Phase II multicenter, uncontrolled trial of sorafenib in patients with metastatic breast cancer

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This trial was conducted to assess the efficacy and safety of sorafenib in patients with metastatic breast cancer. In this multinational, open-label phase II study, patients with metastatic breast cancer that had progressed after at least one prior chemotherapy regimen were continuously treated with oral sorafenib, 400 mg twice daily. The primary endpoint was overall best response; a secondary endpoint was percentage of patients with stable disease for ≥16 weeks. Biomarker analyses were also performed. Of the 56 patients enrolled into the study, 54 were treated with at least one dose of sorafenib. Partial response was observed in one patient (2%) and stable disease in 20 patients (37%); no complete responses were observed. Disease stabilization for ≥16 weeks was seen in 12 patients (22%); stabilization for ≥6 months in seven patients (13%). The most common drug-related grade 3 adverse events were rash/desquamation (6%), hand–foot skin reaction (4%), and fatigue (4%). Baseline vascular endothelial growth factor levels, levels of soluble epidermal growth factor receptor during treatment and both baseline and changes in soluble human epidermal growth factor receptor 2 levels correlated significantly with clinical outcomes. Although the primary endpoint of overall response rate showed minimal improvement on sorafenib 400 mg twice-daily treatment, the rate of disease stabilization was encouraging in patients treated with one or more lines of chemotherapy. The treatment had a clinically manageable toxicity profile. Further investigation of single-agent sorafenib in this patient population is not recommended; however, studies investigating combinations of sorafenib with chemotherapeutic agents are warranted and ongoing. *Anti-Cancer Drugs* 20:616–624 © 2009 Wolters Kluwer Health | Lippincott Williams & Wilkins.

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Introduction

Despite advances in diagnosis and treatment in the last 20 years, metastatic breast cancer (MBC) remains an essentially incurable disease with survival ranging from 18 to 36 months after diagnosis. Although breast cancer mortality has declined, metastatic disease remains incurable and new drug development is critical to further advancement in care [1].

Sorafenib is an orally available bi-aryl urea that targets signaling pathways that may affect tumor cells and tumor vasculature. It inhibits the Ras/Raf/mitogen-activated protein kinase (MAPK) signaling pathway as well as platelet-derived growth factor receptor-α, β, vascular endothelial growth factor receptors (VEGFR-1, 2, and 3), and c-KIT and FLT3 kinases [2]. Preclinical studies in xenograft models have shown a broad-spectrum antitumor activity of sorafenib in colon, breast, and non-small-cell lung cancer xenograft models [2]. These observations have been confirmed in phase II/III clinical trials that have shown the activity as well as the clinically acceptable safety profile of sorafenib either as a single agent [3,4] or in combination with other agents [5].

There are several reasons to consider sorafenib therapy for the treatment of breast cancer. First, MAPK is upregulated in MBC; even tumors that do not possess a mutant Ras gene often express high levels of activated MAPK [6]. Elevated MAPK activity in primary breast tumors may indicate more aggressive cancer, be prognostic for shorter relapse-free survival [7], and may represent a possible mechanism of drug resistance [8]. Second, VEGF may promote tumor growth both by inducing angiogenesis and by direct activation through VEGFR-1 on tumor cells leading to activation of MAPK pathways [9]. Third, recent clinical evidence supports the use of antiangiogenic agents in the treatment of advanced breast cancer [10–12].

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The growth-inhibitory effects of sorafenib in preclinical models and its favorable toxicity profile suggested the potential utility of sorafenib. In this report, we describe the results of a phase II study of sorafenib as a single agent in patients treated with one or more lines of chemotherapy for MBC.

Methods

This multinational, open-label, uncontrolled, single-arm phase II study was conducted in accordance with the Declaration of Helsinki after approval by the local ethics committees of all five participating institutions. Written informed consent was obtained from all patients. The study was sponsored by Bayer HealthCare Pharmaceuticals and Onyx Pharmaceuticals Inc.

Patient eligibility

Patients eligible for the present trial were at least 18 years of age; had histologically documented MBC that had progressed after at least one prior chemotherapy regimen for metastatic disease; had progressed after at least one adjuvant hormonal therapy if estrogen/progesterone receptor positive; and were not eligible for, or had refused trastuzumab therapy if human epidermal growth factor receptor 2 (HER2) positive. Patients were required to have at least one lesion measurable by computed tomography scan or magnetic resonance imaging according to Modified World Health Organization Tumor Response Criteria; at least 4 weeks since the last cytotoxic chemotherapy treatment; adequate hematologic, hepatic, and renal function; and a life expectancy of at least 12 weeks. Exclusion criteria included congestive heart failure more severe than New York Heart Association class II, cardiac arrhythmia requiring antiarrhythmic treatment (excluding beta-blockers or digoxin), active coronary artery disease or ischemia, and any condition that was unstable or could jeopardize safety. Women with active central nervous system metastases were not eligible. Patients who had received prior treatment with any agent that targeted VEGFR or endothelial growth factor receptors, such as bevacizumab, were excluded.

