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Safety, Efficacy, and Biomarker Exploration in a Phase II Study of Bevacizumab, Oxaliplatin, and Gemcitabine in Recurrent Müllerian Carcinoma

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Abstract

Objective—To explore the safety, efficacy, and biomarkers of bevacizumab with gemcitabine and oxaliplatin in women with recurrent platinum-sensitive ovarian carcinoma.

Methods—The patients received bevacizumab (10 mg/kg), gemcitabine (1000 mg/m²), and oxaliplatin (65 mg/m²) on days 1 and 15 in 28-day cycles. The patients with safely accessible tumor underwent intratumoral fluid pressure (IFP) measurements and positron-emission tomographies immediately and 2 weeks after treatment. Blood biomarkers were evaluated at 5 time points.

Results—The trial was closed after enrolling 19 of the 53 projected patient accrual. Thirteen (68.5%) of 19 patients showed a response (1 complete response, 12 partial responses), and 6 patients showed stable disease (31.6%). Median progressive-free survival was 36.9 weeks (258.3 days), and the median overall survival was 112.3 weeks (633 days, not reached). Toxicity was acceptable, and there were no arterial thromboses, serious bleeding, gastrointestinal perforations, or complications from the invasive procedures. Bevacizumab with chemotherapy induced a substantial drop in tumor IFP after treatment. The regimen induced sustained elevation in circulating plasma vascular endothelial growth factor (VEGF), placental growth factor (PlGF), basic fibroblast growth factor (bFGF), soluble vascular endothelial growth factor receptor 2 (sVEGFR2), and circulating progenitor cells. Plasma PlGF, VEGFR2⁺ monocytes, and urinary matrix metalloproteinase 2 activity showed differential associations with treatment outcome when evaluated at baseline and after 14 days of treatment.

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Conclusions—Despite early termination of the study, the results indicate that the regimen was well tolerated and demonstrated activity in platinum-sensitive ovarian cancer. Biomarker evaluations showed that bevacizumab with chemotherapy significantly changed the levels of several circulating cellular and molecular biomarkers. The increases in plasma PlGF and VEGFR2⁺ monocytes showed correlations with outcome. These exploratory data should be further evaluated in future studies of bevacizumab in ovarian cancer.

Keywords

Bevacizumab; Biomarkers; Mullerian cancer

Introduction

Ovarian carcinoma is the second most common gynecologic malignancy, with approximately 22,000 estimated cases in the United States. Initial management of advanced disease consists of aggressive surgical cytoreduction followed by platinum-based chemotherapies. Although 70%-80% of women achieve complete responses (CRs), an equivalent number will experience recurrence. Curing women after a recurrence is elusive, and the goal of therapy becomes to prolong survival and maximize quality of life. Multiple treatment options exist for women with recurrent ovarian cancer, depending on the platinum-free interval, previous therapies and their cumulative toxicities, and patient convenience. For those patients with platinum-sensitive disease (>6-month platinum-free interval), a response rate of 20%-60% would be expected with another platinum-based chemotherapy regimen.¹

Gemcitabine is active as a single agent and in combination, with overall response rates of 13%-22% and stable disease (SD) rates of 40%-50%.²⁻⁴ Oxaliplatin is a third-generation platinum agent that forms DNA adducts resistant to mismatch repair excision. These 2 agents show significant synergy. In the largest reported randomized comparison between oxaliplatin and cisplatin in patients with ovarian cancer, there was a nonstatistically significant survival advantage (36 vs. 25 months) and a better toxicity profile in favor of oxaliplatin.⁵ Phase II trials that evaluated oxaliplatin and gemcitabine reported response rates of 20%-25% and relatively high toxicity.^{6,7}

