. Patients with histologically confirmed breast carcinoma were eligible if they had stage IV disease that was stable or progressing after treatment with 1 or 2 chemotherapy regimens. There was no limit on the number of prior endocrine therapies allowed in either the adjuvant or metastatic setting, nor was endocrine therapy required for eligibility. The use of prior adjuvant chemotherapy did not affect eligibility. Additional criteria included measurable disease by Response Evaluation Criteria in Solid Tumors (29), an Eastern Cooperative Oncology Group performance status of ≤2, and adequate hepatic, renal, and hematologic function. Additionally, if the patient's tumor was HER2 positive, prior therapy with trastuzumab was required. Although EGFR positivity was not required for eligibility, a tissue sample of the patient's breast cancer was obtained for retrospective EGFR assessment. Main exclusion criteria included chemotherapy, radiotherapy, immunotherapy, or investigational therapy within 3 wk or hormonal therapy within 2 wk of initiating study treatment. All patients had to have a baseline computed tomography or magnetic resonance imaging of the brain. Patients with central nervous system disease including primary brain tumor, brain metastases, or history of stroke were also ineligible. Prior treatment with VEGFR inhibitors and/or EGFR targeting therapies, major surgery occurring within 28 d before treatment, nonhealing wound or bone fracture, and baseline proteinuria >500 mg/24 h were additional exclusion criteria. Patients with clinically significant cardiovascular disease (e.g., uncontrolled hypertension, myocardial infarction, unstable angina) were excluded. Concurrent administration of bisphosphonates was allowed throughout the study period. The institutional review boards of the two participating centers reviewed and approved this study protocol. All patients gave written informed consent before participation.

Study design and treatments. This was a nonrandomized, open-label, bi-institutional phase II trial. The primary objective was to determine the response rate (complete or partial) of erlotinib plus bevacizumab according to Response Evaluation Criteria in Solid Tumors (29). Secondary end points included toxicity, time to disease progression, duration of response, and stabilization of disease of ≥26 wk.

Erlotinib was administered p.o. at 150 mg on a continuous daily schedule. Bevacizumab was administered at 15 mg/kg i.v. every 3 wk, with a protocol stipulated window of ±5 d. Toxicity was evaluated according to the National Cancer Institute Common Toxicity Criteria (NCI-CTC v2.0). If toxicity was thought to be related to erlotinib, the daily dose was reduced from 150 to 100 mg/d (first reduction), then to 50 mg/d (second reduction), and finally to 25 mg/d (third reduction). Bevacizumab was continued, whereas the dose of erlotinib was either held or reduced. Grade 2 diarrhea and skin rash did not require temporary discontinuation of erlotinib. Symptomatic patients were treated with loperamide for diarrhea and tetracycline for skin rash. For any grade 3 or 4 erlotinib-related toxicity or medically concerning grade 2 nonhematologic toxicity, erlotinib was held until symptoms resolved to grade 1 or less and then reinstituted at a reduced dose. There was no modification of bevacizumab dose during this study. Erlotinib was continued if the dose of bevacizumab was held secondary to a bevacizumab-related toxicity. For any grade 3 or 4 bevacizumab-related toxicities, bevacizumab was held until symptoms resolved to grade 1 or less, but erlotinib was continued. However, no modification of bevacizumab was allowed during this study. Patients with grade 3 hypertension controlled by oral medications were allowed to continue bevacizumab.

Patient evaluation. Every 9 wk, response was evaluated with radiographic scans reviewed at each site by a designated study radiologist. Response required confirmation with repeat imaging at

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Table 1. Patient characteristics (n = 38)

Characteristic	No. patients (%)
Enrolled and treated	38 (100)
Age, y	
Median	51
Range	35-71
ECOG performance status	
Median	1
Range	0-2
0	14 (37)
1	23 (61)
2	1 (3)
Prior Therapy for MBC	
Chemotherapy	38 (100)
1 prior regimen	22 (57)
2 prior regimens	16 (42)
Hormonal therapy	22 (58)
Trastuzumab	8 (21)
Prior therapy for adjuvant or MBC	
Anthracycline and taxane	25 (66)
Anthracycline only	3 (8)
Taxane only	7 (18)
Metastatic site(s)	
Liver	26 (68)
Lung	20 (53)
Soft tissue & bone only	3 (8)

