Clinical Cancer Research

Pediatric Phase I Trial and Pharmacokinetic Study of Trebananib in Relapsed Solid Tumors, Including Primary Tumors of the Central Nervous System ADVL1115: A Children's Oncology Group Phase I Consortium Report

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Abstract

Purpose: Trebananib is a first-in-class antiangiogenic peptibody (peptide–Fc fusion protein) that inhibits Angiopoietin 1 and 2. A pediatric phase 1 trial was performed to define trebananib dose-limiting toxicities (DLT), recommended phase 2 dose (RP2D), and pharmacokinetics (PK).

Experimental Design: Trebananib was administered by weekly infusion. Three dose levels (10, 15, or 30 mg/kg/dose) were evaluated using a rolling-six design. Part 2 evaluated a cohort of subjects with primary central nervous system (CNS) tumors. Pharmacokinetic sampling and analysis of peripheral blood biomarkers was performed during the first 4 weeks. Response was evaluated after 8 weeks. Correlative studies included angiogenic protein expression and DCE-MRI.

Results: Thirty-seven subjects were enrolled (31 evaluable for toxicity) with median age 12 years (range, 2 to 21). Two of 19 evaluable non-CNS subjects developed DLT at the 30 mg/kg dose level, including venous thrombosis and pleural effusion. In the CNS cohort, 3/12 subjects developed DLT, including decreased platelet count, transient ischemic attack, and cerebral edema with headache and hydrocephalus. Other grade 3 or 4 toxicities included lymphopenia ($n = 4$), anemia, thrombocytopenia, neutropenia, vomiting, and hypertension ($n = 1$ each). Response included stable disease in 7 subjects, no partial or complete responses. Two subjects continued study treatment with prolonged stable disease for 18 cycles (neuroblastoma) and 26 cycles (anaplastic astrocytoma). Pharmacokinetics appeared linear over 3 dose levels. Correlative studies demonstrated increased PlGF and sVCAM-1, but no change in endoglin or perfusion by DCE-MRI.

Conclusions: Trebananib was well tolerated in pediatric patients with recurrent or refractory solid or CNS tumors. RP2D is 30 mg/kg. *Clin Cancer Res; 23(20); 6062–9. ©2017 AACR.*

Introduction

Angiogenesis is a hallmark of malignancy, and anti-angiogenic agents represent a growing class of novel therapeutics. Current FDA-approved antiangiogenic drugs target angiogenesis through inhibition of the VEGF pathway or epidermal growth factor receptor (EGFR; ref. 1). Despite the efficacy of anti-VEGF therapy in a wide variety of tumor types, the clinical benefit of VEGF pathway inhibition remains modest at best (2, 3). One mechanism by which tumors may overcome VEGF inhibition is activation of the angiopoietin pathway (4).

Angiopoietins are cytokines that regulate blood vessel growth in development as well as malignancy, and represent a parallel pathway that does not depend on signaling through VEGF (1, 4). There are four known angiopoietins (abbreviated Ang1-4) that are ligands of the Tie2 receptor tyrosine kinase. In addition to hemangiogenic effects, angiopoietins also regulate lymphangiogenesis (5) and appear to have a role in neurogenesis (6, 7). Elevated levels of angiopoietins, particularly Ang2, are associated with metastatic disease and poor prognosis in a variety of malignancies (8–14). Co-inhibition of Ang1 and Ang2 *in vivo* results in augmented reduction of tumor vascularity compared with inhibition of Ang2 alone, purportedly by preventing Ang1-mediated normalization of tumor vasculature in addition to blocking Ang2-mediated angiogenesis (15), supporting the rationale for therapeutic co-inhibition of Ang1 and Ang2. Additional preclinical studies have demonstrated that angiopoietin inhibition may also sensitize human tumor cells to immune attack (16).