Bevacizumab (Avastin) is a vascular endothelial growth factor (VEGF)-specific antibody. Bevacizumab with chemotherapy increased progression-free (PFS) and/or overall survival (OS) in metastatic colorectal, lung, and breast cancer in phase III trials.⁸⁻¹⁰ VEGF is critical in ovarian tumor angiogenesis and growth, and causes vascular dysfunction and ascites formation.¹¹ Elevated VEGF expression occurs in all stages of ovarian cancer, is associated with poor prognosis, and is confirmed as an independent prognostic indicator of survival by multivariate analysis.¹² Several phase II studies evaluated the effectiveness of bevacizumab as a single agent in recurrent ovarian cancer or in combination with chemotherapy, with a promising efficacy and safety profile.¹³⁻¹⁵ In addition, a combination of bevacizumab with oxaliplatin and gemcitabine has been tested with promising safety and efficacy results in hepatocellular carcinoma.¹⁶

The mechanisms of action and those of resistance to bevacizumab in ovarian cancer remain unclear. In rectal, liver, and brain tumors, bevacizumab or other anti-VEGF agents can “normalize” tumor vasculature by decreasing tumor vascular permeability and interstitial fluid pressure (IFP), and might potentially improve the efficacy of cytotoxic agents.¹⁷⁻²¹ In addition, changes in several blood-circulating factors were identified as potential biomarkers of response and resistance to bevacizumab therapy.²² We, therefore, explored these

biomarker candidates in a phase II study of bevacizumab, oxaliplatin, and gemcitabine in recurrent ovarian cancer.

Patients and Methods

Patients

Phase II study eligibility criteria included the following: platinum-sensitive (>6-month platinum-free interval) recurrent ovarian, fallopian tube, or primary peritoneal carcinoma with an ECOG PS (Eastern Cooperative Oncology Group Performance Status Scale) of 0/1; up to 2 previous treatment regimens (one platinum based); previous gemcitabine/oxaliplatin was allowed but no VEGF inhibitor; adequate bone marrow (platelets $100,000/\text{mm}^3$, hemoglobin 9g/dL, and absolute neutrophil count $1500/\text{mm}^3$), renal (creatinine 2.0 mg/dL or creatinine $1.5 \times$ upper limit of normal, creatinine clearance of >60 mL/min as estimated by the Cockcroft-Gault formula), and liver function (aspartate transaminase and alanine transaminase $2 \times$ upper limit of normal, alkaline phosphatase $2 \times$ upper limit of normal, except if attributed to tumor, and bilirubin $2 \times$ upper limit of normal); normal blood coagulation parameters (prothrombin time such that the international normalized ratio was < 1.5 and a partial thromboplastin time $< 1.2 \times$ control). All the patients had to have measurable disease defined as at least 1 lesion that could be accurately measured in at least one dimension (longest dimension). Each lesion had to be 20 mm when measured by conventional techniques, including palpation, plain x-ray films, computed tomography (CT) and magnetic resonance imaging, or 10mm in 1 dimension when measured by spiral CT. Patients with readily and safely accessible tumors could elect to have IFP measurements. Once initially identified by the primary physician, the imaging was reviewed by the Biopsy Safety Monitoring Committee, which consisted of a member of the division of interventional radiology, study investigators, and the chief of gastrointestinal and general surgery. If the Biopsy Safety Monitoring Committee deemed that the recurrence could be approached and accessed safely, then the patient would be eligible.

Exclusion criteria included (i) use of an experimental agent within 4 weeks of study entry; (ii) bleeding disorder, coagulopathy, or stroke; (iii) a planned major surgical procedure within 28 days or minor surgical procedure within 7 days of study entry; (iv) urine protein: creatinine ratio 1.0 at screening; (v) nonhealing wound, ulcer, or fracture; and (vi) evidence of central nervous system disease. In addition, patients with significant cardiovascular disease (eg, uncontrolled hypertension $>150/100\text{mmHg}$ despite treatment, myocardial infarction within 6 months, unstable angina), New York Heart Association grade II or greater congestive heart failure, serious cardiac arrhythmia that required medication, a history of deep vein thrombosis, or grade II or greater, clinically significant peripheral vascular disease within 1 year of study entry were excluded. Finally, the study excluded patients with other invasive malignancies, with the exception of nonmelanoma skin cancer. International Federation of Gynecology and Obstetrics stage IA or IB endometrioid endometrial carcinoma diagnosed concurrently with the Müllerian tumor was not a contraindication to trial participation. The trial was approved by the Dana Farber/Harvard Cancer Center Institutional Review Board Committee (Clinical Trial identifier NCT00418093).