Study design and treatment

The study was initially planned as a Simon two-stage design with the null hypothesis that the response rate is less than 5% and the alternative hypothesis that the response rate is 17% ($\alpha = 10\%$; 90% power). However, before the study was officially activated, new data that was available from studies in renal cell carcinoma suggested that sorafenib delayed disease progression without improving response rates [13]. Therefore, the study was conducted based on a single-stage design while maintaining the same hypotheses and power but with $\alpha = 5\%$.

Patients were screened no earlier than 28 days before study entry. The treatment period was divided into 28-day cycles, during which patients were treated continuously with oral sorafenib at a dose of 400 mg twice daily until the occurrence of tumor progression, toxicity, or withdrawal of consent. Dose reduction to 400 mg sorafenib once daily was allowed in case of grade 2–3 toxicities. During the first treatment cycle, visits were scheduled on days 1 and 15. Thereafter, visits were scheduled on the first day of each cycle (every 4 weeks).

Efficacy and safety evaluations

The primary endpoint was overall best response. Secondary endpoints included percentage of patients with stable disease for at least 16 weeks, time to and duration of response, time to disease progression (TTP), and overall survival (OS). In this study, TTP was defined as the time from randomization until objective tumor progression; OS was defined as the time from randomization until death. For the efficacy analysis, all patients who received one or more cycles of therapy and had at least one postbaseline tumor measurement were considered evaluable; patients who exhibited objective disease progression before the end of cycle 1 were also included. Tumor response measurements and assessments were conducted at baseline and every 8 weeks (every other cycle) until treatment discontinuation. Response to therapy was assessed using the Modified World Health Organization Tumor Response Criteria.

For safety analyses, all patients who received at least one dose of sorafenib were included. The incidence, severity, and relationship of adverse events to sorafenib were recorded using the National Cancer Institute's Common Toxicity Criteria version 3.0. Adverse events were assessed by an investigator during all visits [baseline (day 1 of cycle 1 before first sorafenib treatment), day 15 of cycle 1 (C1D15), and on the first day of every cycle thereafter] and 30 days after the last dose of sorafenib. A modified scale was used for hand–foot skin reaction (HFSR) to facilitate interpretation [14].

Biomarker samples and assays

In an attempt to identify markers predictive of clinical benefit, correlations between tumors or blood biomarkers (proteins with expected relevance to sorafenib's mechanism of action or to disease biology) and clinical endpoints were explored. Blood samples collected at baseline, C1D15, day 1 of cycle 2 (C2D1), and day 1 of cycle 3 (C3D1) were assayed for circulating protein biomarkers by sandwich enzyme-linked immunosorbent assay. VEGF, the soluble extracellular domain (ECD) of the epidermal growth factor receptor (sEGFR), the soluble ECD of human EGFR-2 (sHER2), and urokinase-type plasminogen activator levels were measured in serum, and plasminogen activator inhibitor-1 was measured in plasma. A tumor sample (paraffin block or unstained paraffin sections mounted on slides) obtained at diagnosis or around the

time of screening was collected from each patient and evaluated by immunohistochemistry for the expression of phosphorylated extracellular signal-regulated kinase (pERK), HER2, estrogen receptor, and progesterone receptor according to published techniques. In view of the fact that the impact of sorafenib on tumor shrinkage was minimal and that sorafenib improved disease stabilization, the secondary endpoints of TTP and OS were chosen for correlative analyses rather than the primary endpoint of overall best response.

Statistical analyses

Based on the two-stage design that was initially planned for the study, the maximum number of patients required for the evaluation of objective tumor response was 50. For the single-stage design the number of patients required was 54.

A Cox regression model (proportional hazards) was used to examine the relationship between each individual biomarker (either baseline levels or change from baseline where applicable) and patient outcome (TTP and OS). Baseline biomarker level analyses included all patients with a baseline sample available, whereas change from baseline analyses included only patients who had both a baseline sample and the relevant during-treatment sample. All analyses were performed using biomarker levels as either a continuous variable or binned into two groups using the 75th percentile. The biomarkers were binned in this manner to separate low from high levels or to separate small from large changes based on the hypothesis that high-level activation of growth-promoting signaling pathways may predispose sensitivity to sorafenib activity. For binned variables, Kaplan–Meier survival functions were generated for each bin of each biomarker-outcome pair.

