Official Protocol Title:	A Phase 2, Double-Blind, Randomized, Placebo-Controlled Study of ACE-011 for the Treatment of Chemotherapy Induced Anemia in Patients with Metastatic Breast Cancer
NCT number:	NCT00931606
Document Date:	13-Mar-2009

CLINICAL STUDY PROTOCOL

A Phase 2, Double-Blind, Randomized, Placebo-Controlled Study of ACE-011 for the Treatment of Chemotherapy Induced Anemia in Patients with Metastatic Breast Cancer

STUDY DRUG: **ACE-011**

PROTOCOL NUMBER: **A011-08**

SPONSOR: **Acceleron Pharma, Inc.**
128 Sidney Street
Cambridge, MA 02139

PPD

MEDICAL MONITOR: **PPD Inc.**

PPD

PROTOCOL DATE: **13 March 2009**

Signature Page

Acceleron Pharma Approval

Signature: PPD **Date:** 17 MAR 2009

Name (print):

Investigator Agreement: I have read the protocol and agree to conduct the study as outlined in the protocol. The study will be conducted in accordance to current United States Food and Drug Administration (FDA) regulations, International Conference of Harmonization (ICH) Guidelines, Good Clinical Practices (GCP), and local ethical and legal requirements.

Signature: _____ **Date:** _____

Name (print): _____

Institution Name and Address:

_____

_____

_____

PROCEDURES IN CASE OF EMERGENCY

Table 1: Emergency Contact Information

Role in Study	Name	Address and Telephone number
PPD Medical Monitor	PPD	PPD Inc. 1800 Perimeter Park Drive, Suite 275 Morrisville, NC 27560-7200 USA PPD
Clinical Trial Manager	PPD	Acceleron Pharma, Inc. 128 Sidney Street Cambridge, MA 02139 USA PPD

2. PROTOCOL SYNOPSIS

Name of Sponsor/Company: Acceleron Pharma, Inc.
128 Sidney Street
Cambridge, MA 02139

Name of Investigational Product: ACE-011

Name of Active Ingredient:
ACE-011 is a fully human fusion protein consisting of the extracellular domain (ECD) of the activin receptor IIA (ActRIIA) linked to the human IgG1 Fc domain.
Mechanism of Action: ACE-011 is a disulfide-linked, glycosylated, dimeric protein. ACE-011 competes with the activin receptor IIA that binds a number of TGF-β superfamily ligands including activin, myostatin (growth differentiation factor [GDF]-8), and GDF-11.

Title of Study: A Phase 2, Double-blind, Randomized, Placebo-Controlled Study of ACE-011 for the Treatment of Chemotherapy Induced Anemia in Patients with Metastatic Breast Cancer.

Study center(s): approximately 25-35 centers

Phase of development: 2

Objectives

Primary:

1. To evaluate the proportion of patients in each ACE-011 dose regimen who achieve a hematopoietic response during the treatment period including up to 2 months after the last dose of ACE-011 treatment of chemotherapy induced anemia in patients with metastatic breast cancer.

Secondary:

1. To evaluate the safety and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer.
2. To evaluate the percentage of patients who achieve an increase in hemoglobin of ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs.
3. To estimate the duration of the hematopoietic response associated with each ACE-011 dose regimen.
4. To estimate the time to the hematopoietic response associated with each ACE-011 dose regimen.
5. To estimate the proportion of patients receiving red blood cell (RBC) transfusions and/or ESAs within each cycle of ACE-011 with each dose regimen.

6. To estimate the effect of each ACE-011 dose regimen on progression free survival (PFS) and objective response rate (ORR).

Exploratory objectives:

1. To estimate the effect of each ACE-011 dose regimen on serum bone biomarkers (OC, BSAP, PINP, CTX, TRACP-5b), urine bone biomarker (uNTX), bone mineral density (DXA scans of lumbar spine and hip) and incidence of skeletal related events (SREs).
2. To evaluate the quality of life during the study in relation to the treatment of chemotherapy induced anemia in patients with metastatic breast cancer during the study.

Methodology: This is a Phase 2, double-blind, randomized, placebo controlled study to evaluate the efficacy, safety, and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer. Patients will be randomized to one of three treatment arms or placebo:

1. ACE-011 0.1 mg/kg subcutaneously (SC) every 28 days for up to 4 doses.
2. ACE-011 0.3 mg/kg SC every 28 days for up to 4 doses.
3. ACE-011 0.5 mg/kg SC every 28 days for up to 4 doses.
4. Placebo SC every 28 days for up to 4 doses.

Number of patients (planned): 105 patients

Diagnosis and main criteria for inclusion

Inclusion Criteria:

1. Women ≥ 18 years of age.
2. Histologically confirmed diagnosis of breast cancer documented by cytology or biopsy.
3. Evidence of metastatic breast cancer with a minimum of one measurable lesion or non measurable lesion per RECIST criteria (Appendix 1).
4. Receiving a multicycle myelosuppressive chemotherapy regimen including one of the following: anthracycline, taxane, gemcitabine or vinorelbine.
5. Planned treatment with the same myelosuppressive chemotherapy regimen for a minimum of 9 weeks after Day 1 of ACE-011/placebo administration.
6. Hemoglobin value between ≥ 6.5 to < 11.0 g/dL (≥ 65 to < 110 g/L).
7. ≥ 30 days must have elapsed (from Day 1) since previous treatment with erythropoiesis-stimulating agent (including concurrent treatment with IV iron) for chemotherapy induced anemia.
8. ≥ 7 days have elapsed (from Day 1) since the last RBC blood transfusion and receipt of ≤ 2 units of blood in the past 30 days.
9. ECOG Performance status of 0 – 2 (Appendix 2).
10. Adequate renal (creatinine ≤ 1.5 x ULN or creatinine clearance ≥ 40 mL/min) and hepatic function (bilirubin ≤ 1.5 x ULN; AST/ALT ≤ 2.5 x ULN).
11. Females of childbearing potential participating in the study are to use adequate birth control measures (i.e. oral contraceptives or two forms of barrier method birth control) during study participation. Females of childbearing potential must have a negative serum pregnancy test within 3 days to ACE-011/placebo administration (Day 1). A female of childbearing potential is a sexually mature woman who has not undergone a hysterectomy or who has not been postmenopausal for at least 24 consecutive months (i.e. who has had menses at some time in the preceding 24 months).
12. Life expectancy of ≥ 6 months.
13. Is willing to adhere to the study visit schedule, understand and comply with all protocol requirements.
14. Understand and sign a written informed consent.

Exclusion Criteria:

1. At the time of screening patients who have any grade ≥ 3 toxicity (as per NCI CTCAE v3.0 (Appendix 3)) (except for the following disease related toxicities: hematological events- anemia, thrombocytopenia or neutropenia or non hematological events- nausea, vomiting, fatigue, or muscle or bone/joint pain).
2. Prior radiation therapy to > 20% of the whole skeleton. Use of palliative radiation if the area being treated is <15% of body surface area with no pelvic radiation or no more than 10% of the bone marrow reserve is radiated is permitted during patient participation in the study, at the discretion of the investigator.
3. Have received more than 5 prior chemotherapy treatment regimens for metastatic breast cancer.
4. Concurrent use of bevacizumab with the chemotherapy regimen during study participation.
5. Clinically significant pulmonary, endocrine, neurologic, gastrointestinal, hepatic or genitourinary disease unrelated to underlying hematologic disorder.
6. Patients with heart failure as classified by the New York Heart Association (NYHA) classification of 3 or higher (Appendix 4).
7. History of thrombosis, deep vein thrombosis (DVT), pulmonary emboli, or embolic stroke.
8. Untreated CNS metastases (exception: CNS metastases treated with whole brain radiotherapy > 6 months prior to randomization).
9. Diagnosis of a myeloid malignancy or known history of myelodysplasia.
10. History of second malignancy within 5 years (except excised and cured basal cell carcinoma, squamous cell carcinoma of the skin or cervical carcinoma in situ).
11. Patients with a recent history (within 14 days of Day 1) of administration of systemic (IV or oral) antibiotics. Patients should be free of infection at least 14 days after the last dose of systemic antibiotic treatment (from Day 1).
12. Uncontrolled hypertension. If hypertension is considered clinically stable, systolic blood pressure (BP) must be < 150 mmHg and diastolic BP must be < 100 mmHg.
13. Known history of hepatitis C antibody (HCV), hepatitis B surface antigen (HBsAg and HB core Ab), or human immunodeficiency virus (HIV) antibody.
14. Deficiency in iron (serum ferritin < 100 ng/mL (< 224.7 pmol/L)), vitamin B_{12}, or folate.
15. History of anemia as a result of inherited hemoglobinopathy such as sickle cell anemia or thalassemia.
16. History of autoimmune or hereditary hemolysis or gastrointestinal bleeding.
17. Patient received treatment with another investigational drug or device within 1 month prior to Day 1, or if the half life of the previous product is known, within 5 times the half life prior to dosing, whichever may be longer.
18. Pregnant or lactating females.
19. History of severe allergic or anaphylactic reactions or hypersensitivity to recombinant proteins or excipients in the investigational product.

Investigational product, dosage and mode of administration:

ACE-011/placebo will be administered as a SC injection, approximately every 28 days, on days 1, 29, 57, and 85 of the treatment period. Each patient will be randomized to one of three treatment arms or placebo:

Dose	Number of Patients
0.1 mg/kg	30
0.3 mg/kg	30
0.5 mg/kg	30
Placebo	15
Total patients enrolled: 105	

ACE-011/placebo dose modification rules are based on hemoglobin values and blood pressure measurements during each treatment cycle. ACE-011/placebo dose interruption and dose reduction rules will be implemented for those patients with hemoglobin of ≥ 11 g/dL, an increase of ≥ 2 g/dL/ 28 days, or hypertension ≥ Grade 2 within each cycle (see Figure 1).

Duration of study: Patients will be followed for a minimum of 7 months following their last dose of study drug. Study participation for each patient is approximately 10.5 months (14 day screening period, 3 month treatment period with a 2 month treatment follow up period, and a 5 month post treatment follow-up period). The start date of the chemotherapy regimen used in this protocol to treat metastatic breast cancer will be recorded. If a patient begins a new chemotherapy regimen during the study, the patient will discontinue further ACE-011/placebo treatment and be followed for survival only. If a patient has a positive anti-drug antibody result at the Day 281/Termination visit, they may be asked to return to the clinical site for additional monthly repeat anti-drug antibody testing for up to 12 months. The study is complete when the last patient enrolled has completed the last protocol required follow up visit.

Criteria for evaluation

Efficacy: The primary endpoint is to establish the proportion of patients who have a hematopoietic response of an increase in hemoglobin of ≥ 0.5 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs.

The proportion of patients with an increase in hemoglobin of ≥ 1g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for 28 consecutive days during the treatment period up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs will be evaluated as a secondary efficacy endpoint.

The hematopoietic endpoints will be determined by the monitoring of hematologic laboratory values, RBC transfusion and/or ESAs administration. The evaluation of response will be from the first dose of ACE-011 through 2 months after the last dose (for each patient). Additional endpoints will be evaluated with the assessment of disease progression by CT/MRI and bone scan and bone mineral density by DXA scans. Bone biomarkers will be evaluated in all

patients and incidence of SREs will be recorded, as applicable. SREs are defined as pathologic fracture, radiation therapy to bone, surgery to bone, or spinal cord compression.

Safety: Throughout the study, safety will be evaluated by the sponsor, medical monitor and principal investigator. Additionally, the DMC will review safety parameters for patients enrolled in the study. All patients will be assessed for safety by monitoring adverse events, clinical laboratory tests, and assessment of quality of life, vital signs and physical examinations on an ongoing basis. ECGs will also be performed during screening, prior to dosing on the first and second cycle, Day 15, and Day 113 (28 days after the last dose of ACE-011) and at Day 281/Termination visit to monitor safety. Blood pressure and lab values will be assessed at each study visit; weekly for the first two treatment (dosing) periods and approximately every one to two weeks for subsequent treatment (dosing) periods.

Pharmacokinetics: The blood samples will be collected at selected study sites from approximately one-third of patients for the evaluation of serum concentrations of ACE-011. Pre-dose PK samples will be collected on Day 1. See Appendix 5 for PK sampling schedule.

Statistical methods

Sample size determination: With an anticipated dropout rate of 20%, a sample size of 30 patients per dose level is planned to achieve at least 80% power with 2 sided significance level of 0.05 to differentiate targeted response rate of p1=0.45 from null hypothesis p0=0.20. An additional 15 patients will be enrolled into the placebo group for safety comparison. A total of 105 patients will be randomized to three ACE-011 dose groups or placebo with a ratio of 2:2:2:1.

Efficacy analysis: The primary endpoint is the proportion of patients who have a hematopoietic response of an increase in hemoglobin of ≥ 0.5 g/dL for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs. The proportion of responders will be determined together with 95% confidence interval.

For binary endpoints, the proportion of responders and 95% confidence interval will be calculated. For time to event endpoints, the Kaplan-Meier method will be used to estimate the survival curve and median time to event. Due to the nature of the study, no multiplicity adjustment will be made for efficacy analysis.

Safety analysis: Data from all patients who receive at least one dose of ACE-011/placebo will be included in the safety analyses. To assess clinical safety, adverse events, vital sign measurements and clinical laboratory information will be summarized. Adverse events and laboratory results will also be summarized by severity grade (NCI CTCAE v3.0). Changes from baseline clinical laboratory values and vital signs will be summarized over time. Descriptive statistics and shift tables will be generated as appropriate.

Schedule of Events

Schedule of Events

	Screen	Treatment Period													Post Treatment Follow up Period				Termination Visit[18]
ACE-011/placebo Dose period		#1				#2			#3			#4	Follow-up						
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	113 (± 7d)	141 (± 7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
Informed consent	X																		
Inclusion/exclusion criteria	X	X																	
Medical history	X																		
Physical examination	X	X				X			X			X	X	X					X
Vital signs[1]	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X					X
FACT-Fatigue	X	X[2]				X[2]			X[2]			X[2]							X
12 lead ECG	X	X[3]		X		X[3]							X						X
Serum BC, transf serum ferriti	X	X[2]				X[2]			X[2]			X[2]							X
Vitamin B12	X																		
Serum folate	X						X						X						X
Coagulation/ Serum chemistry[4]	X	X[5]	X	X	X	X[5]	X	X	X[5]	X	X	X[5]	X	X					X
Hematology[5]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Serum Erythropoietin marker		X[2]				X[2]			X[2]			X[2]		X					X
Peripheral Blood Smear (sample)		X[2]				X[2]			X[2]			X[2]							X
LH, FSH, testosterone, progesterone, estradiol	X	X[2]				X[2]			X[2]			X[2]	X	X					
Anti-drug antibody testing[6]		X				X			X			X	X	X					X
Bone biomarker[7]		X[2]				X[2]			X[2]			X[2]	X	X					X
Back up blood sample for future testing	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X	X	X	X	X
Pregnancy Test	X[8]	X[9]				X[9]			X[9]			X[9]							X
Urinalysis[10]	X	X[2]										X[2]							X
Evaluate transfusion frequency	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X	X	X	X	X

Schedule of Events cont.

	Screen	Treatment Period													Post Treatment Follow up Period				Termination Visit[18]
ACE-011/Placebo Dose period		#1				#2			#3			#4	Follow-up						
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	113 (± 7d	141 (± 7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
Documentation of concomitant medications	X[11]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Evaluation of AEs and SAEs		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Evaluation of Menstrual cycle[12]	X	X[2]				X[2]			X[2]			X[2]	X	X					
CT/MRI scan	X[13]									X[14]			X[14]						
Dual-energy X-ray absorptiometry (DXA[15])	X												X						X
Bone scans[16]	X									X			X						X
ACE-011/placebo administration		X				X			X			X							
Chem 1[17]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

[1] Vital signs include weight, heart rate, seated systolic and diastolic blood pressure, and temperature will be assessed at each visit Height will be collected at screening only

[2] On dosing days (1, 29, 57 and 85), study procedure to be done prior to dosing of ACE-011/placebo

[3] 12 lead ECG is to be performed prior to dosing of ACE-011/placebo including measurement of the QT interval

[4] Coagulation (central lab only) prothrombin time (PT), partial thromboplastin time (PTT), fibrinogen Chemistry (central lab only): AST, ALT, lactate dehydrogenase (LDH), total bilirubin, indirect bilirubin, alkaline phosphatase, total protein, albumin, sodium, potassium, chloride, carbon dioxide, calcium, phosphorus, blood urea nitrogen (BUN), creatinine, uric acid, glucose, amylase, and lipase

[5] Hematology (local lab) hemoglobin and hematocrit Hematology (central lab) complete blood count (CBC) with differential and platelet, CBC includes red blood cells (RBC), white blood cells (WBC), hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), **Reticulocyte should be done on days 1, 8, 15, and days 29, 57, 85, and at the termination visit.** On dosing days (1, 29, 57 and 85), local hemoglobin and hematocrit values are to be drawn and resulted prior to dosing to apply dose modification rules for ACE-011/placebo

[6] If a patient has a positive anti-drug antibody result at the Termination visit, they may be asked to return to the clinical site for additional monthly follow-up, for up to 12 months after their last dose for a repeat anti-drug antibody test

[7] Both a serum and urine samples will be collected at each visit The urine sample should not be from the first morning urine

[8] Serum pregnancy test required of females of child bearing potential only Negative test can be within 3 days of study day 1

[9] For females of child bearing potential only urine (dipstick) pregnancy test to be collected and resulted prior to dosing of ACE-011/placebo

[10] Urinalysis to be performed at screening , prior to administration ACE-011/placebo (Days 1 and 85) and termination visit Assessments (Central lab): pH, specific gravity, protein, glucose, ketones, blood, leukocyte esterase, and nitrite Microscopy required only to follow-up abnormal findings

[11] Screening- any concomitant treatments taken 28 days prior to day of randomization

[12] Females of child bearing potential- collection of menstrual cycle information (date of last menstruation, date of menstrual period onset and duration of menstruation)

[13] A CT/MRI scan acquired within 4 weeks of initiating the current chemotherapy regimen may be used for the screening assessment Head, neck, thoracic, abdominal, and pelvic scans are to be acquired One measurable or non measurable lesion per RECIST is to be identified by the scan Brain (head) CT only to be done to rule out CNS metastases in patients with suspicious clinical symptoms or prior history of CNS metastases

[14] Follow-up CT/MRI scans may be acquired +/- 5 day window for either Day 64 or Day 113 Additional CT/MRI scans acquired during the post treatment follow up period will be performed at the discretion of the investigator

[15] Hip and lumbar spine BMD assessment

[16] Bone scans will be acquired within 4 weeks of initiating the current chemotherapy regimen for those patients who have bone metastases identified Follow up bones scans may be acquired +/- 5 day window for day 64, day 113, and Day 281

[17] Chemotherapy regimens scheduled will be administered to the patient as per standard of care Continuation of the same chemotherapy regimen may be done through the follow up period of the study at the discretion of the investigator

[18] Patients who terminate treatment early due to disease progression and/or begin a new chemotherapy regimen will complete the day 281/Terminiation visit procedures and be followed for survival only (7 months from the last dose of ACE-011/placebo) If a patient withdraws from the study (section 8 4), the Day 281/Termination visit procedures are to be completed and the patient will be followed for survival If withdrawal reason is due to ESA administration, the patient should enter the treatment follow up period

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4. LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

The following abbreviations and specialist terms are used in this study protocol.

Table 2: Abbreviations and specialist terms

Term	Definition
ACTH	Adrenocorticotropic hormone
ActRIIA	Activin receptor IIA
AE	Adverse event
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
AUC	Area under the plasma concentration - time curve
$AUC_{(0\text{-last})}$	AUC from time 0 to time of last quantifiable sample
$AUC_{(0\text{-}\infty)}$	AUC extrapolated to infinity
BMD	Bone mineral density
BP	Blood pressure
BSAP	Bone specific alkaline phosphatase
BUN	Blood urea nitrogen
CBC	Complete Blood Count
CFR	Code of Federal Regulations (US)
CH	Constant domain
CI	Confidence interval
CIA	Chemotherapy induced anemia
CL	Clearance
CL/F	Total clearance
C_{max}	Maximum observed concentration
CNS	Central nervous system
CO_2	Carbon dioxide
CRF	Case report form
CRO	Clinical research organization
CT	Computed tomography
CTCAE	Common Terminology Criteria for Adverse Events
CTX	Serum C-terminal type I collagen telopeptide
DLT	Dose-limiting toxicity
DXA	Dual-energy x-ray absorptiometry
DMC	Data Monitoring Committee
DVT	Deep vein thrombosis
ECD	Extracellular domain
ECG	Electrocardiogram
ECOG	Eastern Cooperative Oncology Group
EDF	Erythroid differentiation factor
ESA	Erythropoiesis-stimulating agents
F	Bioavailability (absolute)
FACT	Functional Assessment of Chronic Illness
FDA	Food and Drug Administration
FSH	Follicle-stimulating hormone
GCP	Good Clinical Practices
GH	Growth hormone

Term	Definition
HBV	Hepatitis B virus
Hct	Hematocrit
HCV	Hepatitis C virus
Hgb	Hemoglobin
HIV	Human immunodeficiency virus
ICF	Informed consent form
ICH	International Conference on Harmonization
IEC	Independent Ethics Committee
IND	Investigational New Drug
IP	Intraperitoneal
IRB	Institutional Review Board
IV	Intravenous
IVRS	Interactive Voice Response System
Kd	Binding coefficient
λz	Elimination rate constant
LDH	Lactate dehydrogenase
LH	Luteinizing hormone
MCH	Mean corpuscular hemoglobin
MCHC	Mean corpuscular hemoglobin concentration
MCV	Mean corpuscular volume
MedDRA	Medical Dictionary for Regulatory Activities
MITT	Modified Intent to Treat (population)
MRI	Magnetic resonance imaging
NOAEL	No adverse events level
NYHA	New York Heart Association
OC	Osteocalcin
ORR	Objective response rate
OVX	Ovariectomized
PD	Progressive disease
PFS	Progression free survival
PHI	Protected health information
PINP	Serum intact procollagen type I N terminal propeptide
PK	Pharmacokinetic
PP	Per-protocol (population)
PT	Prothrombin time
PTH	Parathyroid hormone
PTT	Partial thromboplastin time
RAP-011	Murine version of ACE-011, ActRIIA mFc
RBC	Red blood cell (count)
RECIST	Response Evaluation Criteria in Solid Tumors
SAE	Serious adverse event
SAP	Statistical analysis plan
SC	Subcutaneous
SERM	Selective estrogen receptor modulator
SHAM	Sham-operated
SPEP	Serum protein electrophoresis
SRE	Skeletal-related event
T1/2	Elimination half-life
TIBC	Total iron binding capacity
Tmax	Time to Cmax

Term	Definition
TRACP-5b	Serum tartrate-resistant acid phosphatase isoform-5b
ULN	Upper limit of normal
Vz/F	Volume of distribution
WBC	White blood cell (count)

5. INTRODUCTION

5.1. Indication and Rationale

This study is designed to evaluate the efficacy, safety, and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer.

Treatment of patients with metastatic breast cancer with myelosuppressive chemotherapy is frequently associated with anemia. Chemotherapy induced anemia (CIA) is a significant problem for patients with cancer, causing fatigue and reduced quality of life (1). Patients with CIA are currently treated with blood transfusion and/or erythropoiesis-stimulating agents (2). However, with these treatment options CIA is still an area of unmet medical need. Erythropoiesis-stimulating agents (ESAs) can successfully mitigate transfusion need in a proportion of these patients, but concerns have emerged regarding apparent negative effects on survival and/or tumor progression in certain patient populations. The murine surrogate to ACE-011, RAP-011, has been tested as a single agent in breast cancer cell lines MDA-MB-231 and MCF-7 and no effect on enhanced proliferation of these cell lines has been observed in vitro. Therefore, treatment with ACE-011 may provide a distinct benefit/risk profile to patients with chemotherapy induced anemia.

In both a Phase 1 single dose and multiple dose study of ACE-011 in postmenopausal women, increases in hemoglobin and hematocrit were observed following ACE-011 treatment and remained elevated over the course of study. The observed hemoglobin and hematocrit effects of ACE-011 were dose and time dependent. Please refer to the Investigator Brochure for further information.

Based on the effect of ACE-011 on hematopoiesis and consistent biological phenomena observed in both non clinical and clinical studies, it is hypothesized that the blockade of ActRIIA receptor signaling impedes the terminal differentiation step in erythropoiesis to allow additional rounds of cell replication before cells enter the final differentiation phase. The result is a substantial increase in mature erythrocytes released into the circulation. Since this proposed mechanism is different to that of known ESAs, ACE-011 may provide a different clinical profile in the treatment of CIA.

This study will provide information on the pharmacodynamics (PD) properties regarding the ability of ACE-011 to increase hemoglobin in patients with CIA.

5.2. Description of ACE-011

ACE-011 (ActRIIA-IgG1) is a fully human fusion protein consisting of the extracellular domain (ECD) of activin receptor IIA (ActRIIA) linked to the human IgG1 Fc domain, which includes the heavy chain hinge and constant domains, CH2 and CH3. ACE-011 avidly binds to activin A with a binding coefficient (Kd) approximately 8.9 pM and prevents its binding to endogenous receptors, thereby inhibiting biological effects of activin.

5.3. Activin Biology

Activin A was initially identified as a gonadal differentiation factor involved in modulating follicle stimulating hormone (FSH) secretion from the pituitary (3). Subsequently, the pleiotropic nature of activin A has become more apparent (4). There is a growing body of data suggesting a role for activin A in bone remodeling, specifically as a negative regulator of bone growth (5). Before the two molecules were shown to be identical (6), activin A was also described as erythroid differentiation factor (EDF), effecting red blood cells in the later stages of maturation (7). The mechanism(s) by which activin A influences erythropoiesis remain under investigation and, in fact, there are data from studies in vitro and in animals that support erythropoiesis-stimulatory (8,9) and -inhibitory effects (10).

At the cellular level, activin A binds initially to the high-affinity Type II receptor (ActRIIA). The ligand-bound ActRIIA then recruits the low-affinity Type I receptor (ActRIA or ALK-4). The receptor heterocomplex, through its cytoplasmic protein kinase activity, then activates the Smad signal cascade to eventually influence nuclear transcriptional factors (11, 12). The competitive binding of activin A in the blood by the ACE-011 soluble fusion protein can result in inhibition of ActRIIA receptor signaling pathway by impeding biological processes attributed to activin A.