Treatments

The patients received gemcitabine ($1000 \text{ mg}/\text{m}^2$ infused over 30 minutes), oxaliplatin ($65 \text{ mg}/\text{m}^2$ infused over 120 minutes), and bevacizumab ($10 \text{ mg}/\text{kg}$ infused over 90 minutes), on days 1 and 15 of 28-day cycles until progression. The 4 patients who underwent IFP evaluation on days 1 and 15 were administered bevacizumab after IFP measurement (on day 1) and underwent fluorodeoxyglucose-positron-emission tomography (FDG-PET) on days 2

and 16. In an effort to correlate metabolic activity with IFP, FDG-PETs were performed in those patients undergoing IFP measurements. Dose adjustments were made according to the greatest toxicity as graded by the CTCAE-3 (Common Terminology Criteria for Adverse Events). Hematologic toxicity was based on blood cell counts obtained within 24 hours of retreatment. For ANC 1000–1499 cells/mm³ and/or a platelet count of 75,000–99,000, gemcitabine was reduced to 750 mg/m². For grade 3 neurotoxicity, oxaliplatin was reduced to 50 mg/m. If a patient experienced an ANC of <1000 cells/mm³, a platelet count of <75,000 cells/mm³, or grade 4 neurotoxicity on day 1 or 15, then treatment was delayed up to 2 weeks or the patient was removed from the study if no resolution occurred. The prophylactic use of filgrastim/pegfilgrastim in conjunction with the first course of treatment was not permitted, but use of filgrastim/pegfilgrastim was endorsed in subsequent cycles under specific circumstances: in patients with prolonged grade 4 neutropenia (ANC < 500 cells/mm³ for >5 days or failure of ANC to recover within 28 days), or the occurrence of febrile neutropenia in a previous cycle of treatment. There were no planned dose reductions for bevacizumab. Adverse events that required delays or discontinuation of bevacizumab included grade 3 hypertension (not controlled with medication), hemorrhage, venous thrombosis, proteinuria, and any arterial thromboembolic event, wound dehiscence, or gastrointestinal perforation.

Response and Toxicity

All the patients who received any drug were included in the analysis. CR, partial response (PR), progressive disease (PD), and SD were defined as per response evaluation criteria in solid tumors. Duration of response was determined from the time of first response until progression. PFS was defined as the date of initiation of treatment until disease progression or the date of last contact. Survival was defined as the length of life from initiation of treatment to death or censored to date of last contact. Toxicity was graded per the National Cancer Institute Common Terminology Criteria of Adverse Events version 3.0. The primary endpoint of this trial was PFS. Radiographic assessments were made every 2 cycles. If elevated before treatment, then cancer antigen (CA)-125 needed to normalize to be considered a CR. PR and progression were determined by imaging alone. Secondary endpoints included toxicity, OS, and biomarker evaluation.

IFP Measurements

Once deemed to have an accessible tumor, the patients underwent IFP measurements as described.²³ Under CT guidance, a 17-gauge, coaxial wick-in-needle was placed into the tumor. The needle was connected to a pressure transducer by polyethylene tubing filled with sterile heparinized (70 units/mL) saline solution. Two to 3 valid measurements were taken at varying depths within the tumor that appeared on imaging to be representative and not fluid or necrotic.