Trebananib is a first-in-class peptibody (peptide-Fc fusion protein) that inhibits the angiopoietin pathway. Trebananib binds to Ang1 and Ang2 proteins and prevents their interaction with Tie2 (17). A phase I trial of trebananib in adult cancer patients did not reach an MTD at doses up to 30 mg/kg weekly administered by intravenous infusion. Toxicities included

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Translational Relevance

Angiogenesis is a hallmark of malignancy, and all current FDA-approved antiangiogenic therapeutics function using the mechanism of VEGF pathway inhibition. The angiopoietin pathway acts in parallel to the VEGF pathway to stimulate blood vessel growth in development and cancer. Trebananib is a first-in-class selective inhibitor of Angiopoietin 1 and Angiopoietin 2, limiting cell signaling through the Tie2 receptor tyrosine kinase. This first pediatric trial of trebananib was conducted through the Phase 1 Consortium of the Children's Oncology Group, and describes the safety profile of trebananib in pediatric subjects with relapsed/refractory solid tumors, including primary tumors of the central nervous system. The recommended phase 2 dose of trebananib is 30 mg/kg weekly. Two subjects with very prolonged stable disease on single-agent trebananib suggest that further study of angiopoietin pathway inhibition in pediatric cancer is warranted.

peripheral edema and fatigue (18). Pharmacokinetics in adults were linear, and doses of 3 mg/kg or greater achieved average steady state trough concentrations higher than the optimal biologic dose for tumor inhibition as determined by a preclinical xenograft model (19). However, phase II studies in both ovarian and breast cancer have demonstrated that doses of greater than 10 or 15 mg/kg may provide additional benefit (20–22). Several other clinical trials of trebananib in adult malignancy have been or are being completed. A randomized phase III trial in recurrent ovarian carcinoma demonstrated prolongation of progression-free survival for subjects treated with trebananib in combination with paclitaxel (HR, 0.66; $P < 0.0001$) compared with paclitaxel alone (23), although a follow-up study analysis did not demonstrate significant prolongation in overall survival (24).

We report the results of ADVL1115, a single-agent pediatric phase 1 study of trebananib conducted through the Children's Oncology Group Phase 1 Consortium. The primary objectives were to estimate the MTD and/or recommended Phase 2 dose (RP2D) of trebananib administered as a weekly intravenous infusion to children with recurrent or refractory solid tumors, to describe the toxicities associated with trebananib administration on this schedule, and to characterize its pharmacokinetics and immunogenicity. Following determination of the RP2D, additional objectives included an assessment of the tolerability of the trebananib in children with primary central nervous system (CNS) tumors and measurement of changes in CNS vascular permeability by MRI perfusion. Secondary objectives included an assessment of objective anti-tumor activity within the confines of a phase I trial and measurement of biologic markers of angiogenesis.

Patients and Methods

The protocol was conducted in two parts. The first was a dose-finding phase in children with recurrent or refractory solid tumors and the second was an assessment of the tolerability of the RP2D in children with CNS tumors. Although CNS tumors are the most common solid malignancy in children (25), the rationale for this two-part design was specific concern for potential serious CNS toxicity in the event of intratumoral bleeding or thrombosis in subjects with CNS disease, as well as a desire to evaluate imaging endpoints within the CNS.

Patient eligibility

Patients age 1 to 22 years with solid (Part 1) or CNS (Part 2) tumors refractory to standard treatment for which no curative therapy existed who had measurable or assessable disease, were eligible. Histologic verification of malignancy (at diagnosis or recurrence) was required with the exception of diffuse intrinsic brainstem tumors (DIPG), optic pathway tumors or pineal tumors with elevated serum tumor markers. Other eligibility criteria included Lansky or Karnofsky performance score of ≥ 50; full recovery from the acute toxic effects of prior therapy; adequate bone marrow function (peripheral absolute neutrophil count (ANC) $\geq 1{,}000/mm^3$ and platelet count $\geq 100{,}000/mm^3$), renal function (normal serum creatinine for age and gender, or creatinine clearance $\geq$ 70/mL/min/1.73 m^2), liver function [bilirubin $\leq$ 1.5 times upper limit of normal (ULN) for age, ALT $\leq$110 U/L, serum albumin $\geq$ 2 g/dL], cardiac function (shortening fraction of $\geq$ 27% or ejection fraction of $\geq$ 50% by echocardiogram or radionuclide study, no known cardiac disease), neurologic function (seizures well controlled, nervous system disorders resulting from prior therapy $\leq$ grade 2, no CNS hemorrhage), coagulation (no active bleeding, PT and PTT $\leq$ 1.2 times ULN, INR $\leq$1.2), blood pressure $\leq 95^{th}$ percentile for age, height and gender. Patients were not eligible if they had an uncontrolled infection; non-healing wound, active bleeding; a bleeding diathesis or intratumoral hemorrhage; were pregnant or breastfeeding; were receiving corticosteroids and not on a stable or decreasing dose for at least 7 days; or were receiving other investigational drugs, anticancer agents, anti-graft-versus-host-disease agents post-transplant, anticoagulants, anti-inflammatory or anti-platelet agents, or antihypertensive agents. Patients with CNS tumors were excluded if there was evidence of CNS hemorrhage of more than punctate size and/or more than three foci of punctate hemorrhage on baseline MRI, including ECHO-gradient sequences.