Abbreviation: ECOG, Eastern Cooperative Oncology Group.

least 4 wk after the initial response documentation. In the case of stable disease (SD), follow-up measurements must have met the SD criteria at least once after study entry at a minimum interval of 9 wk. Progressive disease was defined as a ≥20% increase in measurable index lesions from the smallest sum observed (from baseline if no confirmed response) or appearance of any new lesion or site. Patients with confirmed complete response, partial response, or SD continued treatment until disease progression, unacceptable toxicity, or withdrawal from the study. Patients were evaluated for toxicity before each 3-wk treatment cycle.

Statistical considerations. The primary end point of this trial was response rate, defined by complete response + partial response, according to Response Evaluation Criteria in Solid Tumors (29). A Simon optimal two-stage design was used (30). A 20% response rate was considered promising, whereas a 5% response rate was not considered promising. The probabilities of a type I and type II error were both set at 0.10, and 12 patients were accrued to the first stage of the trial. Continuation to the second stage depended on one or more patients from the first stage having a complete response or partial response. If no objective responses were seen in the first stage of this trial (0 of 12), our protocol required that we determine EGFR expression by immunohistochemistry (IHC) in this cohort, and keep patient accrual open until we obtained a total of 12 EGFR positive patients. However, based on 1 partial response in the first stage, an additional 25 patients were accrued, with no requirement to determine EGFR status for eligibility. The planned sample size for the study was 37 patients, 12 in stage 1 and 25 in stage 2. If at least 4 responses were observed among the 37 patients studied, then this regimen would be considered worthy of further testing.

Data on secondary end points included toxicity, SD of ≥26 wk, time to progression, and duration of response. Time to progression was defined as the time from the start of therapy to date of disease progression and was estimated using the Kaplan-Meier method.

Correlative pathology methods. An archival paraffin tissue block (or unstained slides) representative of each patient's primary or MBC was required to perform studies to correlate the antitumor efficacy of erlotinib plus bevacizumab with pretreatment molecular characteristics, such as estrogen receptor (ER), progesterone receptor (PR), HER2 (Herceptest), and EGFR (PharmDx). Antibodies, scoring, and conditions used are summarized in Supplementary Appendix. Using previously published methods, HER1/EGFR DNA sequencing was also done on exons 18, 19, and 21 when sufficient tumor tissue was available (14).

Results

Patient demographics. Thirty-nine patients were registered, 38 were initially treated, and 37 were eligible for statistical analysis. Baseline demographics and disease characteristics of the 38 patients who received at least 1 dose of treatment are listed in Table 1. One patient was never treated because of a suppurative groin infection at the site of prior dendritic cell vaccinations and was ineligible because of this nonhealing wound. A second patient was treated with a single dose of bevacizumab and was subsequently determined to be ineligible because of the presence of a meningioma (a primary central nervous system tumor) on a screening brain magnetic resonance imaging.

The median age of treated participants was 51 years (range, 35-71). All but 1 patient had a baseline Eastern Cooperative Oncology Group score of ≤1. Forty-two percent of patients had 2 prior chemotherapy regimens for MBC and 58% received prior hormonal therapy. Additionally, a majority of patients (66%) received both anthracycline- and taxane-based chemotherapy in either the adjuvant or metastatic setting. The most common metastatic site was the liver (68%); soft tissue and bone were less common (8%).