Results

A total of 56 patients were enrolled from 3 February 2004 to 29 July 2004. Two patients were not treated on account of protocol violation and withdrawal of consent; 54 patients received at least one dose of sorafenib. Two of the 54 treated patients did not have postbaseline evaluations (one patient discontinued because of adverse events and one because of withdrawal of consent); therefore, 54 patients were analyzed for safety and 52 for efficacy. Table 1 shows the baseline characteristics of all patients. Patients had received between one and 13 prior chemotherapy regimens. Of note, 78% of patients were heavily pretreated, having received three or more prior chemotherapy treatments. Some of the more commonly used (in more than 20 patients) antineoplastic and immunomodulating therapies in the palliative setting are shown in Fig. 1. All 54 patients valid for safety analyses eventually discontinued the study drug; 46 (85%) patients discontinued because of progressive disease, four (7%) because of adverse events (drug-related adverse event, $n = 1 - \text{HFSR}$), three (6%) because of death attributed to disease progression, and one (2%) because of withdrawal of consent.

Table 1 Patient baseline characteristics ($N = 54$)

Characteristics	Value
Median age, range (years)	56 (28–77)
ECOG status, *n* (%)	
0	30 (56)
1	22 (41)
2	2 (4)
ER status, *n* (%)	
Positive	35 (65)
Negative	17 (31)
Unknown	2 (4)
PR status, *n* (%)	
Positive	28 (52)
Negative	24 (44)
Unknown	2 (4)
HER2/neu status (FISH)	
Overexpressed	2 (4)
Normal	17 (31)
Missing	7 (13)
Unknown	28 (52)
HER2/neu status (IHC)	
IHC+	23 (43)
IHC++	5 (9)
IHC+++	11 (20)
Missing	1 (2)
Unknown	14 (26)
Patients with triple negative tumors	17 (31)
Number of prior systemic anticancer regimens, *n* (%)	11 (20)
1	4 (7)
2	8 (15)
≥ 3	42 (78)
Number of tumor sites, *n* (%)	
≤ 3	28 (52)
>3	26 (48)
Metastatic sites, *n* (%)	
Liver	37 (69)
Bone	28 (52)
Lymph nodes	23 (43)
Lung	21 (39)
Skin	7 (13)

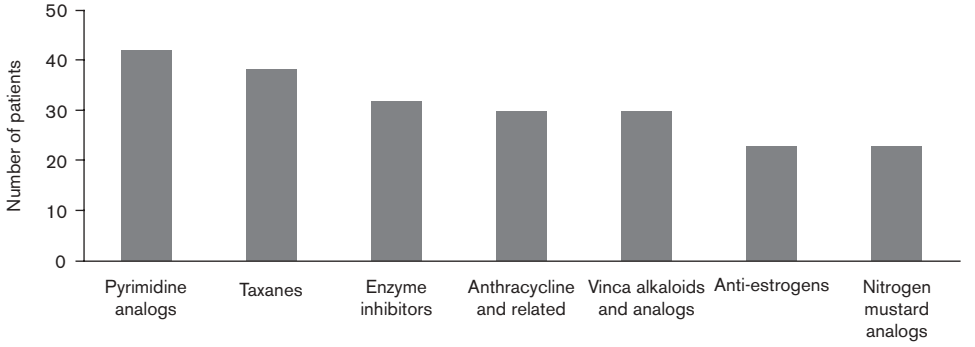
ECOG, Eastern Cooperative Oncology Group; ER, estrogen receptor; FISH, fluorescence in-situ hybridization; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry; PR, progesterone receptor.

All patients received sorafenib per protocol and stayed on therapy for a median of 60 days (range 1–530 days), or two cycles (range one to 18 cycles). The median daily dose of sorafenib was 400 mg twice daily for each cycle.

Efficacy

No complete response was observed; partial response was observed in one patient (2%) and stable disease for at least 8 weeks in 20 patients (38%). In 12 (23%) patients, disease stabilization persisted for 16 weeks or longer, and seven patients (13%) had no disease progression at 6 months. Median TTP was 58 days (95% confidence interval 52–112 days); four patients were censored because of death. Median OS was 259 days (95% confidence interval 177–323 days) with censoring for 19 patients who were alive at the time of analysis. For the patient with a partial response, the time to response was 145 days and the duration of response was 256 days.

Fig. 1



Patients with prior palliative antineoplastic and immunomodulating therapies.