Activin has also been attributed to erythroid differentiation and has been reported to have proerythrocytic effects and to induce terminal differentiation of RBCs. Inhibition of activin may lead to increases in proliferation of the erythroid lineage. Based on its pharmacologic effects, ACE-011 (ActRIIA-IgG1) is being developed for the treatment of bone loss associated with various disease states (e.g., osteoporosis and treatment of osteolytic lesions in patients with multiple myeloma), and treatment of anemia associated with a variety of disorders, such as chemotherapy induced anemia.

The extracellular domain sequence of ActRIIA is completely conserved among numerous species including mouse, rat, cynomolgus monkey and humans, thus ACE-011 is active in these animals. However, in order to reduce the potential immunogenicity of the fully human molecule, ACE-011, in mice and to maximize the opportunity to maintain exposures in chronic models, a murine surrogate molecule was constructed by exchanging the human immunoglobulin Fc sequence portion of ACE-011 with its murine IgG2a homolog. The resultant construct is referred to as RAP-011 (ActRIIA-mIgG2aFc).

5.3.1. Pharmacology Studies

RAP-011 was evaluated in a broad range of animal pharmacology studies to assess the effects of inhibition of activin A on the biological processes attributed to that regulatory protein. RAP-011 has been shown to have significant effects on the red cell compartment. RAP-011 treatment of mice at 10, 30 and 50 mg/kg by intraperitoneal injection (IP) twice per week for 3 months resulted in a 16-26% increase in red blood cell (RBC) counts compared to control animals. Rats treated with RAP-011 at 0.3, 3 and 30 mg/kg once per week for 3 months showed RBC count increases of 6-15% over control animals. Finally, in cynomolgus monkeys treated with 10, 30 or

50 mg/kg of ACE-011 twice per month for 3 months there was a 21- 24% increase in RBC counts compared to controls animals. In this study, the RBC count increase was apparent as early as 2 weeks following the initial dose of ACE-011.

While the mechanism(s) underlying the stimulation effect of ACE-011 on erythropoiesis are not yet fully understood, it is hypothesized that a blockade of ActRIIA receptor signaling impedes the terminal differentiation step in erythropoiesis to allow additional rounds of erythroid cell replication before cells enter the terminal differentiation phase. The result is a substantial increase in mature erythrocytes released into circulation.

RAP-011 administered at 10 mg/kg to mice three days prior to administration of paclitaxel at 30 mg/kg was sufficient to prevent decreases in red blood cell parameters seen three days later. Mice receiving paclitaxel alone had decreased hematocrit from 43% to 38% three days later. RAP-011 administered 3 days prior to paclitaxel injection was sufficient to keep the hematocrit above 42% at three days and up to two weeks after paclitaxel administration. Therefore prophylactic treatment with RAP-011 was able to prevent paclitaxel induced anemia in mice.

RAP-011 has also been shown to significantly increase bone mineral density and strength in normal animals and in a variety of animal models of bone loss (13, 14, 15). In a murine ovariectomy (OVX)-induced model of osteopenia, RAP-011 (10 mg/kg IV, twice per week x 12 weeks) increased trabecular BMD > 25% in 6-12 weeks compared to the phosphate buffered saline (PBS)-treated controls ($p<0.002$). RAP-011 treatment was able to reverse the accelerated bone loss associated with ovariectomy. As expected sham-operated (SHAM) mice (i.e., intact ovaries) started the study with a greater trabecular BMD than OVX mice. PBS-treated SHAM mice showed a 10% decrease in trabecular BMD after 12 weeks of treatment compared to baseline, while SHAM-RAP-011 mice had a 32% increase in trabecular BMD over the same period. These data demonstrate that RAP-011 is able to increase trabecular BMD in both the normal state and in a model of established bone loss. The femurs of both the RAP-011 treated OVX and SHAM mice showed statistically significant increases in all measured parameters of bone strength (i.e., maximum load, stiffness, energy, ultimate strength, and elastic modulus). In summary, treatment with RAP-011 produced bone of superior quality consistent with an anabolic agent.

RAP-011 was evaluated in a mouse model of skeletal metastasis using luciferase-tagged, human MDA-MB-231 breast cancer cells (estrogen receptor negative). Mice were pretreated for 2 weeks with RAP-011 (10 mg/kg, biweekly, SC) prior to intracardiac injection of tumor cells. Treatment with RAP-011 was continued for an additional 5 weeks following tumor implantation. A parallel study was conducted to examine the effect of RAP-011 treatment, as described above, on survival of MDA-MB-231 bearing mice.

The data demonstrate that RAP-011 treatment acts to inhibit MDA-MB-231 metastasis to bone and may also play a role in inhibiting tumor growth in soft tissue. Furthermore, in the presence of tumor cells, RAP-011 is able to prevent lytic disease through an anabolic mechanism of bone formation. Lastly, inhibition of tumor growth and metastasis with RAP-011 leads to a greater than 25% survival benefit in this model.

The efficacy of RAP-011 was examined in an orthotopic model of breast cancer using luciferase-tagged human MCF-7 breast cancer cells (estrogen receptor positive). Mice were pretreated for 2 weeks with RAP-011 (10 mg/kg, biweekly, SC) prior to the implantation of tumor cells into the mammary fat pad of female nude mice. Treatment with RAP-011 was continued for an additional 7 weeks following tumor implantation at which time mice were sacrificed and assessed for tumor burden. RAP-011 effectively lowered the tumor burden in mice as detected by bio-luminescence. In addition, a 60% reduction in tumor weight was observed in the RAP-011 treated mice.

The ability of RAP-011 to repair osteolytic lesions caused by metastatic disease was investigated in mice. In this model athymic nude mice received an intratibial injection of MDA-MB-231-Luc cells to mimic bone metastasis. Mice were treated with a chemotherapy regimen of paclitaxel (20 mg/kg, IP, every three days) starting seven days after tumor injection and continuing for the duration of the study. On study day 42 mice with a detectable tumor burden by bioluminescent imaging were divided into two groups and began treatment with RAP-011 (10 mg/kg, SC, biweekly) or vehicle. Prior to dosing, μCT scans of the tumor bearing tibia were conducted to identify osteolytic lesions. RAP-011 treatment continued for four weeks (to study day 70) and a final μCT scan of the tibia was performed. On study day 70 osteolytic lesions remained the same or had progressed further when compared to study day 42 in the vehicle treated mice. While some mice showed progression of osteolytic lesions (most likely related to tumor burden) on study day 70, a majority of mice treated with RAP-011 demonstrated repair of the osteolytic lesions seen on study day 42. Therefore treatment with RAP-011 has the ability to repair osteolytic lesions caused by tumors and after cytoxic chemotherapy with paclitaxel.

5.3.2. Toxicology

ACE-011 has been evaluated for toxicological effects in two species, Sprague-Dawley rats and cynomolgus monkeys. The dose levels ranged from 0.3 to 30 mg/kg in rats and from 1 to 50 mg/kg in monkeys. These dose ranges were designed to support Phase 1a single-dose levels of 0.01 to 3.0 mg/kg IV and Phase 1b multiple doses of 0.1 to 2 mg/kg (monthly, SC). Weekly dosing in animals was designed to provide continuous, but fluctuating serum concentrations of ACE-011, which would be mimicked by a one-month dosing interval in humans.

Initial studies utilized IV dosing to identify target organs while longer-term studies (3 months in rats; 3 and 6 months in monkeys) utilized SC dosing to mimic the intended dosing regimen for patients. The expected pharmacologic effect, increased red blood cells (RBCs), hemoglobin (Hb), hematocrit (HCT) and reticulocytes, was observed in all studies, presumably based on the ability of ACE-011 to inhibit activin. A second expected pharmacologic effect of ACE-011, based on inhibition of activin, was the reversible reduction in sperm production and testicular tubular damage in rats. Since male cynomolgus monkeys were sexually immature in these studies, it was not possible to monitor this effect in this species.

Effects on the adrenal gland were only seen in rats and were more pronounced in female rats. This toxicological observation may not be relevant for predicting effects in humans since adrenal toxicity was not seen in cynomolgus monkeys. Dose-limiting toxicity and no observable adverse effect levels (NOAELs) were primarily based on renal toxicity in both rats and monkeys. There

was some indication that kidney toxicity was an indirect effect, based on the formation of antibody/antigen complexes, but since these studies were not designed to investigate toxicological mechanisms, a definitive cause of renal impairment and renal damage was not determined. Development of antibodies to ACE-011 was noted in all of these studies, which is an expected immune reaction in rats and monkeys dosed with a human protein.

The no observable adverse effect level (NOAELs) from the 3 month SC studies was 3 and 30 mg/kg in rats and monkeys, respectively. Because the kidney findings were observed at all dose levels, the NOAEL from the 6 month monkey study is < 10 mg/kg.

5.4. Summary of Clinical Experience

5.4.1. A011-01: A Phase 1a Study in Healthy Postmenopausal Women

ACE-011 was first studied in a randomized, Phase 1a, single dose, dose escalation study in healthy, post-menopausal females (16). ACE-011 was diluted in normal saline administered as an IV infusion (over approximately 1 hour) or as a SC injection on Day 1. Dose levels were 0.01, 0.03, 0.1, 0.3, 1.0, and 3.0 mg/kg IV and 0.03 and 0.1 mg/kg SC. A total of 48 subjects were enrolled; 5 active and 1 placebo at each of the IV and SC dose levels. All subjects were followed for 4 months following a single dose administration.

The pharmacokinetics of ACE-011 was linear for all IV and SC doses. Across IV doses ranging from 0.1 through 3.0 mg/kg, the mean clearance (CL) ranged from 0.092 to 0.128 mL/h/kg, the volume of distribution ranged from 73.7 to 110 mL/kg, and the mean T1/2 ranged from 23.7 to 31.8 days, with no apparent dependence on dose. After SC administration of 0.03 and 0.1 mg/kg, ACE-011 was completely absorbed, and the mean T1/2 was approximately 30 days, with no apparent dependence on dose.

The most commonly occurring treatment-emergent adverse events (AEs; i.e., those occurring in more than 1 subject in any treatment group) were headache, infusion site reaction, injection site hemorrhage, and toothache. The majority of the injection site reactions and injection site hemorrhages were in the first IV cohort and were related to infiltration of the IV site. The majority of treatment-emergent AEs were mild in severity and were judged to be unrelated to ACE-011. No deaths, serious AEs (SAEs), or AEs leading to discontinuation were reported. Changes in red blood cells (RBCs), hemoglobin, reticulocytes, liver function tests, glucose, uric acid, amylase and lipase occurred in some subjects; however, none of these laboratory results were reported as adverse events. Mild, transient elevations in pancreas enzymes, liver enzymes, or glucose were reported as AEs in five patients. There were no clinically significant changes from baseline in vital signs, physical examination, endocrine function, or electrocardiogram (ECG) data. Preservation of adrenal cortical function was monitored through evaluation of serum electrolytes (sodium and potassium levels) and through cortisol response to adrenocorticotropic hormone (ACTH) stimulation. No clinically significant findings were observed at doses up to 3.0 mg/kg IV, and ACE-011 was well tolerated in healthy, post-menopausal female volunteers in single dose levels up to 0.1 mg/kg SC and 3.0 mg/kg IV, the highest dose levels tested in this study.

5.4.2. A011-02: A Phase 1b Study in Healthy Postmenopausal Women

ACE-011 was studied in a Phase 1b, single-center, randomized, double-blind, placebo-controlled, multi-dose, dose-escalating study to evaluate the safety, tolerability and pharmacodynamics of ACE-011 in healthy postmenopausal women. Four cohorts of 10 subjects each were planned at the following dose levels: 0.1, 0.3, 1.0, and 2.0 mg/kg administered subcutaneously. Within each cohort, subjects were to be randomized to either active or placebo treatment in an 8:2 ratio. Subjects were to receive one SC injection of ACE-011 or placebo every 28 days for a total of 4 doses. All subjects were to be followed for 12 weeks after the last dose.

The treatment phase of the study was terminated early after a dose-limiting pharmacodynamic effect was observed. While on study, one subject in the 1.0 mg/kg cohort experienced a Serious Adverse Event (SAE) of progressive and persistent hypertension that was attributed to a rapid and significant rise in hemoglobin and hematocrit levels. Due to symptoms of headaches, nausea, eye pain, dizziness and vomiting approximately 1 week following the second dose, the subject was hospitalized for monitoring and evaluation of her hypertension. The head MRI, fundoscopy, and ECG performed were reported as normal and headache symptoms resolved following therapeutic phlebotomy. The SAE resolved and the subject was discharged from the hospital the following day. The hypertension was monitored closely and managed initially with antihypertensive medication. The hypertension resolved by the end of the study without need of any antihypertensive medication. The subject received aspirin prophylactically until the end of the study and continued taking ibuprofen as needed for headaches. Further details can be found in the Investigator Brochure.

Based on the magnitude of the hematopoietic response, the Sponsor suspended dose escalation to the 2.0 mg/kg dose level and further dosing in all cohorts as a result of this dose-limiting pharmacodynamic effect at the 1.0 mg/kg dose level. In total, 31 subjects were enrolled and treated. Dose levels of ACE-011 administered included 0.1, 0.3, and 1.0 mg/kg (Cohorts 1, 2, and 3 respectively). All subjects randomized to active treatment in Cohort 1 received all 4 planned doses of ACE-011. Due to early discontinuation of study drug, subjects randomized to active treatment in Cohort 2 received 3 doses of ACE-011, and subjects randomized to active treatment in Cohort 3 received 2 doses of ACE-011. Subjects randomized to placebo treatment received between 1 and 4 doses of study treatment.

In the analysis of the unblinded data, after the administration of the first dose, a dose and time dependent increase in hemoglobin values was observed (see Table 3 below):

Table 3: A011-02: A Phase 1b Study in Healthy Postmenopausal Women: Hemoglobin Evaluation

Evaluation Time Point	Mean Change from Baseline (g/dL)			
	Placebo N=7*	ACE-011 0.1 mg/kg N=8	ACE-011 0.3 mg/kg N=8	ACE-011 1.0 mg/kg N=8
Baseline Mean, pre-Dose 1	13.20	13.11	13.30	12.71
Day 8	0.17	0.68	0.85	1.21
Day 15	-0.27	0.43	0.44	1.75
Day 29	0.27	0.61	1.21	2.68**
Day 36	0.56	0.64	1.89	2.96
Day 43	-0.02	0.89	1.21	2.85
Day 57	0.27	1.28	1.64**	2.09
Day 64	0.53	1.11	2.49	2.21
Day 71	-0.10	1.34	2.09	1.66
Day 85	0.38	1.18**	2.55	1.86
Day 92	0.00***	1.04	1.60***	3.80***
Day 99	-0.20	1.21	3.20***	1.28***
Day 113	-0.02	1.30	1.29	1.04
Day 141	0.30	0.95	0.34	2.30
Day 169	0.20***	0.06		2.00***

*The number of placebo subjects with data decreases over time as a result of the early discontinuation of the study (i.e., there were placebo subjects in each dosing cohort). There were seven placebo subjects with data at baseline, Days 8, 15, and 36; six subjects with data at Days 29, 43, 57, and 85; five subjects with data on Days 71 and 113; four subjects with data on Day 64; three subjects with data on Day 141; two subjects with data on Day 99; and one subject with data on Days 92 and 169.

** Number of doses administered per treatment group: 0.1 mg/kg 4 doses; 0.3 mg/kg 3 doses; 1.0 mg/kg 2 doses. Data beyond this study day are considered follow-up results.

***n=1

No severe or life-threatening events were reported. The incidence of the most common treatment-emergent AEs (i.e., occurring in more than one subject in any treatment group) is presented below (see Table 4). The most notable AEs were those related to increases in hematologic laboratory measures in the 1.0 mg/kg dose group: hematocrit, hemoglobin, and RBC count. AEs of increased hemoglobin and/or hematocrit were reported for seven of the eight subjects in this dose group. These events were reported as mild or moderate elevations; all were considered probably related to study drug treatment. Three of the subjects in the 1.0 mg/kg group with elevated hemoglobin levels underwent phlebotomies and all hemoglobin elevations were resolved by the end of the follow-up period. No erythroid lineage AEs were reported in the 0.1 or 0.3 mg/kg treatment groups.

Table 4: A011-02: Incidence of Most Common Adverse Events

System Organ Class and Preferred Term	Dose Group			
	Placebo N=7 n (%)	0.1 mg/kg N=8 n (%)	0.3 mg/kg N=8 n (%)	1.0 mg/kg N=8 n (%)
GASTROINTESTINAL DISORDERS				
Diarrhea	0 (0.0%)	2 (25.0%)	0 (0.0%)	0 (0.0%)
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS				
Asthenia	0 (0.0%)	1 (12.5%)	2 (25.0%)	0 (0.0%)
Fatigue	2 (28.6%)	0 (0.0%)	1 (12.5%)	2 (25.0%)
INFECTIONS AND INFESTATIONS				
Viral upper respiratory tract infection	0 (0.0%)	4 (50.0%)	0 (0.0%)	0 (0.0%)
INJURY, POISONING AND PROCEDURAL COMPLICATIONS				
Limb injury	0 (0.0%)	1 (12.5%)	0 (0.0%)	2 (25.0%)
INVESTIGATIONS				
Hematocrit increased	0 (0.0%)	0 (0.0%)	0 (0.0%)	6 (75.0%)
Hemoglobin increased	0 (0.0%)	0 (0.0%)	0 (0.0%)	7 (87.5%)
Red blood cell count increased	0 (0.0%)	0 (0.0%)	0 (0.0%)	3 (37.5%)
METABOLISM AND NUTRITION DISORDERS				
Decreased Appetite	1 (14.3%)	0 (0.0%)	2 (25.0%)	0 (0.0%)
MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS				
Arthralgia	2 (28.6%)	0 (0.0%)	2 (25.0%)	0 (0.0%)
Muscle spasms	0 (0.0%)	1 (12.5%)	0 (0.0%)	2 (25.0%)
NERVOUS SYSTEM DISORDERS				
Dizziness	0 (0.0%)	0 (0.0%)	2 (25.0%)	2 (25.0%)
Headache	2 (28.6%)	4 (50.0%)	3 (37.5%)	2 (25.0%)
Paresthesia	1 (14.3%)	0 (0.0%)	0 (0.0%)	3 (37.5%)
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS				
Oropharyngeal pain	2 (28.6%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
VASCULAR DISORDERS				
Hot flush	0 (0.0%)	2 (25.0%)	0 (0.0%)	1 (12.5%)

Note: Table presents AEs reported in more than one subject in any treatment group.

Headache was reported frequently in all treatment groups with no apparent dose response trend, though the reports in the higher dose groups were more often considered possibly related to treatment. Most headaches were reported as mild (one headache was reported as moderate).

Paresthesia and dizziness were reported more frequently in the ACE-011 groups, though the events were generally not considered drug related. Other frequently reported events (e.g. fatigue, upper respiratory infection) did not appear to increase with dose and were generally mild.

Hemoglobin levels for all subjects with elevations had returned to within normal limits by the end of the study.

Adrenal cortical function was monitored through evaluation of serum electrolytes (sodium and potassium levels) and through cortisol response to adrenocorticotropic hormone (ACTH) stimulation. All ACTH simulation test results were normal.

PK results confirmed that the PK of ACE-011 is linear following the first SC doses of all three dose levels tested, 0.1, 0.3, and 1.0 mg/kg. The terminal half life ($T_{1/2}$) of ACE-011 following the last dose in all three dose groups was identical, with mean $T_{1/2}$ being approximately 23 days. The mean CL/F ranged from 3.05 to 3.90 mL/d/kg, the mean Vz/F ranged from 97.47 to 103.03 mL/kg, and the mean $T_{1/2}$ ranged from 20.92 to 23.34 days in all 3 dose levels, with no apparent dependence on dose.

Bone mineral density was assessed by dual-energy X-ray absorptiometry (DXA). Mean change and mean percent change in BMD results from baseline to study end are summarized in the table below. A dose-dependent increase in the BMD of the total hip from baseline to end of study was observed, with a significant and rapid increase of 2.4% in the 1.0 mg/kg dose group, compared to a 0.7% decrease in the placebo group. BMD results for lumbar spine showed slight increases of 0.4% to 1.0% from baseline to study end in all active treatment groups, compared with a 0.5% decrease in the placebo group (see Table 5 below):

Table 5: A011-02: A Phase 1b Study in Healthy Postmenopausal Women: BMD

	Treatment Group							
	Placebo N=7		**ACE-011 0.1 mg/kg N=8**		**ACE-011 0.3 mg/kg N=8**		**ACE-011 1.0 mg/kg N=8**	
	Mean Change	Mean Percent Change	Mean Change	Mean Percent Change	Mean Change	Mean Percent Change	Mean Change	Mean Percent Change
Lumbar Spine (g/cm^2)	-0.0020	-0.5%	0.0099	0.7%	0.0082	1.0%	0.0051	0.4%
Total Hip (g/cm^2)	-0.0062	-0.7%	0.0050	0.6%	0.0075	0.9%	0.0220	2.4%

Number of doses administered and day of last dose per treatment group: 0.1 mg/kg 4 doses (Day 85); 0.3 mg/kg 3 doses (Day 57); 1.0 mg/kg 2 doses (Day 29). Data beyond this study day are considered follow-up results.

5.4.3. A011-04: A Phase 2a Study in Patients with Osteolytic Lesions of Multiple Myeloma

Study A011-04 is an ongoing, non-IND, Phase 2a, multi-center, randomized, multiple-dose study to evaluate the safety, tolerability and efficacy of ACE-011 in patients with osteolytic lesions of multiple myeloma. Safety evaluations include adverse events (AEs), clinical laboratory tests, standard 12-lead electrocardiogram (ECG), vital signs, Eastern Cooperative Oncology Group

(ECOG) performance status and physical examinations. Additionally, the study includes the assessment of biochemical markers of bone formation and resorption, skeletal related events (SREs), bone mineral density (BMD) by duel-energy x-ray absorptiometry (DXA) and bone pain by visual analog scale (VAS).

In this study, patients are randomized in a 4:1 ratio to one of three dose levels of ACE-011 (0.1, 0.3 and 0.5 mg/kg) or placebo, to be administered to patients every 28 days by subcutaneous injection, for up to four doses over a 3-month period. The test article is being evaluated in combination with the anti-myeloma therapy of melphalan (4 mg/m^2 on days 1-7), prednisolone (40 mg/m^2 on days 1-7) and thalidomide (100 mg per day) (MPT). The sites, Sponsor and Sponsor representatives are blinded to treatment assignment.

As of 10 March 2009, 30 patients have been randomized, enrollment is complete and treatment is ongoing. Fifteen patients have received multiple doses of their assigned treatment; 7 patients have received 4 doses, 3 patients have received 3 doses and 5 patients have received 2 doses. Of the remaining 15 patients, 12 patients have received their first dose of ACE-011 or placebo and 3 patients have not yet received their first dose. There have been no ACE-011 related serious adverse events (SAEs) reported. Per the monitored preliminary data on 18 patients presented to the Independent Data Monitoring Committee (DMC) on 9 February 2009, there were 8 Grade 3/severe AEs reported in 4 patients, including neutropenia, platelet count decreased, pain in extremity, back pain and anemia. All Grade 3 AEs were determined to be not related to ACE-011 and the Grade 3 hematologic adverse events were considered either probably or definitely related to the MPT treatment.

Following preliminary analysis of the blinded central laboratory data, increases in hemoglobin values were observed within 28 days after administration of the first dose of ACE-011/placebo. Per data available, 11 out of 30 patients had $\geq$ 1 g/dL increase in hemoglobin within the first 28 days on the study, 4 patients achieved a $\geq$ 2 g/dL increase within their first 28 days on study, while 8 patients achieved a $\geq$ 2 g/dL increase in hemoglobin through Day 85.

Including all visits as of 10 March 2009 through Day 85, 9 out of 30 patients had a dose interruption due to a hemoglobin value of > 13 g/dL according to the dose modification rules defined in the protocol and 1 of the 9 patients, who had a history of hypertension, also had a dose interruption due to Grade 2 hypertension. Three of the 9 patients that required a dose interruption due to a hemoglobin value of > 13 g/dL, as per protocol, had hemoglobin levels above 13 g/dL prior to their first dose. Taken together, these data, suggest a beneficial pharmacodynamic effect of ACE-011 on erythropoiesis in a patient population with cancer chemotherapy induced anemia. No patients have had a dose reduction in ACE-011/placebo treatment or discontinued treatment.

One SAE of prolonged hospitalization due to pneumonia, that was considered not related to ACE-011, was reported in a PPD enrolled in the study. On PPD, four days after the first dose of ACE-011/placebo and the initial doses of melphalan, prednisolone and thalidomide (MPT), the patient presented with elevated temperatures up to 39º C, cough and fatigue. Chest x-ray performed on PPD revealed a pneumonic infiltration in the lower

lobe of the left lung. On PPD hospitalization of the patient was prolonged due to the event. The patient was treated with moxifloxacin, doripenem, meropenem and fluconazole.

On PPD the pneumonic infiltration resolved, as confirmed by chest x-ray. This SAE resolved upon discharge of the patient from the hospital on PPD

The investigator considered the event of pneumonia not related to ACE-011 and possibly related to MPT. The Sponsor agreed with the Investigator's assessment of causality. Pneumonia is considered a labeled event per the current package insert for thalidomide. The SAE was considered serious, related and expected with respect to thalidomide and unrelated to ACE-011.

The A011-04 study has completed enrollment, while treatment, follow-up and data monitoring are ongoing.

5.5. Potential Risks for Human Use

The 1-month safety study in rats suggested that ACE-011 may affect sperm count and motility in a dose-dependent manner, although these findings appeared to be reversible upon drug discontinuation. These reproductive effects are likely a result of the expected biological activity of activin inhibition.

The 1-month safety study in rats also suggested that ACE-011 induces adrenocortical necrosis in rats. At present, the mechanism by which ACE-011 has induced this lesion in rats and the basis for its apparent species specificity are unknown; the finding has not been observed in mice or cynomolgus monkeys at similar dose levels. Adrenal cortical function was monitored in human subjects receiving ACE-011 by the evaluation of serum electrolytes including sodium and potassium levels and by the evaluation of cortisol response to ACTH stimulation in both the Phase 1a single dose and Phase 1b multiple dose healthy volunteer clinical studies. These parameters were evaluated up to doses of 3.0 mg/kg IV and 1.0 mg/kg SC, and no clinically significant perturbations in adrenal function were observed.