The protocol was reviewed and approved by the Cancer Therapeutics Evaluation Program of the National Cancer Institute (NCI) and institutional review boards of all participating institutions. Informed consent and child assent, when appropriate, were obtained from all participants and/or parents or legal guardians.

Drug administration and study design

Trebananib was supplied by Amgen and distributed to participating institutions by the Cancer Therapeutics Evaluation Program (National Cancer Institute, Bethesda, MD). Drug was administered intravenously once a week. The initial dose was given over 60 minutes. If the first infusion was not associated with an infusion reaction, subsequent doses were given over no less than 30 minutes. A cycle was considered 28 days, and subjects were allowed to continue on therapy until disease progression as long as laboratory eligibility criteria continued to be met.

Dose calculation was based on actual body weight, and intrapatient dose escalation was not allowed.

Three dose levels were planned (10, 15, and 30 mg/kg) using a rolling six design (26). Intermediate dose levels (7 and 20 mg/kg) were planned if an MTD was reached. After the RP2D was determined, additional subjects were enrolled in a PK expansion cohort at the RP2D to evaluate pharmacokinetic parameters in subjects across age groups (both < and > 12 years). Part 2 of the

study was opened to enrollment of subjects with primary CNS malignancy after the RP2D was established in Part 1.

Toxicities were graded according to the NCI Common Terminology Criteria for Adverse Events (CTCAE) version 4. Hematologic dose-limiting toxicity (DLT) was defined as any hematological toxicity possibly, probably or definitely attributable to trebananib requiring treatment interruption for > 14 days, any arterial thromboembolic events, any ≥ grade 3 venous thromboembolic event, or any thrombotic event requiring systemic anticoagulation. Non-hematological DLTs were defined as any grade 3 or 4 non-hematological toxicity possibly, probably or definitely attributable to trebananib, with the specific exclusion of grade 3 nausea and vomiting of <3 days duration; grade 3 liver enzyme elevation < 7 days duration; grade 3 or 4 fever < 5 days duration; grade 3 infection <5 days duration; grade 3 hypophosphatemia, hypokalemia, or hypomagnesemia responsive to oral supplementation; and allergic reactions. A grade 2 non-hematologic toxicity persisting for >7 days and sufficiently medically significant or intolerable to require treatment interruption was considered dose limiting. Hypertension was considered a DLT if blood pressure was >25 mmHg above the 95th percentile for age, height, and gender confirmed by repeated measurement, or >10 mmHg above the 95th percentile for >14 days after beginning antihypertensive therapy. Any subject who experienced a DLT was considered evaluable for adverse effects. Subjects without DLT who received at least 85% of the prescribed dose per protocol guidelines and had appropriate toxicity monitoring studies performed during the first cycle (28 days) of therapy were considered evaluable for adverse effects.

Pretreatment evaluations included a history and a physical exam; routine laboratory evaluations, including a complete blood count, urinalysis, electrolytes, renal and liver function tests, PT, PTT and INR; an echocardiogram; and radiographs to evaluate the tibial growth plate and tumor staging. A history and physical examination were obtained weekly during the first cycle, then before each subsequent cycle. Laboratory evaluations were repeated weekly during the first cycle, then every other week during subsequent cycles. Tibial growth plate evaluations were obtained before cycles 2, 5, and every 6 months thereafter in those with open growth plates. Disease evaluations were obtained every two cycles. Disease response for solid tumors was assessed according to the revised Response Evaluation Criteria in Solid Tumors (RECIST). CNS tumor response was evaluated by MRI, and neuroblastoma was additionally assessed using MIBG for MIBG-avid tumors.