Safety

The overall incidence of drug-related toxicity was 28% grade 1, 43% grade 2, and 19% grade 3. The only grade 4 drug-related adverse event (2%) consisted of an increase in γ-glutamyl transpeptidase. One patient withdrew from the trial because of a drug-related adverse event (grade 3 HFSR). As shown in Table 2, the most common drug-related events were dermatologic, constitutional, and gastrointestinal. An analysis of cycle-specific and cumulative event rates revealed that HFSR and rash/desquamation occurred relatively early during sorafenib therapy. The highest event rate occurred during cycle 1. Hypertension and bleeding were rare (1.9 and 3.7%, respectively) and were grade 1 events.

Table 2 Drug-related adverse events occurring in more than 5% of patients (*N*=54)

	Grade level				
Event, *n* (%)	1	2	3	4	Total
Any event	15 (27.8)	23 (42.6)	10 (18.5)	1 (1.9)[a]	49 (90.7)
Rash/desquamation	8 (14.8)	7 (13.0)	3 (5.6)	–	18 (33.3)
Anorexia	11 (20.4)	4 (7.4)	–	–	15 (27.8)
Hand–foot skin reaction	7 (13.0)	3 (5.6)	2 (3.7)	–	12 (22.2)
Pruritus	12 (22.2)	1 (1.9)	–	–	13 (24.1)
Diarrhea	5 (9.3)	5 (9.3)	–	–	10 (18.5)
Alopecia	7 (13.0)	1 (1.9)	–	–	8 (14.8)
Nausea	5 (9.3)	–	1 (1.9)	–	6 (11.1)
Fatigue	4 (7.4)	–	2 (3.7)	–	6 (11.1)
Mucositis (oral)	5 (9.3)	1 (1.9)	–	–	6 (11.1)
ALT/AST increase	2 (3.7)	1 (1.9)	3 (5.6)	–	6 (11.1)
Gastritis	2 (3.7)	2 (3.7)	1 (1.9)	–	5 (9.3)
Vomiting	3 (5.6)	–	1 (1.9)	–	4 (7.4)
Neuropathy – sensory	2 (3.7)	2 (3.7)	–	–	4 (7.4)
Stomach pain	3 (5.6)	1 (1.9)	–	–	4 (7.4)
Flushing	3 (5.6)	–	–	–	3 (5.6)
Head pain	2 (3.7)	1 (1.9)	–	–	3 (5.6)
Dry skin	3 (5.6)	–	–	–	3 (5.6)
Infection	2 (3.7)	1 (1.9)	–	–	3 (5.6)

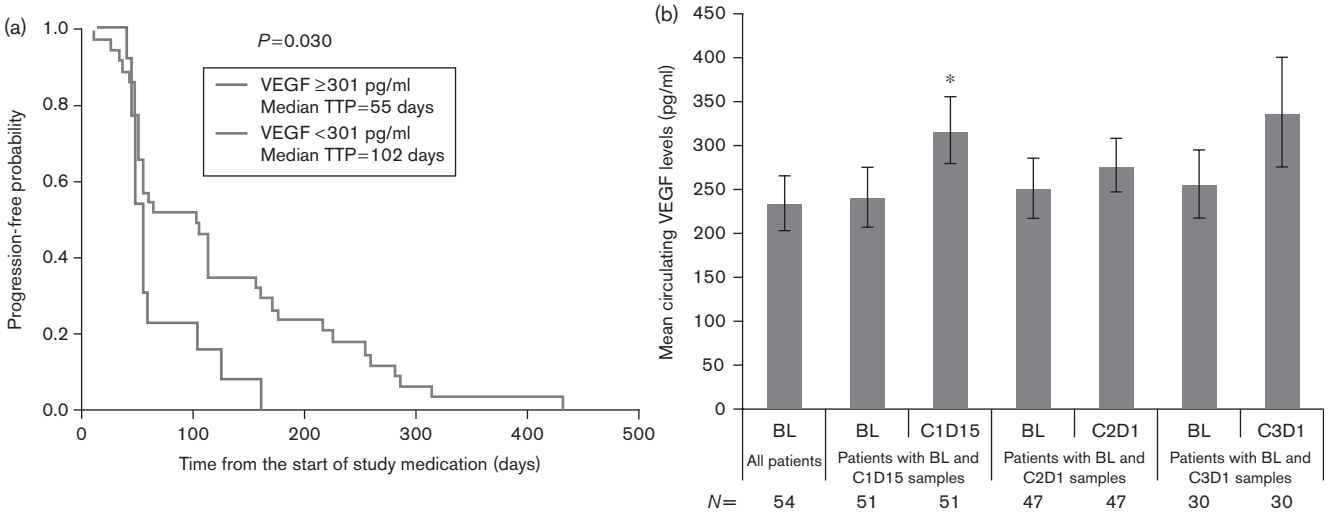
ALT, alanine aminotransferase; AST, aspartate aminotransferase.
[a]Only one patient experienced a grade 4 drug-related adverse event (increase in the levels of γ-glutamyl transpeptidase).