Efficacy. Patients received a median of 3 cycles of treatment (range, 1-85). Thirty-seven patients received at least 1 cycle of erlotinib plus bevacizumab, and of the 37, 1 patient had a confirmed partial response after 3 cycles of therapy (3%; 95% confidence interval, 0-8%; Table 2). There were no complete responses. Fifteen patients (41%; 95% confidence interval, 25-56%) had SD as best response at 9 weeks, and 4 of these 15 patients continued with SD beyond 26 weeks (28, 29, 36, and 46 weeks). Two additional patients discontinued study therapy secondary to toxicity before the initial 9-week protocol-stipulated evaluation but had SD documented around the time of withdrawal from the study. The remaining 19 patients (51%; 95% confidence interval, 35-68%) had progressive disease at or

Table 2. Best response with combined erlotinib and bevacizumab therapy

Best response*	Total population assessed for response (n = 37)
	No. patients (%)
CR	0 (0)
PR	1 (3)
SD at 9 wk	15 (41)
SD at >26 wk	4 (11)
PD at 9 wk	19 (51)

Abbreviations: CR, complete response; PR, partial response; PD, progressive disease.
*Best response assessed by Response Evaluation Criteria in Solid Tumors.

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Table 3. Common treatment-related emergent adverse events on study

Toxicity	Erlotinib + bevacizumab (n = 38)				
	All	Grade 1	Grade 2	Grade 3	Grade 4
	No. (%)	No. (%)	No. (%)	No. (%)	No. (%)
Diarrhea	32 (84)	26 (68)	5 (13)	1 (3)	0 (0)
Rash	29 (76)	14 (37)	15 (39)	0 (0)	0 (0)
Fatigue	24 (63)	20 (53)	4 (11)	0 (0)	0 (0)
Stomatitis	18 (47)	14 (37)	4 (11)	0 (0)	0 (0)
Nausea	13 (34)	11 (29)	1 (3)	1 (3)	0 (0)
Lymphopenia	13 (34)	5 (13)	2 (5)	6 (16)	0 (0)
Myalgia	13 (34)	11 (29)	1 (3)	0 (0)	1 (3)
Dry skin	12 (32)	10 (26)	2 (5)	0 (0)	0 (0)
Proteinuria	8 (21)	6 (16)	2 (5)	0 (0)	0 (0)
Arthralgias	9 (24)	5 (13)	4 (11)	0 (0)	0 (0)
Vomiting	8 (21)	6 (16)	1 (3)	1 (3)	0 (0)
Pruritus	7 (18)	5 (13)	2 (5)	0 (0)	0 (0)
Hypertension	7 (18)	2 (5)	1 (3)	4 (11)	0 (0)
Epistaxis	7 (18)	7 (18)	0 (0)	0 (0)	0 (0)
Dyspepsia	5 (13)	2 (5)	3 (8)	0 (0)	0 (0)
Thrombosis	3 (8)	0 (0)	0 (0)	2 (5)	1 (3)
Dehydration	3 (8)	0 (0)	2 (5)	1 (3)	0 (0)
Dry eyes	3 (8)	2 (5)	1 (3)	0 (0)	0 (0)

NOTE. Toxicity graded according to National Cancer Institute Common Toxicity Criteria v2.0. Table includes worst grade of toxicity per patient.

before 9 weeks. The median time to tumor progression on this trial was 11 weeks (95% confidence interval, 8-18 weeks), with a 52-month response duration in 1 partial responder.

The patient with a partial response had shrinkage of disease in lung and lymph nodes. This 66-year-old female was diagnosed with breast cancer in 1999, and received adjuvant doxorubicin plus cyclophosphamide. In 2003, she developed recurrent metastatic disease, and was treated with 2 cycles of doxorubicin plus docetaxel followed by 11 cycles of paclitaxel. At progression, she was enrolled in this study and remains on therapy after 85 cycles, with a response duration of 52+ months.