Pharmacokinetic studies

Blood samples (2.5–3 mL) for pharmacokinetic studies were obtained before and after the infusion on days 1, 8, 15, and 22 of cycle 1. In addition, samples were collected at 2, 6-8, 24 ± 2, 48 ± 4, 96 ± 4 hours after day 22 infusion, and before cycle 2 day-1 infusion. Serum trebananib concentrations were measured using a proprietary validated ELISA assay at Tandem Laboratory (Trenton, NJ) with recombinant human Ang2 as the capture agent. The limit of quantification (LOQ) of trebananib in human serum was 20 ng/mL (27).

Trebananib serum concentration–time data were analyzed by standard non-compartmental methods using the program Phoenix WinNonlin (Pharsight). AUC_{tau} is the area under the plasma concentration–time curve from the pretreatment sample on day 22 through the pretreatment sample before cycle 2 day-1 infusion.

Immunogenicity studies

Serum samples for immunogenicity studies to assess whether neutralizing antibodies had developed were obtained at baseline, before cycle 3, and 30 days after last dose. Each sample was first screened for the presence of anti-AMG 386 binding antibodies using a validated biosensor-based immunoassay (28). Samples that were confirmed to be positive in the immunoassay were further evaluated for neutralizing activities against AMG 386 using a receptor-binding assay, which measured inhibition of the biological activity of AMG 386 to block the interaction of angiopoietins with a soluble Tie2 receptor. If a sample was positive for binding antibodies and demonstrated neutralizing activity at the same time point, the subject was defined as positive for neutralizing antibodies.

Correlative biology studies

Participation in correlative biology studies, including Tie2-expressing monocytes (TEM) and antiangiogenic proteins (Endoglin, PlGF, and sVCAM-1), was optional for subjects >16 kg in weight. Blood samples (3 mL) were collected in EDTA-containing tubes before drug administration on day 1 cycle 1, 48 ± 4 hours after the third dose (day 24 cycle 1), and before drug on day 1 of cycle 2 treatment for these studies in consenting subjects.

Table 1. Patient characteristics for all eligible study subjects ($n = 37$)

Characteristic	Number (%)
Age, y	
Median	12.1
Range	2.3–21.0
Sex	
Male	23 (62.2)
Female	14 (37.8)
Race	
White	23 (62.2)
Asian	2 (5.4)
Black or African American	6 (16.2)
Unknown	6 (16.2)
Ethnicity	
Non-Hispanic	29 (78.4)
Hispanic	8 (21.6)
Diagnosis	
Adrenal cortical carcinoma	1 (2.7)
Alveolar rhabdomyosarcoma	1 (2.7)
Alveolar soft part sarcoma	1 (2.7)
Astrocytoma, NOS	3 (8.1)
Astrocytoma, anaplastic	4 (10.8)
Carcinoma, NOS	1 (2.7)
Embryonal rhabdomyosarcoma	4 (10.8)
Ependymoma	2 (5.4)
Ewing's sarcoma	3 (8.1)
Ganglioneuroblastoma	1 (2.7)
Glioblastoma multiforme	3 (8.1)
Glioma, malignant	2 (5.4)
Neoplasm, malignant	1 (2.7)
Neuroblastoma	4 (10.8)
Osteosarcoma	4 (10.8)
Peripheral neuroectodermal tumor	1 (2.7)
Synovial sarcoma	1 (2.7)
Prior Therapy	
Chemotherapy Regimens	
Median	2
Range	0–7
Radiotherapy	
Median	1
Range	0–5

Table 2. Dose-limiting toxicity summary

Part	Dose level	Number of patients entered	Number of patients evaluable	Number of pts with DLT	Type of DLT(n)
1	10 mg/kg	6	5	0	
1	15 mg/kg	3	3	0	
1	30 mg/kg	6	6	1	Thromboembolic event (1)
1 PK expansion cohort	30 mg/kg	7	5	1	Pleural effusion (1)
2	30 mg/kg	15	12	3	Platelet count decreased (1), Transient ischemic attacks (1), Edema cerebral (1), Headache (1), Hydrocephalus (1)

Two baseline samples for TEMs studies were obtained, samples were shipped on same day, and TEMs were quantified within 24 to 48 hours using a four-color flow cytometer. A panel of monoclonal antibodies, including CD45-FITC (BD Biosciences), mouse IgG1 Isotype Control-PE (R&D Systems), anti-hTie2-PE (R&D Systems), CD14-PerCP (BD Biosciences), and CD16-Alexa Fluor647 (BD Biosciences) were used to define monocytes and the $Tie2^+$ subset of monocytes. All samples were analyzed by a FACS Calibur cell analyzer and Cellquest Pro acquisition and analysis program (BD Biosciences). The $Tie2^+$ subset of the monocyte population was quantified as a percentage of total nucleated cells (29).