Circulating and tumor biomarkers

An examination of circulating levels of VEGF, a growth factor involved in angiogenesis and which binds to and activates VEGFR signaling, revealed that patients with high baseline VEGF levels (≥301 pg/ml) had significantly shorter TTP than those with lower VEGF levels (median 55 days vs. 102 days; $P = 0.030$; Fig. 2a). In addition, during sorafenib treatment, VEGF increased significantly at C1D15 compared with baseline (Fig. 2b).

Plasma levels of sEGFR and sHER2 were also measured. Higher baseline circulating sHER2 levels or larger increases in sHER2 levels during treatment were significantly associated with poor outcome in this study. Higher baseline sHER2 levels correlated with shorter TTP in an analysis using baseline sHER2 as a continuous variable ($P = 0.043$). Although mean levels of sHER2 did not change significantly during treatment, individual patients with larger increases (≥3.57 ng/ml) in sHER2 from baseline to C2D1 had significantly shorter TTP ($P = 0.005$) and a trend toward shorter OS ($P = 0.087$) than those with either smaller increases or decreases in HER2 at this time point (median OS, 119 vs. 270 days; median TTP, 49 vs. 105 days; Fig. 3a and b). Similarly, larger increases (≥2.34 ng/ml) in sHER2 levels at C3D1 were associated with poor outcome (median OS, 127 vs. 439 days, P=0.026; median TTP, 61 vs. 160 days, $P = 0.004$; Fig. 3c and d). Significant increases in sEGFR levels were observed during sorafenib treatment (Fig. 4a). Although the magnitude of change in sEGFR level from baseline did not correlate significantly with outcome (data not shown), in analyses of sEGFR using the literature-defined lower limit of normal (LLN, 45 ng/ml) [15–17] as a cut-off to bin low versus high sEGFR levels, low sEGFR levels at C2D1 were associated with shorter OS and TTP than high levels (median OS, 94 vs. 270 days, $P = 0.0001$; median TTP, 39 vs. 60 days, $P = 0.007$; Fig. 4b and c). However, the association between C2D1 sEGFR levels and outcome should be interpreted with caution because of the small number of patients with sEGFR lower than LLN ($N = 4$ for OS analysis; $N = 6$ for TTP analysis). Although the soluble ECDs of the

Fig. 2



Correlative analyses of circulating vascular endothelial growth factor (VEGF). (a) Kaplan–Meier analysis of baseline circulating VEGF versus time to disease progression (TTP). The 75th percentile VEGF value at baseline, 301 pg/ml, was used to split patients into high and low VEGF groups. (b) VEGF levels at baseline and during treatment. Error bars represent standard errors. BL, baseline; C1D15, day 15 of cycle 1; C2D1, day 1 of cycle 2; C3D1, day 1 of cycle 3. *$P < 0.05$ compared with baseline in a paired *t*-test.

receptor tyrosine kinases examined here revealed associations with outcome, analysis of pretreatment HER2 and pERK levels in tumor tissue did not (data not shown).

Analysis of circulating levels of urokinase-type plasminogen activator and plasminogen activator inhibitor-1, involved in degradation and remodeling of the extracellular matrix, and pretreatment tumor levels of hormone receptors estrogen receptor and progesterone receptor, did not reveal associations with measures of clinical outcome.