Biomarker Studies

Peripheral blood and urine samples were obtained from all patients enrolled in this study at pretreatment (day 1), and then after 15, 43, and 71 days as well as at the end of study. Plasma analysis was carried out for circulating angiogenic and inflammatory biomarkers VEGF, placental growth factor (PlGF), soluble vascular endothelial growth factor receptor 1 (sVEGFR1), basic fibroblast growth factor (bFGF), interleukin (IL)-1b, IL-6, IL-8, and tumor necrosis factor (TNF)- α (by using multiplex enzyme-linked immunosorbent assay plates from Meso-Scale Discovery) and sVEGFR2 and stromal cell derived factor 1-alpha (SDF1 α) (by using R&D Systems kits). Every sample was run in duplicate. Blood-circulating cells were enumerated in fresh samples by using a standard flow cytometry protocol.²⁴ The quantitative analysis endpoint was the change in the fraction of circulating CD34^{bright}CD45^{dim} CPCs or VEGFR2⁺CD45⁺ monocytic cells among blood mononuclear

cells after treatment. Urinary activities of matrix metalloproteinase (MMP)-2 (65 kDa), MMP-9 (95 kDa), and MMP-9/neutrophil gelatinase-associated lipocalin complex (125 kDa) were evaluated by using gel zymography and were semiquantitatively scored from 1 (absent) to 9 (very strong).²⁵

Statistical Analysis

The primary objective of this study was PFS. The original design assumed that the median PFS for this group of patients with standard treatment is 10 months. Thus, the null hypothesis that the PFS rate at 10 months was 0.5 vs. the alternative that it was higher than 0.5 at the 1-sided .1 significance level when using the exact binomial test. The sample size of 53 evaluable patients was chosen to assure a power of 80% if the true PFS rate at 10 months was 0.65. When assuming an exponential PFS distribution, this was equivalent to a median PFS of 16 months. Secondary endpoints included radiographic responses, toxicity, and OS, as well as circulating and functional biomarkers.

For the correlative studies, biomarker changes from baseline were tested by using the exact paired Wilcoxon test and were reported as the on-study to baseline ratios. Associations of log-transformed biomarker levels with PFS and OS were tested in the Cox proportional hazards models. We report *P* values for the Wald test. Missing measurements of biomarkers were excluded from all analyses. We chose the parameters measured based on their known implication in the pathogenesis of disease and were interested in the results separately for each biomarker. Hence, we did not adjust *P* values with respect to multiple biomarkers. However, in multiple comparisons of on-study vs. baseline biomarker levels, we adjusted the *P* values by using the false discovery control method of Genovese et al,²⁶ with weights proportional to the square root of paired measurements. Because the study accrued only 19 of 53 projected patients, the emphasis was placed on biomarker explorations. With *n* = 19, we had 90% power for testing a correlation when using the Kendall *t* test in a 2-sided test and when assuming true Kendall *t* = 0.5 and a type I error of 5%.

Results

Patient Characteristics

Between November 2005 and October 2008, 19 patients were enrolled and treated with oxaliplatin, gemcitabine, and bevacizumab. The study was closed early because of the slow accrual. Four of 19 patients underwent paired percutaneous biopsies, IFP measurements, and FDG-PET imaging. The clinical characteristics of the patients are shown in Table 1. One patient was removed from the trial after her first cycle of therapy. Shortly after beginning treatment she developed headaches, and an magnetic resonance imaging revealed a 2-cm mass in the cerebellar vermis. Other reasons for patients coming off the study included physician discretion (*n* = 4), toxicity (*n* = 3), consent withdrawal (*n* = 2), completed therapy (*n* = 3), and PD after SD (*n* = 3). All of these patients were analyzed with an intent-to-treat analysis and were included in the outcomes data.