Quantitative determination of proangiogenic and antiangiogenic plasma surrogate markers sVCAM-1, Endoglin and PlGF were measured using commercially available quantitative sandwich enzyme immunoassay kits (ELISA; R&D Systems Inc.). Quantitative differences in the biologic markers measured were compared using a paired *t* test and repeated measures ANOVA. SAS software was used to perform the statistical analyses.

Correlative imaging studies

To evaluate for early changes in tumor perfusion, subjects with CNS tumors had dynamic contrast enhanced (DCE)-MRI evaluation at baseline, 48 to 96 hours after the initial trebananib dose, and then after every 2 cycles of therapy. Combined with a vascular reference concentration function obtained through imaging of a slow-flowing vein in the 3D field of view of the tumor, simple 2-compartment kinetic modeling was performed to estimate the fractional blood volume, transendothelial permeability and extravascular extracellular space fraction (30).

Results

Between March 2012 and August 2013, 37 eligible subjects, were enrolled. No enrolled subjects were found to be ineligible. Six subjects were not fully evaluable for toxicity for the following reasons: one subject never started therapy and five experienced disease progression without DLT before completing the first cycle of treatment and evaluation. Patient characteristics are presented in Table 1.

Six subjects were enrolled and began treatment at the 10 mg/kg dose level, one of whom discontinued study therapy due to the development of metastatic CNS disease. None of the 5 evaluable subjects at this dose level had a DLT. Three subjects were subsequently enrolled and completed the first cycle of treatment at the second dose level, 15 mg/kg, and no DLTs were observed. Six subjects were then enrolled at the 30 mg/kg level. One of them experienced a venous thrombosis around the site of a peripherally inserted central catheter (PICC), which was considered a DLT because the patient required

Table 3. A. Non-dose limiting hematologic toxicities observed in evaluable patients (Part 1, $n = 19$)

	Maximum grade of toxicity across cycle 1 (total, 19 cycles)				Maximum grade of toxicity across cycles 2 to 18 (total, 59 cycles[a])			
Toxicity type	**Grade 1**	**Grade 2**	**Grade 3**	**Grade 4**	**Grade 1**	**Grade 2**	**Grade 3**	**Grade 4**
Anemia	4	2			3	3	1	
Lymphocyte count decreased	1	1	1			2		2
Neutrophil count decreased					1	1	1	
Platelet count decreased						1		1
White blood cell decreased	2					3		

B. Non-dose limiting hematologic toxicities observed in evaluable patients (Part 2, $n = 12$)

	Maximum grade of toxicity across cycle 1 (total, 12 cycles)				Maximum grade of toxicity across cycles 2 to 4 (total, 20 cycles[b])			
Toxicity type	**Grade 1**	**Grade 2**	**Grade 3**	**Grade 4**	**Grade 1**	**Grade 2**	**Grade 3**	**Grade 4**
Anemia	1				2			
Hemoglobin increased	1	1						
Lymphocyte count decreased	4	2			5	2	1	
Lymphocyte count increased						1		
Neutrophil count decreased	1							
Platelet count decreased					1			
White blood cell decreased	3				4	1		

NOTE: This table consists of non-dose limiting hematologic toxicities independent of frequency and attribution.
[a]Includes follow-up cycles.
[b]Includes follow-up cycles.

anti-coagulation therapy. No other DLTs were observed in the dose-finding phase of the trial. Seven subjects were then enrolled in the PK expansion cohort, one of whom never started therapy and one of whom discontinued study therapy due to disease progression. One of five evaluable subjects in the PK expansion cohort experienced a DLT of pleural effusion at the site of progressive metastatic neuroblastoma, which along with disease progression necessitated cessation of trebananib.