Laboratory findings from the Phase 1a single dose study showed elevations in hematology results, pancreas and liver enzymes, and uric acid. The most commonly seen treatment-emergent adverse events in this study were headache, infusion site reaction, injection site hemorrhage, and toothache.

Based on data from the Phase 1b study, the most notable AEs were increases in hematologic laboratory measures in the 1.0 mg/kg dose group: hematocrit, hemoglobin, and RBC count. AEs of increased hemoglobin and/or hematocrit were reported for seven of the eight subjects in this dose group. These events were reported as mild or moderate elevations; all were considered probably related to study drug treatment. No erythroid lineage AEs were reported in the 0.1 or 0.3 mg/kg treatment groups. Hematologic parameters will be monitored carefully in this study.

Headache was reported frequently in all treatment groups with no apparent dose response trend, though the reports in the higher dose groups were more often considered possibly related to treatment. Most headaches were reported as mild (one headache was reported as moderate). Paresthesia and dizziness were reported more frequently in the ACE-011 groups, though the

events were generally not considered drug related. Other frequently reported events (e.g. fatigue, upper respiratory infection) did not appear to increase with dose and were generally mild.

In the Phase 1b multiple-dose healthy volunteer study, some subjects treated with ACE-011 showed elevations in blood pressure. One subject who had received two doses of ACE-011 at the 1.0 mg/kg dose level had a serious adverse event. The patient was hospitalized for further evaluation of increases in blood pressure and symptoms that may have been caused by the increase in blood pressure. The symptoms that the subject experienced included headache, dizziness, nausea, vomiting, and elevated hematology values (increases in the number of red blood cells). This serious adverse event was judged to be probably related to the study drug and resolved following discharge from hospital the following day. Throughout the course of the follow up period hematologic parameters were monitored frequently until the hemoglobin and hematocrit levels returned to within normal limits. Further details are outlined in the Investigator Brochure.

As with all biologics there is the potential for anti-drug antibodies that can be associated with increased drug clearance and hypersensitivity reactions. An immune response to ACE-011 has been seen in monkeys that received the drug for 1 month and longer. Some of those animals developed kidney inflammation possibly related to that immune response. Anti-drug antibody formation will be assessed in this study to examine the immunogenicity of ACE-011, and to monitor reversibility of any AEs over time.

Please refer to the Investigator Brochure for more information.

6. TRIAL OBJECTIVES AND PURPOSE

6.1. Primary objective

1. To evaluate the proportion of patients in each ACE-011 dose regimen who achieve a hematopoietic response during the treatment period including up to 2 months after the last dose of ACE-011 treatment of chemotherapy induced anemia in patients with metastatic breast cancer.

6.2. Secondary objectives

1. To evaluate the safety and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer.
2. To estimate the duration of the hematopoietic response associated with each ACE-011 dose regimen.
3. To estimate the time to the hematopoietic response associated with each ACE-011 dose regimen.
4. To estimate the proportion of patients receiving red blood cell (RBC) transfusions and/or ESAs within each cycle of ACE-011 with each dose regimen.
5. To evaluate the percentage of patients who achieve an increase in hemoglobin of ≥ 1 g/dl, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for a consecutive 28 day period during the treatment period including up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs.
6. To estimate the effect of each ACE-011 dose regimen on progression free survival (PFS) and objective response rate (ORR).

6.3. Exploratory objectives

1. To estimate the effect of each ACE-011 dose regimen on serum bone biomarkers (OC, BSAP, PINP, CTX, TRACP-5b), urine bone biomarker (uNTX), bone mineral density (DXA scans of lumbar spine and hip) and incidence of skeletal related events (SREs).
2. To evaluate the quality of life during the study in relation to the treatment of chemotherapy induced anemia in patients with metastatic breast cancer during the study.

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7. INVESTIGATIONAL PLAN

7.1. Overall Study Design and Plan

This is a Phase 2, double-blind, randomized, placebo-controlled study to evaluate the efficacy, safety, and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer. A total of 105 patients will be enrolled.

The Data Monitoring Committee (DMC) is responsible for reviewing the safety. The DMC will be comprised of a minimum of 3 members. The DMC will meet regularly during the study to review serious adverse events (SAEs), adverse events (AEs), laboratory results, and vital signs. Safety data will be reviewed throughout the study by the DMC, Medical Monitor and the Investigator. DMC responsibilities, membership, meeting frequencies and procedures will be outlined in the DMC charter.

Patients will be evaluated for study inclusion/exclusion criteria by the Investigator. Patients who meet the study entry criteria will be enrolled within 14 days of the screening visit. Central lab values will be utilized for the evaluation of patient eligibility and in the evaluation of patient safety throughout the study. Local lab values for hemoglobin and hematocrit will be collected and reported from screening through the Day 281/Termination visit. Three cohorts of 30 patients each are planned at the following doses of ACE-011: 0.1 mg/kg, 0.3 mg/kg and 0.5 mg/kg. 15 patients will be enrolled into the placebo cohort (as shown in Table 6).

Table 6: Study design

Dose	Number of Patients
0.1 mg/kg	30
0.3 mg/kg	30
0.5 mg/kg	30
Placebo	15
Total patients enrolled: 105	

Each eligible patient will be randomized to one of the four cohorts to receive a dose administered as a subcutaneous injection every 28 days for up to 4 doses. Dosing will be administered on days 1, 29, 57, and 85. Concurrent treatment with a myelosuppressive chemotherapy regimen for metastatic breast cancer will also be administered per standard of care at the site. Patients will be stratified according to administration of either single agent weekly chemotherapy or less frequent chemotherapy regimens.

Blood samples will be collected at selected study sites from approximately one-third of patients for the evaluation of serum concentrations of ACE-011. Pre-dose PK samples will be collected on Day 1. See Appendix 5 for PK sampling schedule.

Patients will be assessed for safety by monitoring adverse events, clinical laboratory tests, assessment of quality of life (FACT- Fatigue (17)), vital signs and physical examinations on an ongoing basis. ECGs will be performed during screening, prior to dosing on the first and second cycle, Day 15, and Day 113 (28 days after the last dose of ACE-011) and at Day 281/ Termination visit to monitor safety. Blood pressure and lab values will be assessed at each study visit during the treatment period; weekly for the first two cycles and approximately every two weeks for subsequent cycles.

Local laboratory hemoglobin values and blood pressure values will be evaluated prior to dosing per the dose modification criteria, as applicable.

Patients will be followed for a minimum of 7 months following their last dose of ACE-011. Study participation for each patient is approximately 10.5 months (14 day screening period, 3 month treatment period with a 2 month treatment follow up period, and a 5 month post treatment follow-up period). If a patient discontinues the study due to progression of disease (metastatic breast cancer) and/or begins another chemotherapy regimen, the patient will complete the Day 281/Termination visit procedures and be followed for survival only (a minimum 7 months from the last dose of ACE-011).

Anti-drug antibody testing will be performed at Day 281/Termination visit and if positive, the patient may be asked to return to the clinical site for additional monthly anti-drug antibody testing for up to 12 months, after their last dose.

Table 7: Schedule of Events

Schedule of Events																			
	Screen	Treatment Period													Post Treatment Follow up Period				Termination Visit[18]
ACE-011/placebo Dose period		#1				#2			#3			#4	Follow-up						
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	113 (± 7d)	141 (± 7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
Informed consent	X																		
Inclusion/exclusion criteria	X	X																	
Medical history	X																		
Physical examination	X	X				X			X			X	X	X					X
Vital signs[1]	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X					X
FACT-Fatigue	X	X[2]				X[2]			X[2]			X[2]							X
12 lead ECG	X	X[3]		X		X[3]							X						X
Serum BC, transf serum ferriti	X	X[2]				X[2]			X[2]			X[2]							X
Vitamin B12	X																		
Serum folate	X						X						X						X
Coagulation/ Serum chemistry[4]	X	X[5]	X	X	X	X[5]	X	X	X[5]	X	X	X[5]	X	X					X
Hematology[5]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Serum Erythropoietin marker		X[2]				X[2]			X[2]			X[2]		X					X
Peripheral Blood Smear (sample)		X[2]				X[2]			X[2]			X[2]							X
LH, FSH, testosterone, progesterone, estradiol	X	X[2]				X[2]			X[2]			X[2]	X	X					
Anti-drug antibody testing [6]		X				X			X			X	X	X					X
Bone biomarker[7]		X[2]				X[2]			X[2]			X[2]	X	X					X
Back up blood sample for future testing	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X	X	X	X	X
Pregnancy Test	X[8]	X[9]				X[9]			X[9]			X[9]							X
Urinalysis[10]	X	X[2]										X[2]							X
Evaluate transfusion frequency	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X	X	X	X	X

Schedule of Events cont.

	Screen	Treatment Period													Post Treatment Follow up Period				Termination Visit[18]
ACE-011/Placebo Dose period		#1				#2			#3			#4	Follow-up						
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	113 (± 7d	141 (± 7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
Documentation of concomitant medications	X[11]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Evaluation of AEs and SAEs		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Evaluation of Menstrual cycle[12]	X	X[2]				X[2]			X[2]			X[2]	X	X					
CT/MRI scan	X[13]									X[14]			X[14]						
Dual-energy X-ray absorptiometry (DXA[15])	X												X						X
Bone scans[16]	X									X			X						X
ACE-011/placebo administration		X				X			X			X							
Chem	[17]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

[1] Vital signs include weight, heart rate, seated systolic and diastolic blood pressure, and temperature will be assessed at each visit Height will be collected at screening only

[2] On dosing days (1, 29, 57 and 85), study procedure to be done prior to dosing of ACE-011/placebo

[3] 12 lead ECG is to be performed prior to dosing of ACE-011/placebo including measurement of the QT interval

[4] Coagulation (central lab only) prothrombin time (PT), partial thromboplastin time (PTT), fibrinogen Chemistry (central lab only): AST, ALT, lactate dehydrogenase (LDH), total bilirubin, indirect bilirubin, alkaline phosphatase, total protein, albumin, sodium, potassium, chloride, carbon dioxide, calcium, phosphorus, blood urea nitrogen (BUN), creatinine, uric acid, glucose, amylase, and lipase

[5] Hematology (local lab) hemoglobin and hematocrit Hematology (central lab) complete blood count (CBC) with differential and platelet, CBC includes red blood cells (RBC), white blood cells (WBC), hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), **Reticulocyte should be done on days 1, 8, 15, and days 29, 57, 85, and at the termination visit.** On dosing days (1, 29, 57 and 85), local hemoglobin and hematocrit values are to be drawn and resulted prior to dosing to apply dose modification rules for ACE-011/placebo

[6] If a patient has a positive anti-drug antibody result at the Termination visit, they may be asked to return to the clinical site for additional monthly follow-up, for up to 12 months after their last dose for a repeat anti-drug antibody test

[7] Both a serum and urine samples will be collected at each visit The urine sample should not be from the first morning urine

[8] Serum pregnancy test required of females of child bearing potential only Negative test can be within 3 days of study day 1

[9] For females of child bearing potential only urine (dipstick) pregnancy test to be collected and resulted prior to dosing of ACE-011/placebo

[10] Urinalysis to be performed at screening , prior to administration ACE-011/placebo (Days 1 and 85) and termination visit Assessments (Central lab): pH, specific gravity, protein, glucose, ketones, blood, leukocyte esterase, and nitrite Microscopy required only to follow-up abnormal findings

[11] Screening- any concomitant treatments taken 28 days prior to day of randomization

[12] Females of child bearing potential- collection of menstrual cycle information (date of last menstruation, date of menstrual period onset and duration of menstruation)

[13] A CT/MRI scan acquired within 4 weeks of initiating the current chemotherapy regimen may be used for the screening assessment Head, neck, thoracic, abdominal, and pelvic scans are to be acquired One measurable or non measurable lesion per RECIST is to be identified by the scan Brain (head) CT only to be done to rule out CNS metastases in patients with suspicious clinical symptoms or prior history of CNS metastases

[14] Follow-up CT/MRI scans may be acquired +/- 5 day window for either Day 64 or Day 113 Additional CT/MRI scans acquired during the post treatment follow up period will be performed at the discretion of the investigator

[15] Hip and lumbar spine BMD assessment

[16] Bone scans will be acquired within 4 weeks of initiating the current chemotherapy regimen for those patients who have bone metastases identified Follow up bones scans may be acquired +/- 5 day window for day 64, day 113, and Day 281

[17] Chemotherapy regimens scheduled will be administered to the patient as per standard of care Continuation of the same chemotherapy regimen may be done through the follow up period of the study at the discretion of the investigator

[18] Patients who terminate treatment early due to disease progression and/or begin a new chemotherapy regimen will complete the day 281/Terminiation visit procedures and be followed for survival only (7 months from the last dose of ACE-011/placebo) If a patient withdraws from the study (section 8 4), the Day 281/Termination visit procedures are to be completed and the patient will be followed for survival If withdrawal reason is due to ESA administration, the patient should enter the treatment follow up period

8. SELECTION AND WITHDRAWL OF PATIENTS

8.1. Number of Patients

The study will include three dose levels of ACE-011 at 0.1 mg/kg, 0.3 mg/kg and 0.5 mg/kg and a placebo group. A total of 105 will be treated with ACE-011 or placebo.

8.2. Entry Criteria

8.2.1. Patient Inclusion Criteria

1. Women ≥ 18 years of age.
2. Histologically confirmed diagnosis of breast cancer documented by cytology or biopsy.
3. Evidence of metastatic breast cancer with a minimum of one measurable lesion or non measurable lesion per RECIST criteria (Appendix 1).
4. Receiving a multicycle myelosuppressive chemotherapy regimen including one of the following: anthracycline, taxane, gemcitabine or vinorelbine.
5. Planned treatment with the same myelosuppressive chemotherapy regimen for a minimum of 9 weeks after Day 1 of ACE-011/placebo administration.
6. Hemoglobin value between ≥ 6.5 to < 11.0 g/dL (≥ 65 to < 110 g/L).
7. ≥ 30 days must have elapsed (from Day 1) since previous treatment with erythropoiesis-stimulating agent (including concurrent treatment with IV iron) for chemotherapy induced anemia.
8. ≥ 7 days have elapsed (from Day 1) since the last RBC blood transfusion and receipt of ≤ 2 units of blood in the past 30 days.
9. ECOG Performance status of 0 – 2 (Appendix 2).
10. Adequate renal (creatinine ≤ 1.5 x ULN or creatinine clearance ≥ 40 mL/min) and hepatic function (bilirubin ≤ 1.5 x ULN; AST/ALT ≤ 2.5 x ULN).
11. Females of childbearing potential participating in the study are to use adequate birth control measures (i.e. oral contraceptives or two forms of barrier method birth control) during study participation. Females of childbearing potential must have a negative serum pregnancy test within 3 days to ACE-011/placebo administration (Day 1). A female of childbearing potential is a sexually mature woman who has not undergone a hysterectomy or who has not been postmenopausal for at least 24 consecutive months (i.e. who has had menses at some time in the preceding 24 months).
12. Life expectancy of ≥ 6 months.
13. Is willing to adhere to the study visit schedule, understand and comply with all protocol requirements.

14. Understand and sign a written informed consent.

8.2.2. Patient Exclusion Criteria

1. At the time of screening patients who have any grade ≥ 3 toxicity (as per NCI CTCAE v3.0 (Appendix 3)) (except for the following disease related toxicities: hematological events- anemia, thrombocytopenia or neutropenia or non hematological events- nausea, vomiting, fatigue, or muscle or bone/joint pain).
2. Prior radiation therapy to > 20% of the whole skeleton. Use of palliative radiation if the area being treated is <15% of body surface area with no pelvic radiation or no more than 10% of the bone marrow reserve is radiated is permitted during patient participation in the study, at the discretion of the investigator.
3. Have received more than 5 prior chemotherapy treatment regimens for metastatic breast cancer.
4. Concurrent use of bevacizumab with the chemotherapy regimen during study participation.
5. Clinically significant pulmonary, endocrine, neurologic, gastrointestinal, hepatic or genitourinary disease unrelated to underlying hematologic disorder.
6. Patients with heart failure as classified by the New York Heart Association (NYHA) classification of 3 or higher (Appendix 4).
7. History of thrombosis, deep vein thrombosis (DVT), pulmonary emboli, or embolic stroke.
8. Untreated CNS metastases (exception: CNS metastases treated with whole brain radiotherapy > 6 months prior to randomization).
9. Diagnosis of a myeloid malignancy or known history of myelodysplasia.
10. History of second malignancy within 5 years (except excised and cured basal cell carcinoma, squamous cell carcinoma of the skin or cervical carcinoma in situ).
11. Patients with a recent history (within 14 days of Day 1) of administration of systemic (IV or oral) antibiotics. Patients should be free of infection at least 14 days after the last dose of systemic antibiotic treatment (from Day 1).
12. Uncontrolled hypertension. If hypertension is considered clinically stable, systolic blood pressure (BP) must be < 150 mmHg and diastolic BP must be < 100 mmHg.
13. Known history of hepatitis C antibody (HCV), hepatitis B surface antigen (HBsAg and HB core Ab), or human immunodeficiency virus (HIV) antibody.
14. Deficiency in iron (serum ferritin< 100 ng/mL (< 224.7 pmol/L)), vitamin B_{12}, or folate.
15. History of anemia as a result of inherited hemoglobinopathy such as sickle cell anemia or thalassemia.
16. History of autoimmune or hereditary hemolysis or gastrointestinal bleeding.
17. Patient received treatment with another investigational drug or device within 1 month prior to Day 1, or if the half life of the previous product is known, within 5 times the half life prior to dosing, whichever may be longer.

18. Pregnant or lactating females.
19. History of severe allergic or anaphylactic reactions or hypersensitivity to recombinant proteins or excipients in the investigational product.

8.3. Patient Withdrawal Criteria

Patients will be informed that they have the right to withdraw from the study at any time for any reason without prejudice to their medical care.

Patients must be withdrawn from the study for any of the following reasons:

- Patient request
- Patient is unwilling or unable to comply with the protocol
- ESA administration during the treatment period
- Medical reason, such as cancer or treatment related toxicity, or at the discretion of the Investigator and/or the Medical Monitor(s)

The reasons for withdrawal must be recorded in the patient's case report form (CRF). The Investigator must notify the Medical Monitor and the contract research organization (CRO) immediately when a patient has been discontinued/withdrawn due to an AE. All patients who are withdrawn from the study should complete the tests and evaluations scheduled for Day 281/Termination at the time of withdrawal. Discontinued/withdrawn patients may be followed for survival only 7 months from the last dose of ACE-011/placebo. Patients who withdraw due to ESA administration during the treatment period may enter the treatment follow-up period.

9. TREATMENT OF PATIENTS

9.1. Concomitant Medications

9.1.1. General Concomitant Medication Usage

During screening, and during the study, patients may take only stable doses of medications for chronic conditions that are not specifically excluded by the protocol (see Sections 8.2.1 Patient Inclusion Criteria and 8.2.2 Patient Exclusion Criteria). The Medical Monitor should be consulted by the Investigator if there are any concerns about what constitutes a stable dose or what is a chronic condition.

9.1.2. Concomitant treatment for anemia

Concurrent therapy with a new prescription medication related to treatment of anemia during the course of the study must first be discussed with the Investigator and Medical Monitor prior to administration, unless appropriate medical care necessitates that medication should begin before the Medical Monitor can be consulted.

9.1.2.1. Iron supplementation

Iron supplementation within 7 days of Day 1 (besides an oral general multivitamin with iron) will not be allowed. If a patient becomes iron replete during study treatment (ferritin < 100 ng/mL (< 224.7 pmol/L) or transferrin < 20%), treatment with iron supplementation is at the discretion of the investigator.

9.1.2.2. Erythropoiesis-stimulating agents (ESAs)

If concurrent treatment with erythropoiesis-stimulating agent is required in the opinion of the investigator, the erythropoiesis-stimulating agent label instructions are to be followed. The patient is to discontinue further treatment with ACE-011/placebo and enter the treatment follow up period of the study. See Table 7 for Schedule of Events.

9.1.2.3. RBC Transfusions

Concurrent treatment for chemotherapy induced anemia with blood transfusions is recommended when hemoglobin value is < 8 g/dL or at investigator discretion if the hemoglobin value is above 8 g/dL and associated with anemia symptoms such as hemodynamic or pulmonary compromise requiring treatment. If a transfusion is given to a patient during the treatment period, ACE-011/placebo should be administered no sooner than 7 days from the date of the transfusion. After the transfusion, the hemoglobin value should be assessed no later than 7 days from the date of the transfusion. On the day of ACE-011/placebo administration, the hemoglobin value and blood pressure will be assessed. See Figure 1 for ACE-011/placebo dose modification rules.

9.2. Chemotherapy

Chemotherapy treatment for metastatic breast cancer is to be given as per standard of care at the site. ACE-011/placebo administration should be given prior to chemotherapy administration on ACE-011/placebo administration days. Continuation of treatment with the same chemotherapy regimen through the treatment or post-treatment follow-up period of the study is at the discretion of the investigator.

9.3. Treatment Compliance

Each dose of ACE-011/placebo will be administered as a SC injection at the clinical site by the study staff and will be documented in the study record. Monitoring for patient compliance with the treatment regimen is therefore unnecessary.

9.4. Randomization

Patients will be randomized to limit scientific bias within the study. Randomization assignments will be generated through a computerized system, provided by an Interactive Web/Voice Response System (IWRS/IVRS). Patients will be stratified according to frequency of the planned chemotherapy regimen (weekly chemotherapy vs. less frequent chemotherapy administration).

9.4.1. Blinding

The Investigators, patients, and sponsor or representative will remain blinded to the treatment arm assignment of each patient. The unblinded statistician will remain uninvolved in the study conduct until database lock and unblinding of the data has occurred.

Safety parameters will be reviewed on an ongoing basis throughout the study to review blinded adverse events, serious adverse events, laboratory listings, and vital signs. If substantial toxicity trends are observed in one treatment group versus another, the DMC may request unblinding the treatment code assignment schema.

9.4.2. Unblinding

In the event of a medical emergency for an individual patient in which knowledge of the study medication is critical to the patient's medical management, the investigator may break the blind for that patient. However, prior to breaking the blind, every effort must be made by the investigator to first discuss the need to break the blind with the sponsor or sponsor representative. Further, it must be determined by the investigator that breaking the treatment blind is necessary information for the medical management of that patient. If the blind is broken, the patient must withdraw from the study by completing the Day 281/Termination visit procedures and be followed for survival only.

9.5. Treatments Administered

9.5.1. Selection of Doses in the Study

Single dose administration of ACE-011 up to 3.0 mg/kg was previously demonstrated to be safe and to have durable effects on markers of bone formation and resorption as well as red blood cells following a single IV administration. A dose-limiting pharmacodynamic effect of ACE-011 with increases in hemoglobin and hematocrit and RBCs was established in healthy post-menopausal women at the 1.0 mg/kg dose level following multiple SC administration. This study will confirm the safety in following repeated SC administration of ACE-011 and further evaluate the potential efficacy in patients with chemotherapy induced anemia. The safety and preliminary efficacy of ACE-011 following multiple doses up to 0.5 mg/kg will be assessed using a parallel randomization design.

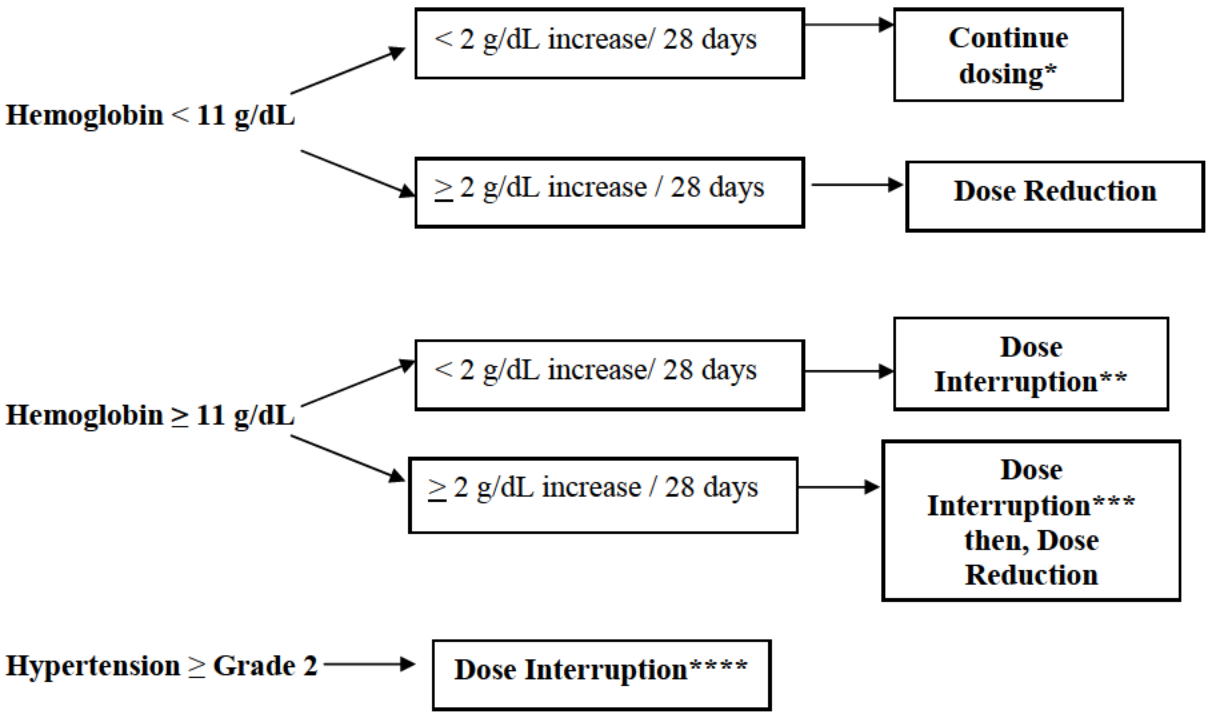
9.5.2. Selection and Timing of Dosing for Each Patient

Patients will be enrolled and receive their assigned dose of ACE-011/placebo every 28 days (i.e. on Days 1, 29, 57, and 85). Patients will be receiving chemotherapy, concurrently for metastatic breast cancer. After completion of the treatment, patients will return to the site monthly for 2 follow-up assessments (i.e., Days 113 and 141). Subsequently, the patient will return to the site for 4 post treatment follow-up assessments (i.e. Days 169, 197, 225, and 253). The patient will return to the site for a termination visit approximately 1 month after the post treatment follow-up period (Day 281). Patients will be discontinued from the ACE-011/placebo treatment for unacceptable toxicity, if the chemotherapy regimen is discontinued or for disease progression that requires the initiation of another chemotherapy treatment.