Efficacy

The overall response rate in this cohort of patients with platinum-sensitive disease was 68% (13/19: 1 CR, 12 PRs). The other 6 patients (31%) had SD. Median PFS was 36.9 weeks (258.3 days), and the median OS was 112.3 weeks (633 days, not reached) at a median follow-up of 23.4 months (Figure 1).

Safety

The oxaliplatin/gemcitabine/bevacizumab regimen was reasonably tolerated other than fatigue and neurotoxicity, and was associated with the typical toxicities of platinum (Table

2). The most common toxicities were grade 2 or 3 fatigue, neutropenia, diarrhea, nausea and/or vomiting, and neuropathy. There was one grade 4 thromboembolic event. On CT to assess response, this patient was noted to have an asymptomatic large left main pulmonary artery thrombosis. Bevacizumab was withheld while she was anticoagulated with Lovenox and then resumed with the next cycle of chemotherapy. One patient was removed from the study during her third cycle of therapy after she developed a partial small bowel obstruction, related to PD, that resolved with additional chemotherapy. Finally, one patient developed a grade 3 allergic reaction during her fifth cycle and subsequently was taken off the trial. The allergic reaction consisted of itchy hives, swollen hands bilaterally, and flushing. These reactions resolved with discontinuation of the infusion and steroid administration. The patients taken off the trial for toxicity were included in the efficacy analysis.

Biomarker Studies

Tumor IFP measurements—Four of the patients underwent tumor interstitial fluid pressure (IFP) measurements. Tumor IFP decreased 2 weeks after bevacizumab in 2 patients with valid serial measurements (Supplementary Table S1).

PET imaging—FDG-PETs showed avid lesions in 3 of 4 patients evaluated. Repeated FDG-PETs showed a decrease in metabolic activity in target tumor sites in all 3 patients at day 16 after bevacizumab with chemotherapy (Figure 2 and Supplementary Table S2).

Circulating biomarkers—Plasma levels of VEGF, PlGF, and sVEGFR2 significantly and persistently increased after bevacizumab and chemotherapy treatment (at days 15 and 43; VEGF and PlGF also were increased at day 71 and end of study), although plasma bFGF was transiently increased only at day 15; plasma sVEGFR1, IL-1b, IL-6, IL-8, TNF- α , and urinary MMP activity did not significantly change at any of these time points (Table 3 and Supplementary Tables S3 and S4). In addition, bevacizumab in combination with chemotherapy significantly and persistently increased the number of CPCs but not circulating VEGFR2⁺ monocytes in peripheral blood mononuclear cells (Table 3 and Supplementary Table S5).

In exploratory biomarker studies, we found a significant correlation between high baseline levels of circulating VEGFR2⁺ monocytes and urinary MMP-2 activity and the hazard of both rapid progression and death in these patients ($P < .05$) (Table 4). Surprisingly, a greater increase in circulating VEGFR2⁺ monocytes at day 14 after bevacizumab with chemotherapy was associated with longer PFS and tended to correlate with longer OS (Table 4). Finally, a greater increase in PlGF was significantly associated with shorter PFS (Table 4). Other biomarkers showed no significant correlation with outcome (Supplementary Table S6), and no correlation was seen with biomarker changes at days 43 or 71 (not shown).

Discussion

Anti-VEGF therapy has shown promising results in delaying tumor growth and in reducing ascites in ovarian cancer. But its addition to chemotherapy regimens showed improvement in survival outcomes only in some cancers (metastatic colorectal, breast, and lung cancer), and not in others (eg, advanced pancreatic cancer or adjuvant in colorectal cancer), and may depend on the mechanism of action of the chemotherapeutics.²⁷ Thus, studying the interaction between VEGF therapy and different cytotoxics in a disease-specific manner and identifying predictive biomarkers have become priorities.²²

Structural and functional abnormalities in tumor vessels result in high IFP, tissue hypoxia, and nonhomogenous delivery of cytotoxics.^{18,28} Blocking VEGF may improve, at least transiently, the efficacy of cytotoxics by vascular normalization.^{18,29,30} As shown in mice,

blocking VEGF also could result in excessive vascular pruning (ie, antivascular effects) and interfere with drug delivery and could increase expression of other proangiogenic and/or proinflammatory factors.^{31,32} Thus, it is critical to determine the effects of anti-VEGF therapy in a disease- and agent-specific manner.