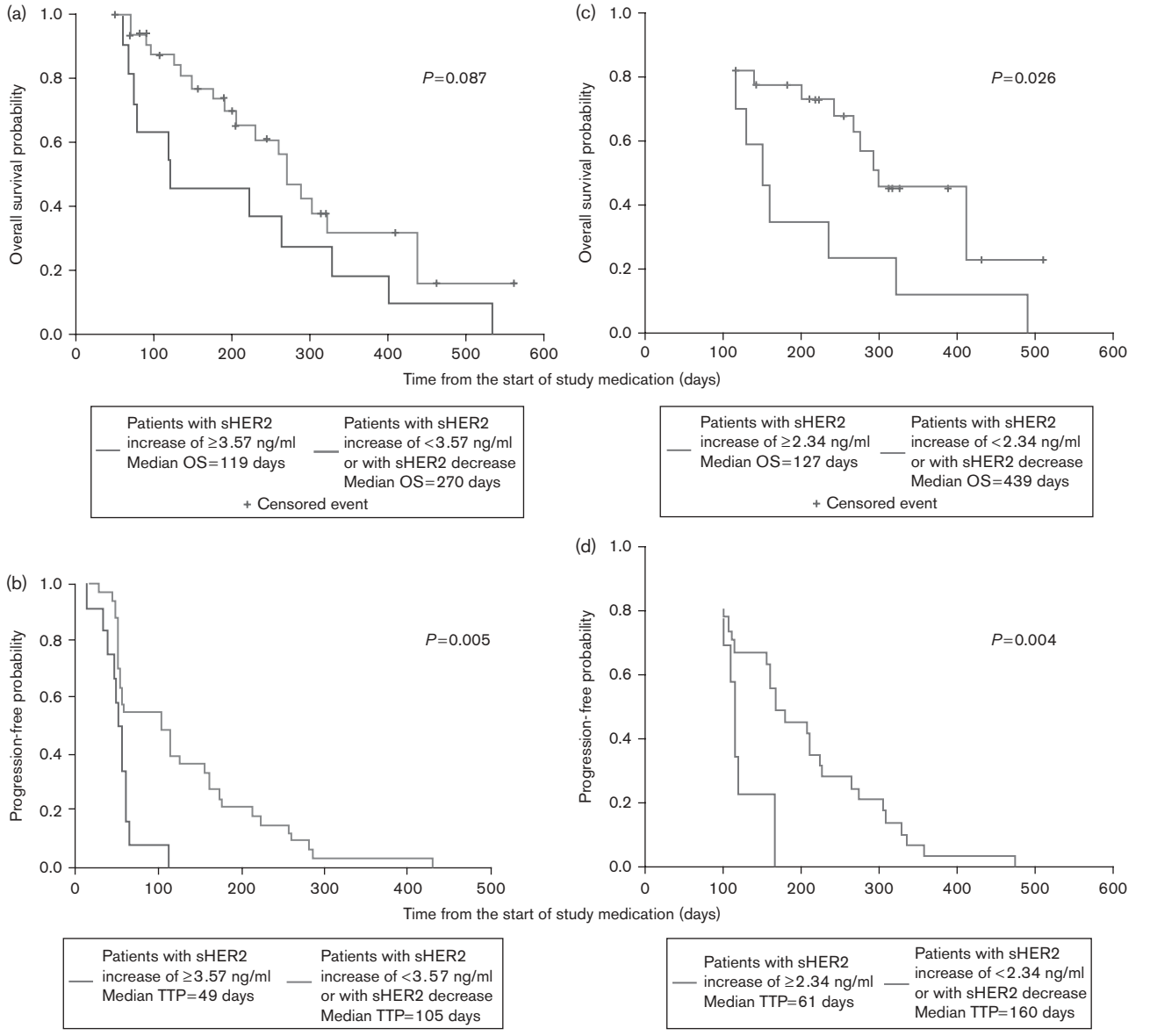
Discussion

The results of this trial indicate that sorafenib had minimal tumor response activity in patients with MBC who received prior therapy [18,19]. In comparison, in a phase I/II trial of bevacizumab involving 75 heavily pretreated patients (40% previously received three or more lines of chemotherapy for MBC), a response rate of 9% was observed; in a phase II trial of sunitinib involving 64 MBC patients (52 had received prior adjuvant chemotherapy and 61 had received previous chemotherapy in the metastatic setting), the response rate was 11%. However, within the limitation of a single-arm study, sorafenib showed encouraging disease stabilization with 24% of patients with stable disease at 4 months and 13% of patients with stable disease at 6 months. This is comparable with disease stabilization observed with other single-agent antiangiogenic agents in advanced breast cancer – with bevacizumab and sunitinib single-agent therapies, 16% of patients had stable disease or disease improvement at 5 months and 6 months, respectively [18,19]. In fact, studies on renal cell carcinoma and hepatocellular carcinoma have shown the efficacy of sorafenib by disease stabilization rather than by tumor shrinkage [3,13,20].

Although antiangiogenic agents have shown modest activity as single agents in advanced breast cancer, these agents may be most promising in combination with standard therapies [11,12]. In designing combination trials, several factors must be taken into consideration, including overlapping toxicities and appropriate measures of product benefit. On the basis of its toxicity profile, within the limitations of an open-label study design, small patient population and short median treatment duration of 60 days, sorafenib is a good candidate for combination therapy given the low incidence of myelosuppression and its acceptable tolerability. Most drug-related adverse events were grade 1 (28%) and 2 (43%). The most common grade 3 adverse reactions of sorafenib seen in the current trial included rash/desquamation (6% of patients), fatigue (4%), and HFSR (4%). There was one report of a grade 4 event (increase of γ-glutamyl-transpeptidase).