9.5.2.1. ACE-011 Dose Modification Rules

Throughout the study blood pressure and hemoglobin values will be evaluated for patient safety. The local hemoglobin values and blood pressure values will be evaluated on each ACE-011/placebo dosing day. The hemoglobin values from the previous 28 days between each ACE-011/placebo dosing day and blood pressure will be reviewed and evaluated for dose modifications. The parameters below outline rules for dosing modifications (Figure 1). Table 8 outlines the dose reduction levels of ACE-011.

Figure 1: ACE-011/placebo Dose Modification Rules (after initial dose of ACE-011/placebo)



* Patients who have received a blood transfusion in the past 28 days should continue with dosing at the same ACE-011 dosing level/placebo if the transfusion was given no sooner than 7 days from the dosing day and the hemoglobin level is < 11 g/dL and hypertension ≤ Grade 1 on the day of dosing.

** ACE-011/placebo should be held until the following scheduled treatment visit.

*** ACE-011/placebo should be held and at the following scheduled treatment visit, a dose reduction of ACE-011/placebo will be administered based on the evaluation of the hemoglobin and blood pressure at that time.

**** ACE-011/placebo should be held until resolution to ≤ Grade 1 (NCI CTCAE v3.0) and may then be resumed at the following scheduled treatment visit.

Table 8: ACE-011/Placebo Dose Reduction Levels

When required per the dose modification rules above (Figure 1), ACE-011/placebo dose(s) should be reduced as follows for dose 2 (Day 29), dose 3 (Day 57) and/or dose 4 (Day 85):

Dose	Dose reduction #1	Dose Reduction #2	Dose Reduction #3
Placebo	N/A	N/A	N/A
0.1 mg/kg	0.05 mg/kg	0.03 mg/kg	0.01 mg/kg
0.3 mg/kg	0.1 mg/kg	0.05 mg/kg	0.03 mg/kg
0.5 mg/kg	0.3 mg/kg	0.1 mg/kg	0.05 mg/kg

Blood pressure and hemoglobin values must be evaluated at each dosing day for consideration of administration of ACE-011/placebo.

The dose level for the initial dose reduction will be maintained unless subsequent dose reductions need to be administered, upon evaluation of the dose modifications listed above. A patient may have up to 3 dose reductions in the study.

Dose reduction steps and the administration of the reduced dose(s) will be conducted in a manner that will preserve the blinded status of the original treatment group of each patient. Patients in the placebo group who are designated by the investigator to undergo a dose reduction will continue to receive placebo.

10. STUDY PROCEDURES AND SCHEDULE

10.1.1. Written Informed consent

Patients will be required to sign an Institutional Review Board/Institutional Ethics Committee (IRB/IEC) approved informed consent form (ICF) prior to any study related procedures, including screening evaluations.

Screen failure information will be maintained, including but not limited to, reason for failure.

10.1.2. Screening (Within 14 days of dosing)

The following will be collected within 14 days prior to the initial dosing Day 1:

- Informed consent
- Assessment of inclusion/exclusion criteria
- Complete Medical History and Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), temperature (°C) and height.
- FACT-Fatigue survey
- 12 lead Electrocardiogram (ECG)- including measurement of the QT interval
- Serum iron, total iron binding capacity (TIBC), transferrin and serum ferritin
- Vitamin B12 and serum folate levels
- Coagulation
- Serum chemistry
- Hematology
- LH, FSH, testosterone, progesterone, and estradiol
- Back-up blood sample to be drawn for future testing
- Serum Pregnancy Test for females of childbearing potential- to be assessed within 3 days of Day 1.
- Urinalysis
- Evaluation of transfusion frequency (including history of transfusion up to 8 weeks prior to Day 1
- Documentation of concomitant medications (any treatments taken 28 days prior to Day 1
- Females of childbearing potential- collection of menstrual cycle information (date of last menstruation, date of menstrual period onset and duration of menstruation)

- CT/MRI scan: Head, neck, thoracic, abdominal, and pelvic scans are to be acquired. A CT/MRI scan acquired within 4 weeks of initiating the current chemotherapy regimen may be used for the screening assessment. One measurable or non-measurable lesion per RECIST is to be identified by the scan.
    - Brain (head) CT only to be done to rule out CNS metastases in patients with suspicious clinical symptoms or prior history of CNS metastases.
- Dual-energy X-ray absorptiometry (DXA) of lumbar spine and hip
- Bone scans (only for patients with bone metastases) acquired within 4 weeks of the initiation of the current chemotherapy regimen may be used for the screening assessment.

10.1.3. Initial Dosing: Day 1

Patients will be dosed on Day 1. Results from screening evaluations must be reviewed prior to randomization to confirm patient eligibility. A patient will be considered randomized once a multi-digit patient identification number has been assigned to the patient through the IVRS. After assignment of the patient identification number, ACE-011/placebo administration may begin.

Chemotherapy regimen will be administered per standard of care at the site. The following tests must be performed prior to ACE-011/placebo administration:

- Confirm eligibility of patient by inclusion/exclusion criteria. The local hemoglobin value should be drawn prior to ACE-011/placebo administration.
- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- FACT-Fatigue survey
- 12 lead ECG- including measurement of the QT interval
- Serum iron, total iron binding capacity, transferrin and serum ferritin
- Coagulation
- Chemistry
- Hematology
- Serum erythropoietin marker
- Peripheral blood smear
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test

- Bone biomarkers- osteocalcin (OC), procollagen type 1 N-terminal propeptide (P1NP), serum bone specific alkaline phosphatase (BSAP), C-terminal type 1 collagen telopeptide (CTX) and Serum tartrate-resistant acid phosphatase isoform-5b (TRACP-5b), and urine N-telopeptide (uNTX).
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions the collection of serum and urine for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Females of childbearing potential- Urine (dipstick) pregnancy test to be collected and resulted prior to ACE-011/placebo dosing
- Urinalysis
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events (after study drug administration)
- Females of childbearing potential- collection of menstrual cycle information (date of last menstruation from day 1, date of menstrual period onset and duration of menstruation)

Patients will be administered ACE-011/placebo before the chemotherapy regimen begins during the study visits.

After ACE-011/placebo and chemotherapy are administered, the patient may leave the clinic, based on the clinical judgment of the staff.

10.1.4. ACE-011/placebo administration : Days 29, 57, and 85 (± 3 days)

Chemotherapy regimen will be administered per standard of care at the site. ACE-011/placebo will be administered approximately every 28 days. The following procedures must be performed prior to ACE-011/placebo administration:

- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- FACT-Fatigue survey
- 12 lead ECG -including measurement of the QT interval (Day 29 only)
- Serum iron, total iron binding capacity, transferrin and serum ferritin
- Coagulation
- Serum chemistry

- Hematology
- Serum erythropoietin marker
- Peripheral blood sample
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test
- Bone biomarkers- OC, BSAP, PINP, CTX, TRACP-5b, and uNTX
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions the collection of serum and urine for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Females of childbearing potential -Urine (dipstick) pregnancy test
- Urinalysis (Day 85 only)
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events
- Females of childbearing potential- collection of menstrual cycle information (date of last menstruation, date of menstrual period onset and duration of menstruation)

Hemoglobin (HgB) and Hematocrit values (Hct) and blood pressure (BP) measurement must be assessed prior to ACE-011/placebo dose administration by review of local laboratory values.

10.1.5. Additional visits during Treatment Period: Days 8, 15, 22, 36 (± 1 day), 43, 64 and 71 (± 2 days)

Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- 12 lead ECG -including measurement of the QT interval (Day 15 only)
- Serum folate (Day 36 only)
- Coagulation
- Serum chemistry
- Hematology
- Back-up blood sample to be drawn for future testing

- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events
- CT/MRI scan (Day 64 only, +/- 5 day window). Same modality as acquired for baseline should be used for follow up scans. Tumor response assessments to be evaluated by the investigator.
- Bone scan (Day 64 only, +/- 5 day window). Follow-up scan to be acquired if performed at baseline.

If patient terminates the treatment early, prior to receiving a fourth dose of ACE-011/placebo, the patient will enter the treatment period follow up. However, if patient terminates ACE-011/placebo early and begins a new chemotherapy regimen, the patient should complete the Day 281/Termination visit procedures and enter the post treatment follow up period.

10.1.6. Treatment Period Follow-up visits : Days 113 and 141 (± 7 days)

Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- 12 lead ECG-including measurement of the QT interval (Day 113 only)
- Serum folate (Day 113 only)
- Coagulation
- Serum chemistry
- Hematology
- Serum erythropoietin marker (Day 141 only)
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test
- Bone biomarkers- OC, BSAP, PINP, CTX, TRACP-5b, and uNTX
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions the collection of serum and urine for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Evaluation of transfusion frequency

- Documentation of concomitant medications
- Evaluation of adverse events
- Females of childbearing potential- collection of menstrual cycle information (date of last menstruation, date of menstrual period onset and duration of menstruation)
- CT/MRI scan (Day 113 only, +/- 5 day window). Same modality as acquired for baseline should be used for follow up scans. Tumor response assessments to be evaluated by the investigator.
- DXA of lumbar spine and hip (Day 113 only)
- Bone scan (Day 113 only, +/- 5 day window). Follow-up scan to be acquired if performed at baseline.

10.1.7. Post Treatment Period Follow-up visits : Days 169, 197, 225, and 253 (± 7 days)

If a CT/MRI and bone scan (if applicable) is performed as per standard of care during the post treatment period follow up, results may be collected within the eCRF to capture the status of the disease. Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Hematology
- Back-up blood sample to be drawn for future testing
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events

10.1.8. Termination Visit: Day 281 (± 7 days)

Day 281 is the final visit. If a patient terminates the study early, the Day 281 procedures should be followed. Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- FACT-Fatigue survey
- 12 lead ECG -including measurement of the QT interval
- Serum iron, total iron binding capacity, transferrin and serum ferritin
- Serum folate
- Coagulation

- Serum chemistry
- Hematology
- Serum erythropoietin marker
- Peripheral blood smear
- Anti-drug antibody test
- Bone biomarkers- OC, BSAP, PINP, CTX, TRACP-5b, and uNTX
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions the collection of serum and urine for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Females of childbearing potential -Urine (dipstick) pregnancy test
- Urinalysis
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events
- DXA of lumbar spine and hip
- Bone scan (Day 281, +/- 5 day window). Follow-up scan to be acquired if performed at baseline.

If a patient has a positive anti-drug antibody result at Day 281/Termination visit, the patient may be asked to return to the clinical site for additional monthly anti-drug antibody testing, 12 months after their last dose.

10.2. Discontinuation of study

The Sponsor may terminate this study, after consultation with the Investigator, at any time for safety or administrative reasons. The Sponsor may terminate the study if the occurrence of serious adverse events (SAEs) or other findings suggest unacceptable risk to the health of the patients.

11. INVESTIGATIONAL PRODUCT MATERIALS AND MANAGEMENT

11.1. Reference Product

The placebo (control agent) to be used in this study will be sterile normal saline (0.9% Sodium Chloride for Injection) administered as a SC injection. Sterile, normal saline will be supplied to the investigational site's pharmacist. The manufacturer's directions for placebo storage and handling are to be followed, as are standard clinical practices for ensuring sterility of the placebo.

11.2. Investigational Product Packaging and Labeling

ACE-011 will be supplied in 2 mL clear glass vials with gray stoppers and red flip-top seals that contain 1 mL of ACE-011. The drug product consists of ACE-011 in PBS at a nominal concentration of approximately 50 mg/mL.

11.3. Investigational Product Storage

ACE-011 is recommended to be stored at ≤ -65℃.

11.4. Investigational Product Preparation

See Investigational Product Preparation Manual.

11.5. Administration

ACE-011/placebo will be administered by subcutaneous injection (SC). Subcutaneous injections will be given in the upper arm and/or thigh. Please refer to Investigational Product Handling and Administration document for further information.

Dose modifications will be determined by the investigator after reference to the dose modification rules listed in Figure 1. In the case of a dose interruption or dose reduction of ACE-011, the investigator or designee will notify the pharmacy staff, who is unblinded, of the treatment decision for appropriate preparation of the study drug to maintain the blind of the study.

11.6. Drug Accountability

Accountability for ACE-011/placebo is the responsibility of the Investigator. Investigational clinical supplies must be received by a designated person at the clinical site and kept in a secured location. The investigational site must maintain accurate records demonstrating dates and amounts of ACE-011/placebo received, to whom it was dispensed (patient-by-patient accounting), and accounts of any ACE-011/placebo accidentally or deliberately destroyed or returned. Unless otherwise notified, all vials of ACE-011/placebo, both used and unused, must be saved for drug accountability. The used vials may be discarded, per the institution's standard

practice, after drug accountability has been completed by the monitor. The Investigator must return all unused vials of ACE-011 to the Sponsor at the end of the study, or the study drug may be destroyed at the clinical site with the permission of the Sponsor. For either scenario, the outcome must be documented on the drug accountability log. The Sponsor will provide direction for the outcome of all unused vials.

11.7. Investigational Product Handling and Disposal

Please refer to the Study Reference Guide, provided under separate cover, for detailed drug handling, administration, and storage instructions.

12. ASSESSMENT OF EFFICACY

Efficacy measurements will include assessments for hematopoietic response which is defined as an increase in hemoglobin of ≥ 0.5 g/dL, ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs, at each dose level. The hematopoietic endpoints will be determined by the monitoring of hematologic laboratory values and RBC transfusion and/or ESA administration. Secondary endpoints associated with assessment of disease progression will be evaluated by CT/MRI scan and bone mineral density by DXA scans, respectively. Bone biomarkers and incidence of skeletal related events (SREs) will also be evaluated for all patients. SREs are defined as pathologic fracture, radiation therapy to bone, surgery to bone, or spinal cord compression. Quality of life assessment (FACT-Fatigue) will be evaluated for each patient.

13. ASSESSMENT OF SAFETY

13.1. Safety Parameters

13.1.1. Adverse Events

All patients will be assessed for safety by monitoring adverse events, clinical laboratory tests, vital signs and physical examinations. AE information will be collected throughout the study. See Section 14 Adverse Events for a full description of AE definitions, AE and SAE reporting procedures, and emergency procedures.

13.1.2. Clinical Laboratory Tests

The following laboratory safety assessments will be drawn at the investigational site's local laboratory according to the laboratory collection recommendations. The samples will then be sent to the central lab for value reporting. Please refer to Lab Reference Manual for further information. Hemoglobin and hematocrit lab tests will be drawn and evaluated by the local laboratory at each visit day throughout the study. The local hemoglobin laboratory value will be used in the evaluation of ACE-011/placebo dose modifications. The sponsor may request additional safety tests be performed based on ongoing data review during the study.

- Iron studies: Serum iron, total iron binding capacity (TIBC), transferrin and serum ferritin on specified days.
- Nutritional tests: Serum folate and vitamin B12
- Coagulation (central lab only): prothrombin time (PT), partial thromboplastin time (PTT), and fibrinogen
- Serum chemistry (central lab only): AST, ALT, lactate dehydrogenase (LDH), total bilirubin, indirect bilirubin, alkaline phosphatase, total protein, albumin, sodium, potassium, chloride, carbon dioxide, calcium, phosphorus, blood urea nitrogen (BUN), creatinine, uric acid, glucose, amylase, and lipase.
- Hematology: complete blood count (CBC) with differential and platelet. CBC includes red blood cells (RBC), white blood cells (WBC), hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), and erythropoietin levels. Reticulocyte percent and peripheral blood smear will be collected on specified days.
- Bone biomarkers: (serum) osteocalcin (OC), procollagen type 1 N-terminal propeptide (P1NP), serum bone specific alkaline phosphatase (BSAP), C-terminal type 1 collagen telopeptide (CTX), tartrate-resistant acid phosphatase isoform-5b (TRACP-5b), and urine N-telopeptide (uNTX).
- Pregnancy test for females of child bearing potential
- Urinalysis: including determination of pH, specific gravity, protein, glucose, ketones, blood, leukocyte esterase and nitrite. Microscopy required only to follow-up abnormal findings.

13.1.3. Other Safety Assessments

- Physical examinations
- Vital signs: weight (kg), heart rate (beats/min), seated blood pressure (mmHg) and oral temperature (°C). Height will be collected at screening only.
- FACT-Fatigue survey
- 12-lead ECG -including measurement of the QT interval
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test
- CT/MRI scan
- DXA
- Bone scans (for those patients with baseline bone metastases)

13.2. Pharmacokinetics

See Appendix 5 for further details regarding PK sampling.

14. ADVERSE EVENT

Adverse Event

An adverse event (AE) is any untoward medical occurrence in a patient administered a pharmaceutical product, which does not necessarily have a causal relationship with the treatment. An AE can be any unfavorable and unintended sign (e.g., including an abnormal laboratory finding), symptom, or disease temporally associated with the use of the study drug, whether or not it is considered to be study drug related. This includes any newly occurring event or previous condition that has increased in severity or frequency since the administration of study drug (treatment-emergent).

Serious Adverse Event

A serious adverse event (SAE) is any AE, occurring at any dose level and regardless of causality that:

- Results in death.
- Is life-threatening: Life-threatening means that the patient was at immediate risk of death from the reaction as it occurred, (i.e., it does not include a reaction which hypothetically might have caused death had it occurred in a more severe form).
- Requires inpatient hospitalization or prolongation of existing hospitalization.
- Results in persistent or significant disability/incapacity.
- Is a congenital anomaly/birth defect.
- Is an important medical event: an important medical event is an event that may not result in death, be life-threatening, or require hospitalization but may be considered an SAE when, based upon appropriate medical judgment, it may jeopardize the patient and may require medical or surgical intervention to prevent one of the outcomes listed in the definitions for SAEs. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.
- A hospitalization for an elective procedure will not be considered a SAE.

Unexpected Adverse Event

An unexpected AE is an AE that is not reported in the Investigator Brochure or in the case of an AE already described, the severity of which is not described in the Investigator Brochure.

14.1. Adverse Event Classification

The following classifications should be used when evaluating the relationship of AEs and SAEs to the investigational drug.

None:	No relationship between the experience and the administration of study drug; related to other etiologies such as concomitant medications or patient's clinical state.
Unlikely:	The current state of knowledge indicates that a relationship is unlikely.
Possibly:	A reaction that follows a plausible temporal sequence from administration of the study drug and follows a known response pattern to the suspected study drug. The reaction might have been produced by the patient's clinical state or other modes of therapy administered to the patient.
Probably:	A reaction that follows a plausible temporal sequence from administration of the study drug and follows a known response pattern to the suspected study drug. The reaction cannot be reasonably explained by the known characteristics of the patient's clinical state or other modes of therapy administered to the patient.
Definitely:	A reaction that follows a plausible temporal sequence from administration of the study drug and follows a known response pattern to the suspected study drug and can be confirmed with a positive re-challenge test or supporting laboratory data.

Severity of AEs will be graded by the investigator using the National Cancer Institute (NCI; US) Common Terminology Criteria for Adverse Events (CTCAE) version 3.0 guidelines (Appendix 3).

14.2. Recording Adverse Events

Patients will be evaluated and questioned generally for AEs during the course of the study, starting from baseline (Day 1). The Medical History CRF should be comprehensive of patient's medical history up to Day 1. All AEs occurring after ACE-011/placebo administration until the Day 281/Termination visit are to be documented on AE CRF. The specific disease or syndrome rather than individual associated signs and symptoms should be identified by the investigator and recorded on the CRF.

Serious Adverse Events (SAEs)

For SAEs, a SAE form must be completed with as much information as possible and submitted in the time frame described in Section 14.3.

When new significant information is obtained as well as when the outcome of an event is known, the investigator should record the information on a new SAE form. If the patient was hospitalized, a copy of the discharge summary must be included as part of the patient medical file. In all instances, the investigator should follow up with patients until the outcome of the SAE is known.

14.3. Reporting Adverse Events

All AEs spontaneously reported by the patient and/or in response to an open question from study personnel or revealed by observation, physical examination or other diagnostic procedures will be recorded on the AE CRF. Any clinically relevant change in laboratory assessments or other clinical findings is considered an AE and must be recorded on the AE CRF. Clinically significant laboratory abnormalities are those that are identified as such by the investigator and/or those that require intervention. All AEs are to be followed until the event resolves or the clinical course is stabilized.

Serious Adverse Events

For SAEs, a SAE form must be completed with as much information as possible and submitted in the time frame described below.

All SAEs that occur during the course of the study (from the signing of the ICF until the last study visit) must be reported by the Investigator to PPD and Sponsor by faxing the SAE form within 24 hours from the point in time when the Investigator becomes aware of the SAE. All SAEs and deaths must be reported whether or not considered causally related to the study drug.

SAE Reporting:

PPD Medical Monitor
PPD Inc.

If there are serious, unexpected AEs associated with the use of the study drug, the Sponsor will notify the appropriate regulatory agency(ies) and all appropriate parties on an expedited basis. It is the responsibility of the Investigator to promptly notify the Institutional Review Board (IRB) of all unexpected SAEs involving risk to human patients.

14.4. Emergency Sponsor Contact

In the event of a medical emergency (i.e., an event that requires immediate attention regarding the treatment of a patient, operation of the clinical study, and/or the use of investigational drug), investigational site personnel must immediately contact the Medical Monitor.

15. STATISTICS

Statistics Analysis Overview

The study will be considered complete with regard to the primary endpoint once all eligible subjects have completed up to 2 months after the last dose of ACE-011. Patients will be randomized to one of three ACE-011 treatment groups (0.1, 0.3 and 0.5 mg/kg) or placebo with a ratio of 2:2:2:1. Patients will be stratified according to administration of either single agent weekly chemotherapy or less frequent chemotherapy regimens.

15.1. Determination of Sample Size

With an anticipated dropout rate of 20%, a sample size of 30 patients per dose level is estimated to achieve at least 80% power with 2 sided significance level of 0.05 to differentiate targeted response rate of p1=0.45 from null hypothesis p0=0.20. An additional 15 patients will be enrolled to the placebo group for safety comparison. A total of 105 patients will be randomized to three ACE-011 dose groups (0.1, 0.3, and 0.5 mg/kg) and placebo with a ratio of 2:2:2:1.

15.2. Analysis Populations

For this study, the following populations will be defined and used in the analysis and presentation of the data.

Modified Intent-to-Treat (MITT) population: The MITT population is defined as all patients randomized who received at least one dose of study drug.

Per-Protocol (PP) population: The PP population is defined as all patients randomized who have confirmed chemotherapy induced anemia, received at least one dose of ACE-011 and on study (treatment) for at least 2 months (57 days).

Safety population: The safety population is defined as all patients who received at least one dose of study drug ACE-011 or placebo.

15.3. Background and Demographic Characteristics

Summary statistics (mean, standard deviation, median, minimum and maximum) will be provided for those variables measured on a continuous scale. The frequency distribution (number and percentage) will be provided for those variables measured on a nominal scale.

15.3.1. Patient Disposition

Patient disposition (analysis population allocation, entered, discontinued, along with primary reason for discontinuation) will be summarized using frequency and percent for each dose group. A summary of patients enrolled by site will be provided.

15.3.2. ACE-011

ACE-011 treatment exposure (total dosage received) will be summarized for each dose group as well as for each cycle for each dose level. Listings for dose adjustment will be provided.

15.3.3. Concomitant Therapy

By-patient listings will be provided for all concomitant medications and therapies taken during the study. All concomitant medications will be coded and categorized by the WHO drug coding system. Usage frequency of each coded concomitant medications will be summarized by each dose group.

15.4. Efficacy Evaluation

15.4.1. Primary Efficacy Analysis

The primary efficacy endpoint is the hematopoietic response rate at each dose level. It is defined as the proportion of patients who have a hematopoietic response of an increase in hemoglobin of ≥ 0.5 g/dL for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs. The ACE-011 treatment period is defined as from the time of first ACE-011 dose to up to 2 months after the last dose of ACE-011 treatment. To be considered as a responder, all hemoglobin measurements during a 28 consecutive day period or longer must have an increase of ≥ 0.5 g/dL from baseline. If RBC transfusions and/or ESAs are required, no hemoglobin measurements within 28 days will be used to determine the hemoglobin response. Primary efficacy analysis will be based on PP population. Primary efficacy analysis will also be conducted on MITT population as supportive to the PP based analysis. Patients who do not receive ACE-011 will be excluded from primary efficacy analysis. Patients who discontinue the ACE-011 treatment prematurely will be counted as non-responder in the calculation of response rate.

A 95% confidence interval (CI) for the response rate for each dose level will be provided to assess whether 95% CI of response rate contains null hypothesis response rate, p0=0.20. The response rate and 95% CI will also be provided for patients with chemotherapy on weekly basis versus those with chemotherapy on a less frequent basis for each dose level as well as overall (all three doses levels analyzed together).

An exploratory analysis with the Kaplan Meier approach will be considered to account for those patients who discontinue the ACE-011 treatment prematurely if the drop out rate is greater than 20%. Exploratory analysis of comparing hematopoietic response rates between active and placebo will be performed using Fisher's exact test. Due to the nature of the study (phase 2 dose determination study), no multiplicity adjustment will made for the multiple comparisons.

15.4.2. Secondary Efficacy Analyses

The secondary efficacy endpoints include:

- Proportion of patients achieving an increase in hemoglobin of ≥ 1 g/dl, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for 28 consecutive days during the treatment period and up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs.
- Duration of hematopoietic response (days) defined as the first time hemoglobin increases at least 0.5 g/dL, ≥ 1 g/dl, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL from baseline to the last time there is hemoglobin ≥ 0.5 g/dL, ≥ 1 g/dl, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL increase from baseline. Duration of response is only calculated for a responder and will be at least 28 days. The duration of response will be at least 28 days is only calculated for a patient who meets the primary efficacy endpoint.
- Time to achieve hematopoietic response (days) defined as time from first dose of ACE-011 to the first time a hemoglobin result at least 0.5 g/dL, ≥ 1 g/dl, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL increase from baseline and maintained for a period of at least 28 consecutive days.
- Proportion of patients receiving red blood cell (RBC) transfusions and/or ESAs in each dose group as well as within each cycle of each ACE-011 dose level.
- Objective tumor response rate for each dose level using RECIST criteria.
- Progression-Free Survival is defined as the time from randomization into the study to the first observation of documented disease progression or death due to any cause. If the subject has not progressed or died at the time of final analysis, PFS will be censored at the time of last tumor assessment.