The underenrollment in our study precluded the projected efficacy analysis. Nevertheless, the activity and safety data for bevacizumab with oxaliplatin and gemcitabine compared favorably with results from previous phase II studies of oxaliplatin and gemcitabine alone.^{6,7} Likely contributing to underenrollment was that there was a competing institutional trial in a similar patient population that evaluated carboplatin and pemetrexed³³ and that, during the course of the study, bevacizumab became available to patients outside participation in clinical trials. Oxaliplatin was chosen for the doublet in this trial given the previous positive results in other phase II trials to explore the possibility that the use of alternative platinum agents in the recurrent setting may provide better responses. Although the use of carboplatin may have improved enrollment, the compelling data from the Arbeitsgemeinschaft Gynäkologische Onkologie Studiengruppe Ovarialkarzinom (AGO-OVAR), National Cancer Institute of Canada Clinical Trials Group (NCIC CTG), and European Organisation for Research and Treatment of Cancer Gynecologic Cancer Group (EORTC GCG) intergroup trial was not available at the time that this trial was opened.³

Biomarkers of anti-VEGF therapy are sorely needed to validate mechanisms of actions and to identify patients likely to respond.²² IFP was measured in 4 patients with no complications. In 2 patients with reliable paired measurements, IFP decreased after combination therapy, which warranted further studies of physiological parameters as biomarkers in this disease. It is impossible to determine if the drop in IFP was induced by bevacizumab, one or both cytotoxics, or the combined treatment. We previously showed that bevacizumab alone as well as paclitaxel alone reduce the tumor IFP in patients with rectal and breast cancer, respectively.^{20,34} Similar to other studies of anti-VEGF agents,²² bevacizumab with chemotherapy significantly increased circulating VEGF and PlGF in patients with ovarian cancer. Interestingly, bevacizumab also significantly increased plasma sVEGFR2, as previously seen in rectal cancer.²⁰ This was in contrast to the significant decrease in plasma sVEGFR2 by anti-VEGF receptor tyrosine kinase inhibitors, which underscores the potential difference between targeting the ligand vs. the receptor activity.²² The pathophysiological significance of these “reactive” changes in circulating biomarkers remains unclear, but preclinical and clinical evidence indicates that these factors are released by the host and not by the cancer cells.^{35,36} Other biomarker changes appear to be more specific to this regimen: plasma bFGF increased after 1 cycle of treatment, and CPCs increased throughout the course of treatment.

High baseline levels of circulating VEGFR2⁺ monocytes and urinary MMP-2 activity were associated with a higher risk of rapid progression and death. Nevertheless, an increase in VEGFR2⁺ monocytes after 1 cycle of treatment associated with a better outcome, and the same trend was seen for plasma TNF- α and CPCs, which suggests a positive result of activation of these inflammatory pathways during combined therapy with bevacizumab and effective chemotherapy (eg, it might reflect tumor cell killing). However, although PlGF increased in most patients after treatment, patients with greater increases in plasma PlGF after 1 cycle of treatment had shorter PFS, which suggests a potential involvement of PlGF (not blocked by bevacizumab) in ovarian cancer resistance to therapy. The role of proangiogenic and inflammatory factors (monocytes or cytokines/chemokines, eg, SDF1 α , TNF- α , IL-6) in tumor resistance to anti-VEGF therapy, supported by preclinical and clinical data,^{22,37} might be differential when these agents are administered alone vs. combined with efficacious cytotoxics. Collectively, these exploratory biomarker data re-

emphasize the need for studying the prognostic vs. predictive value of these biomarker candidates in kinetic studies and an agent- and disease-specific manner.³⁸