Adverse events. The most common adverse events for the 38 patients who received study therapy are listed in Table 3. The majority of patients experienced diarrhea (84%; grade 3 in only 3%), skin rash (76%; grade 1 or 2 only), and fatigue (63%; grade 1 or 2 only). Four patients (11%) developed grade 3 hypertension that was controlled by oral medications. Eight patients (21%) experienced mild proteinuria (grade 1, n = 6; grade 2, n = 2). There were 2 grade 4 events, thrombosis and myalgia. The myalgia was lower back pain and spasm of "unlikely" attribution to the study medications, which ultimately lead to imaging studies that confirmed progression of disease. A total of 5 patients had either a dose delay (n = 2) or dose reduction (n = 3) of erlotinib because of drug-related toxicities. Per protocol, there were no dose reductions of bevacizumab, nor did any patients require a dose delay of bevacizumab for drug-related toxicities.

Six patients withdrew from the study. One patient with grade 3 hypertension that was well-controlled with medication withdrew for personal reasons. Four patients withdrew because of drug-related toxicity, including grade 1 biopsy-proven allergic skin rash (n = 1), grade 2 acneiform rash (n = 1), grade 3 nausea/vomiting despite erlotinib dose reduction (n = 1), and grade 3 diarrhea/dehydration that required hospitalization (n = 1). One patient was diagnosed with a radiation-induced T5 spinal cord myelopathy, with symptoms that predated the start of protocol therapy. She developed progressive lower extremity weakness during study treatment, and required hospitalization for grade 3 motor neuropathy. The protocol therapy was discontinued because of disability. Exacerbation of neuropathy secondary to either bevacizumab or erlotinib was felt unlikely but could not be ruled out. Two patients died of progressive MBC during the 30 days after study termination.

Pathologic correlates. The molecular characteristics of the 38 treated patients are listed in Table 4, including central IHC testing of ER, PR, HER2, and EGFR. The molecular characteristics of the 1 partial responder and the 4 patients with SD of ≥26 weeks are listed in Table 5. The patient with a partial response had triple-negative breast cancer (ER, PR, and HER2 negative); her tumor expressed EGFR at 1+ by IHC.

Table 4. Molecular characteristics by IHC

Characteristic	No. Patients (%)
Tumor tissue available	38 (100)
ER	
Positive (>1% staining)	19 (50)
Negative	19 (50)
PR	
Positive (> 1% staining)	8 (21)
Negative	30 (79)
HER2	
Positive (3+ by IHC)	3 (8)
Negative	35 (92)
EGFR	
0	24 (63)
1+	8 (21)
2+	4 (11)
3+	0 (0)
Insufficient tumor tissue	2 (5)
ER/PR and HER2 negative	19 (50)
ER/PR/HER2 negative and EGFR+ (1-3+ by IHC)	10 (26)

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Table 5. Molecular characteristics of patients by IHC with clinical benefit ≥26 wk or PR (n = 5)

Patient # and response status	ER (%)	PR (%)	HER2	EGFR
1 PR	0	0	0	1+
2 SD for 28 wk	40	<5	3+	0
3 SD for 29 wk	0	0	1+	NA*
4 SD for 36 wk	0	0	0	1+
5 SD for 46 wk	90	0	2+ (FISH nonamplified)	0

Abbreviations: NA, not available; FISH, fluorescence *in situ* hybridization.
*Insufficient tissue available for analysis.

Twenty-five patients had sufficient paraffin-embedded tissue for EGFR tyrosine-kinase domain mutational analysis (exons 18, 19, and 21), including the 1 patient with a partial response and 2 of the patients with SD ≥26 weeks. No mutations were detected in any patient. Two distinct polymorphisms were detected in 6 patients but were felt to be of no clinical significance.

Discussion

Results from our phase II trial do not support the hypothesis, based on preclinical synergy, that the combination of erlotinib plus bevacizumab would be broadly useful in unselected, previously treated MBC patients. However, the 1 patient who had a partial response, with ER, PR, HER2-negative but EGFR 1+ breast cancer, has had a durable response to treatment for >52+ months.