The statistical analysis for binary endpoints will be similar to the primary efficacy analysis. For time to event endpoints, the Kaplan-Meier method will be used to estimate the survival curve and median time to event. These analyses will be performed for each dose group and all ACE-011 groups combined.

15.5. Safety Evaluation

Safety variables will be tabulated and presented for all patients who receive ACE-011. Adverse events will be coded using the Medical Dictionary for Regulatory Activities (MedDRA). Incidence of treatment-emergent adverse events will be presented by system organ class (SOC) and preferred term. Adverse events will also be presented by severity, and relationship to study drug. Change from baseline in clinical laboratory parameters and vital signs will be summarized across time. Shift tables will be presented for selected laboratory parameters. Physical examination results will be presented in listings.

Data from all patients who receive one or more doses of study drug will be included in the safety analyses. To assess clinical safety, adverse events, vital sign measurements and clinical

laboratory information will be summarized by visit. Adverse events and laboratory results will also be summarized by severity grade (NCI CTCAE v3.0). Descriptive statistics will be generated and shift tables provided as appropriate.

15.6. Pharmacokinetic Evaluation

Listing of individual patient serum ACE-011 concentrations, actual blood sampling times and graphs of concentration vs. time will be prepared for each dose group. Summaries of PK parameters will be summarized by dose group. The trough concentrations will be summarized for all patients who provide pharmacokinetic samples by dose group. Exploratory analyses will be performed to correlate trough concentrations with efficacy, bone biomarker data, and safety data, however these analyses will be data-driven and only conducted if warranted by the data.

15.6.1. Anti-drug Antibody Data

The results of anti-drug and neutralizing antibodies will be presented over time. Exploratory analysis will be performed on the potential effect of anti-drug antibody on ACE-011 PK and drug exposure if anti-drug antibody tests are deemed positive.

15.7. Interim Analysis

There is no planned interim analysis for this study.

15.8. Deviation from Original Analysis Plan

Deviations from the statistical analyses outlined in this protocol will be indicated in the statistical analysis plan; any further modifications will be noted in the final clinical study report.

16. DIRECT ACCESS TO SOURCE DATA/DOCUMENTS

16.1. Study Monitoring

The Clinical Monitor will arrange to visit the Investigator sites at regular intervals during the study. The monitoring visits must be conducted according to the applicable ICH and GCP guidelines to ensure protocol adherence, quality of data, drug accountability, compliance with regulatory requirements, and continued adequacy of the investigational site and its facilities. During these visits, CRFs and other data related to the study will be reviewed and any discrepancies or omissions will be resolved. The Clinical Monitor will be given access to study relevant source documents (including medical records) for purposes of source data verification.

During and/or after completion of the study, quality assurance officers named by the Sponsor or the regulatory authorities may wish to perform on-site audits. The Investigator is expected to cooperate with any audit and provide assistance and documentation (including source data) as requested.

16.2. Audits and Inspections

The Investigators and clinical sites will permit trial related monitoring, audits, IRB review, and regulatory inspections as requested by FDA, the Sponsor or designee, including direct access to source data/documents (i.e., original medical records, laboratory reports, hospital documents, progress reports, signed ICFs, etc.) in addition to CRFs.

17. QUALITY CONTROL AND QUALITY ASSURANCE

17.1. Data Quality Control and Quality Assurance

17.1.1. Investigator responsibility

The investigator is responsible for ensuring the study is conducted according to the protocol, Code of Federal Regulations (CFR), GCP, and any other applicable regulatory requirements. These responsibilities outlined in these documents along with the documentation that a signed informed consent must be obtained prior to a patient participation in the study.

17.1.2. Protocol modifications

The investigator may not modify the protocol without agreement from the Sponsor and prior review or approval by the IRB. Any deviations from the protocol should be documented by the investigator or designee.

17.2. Financing and Insurance

Financing and insurance are addressed in a separate document.

17.3. Confidentiality

To maintain patient privacy, all eCRFs, study drug accountability records, study reports and communications will identify the patient by the assigned patient identification number. The Investigator will grant monitor(s) and auditor(s) from the Sponsor or designee and regulatory authority(ies) access to the patient's original medical records for verification of data gathered on the CRFs and to audit the data collection process. The patient's confidentiality will be maintained and will not be made publicly available to the extent permitted by the applicable laws and regulations.

All information regarding the investigational product supplied by the Sponsor to the Investigator is privileged and confidential information. The Investigator agrees to use this information to accomplish the study and will not use it for other purposes without consent from the Sponsor. It is understood that there is an obligation to provide the Sponsor with complete data obtained during the study. The information obtained from the clinical study will be used towards the development of the investigational product and may be disclosed to regulatory authority(ies), other Investigators, corporate partners, or consultants as required.

17.4. Publication Policy

All information concerning ACE-011 is considered confidential and shall remain the sole property of the Sponsor. The Investigator agrees to use this information only in conducting the study and shall not use it for any other purposes without the Sponsor's written approval. The Investigator agrees not to disclose the Sponsor's confidential information to anyone except to

people involved in the study that need such information to assist in conducting the study, and then only on like terms of confidentiality and nonuse.

It is understood by the Investigator that the information developed from this clinical study will be used by the Sponsor in connection with the development of ACE-011, and therefore may be disclosed as required to regulatory agencies. To allow for the use of the information derived from clinical studies, it is understood that there is an obligation to provide the Sponsor with complete test results and all data developed in the study.

No publication or disclosure of study results will be permitted except as specified in a separate, written, agreement between the Sponsor and the Investigator.

17.5. Protocol Amendments

Protocol amendments that impact patient safety change the scope of the investigation, or affect the scientific quality of the study must be approved by the IRB and submitted to the appropriate regulatory authorities before implementation of such modifications to the study.

In the event that the protocol needs to be modified immediately to eliminate an apparent hazard to a patient, the Sponsor will amend and implement the protocol change and subsequently notify the regulatory authorities and/or the IRB, as appropriate.

18. ETHICS

18.1. Institutional Review Board or Independent Ethics Committee

The Investigator must obtain written IRB approval of the protocol, approval for relevant supporting information and all types of patient recruitment and advertisement and ICF prior to starting the study. The IRB will meet all Food and Drug Administration (FDA) requirements governing IRBs (Code of Federal Regulations, Title 21, Part 56).

The Sponsor or designee must approve the ICF submitted to the investigational site's IRB. All patient recruitment and advertisements must be submitted to the Sponsor or designee prior to submission to the IRB, for review.

18.2. Ethical Conduct of the Study

The Sponsor and the Investigator must comply with all instructions, regulations, and agreements in this protocol and in the applicable International Conference on Harmonisation (ICH) and GCP guidelines, and must also conduct the study in accordance with local regulations.

18.3. Written Informed Consent

18.3.1. Informed Consent

Informed written consent is required from each patient prior to any testing under this protocol, including screening tests and evaluations. The consent form, as specified by the clinical site's IRB, must follow the Protection of Human Patients regulations listed in the Code of Federal Regulations, Title 21, Part 50.

The background of the proposed study and the benefits and risks of the procedures and study must be explained to the patients. It is the responsibility of the Investigator to obtain consent and to provide the patient with a copy of the signed and dated ICF. Confirmation of a patient's informed consent must also be documented in the patient's medical record prior to any testing under this protocol, including screening tests and evaluations.

All ICFs used in this study must be approved by the appropriate IRB and by the Sponsor or designee. The ICF must not be altered without the prior agreement of the relevant IRB and the Sponsor.

18.3.2. Patient Data Protection

Prior to any testing under this protocol, including screening tests and evaluations, patients must authorize the release and use of protected health information (PHI), as required by local law.

The patient will not be identified by name in the CRF or in any study reports. These reports will be used for research purposes only. The Sponsor, its designee, and various government health agencies may inspect the records of this study. Every effort will be made to keep the patient's personal medical data confidential.

19. DATA HANDLING AND RECORDKEEPING

19.1. Case Report Form Completion

CRFs will be completed for each randomized patient. It is the Investigator's responsibility to ensure the accuracy, completeness, and timeliness of the data reported in the patient's CRF. Source documentation supporting the CRF data should indicate the patient's participation in the study and should document the dates and details of study procedures, AEs, and patient status.

Investigators will maintain copies of the CRFs at the clinical site. For patients who discontinue or terminate from the study, the CRFs will be completed as much as possible, and the reason for the discontinuation or termination clearly and concisely specified on the appropriate CRF.

19.2. Retention of Records

The Investigator will maintain all study records according to ICH-GCP and applicable regulatory requirements. Records will be retained for at least 2 years after the last marketing application approval or 2 years after formal discontinuation of the clinical development of the investigational product or according to applicable regulatory requirements. If the Investigator withdraws from the responsibility of keeping the study records, custody must be transferred to a person willing to accept the responsibility. The Sponsor must be notified in writing if a custodial change occurs.

20. REFERENCES

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21. APPENDICES

Appendix 1: Response Evaluation Criteria in Solid Tumors

Response Evaluation Criteria in Solid Tumors (RECIST) v1.1	
Terms	Definition
Measurable disease	The presence of at least one measurable lesion by CT or MRI. If the measurable disease is restricted to a solitary lesion, its neoplastic nature should be confirmed by cytology/histology. All measurements should be taken and recorded in metric notation, using a ruler or calipers. All baseline evaluations should be performed as closely as possible to the beginning of treatment and never more than 4 weeks before the beginning of the treatment.
Measurable lesions	Lesions that can be accurately measured in least one dimension by CT scan with longest diameter $\geq$ 10 mm (CT scan thickness no greater than 5 mm). The same method of assessment and the same technique should be used to characterize each identified and reported lesion at baseline and during follow-up.
Non-measurable lesion	All other lesions, including small lesions (longest diameter <10mm or pathological lymph nodes with P10 to <15mm short axis) as well as truly non-measurable lesions. Lesions considered truly non-measurable include: leptomeningeal disease, ascites, pleural or pericardial effusion, inflammatory breast disease, lymphangitic involvement of skin or lung, abdominal masses/abdominal organomegaly identified by physical exam that is not measurable by reproducible imaging techniques.

RECIST Criteria-Response Evaluation

Evaluation of target lesions	
Complete Response (CR)	Disappearance of all target lesions. Any pathological lymph nodes (whether target or non-target) must have reduction in short axis to <10 mm.
Partial Response (PR):	At least a 30% decrease in the sum of diameters of target lesions, taking as reference the baseline sum diameters.
Stable Disease (SD):	Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum diameters while on study.
Progressive Disease (PD):	At least a 20% increase in the sum of diameters of target lesions, taking as reference the smallest sum on study (this includes the baseline sum if that is the smallest on study). In addition to the relative increase of 20%, the sum must also demonstrate an absolute increase of at least 5 mm. (Note: the appearance of one or more new lesions is also considered progression).

Evaluation of non-target lesions	
Complete Response (CR):	Disappearance of all non-target lesions and normalisation of tumour marker level. All lymph nodes must be non-pathological in size (<10mm short axis).
Non-CR/Non-PD	Persistence of one or more non-target lesion(s) and/or maintenance of tumour marker level above the normal limits.
Progressive Disease (PD):	Unequivocal progression (see comments below) of existing non-target lesions. (Note: the appearance of one or more new lesions is also considered progression).

RECIST Criteria-Response Evaluation cont-

Evaluation of best overall response	The best overall response is the best response recorded from the start of the treatment until disease progression/recurrence (taking as reference for PD the smallest measurements recorded since the treatment started). In general, the patient's best response assignment will depend on the achievement of both measurement and confirmation criteria. Patients with a global deterioration of health status requiring discontinuation of treatment without objective evidence of disease progression at that time should be classified as having "symptomatic deterioration". Every effort should be made to document the objective progression even after discontinuation of treatment. In some circumstances it may be difficult to distinguish residual disease from normal tissue. When the evaluation of complete response depends on this determination, it is recommended that the residual lesion be investigated (fine needle aspirate/biopsy) to confirm the complete response status.

Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, Dancey J, Arbuck S, Gwyther S, Mooney M, Rubinstein L, Shankar L, Dodd L, Kaplan R, Lacombe D, Verweij J.New response evaluation criteria in solid tumours: Revised RECIST guideline (version 1.1) (2009). European Journal of Cancer 45, 228-247.

Appendix 2: ECOG Performance Status

The ECOG scale is used to assess a patient's quality of life in an evaluation by a health professional of the daily activities and how the activities are affected by the disease of the patient.

Grade	ECOG
0	Fully active, able to carry on all pre-disease performance without restriction
1	Restricted in physically strenuous activity but ambulatory and able to carry out work of a light or sedentary nature, e.g., light house work, office work
2	Ambulatory and capable of all selfcare but unable to carry out any work activities. Up and about more than 50% of waking hours
3	Capable of only limited selfcare, confined to bed or chair more than 50% of waking hours
4	Completely disabled. Cannot carry on any selfcare. Totally confined to bed or chair
5	Dead

Oken, M.M., Creech, R.H., Tormey, D.C., Horton, J., Davis, T.E., McFadden, E.T., Carbone, P.P. (1982) Toxicity And Response Criteria Of The Eastern Cooperative Oncology Group. Am J Clin Onco 5:649-655.

Appendix 3: National Cancer Institute (NCI) Common Terminology Criteria for Adverse Events (CTCAE) Version 3.0

See http://ctep.cancer.gov/reporting/ctc.html

Appendix 4: New York Heart Association- Classification of heart failure

Class 1 - Class 1 heart failure - no limitation of activities. No symptoms from ordinary activities.
Class 2 - Class 2 heart failure- mild limitation of activity. Comfortable with rest or mild exertion.
Class 3 - Class 3 heart failure- marked limitation of activity and be comfortable only at rest.
Class 4 –Class 4 heart failure- complete rest, confined to bed or chair; any physical activity brings on discomfort and symptoms occur at rest.

National Heart, Lung, and Blood Institute, National Institutes of Health. New York Heart Association Classification. 2008.

Appendix 5- Pharmacokinetic Sampling

Pharmacokinetics

Blood samples will be collected from approximately one-third of patients at selected study sites for the evaluation of serum concentrations of ACE-011. Pre-dose PK samples will be collected on Day 1.

Blood samples for PK should be collected in a fasted state (defined by no food or drink except water for at least 4 hours prior to the study procedure). The sample should be collected prior to ACE-011/placebo dosing. Chemotherapy regimens scheduled will be administered to the patient as per standard of care. Collection, handling, and shipping procedures for blood samples are provided in the Study Reference Guide.

Schedule of Events

	Screen	Treatment period													Post Treatment Follow up Period				Termination Visit
ACE-011/Placebo Dose period		#1				#2			#3			#4	Follow up						
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	113 (± 7d	141 (± 7d	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
PK Sampling		X				X	X			X			X						
ACE-011/placebo administration		X				X			X			X							
Chemo regimen		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

Protocol Amendment – Summary of Changes

CLINICAL STUDY PROTOCOL

A Phase 2, Double-Blind, Randomized, Placebo-Controlled Study of ACE-011 for the Treatment of Chemotherapy Induced Anemia in Patients with Metastatic Breast Cancer

STUDY DRUG: **ACE-011**

PROTOCOL NUMBER: **A011-08**

SPONSOR: **Acceleron Pharma, Inc.**
128 Sidney Street
Cambridge, MA 02139

PPD

MEDICAL MONITOR: **PPD Inc.**

PPD

PROTOCOL DATE: **13 March 2009**
AMENDMENT #1 : **22 April 2009**

Summary:
Amendment 01 includes the following changes:

- Added description for treatment discontinuation of dosing of ACE-011/placebo due to grade ≥ 3 toxicities related to ACE-011/placebo treatment, with the exception of patients with grade 3 hypertension that decreases to grade ≤ 1 within 7 days. A dose modification rule for the exception of grade 3 hypertension is also included.
- Added description for treatment discontinuation of patients with a hemoglobin level above the upper limit of normal during the study.

Protocol Location	Change
2. Protocol Synopsis- Investigational Product, Dosage, and Mode of Administration	Additional text was added that describes the treatment discontinuation of patients for grade ≥ 3 toxicities related to ACE-011/placebo treatment, with the exception of patients with grade 3 hypertension that decreases to grade ≤ 1 within 7 days. In this case, dose reduction rules apply. Additional text was added that describes the treatment discontinuation of patients with a hemoglobin level above the upper limit of normal during the study.
Schedule of Events	Footnote added to clarify that patients who discontinue due to ACE-011/placebo related toxicities will enter the treatment follow-up period.
9.5.2.1 ACE-011 Treatment Related Toxicity	New section added to describe the treatment discontinuation of patients for grade ≥ 3 toxicities related to ACE-011/placebo treatment, with the exception of patients with grade 3 hypertension that decreases to grade ≤ 1 within 7 days. In this case, dose reduction rules apply. Additional text was added that describes the treatment discontinuation of patients with a hemoglobin level above the upper limit of normal during the study. Added text to clarify that patients who discontinue due to ACE-011/placebo related toxicities will enter the treatment follow-up period.
Figure 1: ACE-011/placebo Dose Modification Rules (after initial dose of ACE-011/placebo)	Dose modification rule added for grade 3 hypertension.

CLINICAL STUDY PROTOCOL

A Phase 2, Double-Blind, Randomized, Placebo-Controlled Study of ACE-011 for the Treatment of Chemotherapy Induced Anemia in Patients with Metastatic Breast Cancer

STUDY DRUG: **ACE-011**

PROTOCOL NUMBER: **A011-08**

SPONSOR: **Acceleron Pharma, Inc.**
128 Sidney Street
Cambridge, MA 02139

PPD

MEDICAL MONITOR: **PPD Inc.**
PPD

PROTOCOL DATE: **13 March 2009**
AMENDMENT #1 : **22 April 2009**

Signature Page

Acceleron Pharma Approval

Signature: PPD **Date:** 23 April 2009

Name (print): _____

Investigator Agreement: I have read the protocol and agree to conduct the study as outlined in the protocol. The study will be conducted in accordance to current United States Food and Drug Administration (FDA) regulations, International Conference of Harmonization (ICH) Guidelines, Good Clinical Practices (GCP), and local ethical and legal requirements.

Signature: _____ **Date:** _____

Name (print): _____

Institution Name and Address:

_____

_____

_____

PROCEDURES IN CASE OF EMERGENCY

Table 1: Emergency Contact Information

Role in Study	Name	Address and Telephone number
PPD Medical Monitor	PPD	PPD Inc. 1800 Perimeter Park Drive, Suite 275 Morrisville, NC 27560-7200 USA PPD
Clinical Trial Manager	PPD	Acceleron Pharma, Inc. 128 Sidney Street Cambridge, MA 02139 USA PPD

2. PROTOCOL SYNOPSIS

Name of Sponsor/Company: Acceleron Pharma, Inc.
128 Sidney Street
Cambridge, MA 02139

Name of Investigational Product: ACE-011

Name of Active Ingredient:
ACE-011 is a fully human fusion protein consisting of the extracellular domain (ECD) of the activin receptor IIA (ActRIIA) linked to the human IgG1 Fc domain.
Mechanism of Action: ACE-011 is a disulfide-linked, glycosylated, dimeric protein. ACE-011 competes with the activin receptor IIA that binds a number of TGF-β superfamily ligands including activin, myostatin (growth differentiation factor [GDF]-8), and GDF-11.

Title of Study: A Phase 2, Double-blind, Randomized, Placebo-Controlled Study of ACE-011 for the Treatment of Chemotherapy Induced Anemia in Patients with Metastatic Breast Cancer.

Study center(s): approximately 25-35 centers

Phase of development: 2

Objectives

Primary:

1. To evaluate the proportion of patients in each ACE-011 dose regimen who achieve a hematopoietic response during the treatment period including up to 2 months after the last dose of ACE-011 treatment of chemotherapy induced anemia in patients with metastatic breast cancer.

Secondary:

1. To evaluate the safety and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer.
2. To evaluate the percentage of patients who achieve an increase in hemoglobin of ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs.
3. To estimate the duration of the hematopoietic response associated with each ACE-011 dose regimen.
4. To estimate the time to the hematopoietic response associated with each ACE-011 dose regimen.
5. To estimate the proportion of patients receiving red blood cell (RBC) transfusions and/or ESAs within each cycle of ACE-011 with each dose regimen.

6. To estimate the effect of each ACE-011 dose regimen on progression free survival (PFS) and objective response rate (ORR).

Exploratory objectives:

1. To estimate the effect of each ACE-011 dose regimen on serum bone biomarkers (OC, BSAP, PINP, CTX, TRACP-5b), urine bone biomarker (uNTX), bone mineral density (DXA scans of lumbar spine and hip) and incidence of skeletal related events (SREs).
2. To evaluate the quality of life during the study in relation to the treatment of chemotherapy induced anemia in patients with metastatic breast cancer during the study.

Methodology: This is a Phase 2, double-blind, randomized, placebo controlled study to evaluate the efficacy, safety, and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer. Patients will be randomized to one of three treatment arms or placebo:

1. ACE-011 0.1 mg/kg subcutaneously (SC) every 28 days for up to 4 doses.
2. ACE-011 0.3 mg/kg SC every 28 days for up to 4 doses.
3. ACE-011 0.5 mg/kg SC every 28 days for up to 4 doses.
4. Placebo SC every 28 days for up to 4 doses.

Number of patients (planned): 105 patients

Diagnosis and main criteria for inclusion

Inclusion Criteria:

1. Women ≥ 18 years of age.
2. Histologically confirmed diagnosis of breast cancer documented by cytology or biopsy.
3. Evidence of metastatic breast cancer with a minimum of one measurable lesion or non measurable lesion per RECIST criteria (Appendix 1).
4. Receiving a multicycle myelosuppressive chemotherapy regimen including one of the following: anthracycline, taxane, gemcitabine or vinorelbine.
5. Planned treatment with the same myelosuppressive chemotherapy regimen for a minimum of 9 weeks after Day 1 of ACE-011/placebo administration.
6. Hemoglobin value between ≥ 6.5 to < 11.0 g/dL (≥ 65 to < 110 g/L).
7. ≥ 30 days must have elapsed (from Day 1) since previous treatment with erythropoiesis-stimulating agent (including concurrent treatment with IV iron) for chemotherapy induced anemia.
8. ≥ 7 days have elapsed (from Day 1) since the last RBC blood transfusion and receipt of ≤ 2 units of blood in the past 30 days.
9. ECOG Performance status of 0 – 2 (Appendix 2).
10. Adequate renal (creatinine ≤ 1.5 x ULN or creatinine clearance ≥ 40 mL/min) and hepatic function (bilirubin ≤ 1.5 x ULN; AST/ALT ≤ 2.5 x ULN).
11. Females of childbearing potential participating in the study are to use adequate birth control measures (i.e. oral contraceptives or two forms of barrier method birth control) during study participation. Females of childbearing potential must have a negative serum pregnancy test within 3 days to ACE-011/placebo administration (Day 1). A female of childbearing potential is a sexually mature woman who has not undergone a hysterectomy or who has not been postmenopausal for at least 24 consecutive months (i.e. who has had menses at some time in the preceding 24 months).
12. Life expectancy of ≥ 6 months.
13. Is willing to adhere to the study visit schedule, understand and comply with all protocol requirements.
14. Understand and sign a written informed consent.

Exclusion Criteria:

1. At the time of screening patients who have any grade ≥ 3 toxicity (as per NCI CTCAE v3.0 (Appendix 3)) (except for the following disease related toxicities: hematological events- anemia, thrombocytopenia or neutropenia or non hematological events- nausea, vomiting, fatigue, or muscle or bone/joint pain).
2. Prior radiation therapy to > 20% of the whole skeleton. Use of palliative radiation if the area being treated is <15% of body surface area with no pelvic radiation or no more than 10% of the bone marrow reserve is radiated is permitted during patient participation in the study, at the discretion of the investigator.
3. Have received more than 5 prior chemotherapy treatment regimens for metastatic breast cancer.
4. Concurrent use of bevacizumab with the chemotherapy regimen during study participation.
5. Clinically significant pulmonary, endocrine, neurologic, gastrointestinal, hepatic or genitourinary disease unrelated to underlying hematologic disorder.
6. Patients with heart failure as classified by the New York Heart Association (NYHA) classification of 3 or higher (Appendix 4).
7. History of thrombosis, deep vein thrombosis (DVT), pulmonary emboli, or embolic stroke.
8. Untreated CNS metastases (exception: CNS metastases treated with whole brain radiotherapy > 6 months prior to randomization).
9. Diagnosis of a myeloid malignancy or known history of myelodysplasia.
10. History of second malignancy within 5 years (except excised and cured basal cell carcinoma, squamous cell carcinoma of the skin or cervical carcinoma in situ).
11. Patients with a recent history (within 14 days of Day 1) of administration of systemic (IV or oral) antibiotics. Patients should be free of infection at least 14 days after the last dose of systemic antibiotic treatment (from Day 1).
12. Uncontrolled hypertension. If hypertension is considered clinically stable, systolic blood pressure (BP) must be < 150 mmHg and diastolic BP must be < 100 mmHg.
13. Known history of hepatitis C antibody (HCV), hepatitis B surface antigen (HBsAg and HB core Ab), or human immunodeficiency virus (HIV) antibody.
14. Deficiency in iron (serum ferritin < 100 ng/mL (< 224.7 pmol/L)), vitamin B_{12}, or folate.
15. History of anemia as a result of inherited hemoglobinopathy such as sickle cell anemia or thalassemia.
16. History of autoimmune or hereditary hemolysis or gastrointestinal bleeding.
17. Patient received treatment with another investigational drug or device within 1 month prior to Day 1, or if the half life of the previous product is known, within 5 times the half life prior to dosing, whichever may be longer.
18. Pregnant or lactating females.
19. History of severe allergic or anaphylactic reactions or hypersensitivity to recombinant proteins or excipients in the investigational product.

Investigational product, dosage and mode of administration:

ACE-011/placebo will be administered as a SC injection, approximately every 28 days, on days 1, 29, 57, and 85 of the treatment period. Each patient will be randomized to one of three treatment arms or placebo:

Dose	Number of Patients
0.1 mg/kg	30
0.3 mg/kg	30
0.5 mg/kg	30
Placebo	15
Total patients enrolled: 105	

Patients with grade ≥3 toxicity related to ACE-011 will be discontinued from ACE-011/ placebo treatment except for the following. Patients with grade 3 hypertension that decreases to grade ≤ 1 within 7 days after treatment with anti-hypertensive therapy may continue on ACE-011/placebo treatment. Dose reduction rules will apply at the subsequent scheduled dosing day(s). Patients with a hemoglobin level above the upper limit of normal at any time during the study after Day 1 will be discontinued.