In summary, the combination of bevacizumab with oxaliplatin and gemcitabine will likely not be studied further in women with ovarian carcinoma. Given the success and tolerability of carboplatin and gemcitabine combinations in this patient population, the Ovarian Cancer Evaluation of Bevacizumab and Safety (OCEANS) trial that is evaluating this doublet plus bevacizumab is more likely to provide informative efficacy data. With that said, the biomarker results are provocative and consistent with results from other studies of anti-VEGF therapy in other cancers. The regimen significantly increased circulating VEGF, PIGF, sVEGFR2, bFGF, and CPCs. A worse outcome was seen in patients with higher baseline VEGFR2⁺ monocytes and urinary MMP-2 activity, and in those with greater increases in plasma PIGF and decreases in VEGFR2⁺ monocytes after therapy. The biomarker data from this hypothesis-generating study should be validated in women with ovarian cancer in larger trials by using other chemotherapy regimens.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Figure 1.
Kaplan-Meier Plots of Progression-Free Survival (PFS) in Patients with Ovarian Carcinoma Treated with Bevacizumab and Chemotherapy; PFS with Median and 95% Confidence Intervals

Figure 2. Representative Positron Emission Tomographic (PET) Response after Bevacizumab and Chemotherapy. A, B: Treatment With Bevacizumab, Oxaliplatin, and Gemcitabine Resulted in a Marked Decrease in fluorodeoxyglucose (FDG) Uptake in the Tumor at 2 Weeks (B) Compared With Baseline (A)

Table 1

Patient Characteristics

Demographic	Patients (n = 19)
Median Age (range), y	61 (48–71)
Stage, n (%)	
IC	1 (5)
IIIA	3 (16)
IIIC	10 (53)
IV	1 (5)
Grade, n (%)	
2	3 (16)
3	14 (73)
Not known	2 (11)
Histology, n (%)	
Papillary serous	11 (58)
Endometrioid	2 (11)
Clear cell	1 (5)
Other	5 (26)
No. Previous Regimens, n (%)	
1	9 (47)
2	10 (53)
Average Cycles on Trial (range)	4.5 (1-6)

Table 2Toxicity^a

Toxicity	Grade		
	2	3	4
Hematologic Toxicity			
Anemia	1	0	0
Neutropenia	1	5	0
Nonhematologic Toxicity			
Thrombo-embolic	0	0	1 ^b
Fatigue	4	3	0
Nausea/vomiting	3	2 ^c	0
Neuropathy	2	3	0
Liver	0	1	0
Hypertension	1	1	0
Allergic	0	1 ^d	0
Headache	0	1	0
Diarrhea	5	0	0
Dyspepsia	3	0	0
Pain	1	0	0
Anorexia	1	0	0
Dehydration	2	0	0
Rash	1	0	0
Hyperglycemia	1	0	0
Rhinorrhea	2	0	0
Mucositis	1	0	0

^aToxicity as defined by Common Terminology Criteria for Adverse Events, version 3.^bAsymptomatic pulmonary embolism.^cPartial small bowel obstruction.^dItchy hives, swelling of hands bilaterally, flushing, presumed related to oxaliplatin, however, skin testing was negative for carboplatin and cisplatin. There were no fatal toxicities.