In the CNS cohort (Part 2), three of 12 evaluable subjects with primary CNS tumors experienced a DLT. One subject experienced grade 2 thrombocytopenia resulting in >14-day delay in starting cycle 2 therapy, one experienced a grade 2 transient ischemic attack, and one experienced an acute life-threatening event consisting of hydrocephalus, headache and cerebral edema associated with tumor progression. The latter event was considered possibly related to study treatment as it occurred 3 days after the first dose of trebananib.

Toxicities are summarized in Tables 2–4: Table 2 summarizes all DLTs observed, Table 3 shows all hematological toxicities irrespective of attribution, and Table 4 shows all non-hematologic toxicities attributed to trebananib.

Therapy was generally well tolerated, with a toxicity profile distinct from agents which target the VEGF pathway at the dose levels studied. Specifically, no bleeding events and only one event of grade 3 hypertension were observed. The expected toxicity of limb edema was rare and mild when it occurred. Pleural effusions were observed exclusively in subjects with pleural disease.

Response evaluation

Subjects completed a median of two cycles of therapy (range, 0 to 26). There were no complete or partial responses observed. Stable disease was observed in seven subjects, two of whom experienced stable disease >4 months verified by central radiologic review (one with neuroblastoma who completed 18 cycles, and one with anaplastic astrocytoma who completed 26 cycles of therapy). One additional subject with high-grade glioma experienced acute neurologic changes during the second cycle of therapy that were initially attributed to progressive disease. This patient remained off study therapy after the event, but resumed trebananib therapy through a compassionate use program when later imaging documented non-progression.

Pharmacokinetic and immunogenicity studies

Trebananib trough concentrations and other pharmacokinetic parameters were evaluable in 31 and 26 subjects, respectively. Trough concentrations reached a plateau on day 15 of cycle 1. Consequently, sample collection on day 22 permitted estimation of steady-state clearance (CL_{ss}). Steady-state pharmacokinetics and trough concentration data are summarized in Table 5. C_{max}, C_{trough}, and AUC_{tau}, increased in proportion to dose across the range of dose levels examined. The mean ± SD terminal elimination half-life and CL_{ss} values were 62.8 ± 16.5 hours and 68.0 ± 28.3 mL/h, respectively. CL_{ss} values for children ≤ age 12 and > age 12 were 52.9 ± 25.6 mL/h and 86.7 ± 17.3 mL/h, respectively. However, only weight maintained a statistically significant association with clearance after simultaneously adjusting for age,

Table 4. A. Non-dose limiting non-hematologic toxicities related to protocol therapy and observed in more than 10%[a] of evaluable patients (Part 1, $n = 19$)

	Maximum grade of toxicity across cycle 1 (total, 19 cycles)				Maximum grade of toxicity across cycles 2 to 18 (total, 59 cycles[b])			
Toxicity type	**Grade 1**	**Grade 2**	**Grade 3**	**Grade 4**	**Grade 1**	**Grade 2**	**Grade 3**	**Grade 4**
Alanine aminotransferase increased	2							
Alkaline phosphatase increased	2							
Anorexia	2				2			
Cough	2							
Edema limbs	3				3			
Fatigue	3	2			3			
Fever	2					1		
Headache	3							
Hematuria	2				2			
Hyperglycemia	2				1			
Hypertension	2	1			2		1	
Hypokalemia	2				2			
Hyponatremia	3				3			
Hypophosphatemia	3	2			1	1		
Nausea	4				1			
Pain in extremity	1	1			1			
Proteinuria	3				2	1		

B. Non-dose limiting non-hematologic toxicities related to protocol therapy and observed in more than 10%[c] of evaluable patients (Part 2, $n = 12$)

	Maximum grade of toxicity across cycle 1 (total, 17 cycles)				Maximum grade of toxicity across cycles 2 to 4 (total, 20 cycles[d])			
Toxicity type	**Grade 1**	**Grade 2**	**Grade 3**	**Grade 4**	**Grade 1**	**Grade 2**	**Grade 3**	**Grade 4**
Aspartate aminotransferase increased	1	1						
Blood bilirubin increased		2				1		
Fatigue	2				1			
Hypertension	2	1						
Hypokalemia	3				1			
Proteinuria	1	1				1		
Vomiting		1	1					

[a]Toxicities that occurred in more than 10% of patients as determined in the first cycle of protocol therapy.
[b]Includes follow-up cycles.
[c]Toxicities that occurred in more than 10% of patients as determined in the first cycle of protocol therapy.
[d]Includes follow-up cycles.