ACE-011/placebo dose modification rules are based on hemoglobin values and blood pressure measurements during each treatment cycle. ACE-011/placebo dose interruption and dose reduction rules will be implemented for those patients with hemoglobin of ≥ 11 g/dL, an increase of ≥ 2 g/dL/ 28 days, or hypertension ≥ Grade 2 within each cycle (see Figure 1).

Duration of study: Patients will be followed for a minimum of 7 months following their last dose of study drug. Study participation for each patient is approximately 10.5 months (14 day screening period, 3 month treatment period with a 2 month treatment follow up period, and a 5 month post treatment follow-up period). The start date of the chemotherapy regimen used in this protocol to treat metastatic breast cancer will be recorded. If a patient begins a new chemotherapy regimen during the study, the patient will discontinue further ACE-011/placebo treatment and be followed for survival only. If a patient has a positive anti-drug antibody result at the Day 281/Termination visit, they may be asked to return to the clinical site for additional monthly repeat anti-drug antibody testing for up to 12 months. The study is complete when the last patient enrolled has completed the last protocol required follow up visit.

Criteria for evaluation

Efficacy: The primary endpoint is to establish the proportion of patients who have a hematopoietic response of an increase in hemoglobin of ≥ 0.5 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs.

The proportion of patients with an increase in hemoglobin of ≥ 1g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for 28 consecutive days during the treatment period up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs

will be evaluated as a secondary efficacy endpoint.

The hematopoietic endpoints will be determined by the monitoring of hematologic laboratory values, RBC transfusion and/or ESAs administration. The evaluation of response will be from the first dose of ACE-011 through 2 months after the last dose (for each patient). Additional endpoints will be evaluated with the assessment of disease progression by CT/MRI and bone scan and bone mineral density by DXA scans. Bone biomarkers will be evaluated in all patients and incidence of SREs will be recorded, as applicable. SREs are defined as pathologic fracture, radiation therapy to bone, surgery to bone, or spinal cord compression.

Safety: Throughout the study, safety will be evaluated by the sponsor, medical monitor and principal investigator. Additionally, the DMC will review safety parameters for patients enrolled in the study. All patients will be assessed for safety by monitoring adverse events, clinical laboratory tests, and assessment of quality of life, vital signs and physical examinations on an ongoing basis. ECGs will also be performed during screening, prior to dosing on the first and second cycle, Day 15, and Day 113 (28 days after the last dose of ACE-011) and at Day 281/Termination visit to monitor safety. Blood pressure and lab values will be assessed at each study visit; weekly for the first two treatment (dosing) periods and approximately every one to two weeks for subsequent treatment (dosing) periods.

Pharmacokinetics: The blood samples will be collected at selected study sites from approximately one-third of patients for the evaluation of serum concentrations of ACE-011. Pre-dose PK samples will be collected on Day 1. See Appendix 5 for PK sampling schedule.

Statistical methods

Sample size determination: With an anticipated dropout rate of 20%, a sample size of 30 patients per dose level is planned to achieve at least 80% power with 2 sided significance level of 0.05 to differentiate targeted response rate of p1=0.45 from null hypothesis p0=0.20. An additional 15 patients will be enrolled into the placebo group for safety comparison. A total of 105 patients will be randomized to three ACE-011 dose groups or placebo with a ratio of 2:2:2:1.

Efficacy analysis: The primary endpoint is the proportion of patients who have a hematopoietic response of an increase in hemoglobin of ≥ 0.5 g/dL for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs. The proportion of responders will be determined together with 95% confidence interval.

For binary endpoints, the proportion of responders and 95% confidence interval will be calculated. For time to event endpoints, the Kaplan-Meier method will be used to estimate the survival curve and median time to event. Due to the nature of the study, no multiplicity adjustment will be made for efficacy analysis.

Safety analysis: Data from all patients who receive at least one dose of ACE-011/placebo will be included in the safety analyses. To assess clinical safety, adverse events, vital sign measurements and clinical laboratory information will be summarized. Adverse events and laboratory results will also be summarized by severity grade (NCI CTCAE v3.0). Changes from baseline clinical laboratory values and vital signs will be summarized over time. Descriptive statistics and shift tables will be generated as appropriate.

Schedule of Events

	Screen	Treatment Period													Post Treatment Follow up Period				Termination Visit[19]
ACE-011/placebo Dose period		#1				#2			#3			#4	Follow-up						
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	113 (± 7d)	141 (± 7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
Informed consent	X																		
Inclusion/exclusion criteria	X	X																	
Medical history	X																		
Physical examination	X	X				X			X			X	X	X					X
Vital signs[1]	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X					X
FACT-Fatigue	X	X[2]				X[2]			X[2]			X[2]							X
12 lead ECG	X	X[3]		X		X[3]							X						X
Serum iron, TIBC, transferrin and serum ferritin	X	X[2]				X[2]			X[2]			X[2]							X
Vitamin B12	X																		
Serum folate	X						X						X						X
Coagulation/ Serum chemistry[4]	X	X[5]	X	X	X	X[5]	X	X	X[5]	X	X	X[5]	X	X					X
Hematology[5]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Serum Erythropoietin marker		X[2]				X[2]			X[2]			X[2]		X					X
Peripheral Blood Smear (sample)		X[2]				X[2]			X[2]			X[2]							X
LH, FSH, testosterone, progesterone, estradiol	X	X[2]				X[2]			X[2]			X[2]	X	X					
Anti-drug antibody testing[6]		X				X			X			X	X	X					X
Bone biomarker[7]		X[2]				X[2]			X[2]			X[2]	X	X					X
Back up blood sample for future testing	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X	X	X	X	X
Pregnancy Test	X[8]	X[9]				X[9]			X[9]			X[9]							X
Urinalysis[10]	X	X[2]										X[2]							X
Evaluate transfusion frequency	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X	X	X	X	X

Schedule of Events cont.

	Screen	Treatment Period													Post Treatment Follow up Period				Termination Visit[19]
ACE-011/Placebo Dose period		#1				#2			#3			#4	Follow-up						
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	113 (± 7d	141 (± 7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
Documentation of concomitant medications	X[11]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Evaluation of AEs and SAEs		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Evaluation of Menstrual cycle[12]	X	X[2]				X[2]			X[2]			X[2]	X	X					
CT/MRI scan	X[13]									X[14]			X[14]						
Dual-energy X-ray absorptiometry (DXA[15])	X												X						X
Bone scans[16]	X									X			X						X
ACE-011/placebo administration[17]		X				X			X			X							
Chemo regimen[18]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

[1] Vital signs include weight, heart rate, seated systolic and diastolic blood pressure, and temperature will be assessed at each visit Height will be collected at screening only

[2] On dosing days (1, 29, 57 and 85), study procedure to be done prior to dosing of ACE-011/placebo

[3] 12 lead ECG is to be performed prior to dosing of ACE-011/placebo including measurement of the QT interval

[4] Coagulation (central lab only) prothrombin time (PT), partial thromboplastin time (PTT), fibrinogen Chemistry (central lab only): AST, ALT, lactate dehydrogenase (LDH), total bilirubin, indirect bilirubin, alkaline phosphatase, total protein, albumin, sodium, potassium, chloride, carbon dioxide, calcium, phosphorus, blood urea nitrogen (BUN), creatinine, uric acid, glucose, amylase, and lipase

[5] Hematology (local lab) hemoglobin and hematocrit Hematology (central lab) complete blood count (CBC) with differential and platelet, CBC includes red blood cells (RBC), white blood cells (WBC), hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), **Reticulocyte should be done on days 1, 8, 15, and days 29, 57, 85, and at the termination visit.** On dosing days (1, 29, 57 and 85), local hemoglobin and hematocrit values are to be drawn and resulted prior to dosing to apply dose modification rules for ACE-011/placebo

[6] If a patient has a positive anti-drug antibody result at the Termination visit, they may be asked to return to the clinical site for additional monthly follow-up, for up to 12 months after their last dose for a repeat anti-drug antibody test

[7] Both a serum and urine samples will be collected at each visit The urine sample should not be from the first morning urine

[8] Serum pregnancy test required of females of child bearing potential only Negative test can be within 3 days of study day 1

[9] For females of child bearing potential only urine (dipstick) pregnancy test to be collected and resulted prior to dosing of ACE-011/placebo

[10] Urinalysis to be performed at screening , prior to administration ACE-011/placebo (Days 1 and 85) and termination visit Assessments (Central lab): pH, specific gravity, protein, glucose, ketones, blood, leukocyte esterase, and nitrite Microscopy required only to follow-up abnormal findings

[11] Screening- any concomitant treatments taken 28 days prior to day of randomization

[12] Females of child bearing potential- collection of menstrual cycle information (date of last menstruation, date of menstrual period onset and duration of menstruation)

[13] A CT/MRI scan acquired within 4 weeks of initiating the current chemotherapy regimen may be used for the screening assessment Head, neck, thoracic, abdominal, and pelvic scans are to be acquired One measurable or non measurable lesion per RECIST is to be identified by the scan Brain (head) CT only to be done to rule out CNS metastases in patients with suspicious clinical symptoms or prior history of CNS metastases

[14] Follow-up CT/MRI scans may be acquired +/- 5 day window for either Day 64 or Day 113 Additional CT/MRI scans acquired during the post treatment follow up period will be performed at the discretion of the investigator

[15] Hip and lumbar spine BMD assessment

[16] Bone scans will be acquired within 4 weeks of initiating the current chemotherapy regimen for those patients who have bone metastases identified Follow up bones scans may be acquired +/- 5 day window for day 64, day 113, and Day 281

[17] Patients who discontinue ACE-011/placebo due to ACE-011/placebp related toxicities (section 9 5 2 1) will enter the treatment follow-up period

[18] Chemotherapy regimens scheduled will be administered to the patient as per standard of care Continuation of the same chemotherapy regimen may be done through the follow up period of the study at the discretion of the investigator

[19] Patients who terminate treatment early due to disease progression and/or begin a new chemotherapy regimen will complete the day 281/Terminiation visit procedures and be followed for survival only (7 months from the last dose of ACE-011/placebo) If a patient withdraws from the study (section 8 4), the Day 281/Termination visit procedures are to be completed and the patient will be followed for survival If withdrawal reason is due to ESA administration, the patient should enter the treatment follow up period

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4. LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

The following abbreviations and specialist terms are used in this study protocol.

Table 2: Abbreviations and Specialist Terms

Term	Definition
ACTH	Adrenocorticotropic hormone
ActRIIA	Activin receptor IIA
AE	Adverse event
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
AUC	Area under the plasma concentration - time curve
$AUC_{(0\text{-last})}$	AUC from time 0 to time of last quantifiable sample
$AUC_{(0\text{-}\infty)}$	AUC extrapolated to infinity
BMD	Bone mineral density
BP	Blood pressure
BSAP	Bone specific alkaline phosphatase
BUN	Blood urea nitrogen
CBC	Complete Blood Count
CFR	Code of Federal Regulations (US)
CH	Constant domain
CI	Confidence interval
CIA	Chemotherapy induced anemia
CL	Clearance
CL/F	Total clearance
C_{max}	Maximum observed concentration
CNS	Central nervous system
CO_2	Carbon dioxide
CRF	Case report form
CRO	Clinical research organization
CT	Computed tomography
CTCAE	Common Terminology Criteria for Adverse Events
CTX	Serum C-terminal type I collagen telopeptide
DLT	Dose-limiting toxicity
DXA	Dual-energy x-ray absorptiometry
DMC	Data Monitoring Committee
DVT	Deep vein thrombosis
ECD	Extracellular domain
ECG	Electrocardiogram
ECOG	Eastern Cooperative Oncology Group
EDF	Erythroid differentiation factor
ESA	Erythropoiesis-stimulating agents
F	Bioavailability (absolute)
FACT	Functional Assessment of Chronic Illness
FDA	Food and Drug Administration
FSH	Follicle-stimulating hormone
GCP	Good Clinical Practices
GH	Growth hormone

Term	Definition
HBV	Hepatitis B virus
Hct	Hematocrit
HCV	Hepatitis C virus
Hgb	Hemoglobin
HIV	Human immunodeficiency virus
ICF	Informed consent form
ICH	International Conference on Harmonization
IEC	Independent Ethics Committee
IND	Investigational New Drug
IP	Intraperitoneal
IRB	Institutional Review Board
IV	Intravenous
IVRS	Interactive Voice Response System
Kd	Binding coefficient
λz	Elimination rate constant
LDH	Lactate dehydrogenase
LH	Luteinizing hormone
MCH	Mean corpuscular hemoglobin
MCHC	Mean corpuscular hemoglobin concentration
MCV	Mean corpuscular volume
MedDRA	Medical Dictionary for Regulatory Activities
MITT	Modified Intent to Treat (population)
MRI	Magnetic resonance imaging
NOAEL	No adverse events level
NYHA	New York Heart Association
OC	Osteocalcin
ORR	Objective response rate
OVX	Ovariectomized
PD	Progressive disease
PFS	Progression free survival
PHI	Protected health information
PINP	Serum intact procollagen type I N terminal propeptide
PK	Pharmacokinetic
PP	Per-protocol (population)
PT	Prothrombin time
PTH	Parathyroid hormone
PTT	Partial thromboplastin time
RAP-011	Murine version of ACE-011, ActRIIA mFc
RBC	Red blood cell (count)
RECIST	Response Evaluation Criteria in Solid Tumors
SAE	Serious adverse event
SAP	Statistical analysis plan
SC	Subcutaneous
SERM	Selective estrogen receptor modulator
SHAM	Sham-operated
SPEP	Serum protein electrophoresis
SRE	Skeletal-related event
T1/2	Elimination half-life
TIBC	Total iron binding capacity
Tmax	Time to Cmax

Term	Definition
TRACP-5b	Serum tartrate-resistant acid phosphatase isoform-5b
ULN	Upper limit of normal
Vz/F	Volume of distribution
WBC	White blood cell (count)

5. INTRODUCTION

5.1. Indication and Rationale

This study is designed to evaluate the efficacy, safety, and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer.

Treatment of patients with metastatic breast cancer with myelosuppressive chemotherapy is frequently associated with anemia. Chemotherapy induced anemia (CIA) is a significant problem for patients with cancer, causing fatigue and reduced quality of life (1). Patients with CIA are currently treated with blood transfusion and/or erythropoiesis-stimulating agents (2). However, with these treatment options CIA is still an area of unmet medical need. Erythropoiesis-stimulating agents (ESAs) can successfully mitigate transfusion need in a proportion of these patients, but concerns have emerged regarding apparent negative effects on survival and/or tumor progression in certain patient populations. The murine surrogate to ACE-011, RAP-011, has been tested as a single agent in breast cancer cell lines MDA-MB-231 and MCF-7 and no effect on enhanced proliferation of these cell lines has been observed in vitro. Therefore, treatment with ACE-011 may provide a distinct benefit/risk profile to patients with chemotherapy induced anemia.

In both a Phase 1 single dose and multiple dose study of ACE-011 in postmenopausal women, increases in hemoglobin and hematocrit were observed following ACE-011 treatment and remained elevated over the course of study. The observed hemoglobin and hematocrit effects of ACE-011 were dose and time dependent. Please refer to the Investigator Brochure for further information.

Based on the effect of ACE-011 on hematopoiesis and consistent biological phenomena observed in both non clinical and clinical studies, it is hypothesized that the blockade of ActRIIA receptor signaling impedes the terminal differentiation step in erythropoiesis to allow additional rounds of cell replication before cells enter the final differentiation phase. The result is a substantial increase in mature erythrocytes released into the circulation. Since this proposed mechanism is different to that of known ESAs, ACE-011 may provide a different clinical profile in the treatment of CIA.

This study will provide information on the pharmacodynamics (PD) properties regarding the ability of ACE-011 to increase hemoglobin in patients with CIA.

5.2. Description of ACE-011

ACE-011 (ActRIIA-IgG1) is a fully human fusion protein consisting of the extracellular domain (ECD) of activin receptor IIA (ActRIIA) linked to the human IgG1 Fc domain, which includes the heavy chain hinge and constant domains, CH2 and CH3. ACE-011 avidly binds to activin A with a binding coefficient (Kd) approximately 8.9 pM and prevents its binding to endogenous receptors, thereby inhibiting biological effects of activin.

5.3. Activin Biology

Activin A was initially identified as a gonadal differentiation factor involved in modulating follicle stimulating hormone (FSH) secretion from the pituitary (3). Subsequently, the pleiotropic nature of activin A has become more apparent (4). There is a growing body of data suggesting a role for activin A in bone remodeling, specifically as a negative regulator of bone growth (5). Before the two molecules were shown to be identical (6), activin A was also described as erythroid differentiation factor (EDF), effecting red blood cells in the later stages of maturation (7). The mechanism(s) by which activin A influences erythropoiesis remain under investigation and, in fact, there are data from studies in vitro and in animals that support erythropoiesis-stimulatory (8,9) and -inhibitory effects (10).

At the cellular level, activin A binds initially to the high-affinity Type II receptor (ActRIIA). The ligand-bound ActRIIA then recruits the low-affinity Type I receptor (ActRIA or ALK-4). The receptor heterocomplex, through its cytoplasmic protein kinase activity, then activates the Smad signal cascade to eventually influence nuclear transcriptional factors (11, 12). The competitive binding of activin A in the blood by the ACE-011 soluble fusion protein can result in inhibition of ActRIIA receptor signaling pathway by impeding biological processes attributed to activin A.

Activin has also been attributed to erythroid differentiation and has been reported to have proerythrocytic effects and to induce terminal differentiation of RBCs. Inhibition of activin may lead to increases in proliferation of the erythroid lineage. Based on its pharmacologic effects, ACE-011 (ActRIIA-IgG1) is being developed for the treatment of bone loss associated with various disease states (e.g., osteoporosis and treatment of osteolytic lesions in patients with multiple myeloma), and treatment of anemia associated with a variety of disorders, such as chemotherapy induced anemia.

The extracellular domain sequence of ActRIIA is completely conserved among numerous species including mouse, rat, cynomolgus monkey and humans, thus ACE-011 is active in these animals. However, in order to reduce the potential immunogenicity of the fully human molecule, ACE-011, in mice and to maximize the opportunity to maintain exposures in chronic models, a murine surrogate molecule was constructed by exchanging the human immunoglobulin Fc sequence portion of ACE-011 with its murine IgG2a homolog. The resultant construct is referred to as RAP-011 (ActRIIA-mIgG2aFc).

5.3.1. Pharmacology Studies

RAP-011 was evaluated in a broad range of animal pharmacology studies to assess the effects of inhibition of activin A on the biological processes attributed to that regulatory protein. RAP-011 has been shown to have significant effects on the red cell compartment. RAP-011 treatment of mice at 10, 30 and 50 mg/kg by intraperitoneal injection (IP) twice per week for 3 months resulted in a 16-26% increase in red blood cell (RBC) counts compared to control animals. Rats treated with RAP-011 at 0.3, 3 and 30 mg/kg once per week for 3 months showed RBC count increases of 6-15% over control animals. Finally, in cynomolgus monkeys treated with 10, 30 or

50 mg/kg of ACE-011 twice per month for 3 months there was a 21- 24% increase in RBC counts compared to controls animals. In this study, the RBC count increase was apparent as early as 2 weeks following the initial dose of ACE-011.

While the mechanism(s) underlying the stimulation effect of ACE-011 on erythropoiesis are not yet fully understood, it is hypothesized that a blockade of ActRIIA receptor signaling impedes the terminal differentiation step in erythropoiesis to allow additional rounds of erythroid cell replication before cells enter the terminal differentiation phase. The result is a substantial increase in mature erythrocytes released into circulation.

RAP-011 administered at 10 mg/kg to mice three days prior to administration of paclitaxel at 30 mg/kg was sufficient to prevent decreases in red blood cell parameters seen three days later. Mice receiving paclitaxel alone had decreased hematocrit from 43% to 38% three days later. RAP-011 administered 3 days prior to paclitaxel injection was sufficient to keep the hematocrit above 42% at three days and up to two weeks after paclitaxel administration. Therefore prophylactic treatment with RAP-011 was able to prevent paclitaxel induced anemia in mice.

RAP-011 has also been shown to significantly increase bone mineral density and strength in normal animals and in a variety of animal models of bone loss (13, 14, 15). In a murine ovariectomy (OVX)-induced model of osteopenia, RAP-011 (10 mg/kg IV, twice per week x 12 weeks) increased trabecular BMD > 25% in 6-12 weeks compared to the phosphate buffered saline (PBS)-treated controls ($p<0.002$). RAP-011 treatment was able to reverse the accelerated bone loss associated with ovariectomy. As expected sham-operated (SHAM) mice (i.e., intact ovaries) started the study with a greater trabecular BMD than OVX mice. PBS-treated SHAM mice showed a 10% decrease in trabecular BMD after 12 weeks of treatment compared to baseline, while SHAM-RAP-011 mice had a 32% increase in trabecular BMD over the same period. These data demonstrate that RAP-011 is able to increase trabecular BMD in both the normal state and in a model of established bone loss. The femurs of both the RAP-011 treated OVX and SHAM mice showed statistically significant increases in all measured parameters of bone strength (i.e., maximum load, stiffness, energy, ultimate strength, and elastic modulus). In summary, treatment with RAP-011 produced bone of superior quality consistent with an anabolic agent.

RAP-011 was evaluated in a mouse model of skeletal metastasis using luciferase-tagged, human MDA-MB-231 breast cancer cells (estrogen receptor negative). Mice were pretreated for 2 weeks with RAP-011 (10 mg/kg, biweekly, SC) prior to intracardiac injection of tumor cells. Treatment with RAP-011 was continued for an additional 5 weeks following tumor implantation. A parallel study was conducted to examine the effect of RAP-011 treatment, as described above, on survival of MDA-MB-231 bearing mice.

The data demonstrate that RAP-011 treatment acts to inhibit MDA-MB-231 metastasis to bone and may also play a role in inhibiting tumor growth in soft tissue. Furthermore, in the presence of tumor cells, RAP-011 is able to prevent lytic disease through an anabolic mechanism of bone formation. Lastly, inhibition of tumor growth and metastasis with RAP-011 leads to a greater than 25% survival benefit in this model.

The efficacy of RAP-011 was examined in an orthotopic model of breast cancer using luciferase-tagged human MCF-7 breast cancer cells (estrogen receptor positive). Mice were pretreated for 2 weeks with RAP-011 (10 mg/kg, biweekly, SC) prior to the implantation of tumor cells into the mammary fat pad of female nude mice. Treatment with RAP-011 was continued for an additional 7 weeks following tumor implantation at which time mice were sacrificed and assessed for tumor burden. RAP-011 effectively lowered the tumor burden in mice as detected by bio-luminescence. In addition, a 60% reduction in tumor weight was observed in the RAP-011 treated mice.

The ability of RAP-011 to repair osteolytic lesions caused by metastatic disease was investigated in mice. In this model athymic nude mice received an intratibial injection of MDA-MB-231-Luc cells to mimic bone metastasis. Mice were treated with a chemotherapy regimen of paclitaxel (20 mg/kg, IP, every three days) starting seven days after tumor injection and continuing for the duration of the study. On study day 42 mice with a detectable tumor burden by bioluminescent imaging were divided into two groups and began treatment with RAP-011 (10 mg/kg, SC, biweekly) or vehicle. Prior to dosing, μCT scans of the tumor bearing tibia were conducted to identify osteolytic lesions. RAP-011 treatment continued for four weeks (to study day 70) and a final μCT scan of the tibia was performed. On study day 70 osteolytic lesions remained the same or had progressed further when compared to study day 42 in the vehicle treated mice. While some mice showed progression of osteolytic lesions (most likely related to tumor burden) on study day 70, a majority of mice treated with RAP-011 demonstrated repair of the osteolytic lesions seen on study day 42. Therefore treatment with RAP-011 has the ability to repair osteolytic lesions caused by tumors and after cytoxic chemotherapy with paclitaxel.

5.3.2. Toxicology

ACE-011 has been evaluated for toxicological effects in two species, Sprague-Dawley rats and cynomolgus monkeys. The dose levels ranged from 0.3 to 30 mg/kg in rats and from 1 to 50 mg/kg in monkeys. These dose ranges were designed to support Phase 1a single-dose levels of 0.01 to 3.0 mg/kg IV and Phase 1b multiple doses of 0.1 to 2 mg/kg (monthly, SC). Weekly dosing in animals was designed to provide continuous, but fluctuating serum concentrations of ACE-011, which would be mimicked by a one-month dosing interval in humans.

Initial studies utilized IV dosing to identify target organs while longer-term studies (3 months in rats; 3 and 6 months in monkeys) utilized SC dosing to mimic the intended dosing regimen for patients. The expected pharmacologic effect, increased red blood cells (RBCs), hemoglobin (Hb), hematocrit (HCT) and reticulocytes, was observed in all studies, presumably based on the ability of ACE-011 to inhibit activin. A second expected pharmacologic effect of ACE-011, based on inhibition of activin, was the reversible reduction in sperm production and testicular tubular damage in rats. Since male cynomolgus monkeys were sexually immature in these studies, it was not possible to monitor this effect in this species.

Effects on the adrenal gland were only seen in rats and were more pronounced in female rats. This toxicological observation may not be relevant for predicting effects in humans since adrenal toxicity was not seen in cynomolgus monkeys. Dose-limiting toxicity and no observable adverse effect levels (NOAELs) were primarily based on renal toxicity in both rats and monkeys. There

was some indication that kidney toxicity was an indirect effect, based on the formation of antibody/antigen complexes, but since these studies were not designed to investigate toxicological mechanisms, a definitive cause of renal impairment and renal damage was not determined. Development of antibodies to ACE-011 was noted in all of these studies, which is an expected immune reaction in rats and monkeys dosed with a human protein.

The no observable adverse effect level (NOAELs) from the 3 month SC studies was 3 and 30 mg/kg in rats and monkeys, respectively. Because the kidney findings were observed at all dose levels, the NOAEL from the 6 month monkey study is < 10 mg/kg.

5.4. Summary of Clinical Experience

5.4.1. A011-01: A Phase 1a Study in Healthy Postmenopausal Women

ACE-011 was first studied in a randomized, Phase 1a, single dose, dose escalation study in healthy, post-menopausal females (16). ACE-011 was diluted in normal saline administered as an IV infusion (over approximately 1 hour) or as a SC injection on Day 1. Dose levels were 0.01, 0.03, 0.1, 0.3, 1.0, and 3.0 mg/kg IV and 0.03 and 0.1 mg/kg SC. A total of 48 subjects were enrolled; 5 active and 1 placebo at each of the IV and SC dose levels. All subjects were followed for 4 months following a single dose administration.