In addition to our study, other trials of EGFR/HER1-targeted therapy in unselected breast cancer patients have shown minimal activity, including EGFR tyrosine kinase inhibitors [erlotinib (18) and gefitinib (17, 31)] and anti-EGFR monoclonal antibodies (cetuximab; ref. 32). Although activity has been shown by targeting the ErbB family in breast cancer with monoclonal antibodies (anti-HER2 therapy with trastuzumab; refs. 9, 10, 33) and tyrosine kinase inhibitors [dual anti-HER1 and anti-HER2 therapy with lapatinib (Tykerb; GlaxoSmith Kline; refs. 34, 35)], this activity has been limited to patients with HER2-positive tumors. In this same population, gefitinib in combination with trastuzumab did not increase antitumor activity despite evidence of preclinical synergy (36). Neoadjuvant gefitinib plus anastrozole also did not increase clinical response rates or decrease tumor cell proliferation in patients with ER-positive breast cancer despite preclinical evidence that anti-EGFR therapy may reverse resistance to endocrine therapy(37).

Several molecular subtypes of breast cancer have been defined by microarray studies (38). High expression of ER is identified in two of the molecular subtypes, and HER2 expression in combination with ER expression defines a third subtype. In contrast, the basal-like subtype has a low expression of both ER and HER2, although many basal-like tumors express EGFR by IHC (39). In preclinical models, basal-like cell lines are more sensitive to EGFR inhibitors (40). It is intriguing that our patient with a long-term partial response has a tumor that fits within this basal-like subtype. However, 50% of our cohort had ER, PR, and HER2-negative breast cancers; half of these patients with basal-like tumors were EGFR positive. Presently, the role for anti-EGFR/HER1 therapy in breast cancer remains an area of active investigation, and ongoing trials are evaluating anti-EGFR therapy plus chemotherapy in patients with basal-like tumors(41).

Phase II trials in other tumor types, including NSCLC (27), renal cell carcinoma (26), and carcinomas of unknown primary site(28), have suggested promising activity for targeting the EGFR and VEGF pathways with erlotinib plus bevacizumab. However, a randomized phase 2 trial in renal cell carcinoma did not confirm the increased activity for this combination compared with bevacizumab alone (42). Although preclinical data (24) and small phase 2 trials suggest activity for this regimen, randomized trials are essential to avoid selection bias and confirm increased efficacy. In support of our findings in breast cancer, a trial of ZD6474 (Astra Zeneca), an oral multitargeted tyrosine kinase inhibitor that inhibits both EGFR/HER1 and VEGFR-2, was also inactive in a similar population of patients with metastatic disease (43).

Selection of patients for targeted therapy remains a challenge because we presently lack reliable biomarkers to predict activity for anti-EGFR and antiangiogenic therapy (44). Patients in our study were not enrolled based on EGFR or HER2 status of tumor tissue, although these tissue-based studies were done retrospectively in the study patients. EGFR and HER2 positivity by IHC did not correlate with the activity of this regimen, although definitive conclusions are limited by the small number of patients and overall lack of activity for the combination in this trial. Based on increased activity of erlotinib in NSCLC patients with EGFR gene mutations in exons 18, 19, or 21(13, 14), DNA sequencing of these domains was done in our study when sufficient tumor tissue was available. No EGFR mutations were discovered in the 25 patients tested, including the 1 patient with a partial response to treatment and 2 of 4 patients with SD at 26 weeks.

Circulating tumor cells and endothelial cells are promising surrogate biomarkers of response to chemotherapy and antiangiogenic therapy, respectively (45, 46). In our trial, a companion study of baseline and serial levels of circulating tumor cells and circulating endothelial cells was done. Preliminary results suggest that decreasing circulating endothelial cells at week 3 predicts for progression-free survival at the time of first response evaluation (week 9; ref. 47).

The limited activity of erlotinib plus bevacizumab in this study does not support further investigation of that combination in unselected breast cancer patients. However, preclinical (48) and preliminary clinical data (49) show activity for combined targeting of the HER2 and VEGF pathways in HER2-positive breast cancer. Phase II studies of anti-HER2

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(trastuzumab, lapatinib) and anti-VEGF (bevacizumab, pazopanib) therapy are under way.

Disclosure of Potential Conflicts of Interest

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