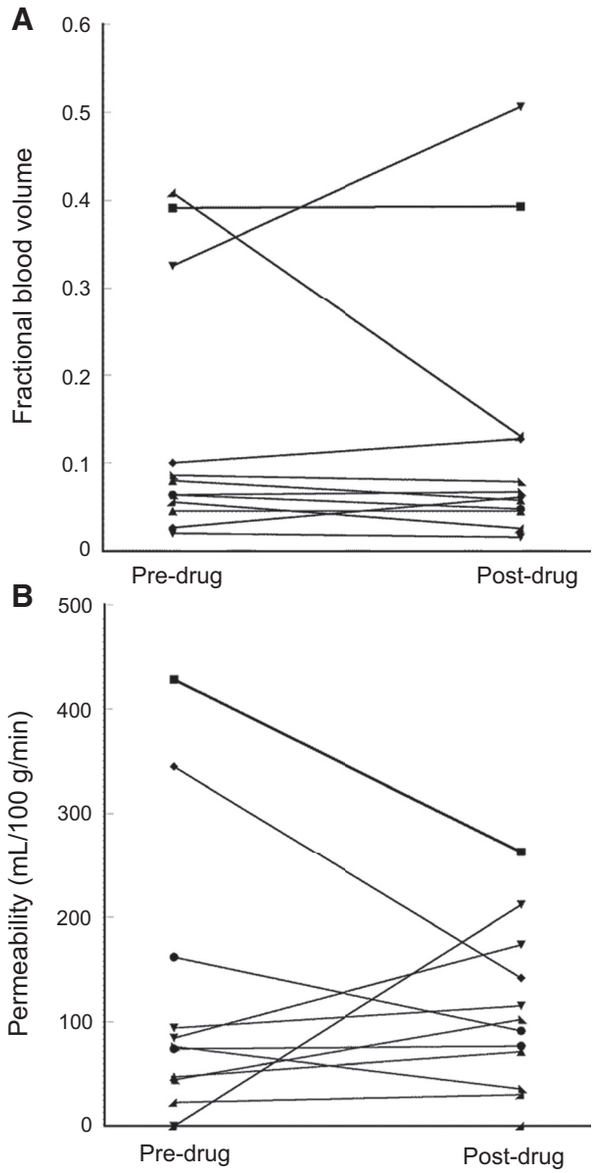

Figure 1.
Changes in fractional blood volume (**A**) and permeability (**B**) of brain tumors before (pre) and 48–96 hours after (post) the first dose of trebananib.

weight, and baseline creatinine in a multiple regression model ($P = 0.01$; alpha $= 0.05$).

Immunogenicity studies were available for 32 subjects. Six subjects (19%) demonstrated trebananib-binding antibodies, and no subjects developed trebananib-neutralizing antibodies.

Correlative biology studies

Correlative biology studies of antiangiogenic proteins were available for two-time points (cycle 1 baseline and day 24) in 18 patients and for three-time points (including cycle 2 day 1) in 15 patients. Compared with baseline, plasma sVCAM-1 and PlGF were increased after AMG 386 treatment ($P < 0.05$). No significant change in Endoglin was observed. Tie2 expressing monocyte (TEM) samples were available at baseline for 20 patients, at two-time points for 8 patients, and at all three-time points for 7 patients. No significant change in the percentage of TEMs was observed in study patients, but study patients did have a significantly higher baseline percentage of TEMs compared with healthy controls ($P = 0.02$).

Functional imaging studies

Paired DCE-MRI images at baseline and after the first dose of trebananib were available for 12 patients in part 2 of the study. A $t2^*$ perfusion scan was performed on 3 of the 12, and t1 permeability on the remaining 9. There was no statistically significant observable change in fractional blood volume or scaled permeability for this time period ($P > 0.05$, see Fig. 1).

Discussion

This pediatric phase I study demonstrated that trebananib was well tolerated as a single-agent administered intravenously on a weekly schedule. An MTD was not reached in this study, and doses of up to 30 mg/kg may be considered for further study in pediatric patients.