Table 3

Circulating Markers That Change Significantly After Bevacizumab With Chemotherapy (Posttreatment) Compared With Pretreatment Values; Data Are Shown as Medians (Interquartile Ranges)

Marker	Pretreatment	Day 15	Day 43	Day 71	End of Study
Plasma PlGF (pg/mL)	21 (18, 27) (n = 19)	36 (32, 42) (n = 19)	41 (37, 52) (n = 17)	44 (41, 51) (n = 17)	44 (37, 59) (n = 8)
<i>P</i> value ^a	N/A	<.0001	<.0001	<.0001	.0078
<i>P</i> -adj value ^b	N/A	<.0001	<.0001	<.0001	.011
Plasma VEGF (pg/mL)	140 (120, 190) (n = 19)	1180 (1020, 1290) (n = 19)	1630 (1460, 1780) (n = 17)	1890 (1570, 2090) (n = 17)	2180 (1870, 2240) (n = 8)
<i>P</i> value	N/A	<.0001	<.0001	<.0001	.0078
<i>P</i> -adj value	N/A	<.0001	<.0001	<.0001	.011
Plasma bFGF (pg/mL)	39 (21, 67) (n = 19)	73 (50, 103) (n = 19)	50 (21, 62) (n = 17)	50 (43, 102) (n = 17)	54 (40, 90) (n = 8)
<i>P</i> value	N/A	.0062	.24	.050	.55
<i>P</i> -adj value	N/A	.021	.30	.095	.75
Plasma sVEGFR2 (pg/mL)	8180 (6919, 9302) (n = 19)	9728 (7192, 11332) (n = 19)	9269 (7501, 11503) (n = 17)	9432 (6631, 10657) (n = 17)	7625 (5943, 9399) (n = 8)
<i>P</i> value	N/A	.011	.0008	.46	.81
<i>P</i> -adj value	N/A	.019	.0031	.57	1.0
CPCs (% PBMCs)	0.04 (0.04, 0.07) (n = 18)	0.14 (0.09, 0.19) (n = 19)	0.15 (0.11, 0.20) (n = 17)	0.12 (0.07, 0.15) (n = 17)	0.06 (0.05, 0.10) (n = 8)
<i>P</i> value	N/A	.0005	<.0001	<.0001	.31
<i>P</i> -adj value	N/A	.0006	.0001	.0001	.44

Abbreviations: adj = adjusted; bFGF = basic fibroblast growth factor; CPC = circulating progenitor cells; N/A = not available; PBMC = peripheral blood mononuclear cells; PlGF = placental growth factor; sVEGFR2 = soluble vascular endothelial growth factor receptor 2; VEGF = vascular endothelial growth factor.

^a *P* values are from the paired exact Wilcoxon test, unadjusted.

^b *P* values are from the paired exact Wilcoxon test, adjusted to control the false discovery rate over time, with weights proportional to the square root of number of the measurements.

Table 4

Baseline Blood Biomarkers Significantly Associated With Time to Tumor Progression and/or Mortality in Patients With Advanced Ovarian Cancer Who Received Bevacizumab With Chemotherapy; Data Are Shown as Hazard Ratios for Progression and/or Mortality, With 95% Confidence Intervals (In Square Brackets) for Rapid Tumor Progression or Mortality^a

Biomarker	Tumor progression		Mortality	
	Baseline	Day 14	Baseline	Day 14
Plasma PIGF	0.48 [0.10, 2.35] (n = 19); P= .37	7.87 [1.65, 37.45] (n = 19); P= .0095	0.12 [0.01, 1.41] (n = 19); P= .091	3.18 [0.18, 57.33] (n = 19); P= .43
Urinary MMP-2 Activity	1.58 [1.02, 2.43] (n = 12); P= .040	0.88 [0.59, 1.33] (n = 11); P= .55	2.57 [1.02, 6.47] (n = 12); P= .045	1.18 [0.66, 2.09] (n = 11); P= .58
VEGFR2⁺ Monocytes	1.90 [1.14, 3.17] (n = 19); P= .013	0.44 [0.21, 0.92] (n = 19); P= .028	2.38 [1.23, 4.59] (n = 19); P= .0098	0.24 [0.05, 1.14] (n = 19); P= .073

Abbreviations: MMP = matrix metalloproteinase; PIGF = placental growth factor; VEGFR2⁺ = vascular endothelial growth factor receptor 2.

^aP values are from the Wald test in proportional hazards model.