The pharmacokinetics of ACE-011 was linear for all IV and SC doses. Across IV doses ranging from 0.1 through 3.0 mg/kg, the mean clearance (CL) ranged from 0.092 to 0.128 mL/h/kg, the volume of distribution ranged from 73.7 to 110 mL/kg, and the mean T1/2 ranged from 23.7 to 31.8 days, with no apparent dependence on dose. After SC administration of 0.03 and 0.1 mg/kg, ACE-011 was completely absorbed, and the mean T1/2 was approximately 30 days, with no apparent dependence on dose.

The most commonly occurring treatment-emergent adverse events (AEs; i.e., those occurring in more than 1 subject in any treatment group) were headache, infusion site reaction, injection site hemorrhage, and toothache. The majority of the injection site reactions and injection site hemorrhages were in the first IV cohort and were related to infiltration of the IV site. The majority of treatment-emergent AEs were mild in severity and were judged to be unrelated to ACE-011. No deaths, serious AEs (SAEs), or AEs leading to discontinuation were reported. Changes in red blood cells (RBCs), hemoglobin, reticulocytes, liver function tests, glucose, uric acid, amylase and lipase occurred in some subjects; however, none of these laboratory results were reported as adverse events. Mild, transient elevations in pancreas enzymes, liver enzymes, or glucose were reported as AEs in five patients. There were no clinically significant changes from baseline in vital signs, physical examination, endocrine function, or electrocardiogram (ECG) data. Preservation of adrenal cortical function was monitored through evaluation of serum electrolytes (sodium and potassium levels) and through cortisol response to adrenocorticotropic hormone (ACTH) stimulation. No clinically significant findings were observed at doses up to 3.0 mg/kg IV, and ACE-011 was well tolerated in healthy, post-menopausal female volunteers in single dose levels up to 0.1 mg/kg SC and 3.0 mg/kg IV, the highest dose levels tested in this study.

5.4.2. A011-02: A Phase 1b Study in Healthy Postmenopausal Women

ACE-011 was studied in a Phase 1b, single-center, randomized, double-blind, placebo-controlled, multi-dose, dose-escalating study to evaluate the safety, tolerability and pharmacodynamics of ACE-011 in healthy postmenopausal women. Four cohorts of 10 subjects each were planned at the following dose levels: 0.1, 0.3, 1.0, and 2.0 mg/kg administered subcutaneously. Within each cohort, subjects were to be randomized to either active or placebo treatment in an 8:2 ratio. Subjects were to receive one SC injection of ACE-011 or placebo every 28 days for a total of 4 doses. All subjects were to be followed for 12 weeks after the last dose.

The treatment phase of the study was terminated early after a dose-limiting pharmacodynamic effect was observed. While on study, one subject in the 1.0 mg/kg cohort experienced a Serious Adverse Event (SAE) of progressive and persistent hypertension that was attributed to a rapid and significant rise in hemoglobin and hematocrit levels. Due to symptoms of headaches, nausea, eye pain, dizziness and vomiting approximately 1 week following the second dose, the subject was hospitalized for monitoring and evaluation of her hypertension. The head MRI, fundoscopy, and ECG performed were reported as normal and headache symptoms resolved following therapeutic phlebotomy. The SAE resolved and the subject was discharged from the hospital the following day. The hypertension was monitored closely and managed initially with antihypertensive medication. The hypertension resolved by the end of the study without need of any antihypertensive medication. The subject received aspirin prophylactically until the end of the study and continued taking ibuprofen as needed for headaches. Further details can be found in the Investigator Brochure.

Based on the magnitude of the hematopoietic response, the Sponsor suspended dose escalation to the 2.0 mg/kg dose level and further dosing in all cohorts as a result of this dose-limiting pharmacodynamic effect at the 1.0 mg/kg dose level. In total, 31 subjects were enrolled and treated. Dose levels of ACE-011 administered included 0.1, 0.3, and 1.0 mg/kg (Cohorts 1, 2, and 3 respectively). All subjects randomized to active treatment in Cohort 1 received all 4 planned doses of ACE-011. Due to early discontinuation of study drug, subjects randomized to active treatment in Cohort 2 received 3 doses of ACE-011, and subjects randomized to active treatment in Cohort 3 received 2 doses of ACE-011. Subjects randomized to placebo treatment received between 1 and 4 doses of study treatment.

In the analysis of the unblinded data, after the administration of the first dose, a dose and time dependent increase in hemoglobin values was observed (see Table 3 below):

Table 3: A011-02: A Phase 1b Study in Healthy Postmenopausal Women: Hemoglobin Evaluation

Evaluation Time Point	Mean Change from Baseline (g/dL)			
	Placebo N=7*	ACE-011 0.1 mg/kg N=8	ACE-011 0.3 mg/kg N=8	ACE-011 1.0 mg/kg N=8
Baseline Mean, pre-Dose 1	13.20	13.11	13.30	12.71
Day 8	0.17	0.68	0.85	1.21
Day 15	-0.27	0.43	0.44	1.75
Day 29	0.27	0.61	1.21	2.68**
Day 36	0.56	0.64	1.89	2.96
Day 43	-0.02	0.89	1.21	2.85
Day 57	0.27	1.28	1.64**	2.09
Day 64	0.53	1.11	2.49	2.21
Day 71	-0.10	1.34	2.09	1.66
Day 85	0.38	1.18**	2.55	1.86
Day 92	0.00***	1.04	1.60***	3.80***
Day 99	-0.20	1.21	3.20***	1.28***
Day 113	-0.02	1.30	1.29	1.04
Day 141	0.30	0.95	0.34	2.30
Day 169	0.20***	0.06		2.00***

*The number of placebo subjects with data decreases over time as a result of the early discontinuation of the study (i.e., there were placebo subjects in each dosing cohort). There were seven placebo subjects with data at baseline, Days 8, 15, and 36; six subjects with data at Days 29, 43, 57, and 85; five subjects with data on Days 71 and 113; four subjects with data on Day 64; three subjects with data on Day 141; two subjects with data on Day 99; and one subject with data on Days 92 and 169.

** Number of doses administered per treatment group: 0.1 mg/kg 4 doses; 0.3 mg/kg 3 doses; 1.0 mg/kg 2 doses. Data beyond this study day are considered follow-up results.

***n=1

No severe or life-threatening events were reported. The incidence of the most common treatment-emergent AEs (i.e., occurring in more than one subject in any treatment group) is presented below (see Table 4). The most notable AEs were those related to increases in hematologic laboratory measures in the 1.0 mg/kg dose group: hematocrit, hemoglobin, and RBC count. AEs of increased hemoglobin and/or hematocrit were reported for seven of the eight subjects in this dose group. These events were reported as mild or moderate elevations; all were considered probably related to study drug treatment. Three of the subjects in the 1.0 mg/kg group with elevated hemoglobin levels underwent phlebotomies and all hemoglobin elevations were resolved by the end of the follow-up period. No erythroid lineage AEs were reported in the 0.1 or 0.3 mg/kg treatment groups.

Table 4: A011-02: Incidence of Most Common Adverse Events

System Organ Class and Preferred Term	Dose Group			
	Placebo N=7 n (%)	0.1 mg/kg N=8 n (%)	0.3 mg/kg N=8 n (%)	1.0 mg/kg N=8 n (%)
GASTROINTESTINAL DISORDERS				
Diarrhea	0 (0.0%)	2 (25.0%)	0 (0.0%)	0 (0.0%)
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS				
Asthenia	0 (0.0%)	1 (12.5%)	2 (25.0%)	0 (0.0%)
Fatigue	2 (28.6%)	0 (0.0%)	1 (12.5%)	2 (25.0%)
INFECTIONS AND INFESTATIONS				
Viral upper respiratory tract infection	0 (0.0%)	4 (50.0%)	0 (0.0%)	0 (0.0%)
INJURY, POISONING AND PROCEDURAL COMPLICATIONS				
Limb injury	0 (0.0%)	1 (12.5%)	0 (0.0%)	2 (25.0%)
INVESTIGATIONS				
Hematocrit increased	0 (0.0%)	0 (0.0%)	0 (0.0%)	6 (75.0%)
Hemoglobin increased	0 (0.0%)	0 (0.0%)	0 (0.0%)	7 (87.5%)
Red blood cell count increased	0 (0.0%)	0 (0.0%)	0 (0.0%)	3 (37.5%)
METABOLISM AND NUTRITION DISORDERS				
Decreased Appetite	1 (14.3%)	0 (0.0%)	2 (25.0%)	0 (0.0%)
MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS				
Arthralgia	2 (28.6%)	0 (0.0%)	2 (25.0%)	0 (0.0%)
Muscle spasms	0 (0.0%)	1 (12.5%)	0 (0.0%)	2 (25.0%)
NERVOUS SYSTEM DISORDERS				
Dizziness	0 (0.0%)	0 (0.0%)	2 (25.0%)	2 (25.0%)
Headache	2 (28.6%)	4 (50.0%)	3 (37.5%)	2 (25.0%)
Paresthesia	1 (14.3%)	0 (0.0%)	0 (0.0%)	3 (37.5%)
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS				
Oropharyngeal pain	2 (28.6%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
VASCULAR DISORDERS				
Hot flush	0 (0.0%)	2 (25.0%)	0 (0.0%)	1 (12.5%)

Note: Table presents AEs reported in more than one subject in any treatment group.

Headache was reported frequently in all treatment groups with no apparent dose response trend, though the reports in the higher dose groups were more often considered possibly related to treatment. Most headaches were reported as mild (one headache was reported as moderate).

Paresthesia and dizziness were reported more frequently in the ACE-011 groups, though the events were generally not considered drug related. Other frequently reported events (e.g. fatigue, upper respiratory infection) did not appear to increase with dose and were generally mild.

Hemoglobin levels for all subjects with elevations had returned to within normal limits by the end of the study.

Adrenal cortical function was monitored through evaluation of serum electrolytes (sodium and potassium levels) and through cortisol response to adrenocorticotropic hormone (ACTH) stimulation. All ACTH simulation test results were normal.

PK results confirmed that the PK of ACE-011 is linear following the first SC doses of all three dose levels tested, 0.1, 0.3, and 1.0 mg/kg. The terminal half life ($T_{1/2}$) of ACE-011 following the last dose in all three dose groups was identical, with mean $T_{1/2}$ being approximately 23 days. The mean CL/F ranged from 3.05 to 3.90 mL/d/kg, the mean Vz/F ranged from 97.47 to 103.03 mL/kg, and the mean $T_{1/2}$ ranged from 20.92 to 23.34 days in all 3 dose levels, with no apparent dependence on dose.

Bone mineral density was assessed by dual-energy X-ray absorptiometry (DXA). Mean change and mean percent change in BMD results from baseline to study end are summarized in the table below. A dose-dependent increase in the BMD of the total hip from baseline to end of study was observed, with a significant and rapid increase of 2.4% in the 1.0 mg/kg dose group, compared to a 0.7% decrease in the placebo group. BMD results for lumbar spine showed slight increases of 0.4% to 1.0% from baseline to study end in all active treatment groups, compared with a 0.5% decrease in the placebo group (see Table 5 below):

Table 5: A011-02: A Phase 1b Study in Healthy Postmenopausal Women: BMD

	Treatment Group							
	Placebo N=7		**ACE-011 0.1 mg/kg N=8**		**ACE-011 0.3 mg/kg N=8**		**ACE-011 1.0 mg/kg N=8**	
	Mean Change	Mean Percent Change	Mean Change	Mean Percent Change	Mean Change	Mean Percent Change	Mean Change	Mean Percent Change
Lumbar Spine (g/cm^2)	-0.0020	-0.5%	0.0099	0.7%	0.0082	1.0%	0.0051	0.4%
Total Hip (g/cm^2)	-0.0062	-0.7%	0.0050	0.6%	0.0075	0.9%	0.0220	2.4%

Number of doses administered and day of last dose per treatment group: 0.1 mg/kg 4 doses (Day 85); 0.3 mg/kg 3 doses (Day 57); 1.0 mg/kg 2 doses (Day 29). Data beyond this study day are considered follow-up results.

5.4.3. A011-04: A Phase 2a Study in Patients with Osteolytic Lesions of Multiple Myeloma

Study A011-04 is an ongoing, non-IND, Phase 2a, multi-center, randomized, multiple-dose study to evaluate the safety, tolerability and efficacy of ACE-011 in patients with osteolytic lesions of multiple myeloma. Safety evaluations include adverse events (AEs), clinical laboratory tests, standard 12-lead electrocardiogram (ECG), vital signs, Eastern Cooperative Oncology Group

(ECOG) performance status and physical examinations. Additionally, the study includes the assessment of biochemical markers of bone formation and resorption, skeletal related events (SREs), bone mineral density (BMD) by duel-energy x-ray absorptiometry (DXA) and bone pain by visual analog scale (VAS).

In this study, patients are randomized in a 4:1 ratio to one of three dose levels of ACE-011 (0.1, 0.3 and 0.5 mg/kg) or placebo, to be administered to patients every 28 days by subcutaneous injection, for up to four doses over a 3-month period. The test article is being evaluated in combination with the anti-myeloma therapy of melphalan (4 mg/m^2 on days 1-7), prednisolone (40 mg/m^2 on days 1-7) and thalidomide (100 mg per day) (MPT). The sites, Sponsor and Sponsor representatives are blinded to treatment assignment.

As of 10 March 2009, 30 patients have been randomized, enrollment is complete and treatment is ongoing. Fifteen patients have received multiple doses of their assigned treatment; 7 patients have received 4 doses, 3 patients have received 3 doses and 5 patients have received 2 doses. Of the remaining 15 patients, 12 patients have received their first dose of ACE-011 or placebo and 3 patients have not yet received their first dose. There have been no ACE-011 related serious adverse events (SAEs) reported. Per the monitored preliminary data on 18 patients presented to the Independent Data Monitoring Committee (DMC) on 9 February 2009, there were 8 Grade 3/severe AEs reported in 4 patients, including neutropenia, platelet count decreased, pain in extremity, back pain and anemia. All Grade 3 AEs were determined to be not related to ACE-011 and the Grade 3 hematologic adverse events were considered either probably or definitely related to the MPT treatment.

Following preliminary analysis of the blinded central laboratory data, increases in hemoglobin values were observed within 28 days after administration of the first dose of ACE-011/placebo. Per data available, 11 out of 30 patients had $\geq$ 1 g/dL increase in hemoglobin within the first 28 days on the study, 4 patients achieved a $\geq$ 2 g/dL increase within their first 28 days on study, while 8 patients achieved a $\geq$ 2 g/dL increase in hemoglobin through Day 85.

Including all visits as of 10 March 2009 through Day 85, 9 out of 30 patients had a dose interruption due to a hemoglobin value of > 13 g/dL according to the dose modification rules defined in the protocol and 1 of the 9 patients, who had a history of hypertension, also had a dose interruption due to Grade 2 hypertension. Three of the 9 patients that required a dose interruption due to a hemoglobin value of > 13 g/dL, as per protocol, had hemoglobin levels above 13 g/dL prior to their first dose. Taken together, these data, suggest a beneficial pharmacodynamic effect of ACE-011 on erythropoiesis in a patient population with cancer chemotherapy induced anemia. No patients have had a dose reduction in ACE-011/placebo treatment or discontinued treatment.

One SAE of prolonged hospitalization due to pneumonia, that was considered not related to ACE-011, was reported in a PPD enrolled in the study. On PPD four days after the first dose of ACE-011/placebo and the initial doses of melphalan, prednisolone and thalidomide (MPT), the patient presented with elevated temperatures up to 39º C, cough and fatigue. Chest x-ray performed on PPD revealed a pneumonic infiltration in the lower

lobe of the left lung. On PPD hospitalization of the patient was prolonged due to the event. The patient was treated with moxifloxacin, doripenem, meropenem and fluconazole.

On PPD the pneumonic infiltration resolved, as confirmed by chest x-ray. This SAE resolved upon discharge of the patient from the hospital on PPD.

The investigator considered the event of pneumonia not related to ACE-011 and possibly related to MPT. The Sponsor agreed with the Investigator's assessment of causality. Pneumonia is considered a labeled event per the current package insert for thalidomide. The SAE was considered serious, related and expected with respect to thalidomide and unrelated to ACE-011.

The A011-04 study has completed enrollment, while treatment, follow-up and data monitoring are ongoing.

5.5. Potential Risks for Human Use

The 1-month safety study in rats suggested that ACE-011 may affect sperm count and motility in a dose-dependent manner, although these findings appeared to be reversible upon drug discontinuation. These reproductive effects are likely a result of the expected biological activity of activin inhibition.

The 1-month safety study in rats also suggested that ACE-011 induces adrenocortical necrosis in rats. At present, the mechanism by which ACE-011 has induced this lesion in rats and the basis for its apparent species specificity are unknown; the finding has not been observed in mice or cynomolgus monkeys at similar dose levels. Adrenal cortical function was monitored in human subjects receiving ACE-011 by the evaluation of serum electrolytes including sodium and potassium levels and by the evaluation of cortisol response to ACTH stimulation in both the Phase 1a single dose and Phase 1b multiple dose healthy volunteer clinical studies. These parameters were evaluated up to doses of 3.0 mg/kg IV and 1.0 mg/kg SC, and no clinically significant perturbations in adrenal function were observed.

Laboratory findings from the Phase 1a single dose study showed elevations in hematology results, pancreas and liver enzymes, and uric acid. The most commonly seen treatment-emergent adverse events in this study were headache, infusion site reaction, injection site hemorrhage, and toothache.

Based on data from the Phase 1b study, the most notable AEs were increases in hematologic laboratory measures in the 1.0 mg/kg dose group: hematocrit, hemoglobin, and RBC count. AEs of increased hemoglobin and/or hematocrit were reported for seven of the eight subjects in this dose group. These events were reported as mild or moderate elevations; all were considered probably related to study drug treatment. No erythroid lineage AEs were reported in the 0.1 or 0.3 mg/kg treatment groups. Hematologic parameters will be monitored carefully in this study.

Headache was reported frequently in all treatment groups with no apparent dose response trend, though the reports in the higher dose groups were more often considered possibly related to treatment. Most headaches were reported as mild (one headache was reported as moderate). Paresthesia and dizziness were reported more frequently in the ACE-011 groups, though the

events were generally not considered drug related. Other frequently reported events (e.g. fatigue, upper respiratory infection) did not appear to increase with dose and were generally mild.

In the Phase 1b multiple-dose healthy volunteer study, some subjects treated with ACE-011 showed elevations in blood pressure. One subject who had received two doses of ACE-011 at the 1.0 mg/kg dose level had a serious adverse event. The patient was hospitalized for further evaluation of increases in blood pressure and symptoms that may have been caused by the increase in blood pressure. The symptoms that the subject experienced included headache, dizziness, nausea, vomiting, and elevated hematology values (increases in the number of red blood cells). This serious adverse event was judged to be probably related to the study drug and resolved following discharge from hospital the following day. Throughout the course of the follow up period hematologic parameters were monitored frequently until the hemoglobin and hematocrit levels returned to within normal limits. Further details are outlined in the Investigator Brochure.

As with all biologics there is the potential for anti-drug antibodies that can be associated with increased drug clearance and hypersensitivity reactions. An immune response to ACE-011 has been seen in monkeys that received the drug for 1 month and longer. Some of those animals developed kidney inflammation possibly related to that immune response. Anti-drug antibody formation will be assessed in this study to examine the immunogenicity of ACE-011, and to monitor reversibility of any AEs over time.

Please refer to the Investigator Brochure for more information.

6. TRIAL OBJECTIVES AND PURPOSE

6.1. Primary objective

1. To evaluate the proportion of patients in each ACE-011 dose regimen who achieve a hematopoietic response during the treatment period including up to 2 months after the last dose of ACE-011 treatment of chemotherapy induced anemia in patients with metastatic breast cancer.

6.2. Secondary objectives

1. To evaluate the safety and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer.
2. To estimate the duration of the hematopoietic response associated with each ACE-011 dose regimen.
3. To estimate the time to the hematopoietic response associated with each ACE-011 dose regimen.
4. To estimate the proportion of patients receiving red blood cell (RBC) transfusions and/or ESAs within each cycle of ACE-011 with each dose regimen.
5. To evaluate the percentage of patients who achieve an increase in hemoglobin of ≥ 1 g/dl, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for a consecutive 28 day period during the treatment period including up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs.
6. To estimate the effect of each ACE-011 dose regimen on progression free survival (PFS) and objective response rate (ORR).

6.3. Exploratory objectives

1. To estimate the effect of each ACE-011 dose regimen on serum bone biomarkers (OC, BSAP, PINP, CTX, TRACP-5b), urine bone biomarker (uNTX), bone mineral density (DXA scans of lumbar spine and hip) and incidence of skeletal related events (SREs).
2. To evaluate the quality of life during the study in relation to the treatment of chemotherapy induced anemia in patients with metastatic breast cancer during the study.

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7. INVESTIGATIONAL PLAN

7.1. Overall Study Design and Plan

This is a Phase 2, double-blind, randomized, placebo-controlled study to evaluate the efficacy, safety, and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer. A total of 105 patients will be enrolled.

The Data Monitoring Committee (DMC) is responsible for reviewing the safety. The DMC will be comprised of a minimum of 3 members. The DMC will meet regularly during the study to review serious adverse events (SAEs), adverse events (AEs), laboratory results, and vital signs. Safety data will be reviewed throughout the study by the DMC, Medical Monitor and the Investigator. DMC responsibilities, membership, meeting frequencies and procedures will be outlined in the DMC charter.

Patients will be evaluated for study inclusion/exclusion criteria by the Investigator. Patients who meet the study entry criteria will be enrolled within 14 days of the screening visit. Central lab values will be utilized for the evaluation of patient eligibility and in the evaluation of patient safety throughout the study. Local lab values for hemoglobin and hematocrit will be collected and reported from screening through the Day 281/Termination visit. Three cohorts of 30 patients each are planned at the following doses of ACE-011: 0.1 mg/kg, 0.3 mg/kg and 0.5 mg/kg. 15 patients will be enrolled into the placebo cohort (as shown in Table 6).

Table 6: Study Design

Dose	Number of Patients
0.1 mg/kg	30
0.3 mg/kg	30
0.5 mg/kg	30
Placebo	15
Total patients enrolled: 105	

Each eligible patient will be randomized to one of the four cohorts to receive a dose administered as a subcutaneous injection every 28 days for up to 4 doses. Dosing will be administered on days 1, 29, 57, and 85. Concurrent treatment with a myelosuppressive chemotherapy regimen for metastatic breast cancer will also be administered per standard of care at the site. Patients will be stratified according to administration of either single agent weekly chemotherapy or less frequent chemotherapy regimens.

Blood samples will be collected at selected study sites from approximately one-third of patients for the evaluation of serum concentrations of ACE-011. Pre-dose PK samples will be collected on Day 1. See Appendix 5 for PK sampling schedule.

Patients will be assessed for safety by monitoring adverse events, clinical laboratory tests, assessment of quality of life (FACT- Fatigue (17)), vital signs and physical examinations on an ongoing basis. ECGs will be performed during screening, prior to dosing on the first and second cycle, Day 15, and Day 113 (28 days after the last dose of ACE-011) and at Day 281/ Termination visit to monitor safety. Blood pressure and lab values will be assessed at each study visit during the treatment period; weekly for the first two cycles and approximately every two weeks for subsequent cycles.

Local laboratory hemoglobin values and blood pressure values will be evaluated prior to dosing per the dose modification criteria, as applicable.

Patients will be followed for a minimum of 7 months following their last dose of ACE-011. Study participation for each patient is approximately 10.5 months (14 day screening period, 3 month treatment period with a 2 month treatment follow up period, and a 5 month post treatment follow-up period). If a patient discontinues the study due to progression of disease (metastatic breast cancer) and/or begins another chemotherapy regimen, the patient will complete the Day 281/Termination visit procedures and be followed for survival only (a minimum 7 months from the last dose of ACE-011).

Anti-drug antibody testing will be performed at Day 281/Termination visit and if positive, the patient may be asked to return to the clinical site for additional monthly anti-drug antibody testing for up to 12 months, after their last dose.

Table 7: Schedule of Events

Schedule of Events																			
	Screen	Treatment Period													Post Treatment Follow up Period				Termination Visit[19]
ACE-011/placebo Dose period		#1				#2			#3			#4	Follow-up						
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	113 (± 7d)	141 (± 7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
Informed consent	X																		
Inclusion/exclusion criteria	X	X																	
Medical history	X																		
Physical examination	X	X				X			X			X	X	X					X
Vital signs[1]	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X					X
FACT-Fatigue	X	X[2]				X[2]			X[2]			X[2]							X
12 lead ECG	X	X[3]		X		X[3]							X						X
Serum iron, TIBC, transferrin and serum ferritin	X	X[2]				X[2]			X[2]			X[2]							X
Vitamin B12	X																		
Serum folate	X						X						X						X
Coagulation/ Serum chemistry[4]	X	X[5]	X	X	X	X[5]	X	X	X[5]	X	X	X[5]	X	X					X
Hematology[5]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Serum Erythropoietin marker		X[2]				X[2]			X[2]			X[2]		X					X
Peripheral Blood Smear (sample)		X[2]				X[2]			X[2]			X[2]							X
LH, FSH, testosterone, progesterone, estradiol	X	X[2]				X[2]			X[2]			X[2]	X	X					
Anti-drug antibody testing [6]		X				X			X			X	X	X					X
Bone biomarker[7]		X[2]				X[2]			X[2]			X[2]	X	X					X
Back up blood sample for future testing	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X	X	X	X	X
Pregnancy Test	X[8]	X[9]				X[9]			X[9]			X[9]							X
Urinalysis[10]	X	X[2]										X[2]							X
Evaluate transfusion frequency	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X	X	X	X	X

Schedule of Events cont.