Recent publications have highlighted the importance of biomarkers predictive of drug response in oncology, most notably the finding that activating mutations in the KRas gene effectively abolish response to treatment with the EGFR-inhibiting antibody drugs – cetuximab and panitumumab – in colorectal cancer [21,22]. In this study, a number of protein biomarkers were examined, chosen based on specific relevance to signaling pathways inhibited by sorafenib and to breast cancer biology. MBC

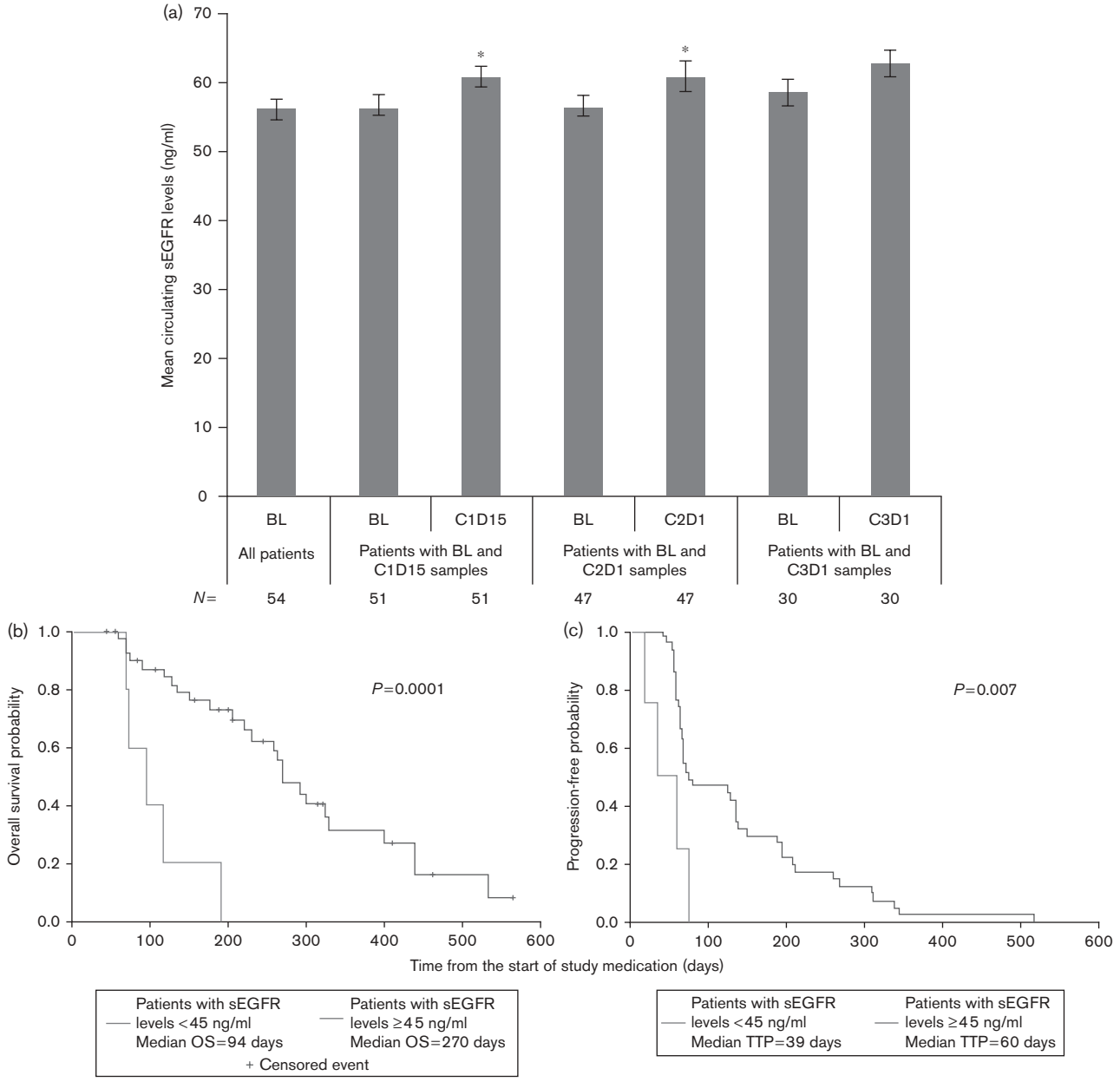
Fig. 3



Kaplan–Meier analyses of change in circulating soluble extracellular domain of human epidermal growth factor receptor-2 (sHER2) versus time to disease progression (TTP) and overall survival (OS). The 75th percentile value for the change in sHER2 at each time point [3.57 ng/ml at day 1 of cycle 2 (C2D1) and 2.34 ng/ml at day 1 of cycle 3 (C3D1)] was used to split patients into bins for analysis. (a) Change in sHER2 from baseline to C2D1 versus OS. (b) Change in sHER2 from baseline to C2D1 versus TTP. (c) Change in sHER2 from baseline to C3D1 versus OS. (d) Change in sHER2 from baseline to C3D1 versus TTP.