Analysis of pharmacokinetic parameters in this pediatric population demonstrated that half-life, C_{max}, C_{trough}, and $AUC_{0\text{-}tau}$ were lower and weight-corrected CL_{ss} was higher than those observed in adult patients (18). This is consistent with published reports of higher weight-corrected clearance of some biotherapeutics in pediatrics, especially for agents that have a significant portion of clearance through a renal mechanism (31). The role of renal clearance of trebananib is supported by studies in adults demonstrating that trebananib clearance is proportional to glomerular filtration rate (32, 33). Despite higher weight corrected CL_{ss} in this pediatric population, the trebananib trough concentrations at all dose levels tested exceeded the minimum target for pharmacological exposure determined by pre-clinical studies (19).

Toxicity associated with trebananib use in this heavily pretreated pediatric population was generally mild and distinct from the toxicities observed in studies of inhibitors of the VEGF pathway (34). No episodes of bleeding were observed, nor was clinically significant hypertension or proteinuria. The distinct toxicity profile suggests a possible role for combination therapy.

Table 5. Summary of trebananib pharmacokinetics (Values are Mean ± SD with %CV in parentheses)

Dose level (mg/kg)	10 mg/kg ($n = 5$)	15 mg/kg ($n = 3$)	30 mg/kg ($n = 18$)
Half-life (h)	63.4 ± 14.7 (23.2)	69.8 ± 19.8 (28.3)	61.4 ± 17.1 (27.8)
C_{max} (μg/mL)	229 ± 61 (26.8)	257 ± 50 (19.3)	633 ± 166 (26.2)
C_{trough} (μg/mL)	11.8 ± 6.8 (57.3)	13.7 ± 6.3 (45.5)	28.5 ± 19.2 (67.2, $n = 23$)
AUC_{tau} (hr·μg/mL)	6,970 ± 2,590 (37.2)	8,260 ± 1,900 (23)	18,630 ± 6,380 (34.3)
CL_{ss} (mL/h)	68.4 ± 39.1 (57.2)	56.0 ± 28.6 (51.0)	69.8 ± 26.4 (37.8)
CL_{ss} (mL/h/kg)	1.63 ± 0.68 (41.6)	1.88 ± 0.39 (20.8)	1.82 ± 0.714 (39.2)

Preclinical studies have supported the potential for efficacy of co-inhibition of VEGF and Angiopoietin pathways (35), trebananib has been given safely in combination with VEGFR kinase inhibitors (36), and further studies are ongoing to evaluate efficacy of trebananib in combination with VEGF inhibition and chemotherapy. A randomized double-blind placebo controlled trial demonstrated that trebananib prolonged survival of women with recurrent ovarian carcinoma when given in combination with paclitaxel (37).

Changes in circulating angiogenic proteins sVCAM-1 and PlGF were observed in response to treatment with trebananib, which is similar to those observed in anti-VEGF therapy (38). Because of limited biologic correlative study participation, it was not possible to evaluate the correlation between disease stabilization and biomarker response. However, the most striking finding was the difference between baseline levels of TEMs observed between study patients in comparison to healthy controls. This suggests that TEMs may be a systemic marker of tumor angiogenic signaling. Recent preclinical studies have suggested that the angiopoietin/Tie2 pathway may play a role in immune modulation, suggesting a possible use in the context of cancer immunotherapy (16). Additional biologic correlative studies are needed to identify patients most likely to respond to antiangiopoietin therapy.

There were a wide variety of diagnoses in the patients included on this study, and no conclusions can be drawn regarding efficacy. Although antiangiogenic therapy is of interest in medulloblastoma, there were no patients with medulloblastoma enrolled on this study, likely due to the concurrent availability of a phase II trial of combination therapy for this diagnosis (NCT01217437). Prolonged stable disease observed in two patients, including one with neuroblastoma and one with anaplastic astrocytoma, is encouraging in response to single-agent therapy, as both of these diseases would be expected to be rapidly progressive within weeks in the absence of effective therapy. Further studies to evaluate the efficacy of trebananib for the treatment of pediatric solid tumors in combination therapy is warranted.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

Disclaimer

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Authors' Contributions

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