is associated with elevated expression of tumor EGFR [23,24] and HER2 [25,26]. Co-expression of both tumor receptors [27], sustained high serum levels of sHER2 [28], or conversion to serum sHER2 positivity [29] have been associated with advanced disease and poor prognosis. Consistent with these observations, in this study, serum levels of sHER2 at baseline and posttreatment increases in sHER2 levels correlated with poorer clinical outcomes. The role of sEGFR as well as the relationship between tumor EGFR levels and sEGFR levels is unclear [16]. Counter-intuitively, a significant proportion of patients with MBC tested had decreased sEGFR levels compared with healthy volunteers; and in one study, patients with decreased sEGFR had worse outcomes with hormone therapy than those with normal sEGFR [16]. In this study, patients with sEGFR less than LLN at C2D1 had poorer outcomes than those with levels greater than LLN, supporting these published observations. A number of different cancer types exhibit elevated circulating VEGF as compared with healthy volunteers, and higher

Fig. 4



Correlative analyses of the soluble extracellular domain of the epidermal growth factor receptor (sEGFR) in plasma. (a) sEGFR levels at baseline and during treatment. Error bars represent standard errors. *$P < 0.05$ compared with baseline in a paired *t*-test. (b) Kaplan–Meier analysis of C2D1 levels of plasma sEGFR versus overall survival (OS). The literature-defined lower limit of normal (LLN) for sEGFR, 45 ng/ml [15–17], was used to split patients into bins for analysis. (c) Kaplan–Meier analysis of C2D1 levels of plasma sEGFR versus time to disease progression (TTP). The literature-defined LLN for sEGFR, 45 ng/ml [15–17], was used to split patients into bins for analysis. C1D15, day 15 of cycle 1; C2D1, day 1 of cycle 2; C3D1, day 1 of cycle 3; BL, baseline.

VEGF levels have been associated with worse outcomes in a number of studies, including a phase III clinical study of sorafenib in renal-cell carcinoma [30,31]. The finding from this study that patients with elevated pretreatment levels of VEGF have a worse outcome than those with lower levels is certainly consistent with the prognostic role of VEGF. However, because of the lack of a control arm in this study, it is not possible to determine whether this observed correlation (and all others described here) might be predictive of sorafenib's treatment effect, indicative of a prognostic biomarker or the course of disease, or a combination of the two. Increases in VEGF during the course of treatment have been observed in studies of sorafenib [30,31] and a variety of VEGF signaling inhibitors [32–36]; similar results were observed in this study as well. Although serum VEGF levels are

impacted by the release of VEGF from platelets during coagulation and thus do not represent only tumor-released VEGF [37], the results from this study remain consistent with previous findings. It should be noted that the biomarker analyses performed in this study were exploratory and hypothesis generating, and interpretation of the results should be tempered by the limited number of patients and the lack of a control arm. Further biomarker investigations in larger, placebo-controlled studies are warranted to confirm the observed correlations and determine their clinical utility.

Although significant advances have been made in our understanding and treatment of breast cancer during the last 20 years which have resulted in a decline in breast cancer mortality, metastatic disease remains incurable, and new drug development is critical to further advancement in patient care. The efficacy results of this study might have been influenced by the heavily pretreated patient population and lack of preselection with regard to receptor status; nonetheless, further investigation of single-agent sorafenib in pretreated patients with MBC is not recommended. Based on studies in other tumor types [5,38,39], we believe that the most promising role for sorafenib in the treatment of MBC would be in combination with standard therapies. Studies evaluating the combination of sorafenib with agents such as letrozole, anastrozole, exemestane, bevacizumab, vinorelbine, paclitaxel, fulvestrant, and abraxane in the metastatic setting are under way. For the adjuvant treatment of breast cancer, sorafenib is being investigated as a single agent and also in combination with doxorubicin and cyclophosphamide with or without paclitaxel, and epirubicin and cyclophosphamide with or without paclitaxel.

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