	Screen	Treatment Period													Post Treatment Follow up Period				Termination Visit[19]
ACE-011/Placebo Dose period		#1				#2			#3			#4	Follow-up						
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	113 (± 7d	141 (± 7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
Documentation of concomitant medications	X[11]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Evaluation of AEs and SAEs		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Evaluation of Menstrual cycle[12]	X	X[2]				X[2]			X[2]			X[2]	X	X					
CT/MRI scan	X[13]									X[14]			X[14]						
Dual-energy X-ray absorptiometry (DXA[15])	X												X						X
Bone scans[16]	X									X			X						X
ACE-011/placebo administration[17]		X				X			X			X							
Chemo regimen[18]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

[1] Vital signs include weight, heart rate, seated systolic and diastolic blood pressure, and temperature will be assessed at each visit Height will be collected at screening only
[2] On dosing days (1, 29, 57 and 85), study procedure to be done prior to dosing of ACE-011/placebo
[3] 12 lead ECG is to be performed prior to dosing of ACE-011/placebo including measurement of the QT interval
[4] Coagulation (central lab only) prothrombin time (PT), partial thromboplastin time (PTT), fibrinogen Chemistry (central lab only): AST, ALT, lactate dehydrogenase (LDH), total bilirubin, indirect bilirubin, alkaline phosphatase, total protein, albumin, sodium, potassium, chloride, carbon dioxide, calcium, phosphorus, blood urea nitrogen (BUN), creatinine, uric acid, glucose, amylase, and lipase
[5] Hematology (local lab) hemoglobin and hematocrit Hematology (central lab) complete blood count (CBC) with differential and platelet, CBC includes red blood cells (RBC), white blood cells (WBC), hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), **Reticulocyte should be done on days 1, 8, 15, and days 29, 57, 85, and at the termination visit.** On dosing days (1, 29, 57 and 85), local hemoglobin and hematocrit values are to be drawn and resulted prior to dosing to apply dose modification rules for ACE-011/placebo
[6] If a patient has a positive anti-drug antibody result at the Termination visit, they may be asked to return to clinic for additional monthly follow-up, for up to 12 months after their last dose for a repeat anti-drug antibody test
[7] Both a serum and urine samples will be collected at each visit The urine sample should not be from the first morning urine
[8] Serum pregnancy test required of females of child bearing potential only Negative test can be within 3 days of study day 1
[9] For females of child bearing potential only urine (dipstick) pregnancy test to be collected and resulted prior to dosing of ACE-011/placebo
[10] Urinalysis to be performed at screening , prior to administration ACE-011/placebo (Days 1 and 85) and termination visit Assessments (Central lab): pH, specific gravity, protein, glucose, ketones, blood, leukocyte esterase, and nitrite Microscopy required only to follow-up abnormal findings
[11] Screening- any concomitant treatments taken 28 days prior to day of randomization
[12] Females of child bearing potential- collection of menstrual cycle information (date of last menstruation, date of menstrual period onset and duration of menstruation)
[13] A CT/MRI scan acquired within 4 weeks of initiating the current chemotherapy regimen may be used for the screening assessment Head, neck, thoracic, abdominal, and pelvic scans are to be acquired One measurable or non measurable lesion per RECIST is to be identified by the scan Brain (head) CT only to be done to rule out CNS metastases in patients with suspicious clinical symptoms or prior history of CNS metastases
[14] Follow-up CT/MRI scans may be acquired +/- 5 day window for either Day 64 or Day 113 Additional CT/MRI scans acquired during the post treatment follow up period will be performed at the discretion of the investigator
[15] Hip and lumbar spine BMD assessment
[16] Bone scans will be acquired within 4 weeks of initiating the current chemotherapy regimen for those patients who have bone metastases identified Follow up bones scans may be acquired +/- 5 day window for day 64, day 113, and Day 281
[17] Patients who discontinue ACE-011/placebo due to ACE-011/placebo related toxicities (section 9 5 2 1) will enter the treatment follow-up period
[18] Chemotherapy regimens scheduled will be administered to the patient as per standard of care Continuation of the same chemotherapy regimen may be done through the follow up period of the study at the discretion of the investigator
[19] Patients who terminate treatment early due to disease progression and/or begin a new chemotherapy regimen will complete the day 281/Terminiation visit procedures and be followed for survival only (7 months from the last dose of ACE-011/placebo) If a patient withdraws from the study (section 8 4), the Day 281/Termination visit procedures are to be completed and the patient will be followed for survival If withdrawal reason is due to ESA administration, the patient should enter the treatment follow up period

8. SELECTION AND WITHDRAWL OF PATIENTS

8.1. Number of Patients

The study will include three dose levels of ACE-011 at 0.1 mg/kg, 0.3 mg/kg and 0.5 mg/kg and a placebo group. A total of 105 will be treated with ACE-011 or placebo.

8.2. Entry Criteria

8.2.1. Patient Inclusion Criteria

1. Women ≥ 18 years of age.
2. Histologically confirmed diagnosis of breast cancer documented by cytology or biopsy.
3. Evidence of metastatic breast cancer with a minimum of one measurable lesion or non measurable lesion per RECIST criteria (Appendix 1).
4. Receiving a multicycle myelosuppressive chemotherapy regimen including one of the following: anthracycline, taxane, gemcitabine or vinorelbine.
5. Planned treatment with the same myelosuppressive chemotherapy regimen for a minimum of 9 weeks after Day 1 of ACE-011/placebo administration.
6. Hemoglobin value between ≥ 6.5 to < 11.0 g/dL (≥ 65 to < 110 g/L).
7. ≥ 30 days must have elapsed (from Day 1) since previous treatment with erythropoiesis-stimulating agent (including concurrent treatment with IV iron) for chemotherapy induced anemia.
8. ≥ 7 days have elapsed (from Day 1) since the last RBC blood transfusion and receipt of ≤ 2 units of blood in the past 30 days.
9. ECOG Performance status of 0 – 2 (Appendix 2).
10. Adequate renal (creatinine ≤ 1.5 x ULN or creatinine clearance ≥ 40 mL/min) and hepatic function (bilirubin ≤ 1.5 x ULN; AST/ALT ≤ 2.5 x ULN).
11. Females of childbearing potential participating in the study are to use adequate birth control measures (i.e. oral contraceptives or two forms of barrier method birth control) during study participation. Females of childbearing potential must have a negative serum pregnancy test within 3 days to ACE-011/placebo administration (Day 1). A female of childbearing potential is a sexually mature woman who has not undergone a hysterectomy or who has not been postmenopausal for at least 24 consecutive months (i.e. who has had menses at some time in the preceding 24 months).
12. Life expectancy of ≥ 6 months.
13. Is willing to adhere to the study visit schedule, understand and comply with all protocol requirements.

14. Understand and sign a written informed consent.

8.2.2. Patient Exclusion Criteria

1. At the time of screening patients who have any grade ≥ 3 toxicity (as per NCI CTCAE v3.0 (Appendix 3)) (except for the following disease related toxicities: hematological events- anemia, thrombocytopenia or neutropenia or non hematological events- nausea, vomiting, fatigue, or muscle or bone/joint pain).
2. Prior radiation therapy to > 20% of the whole skeleton. Use of palliative radiation if the area being treated is <15% of body surface area with no pelvic radiation or no more than 10% of the bone marrow reserve is radiated is permitted during patient participation in the study, at the discretion of the investigator.
3. Have received more than 5 prior chemotherapy treatment regimens for metastatic breast cancer.
4. Concurrent use of bevacizumab with the chemotherapy regimen during study participation.
5. Clinically significant pulmonary, endocrine, neurologic, gastrointestinal, hepatic or genitourinary disease unrelated to underlying hematologic disorder.
6. Patients with heart failure as classified by the New York Heart Association (NYHA) classification of 3 or higher (Appendix 4).
7. History of thrombosis, deep vein thrombosis (DVT), pulmonary emboli, or embolic stroke.
8. Untreated CNS metastases (exception: CNS metastases treated with whole brain radiotherapy > 6 months prior to randomization).
9. Diagnosis of a myeloid malignancy or known history of myelodysplasia.
10. History of second malignancy within 5 years (except excised and cured basal cell carcinoma, squamous cell carcinoma of the skin or cervical carcinoma in situ).
11. Patients with a recent history (within 14 days of Day 1) of administration of systemic (IV or oral) antibiotics. Patients should be free of infection at least 14 days after the last dose of systemic antibiotic treatment (from Day 1).
12. Uncontrolled hypertension. If hypertension is considered clinically stable, systolic blood pressure (BP) must be < 150 mmHg and diastolic BP must be < 100 mmHg.
13. Known history of hepatitis C antibody (HCV), hepatitis B surface antigen (HBsAg and HB core Ab), or human immunodeficiency virus (HIV) antibody.
14. Deficiency in iron (serum ferritin< 100 ng/mL (< 224.7 pmol/L)), vitamin B_{12}, or folate.
15. History of anemia as a result of inherited hemoglobinopathy such as sickle cell anemia or thalassemia.
16. History of autoimmune or hereditary hemolysis or gastrointestinal bleeding.
17. Patient received treatment with another investigational drug or device within 1 month prior to Day 1, or if the half life of the previous product is known, within 5 times the half life prior to dosing, whichever may be longer.

18. Pregnant or lactating females.
19. History of severe allergic or anaphylactic reactions or hypersensitivity to recombinant proteins or excipients in the investigational product.

8.3. Patient Withdrawal Criteria

Patients will be informed that they have the right to withdraw from the study at any time for any reason without prejudice to their medical care.

Patients must be withdrawn from the study for any of the following reasons:

- Patient request
- Patient is unwilling or unable to comply with the protocol
- ESA administration during the treatment period
- Medical reason, such as cancer or treatment related toxicity, or at the discretion of the Investigator and/or the Medical Monitor(s)

The reasons for withdrawal must be recorded in the patient's case report form (CRF). The Investigator must notify the Medical Monitor and the contract research organization (CRO) immediately when a patient has been discontinued/withdrawn due to an AE. All patients who are withdrawn from the study should complete the tests and evaluations scheduled for Day 281/Termination at the time of withdrawal. Discontinued/withdrawn patients may be followed for survival only 7 months from the last dose of ACE-011/placebo. Patients who withdraw due to ESA administration during the treatment period may enter the treatment follow-up period.

9. TREATMENT OF PATIENTS

9.1. Concomitant Medications

9.1.1. General Concomitant Medication Usage

During screening, and during the study, patients may take only stable doses of medications for chronic conditions that are not specifically excluded by the protocol (see Sections 8.2.1 Patient Inclusion Criteria and 8.2.2 Patient Exclusion Criteria). The Medical Monitor should be consulted by the Investigator if there are any concerns about what constitutes a stable dose or what is a chronic condition.

9.1.2. Concomitant treatment for anemia

Concurrent therapy with a new prescription medication related to treatment of anemia during the course of the study must first be discussed with the Investigator and Medical Monitor prior to administration, unless appropriate medical care necessitates that medication should begin before the Medical Monitor can be consulted.

9.1.2.1. Iron supplementation

Iron supplementation within 7 days of Day 1 (besides an oral general multivitamin with iron) will not be allowed. If a patient becomes iron replete during study treatment (ferritin < 100 ng/mL (< 224.7 pmol/L) or transferrin < 20%), treatment with iron supplementation is at the discretion of the investigator.

9.1.2.2. Erythropoiesis-stimulating agents (ESAs)

If concurrent treatment with erythropoiesis-stimulating agent is required in the opinion of the investigator, the erythropoiesis-stimulating agent label instructions are to be followed. The patient is to discontinue further treatment with ACE-011/placebo and enter the treatment follow up period of the study. See Table 7 for Schedule of Events.

9.1.2.3. RBC Transfusions

Concurrent treatment for chemotherapy induced anemia with blood transfusions is recommended when hemoglobin value is < 8 g/dL or at investigator discretion if the hemoglobin value is above 8 g/dL and associated with anemia symptoms such as hemodynamic or pulmonary compromise requiring treatment. If a transfusion is given to a patient during the treatment period, ACE-011/placebo should be administered no sooner than 7 days from the date of the transfusion. After the transfusion, the hemoglobin value should be assessed no later than 7 days from the date of the transfusion. On the day of ACE-011/placebo administration, the hemoglobin value and blood pressure will be assessed. See Figure 1 for ACE-011/placebo dose modification rules.

9.2. Chemotherapy

Chemotherapy treatment for metastatic breast cancer is to be given as per standard of care at the site. ACE-011/placebo administration should be given prior to chemotherapy administration on ACE-011/placebo administration days. Continuation of treatment with the same chemotherapy regimen through the treatment or post-treatment follow-up period of the study is at the discretion of the investigator.

9.3. Treatment Compliance

Each dose of ACE-011/placebo will be administered as a SC injection at the clinical site by the study staff and will be documented in the study record. Monitoring for patient compliance with the treatment regimen is therefore unnecessary.

9.4. Randomization

Patients will be randomized to limit scientific bias within the study. Randomization assignments will be generated through a computerized system, provided by an Interactive Web/Voice Response System (IWRS/IVRS). Patients will be stratified according to frequency of the planned chemotherapy regimen (weekly chemotherapy vs. less frequent chemotherapy administration).

9.4.1. Blinding

The Investigators, patients, and sponsor or representative will remain blinded to the treatment arm assignment of each patient. The unblinded statistician will remain uninvolved in the study conduct until database lock and unblinding of the data has occurred.

Safety parameters will be reviewed on an ongoing basis throughout the study to review blinded adverse events, serious adverse events, laboratory listings, and vital signs. If substantial toxicity trends are observed in one treatment group versus another, the DMC may request unblinding the treatment code assignment schema.

9.4.2. Unblinding

In the event of a medical emergency for an individual patient in which knowledge of the study medication is critical to the patient's medical management, the investigator may break the blind for that patient. However, prior to breaking the blind, every effort must be made by the investigator to first discuss the need to break the blind with the sponsor or sponsor representative. Further, it must be determined by the investigator that breaking the treatment blind is necessary information for the medical management of that patient. If the blind is broken, the patient must withdraw from the study by completing the Day 281/Termination visit procedures and be followed for survival only.

9.5. Treatments Administered

9.5.1. Selection of Doses in the Study

Single dose administration of ACE-011 up to 3.0 mg/kg was previously demonstrated to be safe and to have durable effects on markers of bone formation and resorption as well as red blood cells following a single IV administration. A dose-limiting pharmacodynamic effect of ACE-011 with increases in hemoglobin and hematocrit and RBCs was established in healthy post-menopausal women at the 1.0 mg/kg dose level following multiple SC administration. This study will confirm the safety in following repeated SC administration of ACE-011 and further evaluate the potential efficacy in patients with chemotherapy induced anemia. The safety and preliminary efficacy of ACE-011 following multiple doses up to 0.5 mg/kg will be assessed using a parallel randomization design.

9.5.2. Selection and Timing of Dosing for Each Patient

Patients will be enrolled and receive their assigned dose of ACE-011/placebo every 28 days (i.e. on Days 1, 29, 57, and 85). Patients will be receiving chemotherapy, concurrently for metastatic breast cancer. After completion of the treatment, patients will return to the site monthly for 2 follow-up assessments (i.e., Days 113 and 141). Subsequently, the patient will return to the site for 4 post treatment follow-up assessments (i.e. Days 169, 197, 225, and 253). The patient will return to the site for a termination visit approximately 1 month after the post treatment follow-up period (Day 281). Patients will be discontinued from the ACE-011/placebo treatment for unacceptable toxicity, if the chemotherapy regimen is discontinued or for disease progression that requires the initiation of another chemotherapy treatment.

9.5.2.1. ACE-011 Treatment Related Toxicity

Patients with grade ≥3 toxicity related to ACE-011 will be discontinued from ACE-011/placebo treatment except for the following. Patients with grade 3 hypertension that decreases to grade ≤ 1 within 7 days after treatment with anti-hypertensive therapy may continue on ACE-011/placebo treatment. Dose reduction rules will apply at the subsequent scheduled dosing day(s).

Patients with a hemoglobin level above the upper limit of normal at any time during the study after Day 1 will be discontinued.

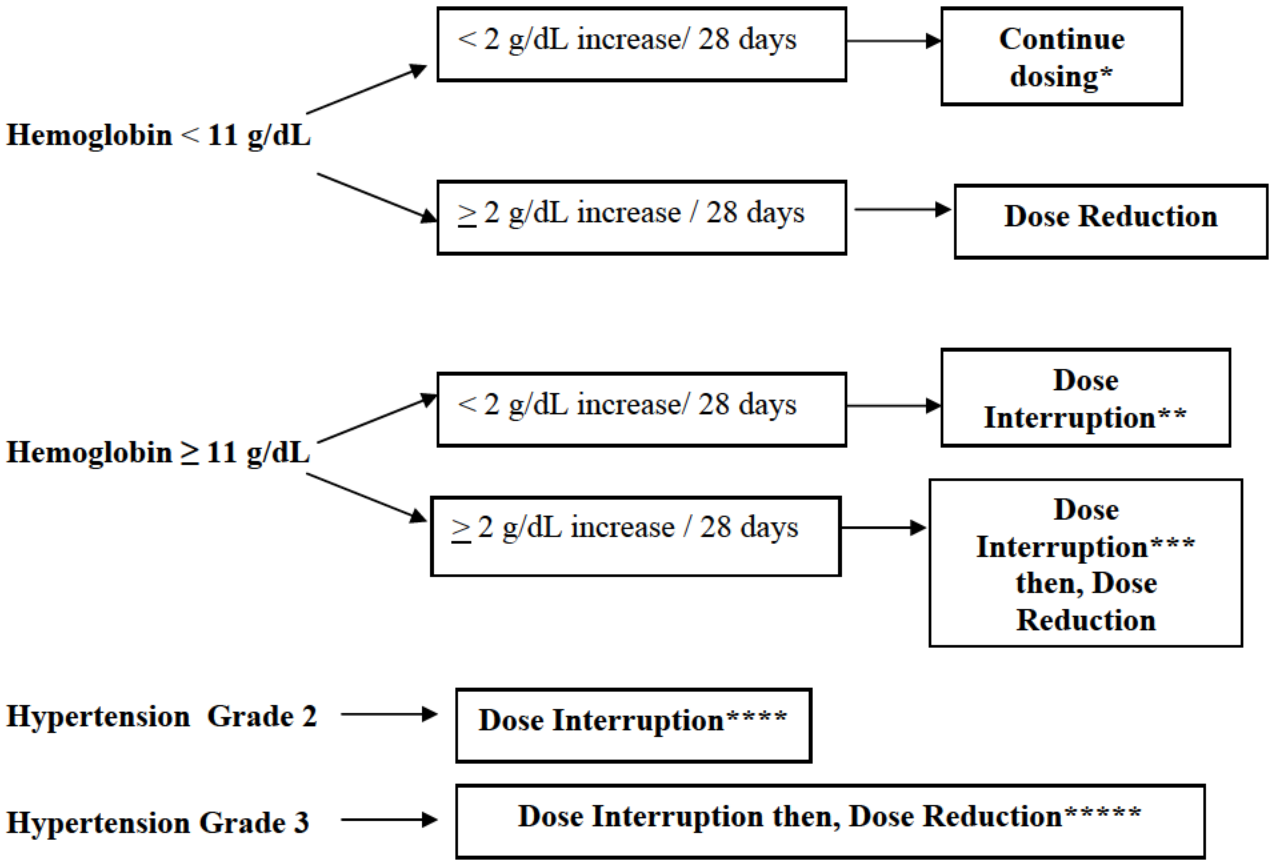
Patients who discontinue treatment with ACE-011/placebo will enter the treatment follow-up period.

9.5.2.2. ACE-011 Dose Modification Rules

Throughout the study blood pressure and hemoglobin values will be evaluated for patient safety. The local hemoglobin values and blood pressure values will be evaluated on each ACE-011/placebo dosing day. The hemoglobin values from the previous 28 days between each

ACE-011/placebo dosing day and blood pressure will be reviewed and evaluated for dose modifications. The parameters below outline rules for dosing modifications (Figure 1). Table 8 outlines the dose reduction levels of ACE-011.

Figure 1: ACE-011/placebo Dose Modification Rules (after initial dose of ACE-011/placebo)



* Patients who have received a blood transfusion in the past 28 days should continue with dosing at the same ACE-011/placebo dosing level if the transfusion was given no sooner than 7 days from the dosing day and the hemoglobin level is < 11 g/dL and hypertension ≤ Grade 1 on the day of dosing

** ACE-011/placebo should be held until the following scheduled treatment visit

*** ACE-011/placebo should be held and at the following scheduled treatment visit, a dose reduction of ACE-011/placebo will be administered based on the evaluation of the hemoglobin and blood pressure at that time

**** ACE-011/placebo should be held until resolution to ≤ Grade 1 (NCI CTCAE v3 0) and may then be resumed at the following scheduled treatment visit.

***** ACE-011/placebo should be held until resolution to ≤Grade 1 (NCI CTCAE v3 0) within 7 days after treatment with anti- hypertensive therapy and may continue on a reduced dose of ACE-011/placebo treatment for the subsequent scheduled dosing day

Table 8: ACE-011/Placebo Dose Reduction Levels

When required per the dose modification rules above (Figure 1), ACE-011/placebo dose(s) should be reduced as follows for dose 2 (Day 29), dose 3 (Day 57) and/or dose 4 (Day 85):

Dose	Dose reduction #1	Dose Reduction #2	Dose Reduction #3
Placebo	N/A	N/A	N/A
0.1 mg/kg	0.05 mg/kg	0.03 mg/kg	0.01 mg/kg
0.3 mg/kg	0.1 mg/kg	0.05 mg/kg	0.03 mg/kg
0.5 mg/kg	0.3 mg/kg	0.1 mg/kg	0.05 mg/kg

Blood pressure and hemoglobin values must be evaluated at each dosing day for consideration of administration of ACE-011/placebo.

The dose level for the initial dose reduction will be maintained unless subsequent dose reductions need to be administered, upon evaluation of the dose modifications listed above. A patient may have up to 3 dose reductions in the study.

Dose reduction steps and the administration of the reduced dose(s) will be conducted in a manner that will preserve the blinded status of the original treatment group of each patient. Patients in the placebo group who are designated by the investigator to undergo a dose reduction will continue to receive placebo.

10. STUDY PROCEDURES AND SCHEDULE

10.1.1. Written Informed consent

Patients will be required to sign an Institutional Review Board/Institutional Ethics Committee (IRB/IEC) approved informed consent form (ICF) prior to any study related procedures, including screening evaluations.

Screen failure information will be maintained, including but not limited to, reason for failure.

10.1.2. Screening (Within 14 days of dosing)

The following will be collected within 14 days prior to the initial dosing Day 1:

- Informed consent
- Assessment of inclusion/exclusion criteria
- Complete Medical History and Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), temperature (°C) and height.
- FACT-Fatigue survey
- 12 lead Electrocardiogram (ECG)- including measurement of the QT interval
- Serum iron, total iron binding capacity (TIBC), transferrin and serum ferritin
- Vitamin B12 and serum folate levels
- Coagulation
- Serum chemistry
- Hematology
- LH, FSH, testosterone, progesterone, and estradiol
- Back-up blood sample to be drawn for future testing
- Serum Pregnancy Test for females of childbearing potential- to be assessed within 3 days of Day 1.
- Urinalysis
- Evaluation of transfusion frequency (including history of transfusion up to 8 weeks prior to Day 1
- Documentation of concomitant medications (any treatments taken 28 days prior to Day 1
- Females of childbearing potential- collection of menstrual cycle information (date of last menstruation, date of menstrual period onset and duration of menstruation)

- CT/MRI scan: Head, neck, thoracic, abdominal, and pelvic scans are to be acquired. A CT/MRI scan acquired within 4 weeks of initiating the current chemotherapy regimen may be used for the screening assessment. One measurable or non-measurable lesion per RECIST is to be identified by the scan.
    - Brain (head) CT only to be done to rule out CNS metastases in patients with suspicious clinical symptoms or prior history of CNS metastases.
- Dual-energy X-ray absorptiometry (DXA) of lumbar spine and hip
- Bone scans (only for patients with bone metastases) acquired within 4 weeks of the initiation of the current chemotherapy regimen may be used for the screening assessment.

10.1.3. Initial Dosing: Day 1

Patients will be dosed on Day 1. Results from screening evaluations must be reviewed prior to randomization to confirm patient eligibility. A patient will be considered randomized once a multi-digit patient identification number has been assigned to the patient through the IVRS. After assignment of the patient identification number, ACE-011/placebo administration may begin.

Chemotherapy regimen will be administered per standard of care at the site. The following tests must be performed prior to ACE-011/placebo administration:

- Confirm eligibility of patient by inclusion/exclusion criteria. The local hemoglobin value should be drawn prior to ACE-011/placebo administration.
- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- FACT-Fatigue survey
- 12 lead ECG- including measurement of the QT interval
- Serum iron, total iron binding capacity, transferrin and serum ferritin
- Coagulation
- Chemistry
- Hematology
- Serum erythropoietin marker
- Peripheral blood smear
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test

- Bone biomarkers- osteocalcin (OC), procollagen type 1 N-terminal propeptide (P1NP), serum bone specific alkaline phosphatase (BSAP), C-terminal type 1 collagen telopeptide (CTX) and Serum tartrate-resistant acid phosphatase isoform-5b (TRACP-5b), and urine N-telopeptide (uNTX).
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions the collection of serum and urine for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Females of childbearing potential- Urine (dipstick) pregnancy test to be collected and resulted prior to ACE-011/placebo dosing
- Urinalysis
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events (after study drug administration)
- Females of childbearing potential- collection of menstrual cycle information (date of last menstruation from day 1, date of menstrual period onset and duration of menstruation)

Patients will be administered ACE-011/placebo before the chemotherapy regimen begins during the study visits.

After ACE-011/placebo and chemotherapy are administered, the patient may leave the clinic, based on the clinical judgment of the staff.

10.1.4. ACE-011/placebo administration : Days 29, 57, and 85 (± 3 days)

Chemotherapy regimen will be administered per standard of care at the site. ACE-011/placebo will be administered approximately every 28 days. The following procedures must be performed prior to ACE-011/placebo administration:

- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- FACT-Fatigue survey
- 12 lead ECG -including measurement of the QT interval (Day 29 only)
- Serum iron, total iron binding capacity, transferrin and serum ferritin
- Coagulation
- Serum chemistry

- Hematology
- Serum erythropoietin marker
- Peripheral blood sample
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test
- Bone biomarkers- OC, BSAP, PINP, CTX, TRACP-5b, and uNTX
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions the collection of serum and urine for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Females of childbearing potential -Urine (dipstick) pregnancy test
- Urinalysis (Day 85 only)
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events
- Females of childbearing potential- collection of menstrual cycle information (date of last menstruation, date of menstrual period onset and duration of menstruation)

Hemoglobin (HgB) and Hematocrit values (Hct) and blood pressure (BP) measurement must be assessed prior to ACE-011/placebo dose administration by review of local laboratory values.

10.1.5. Additional visits during Treatment Period: Days 8, 15, 22, 36 (± 1 day), 43, 64 and 71 (± 2 days)

Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- 12 lead ECG -including measurement of the QT interval (Day 15 only)
- Serum folate (Day 36 only)
- Coagulation
- Serum chemistry
- Hematology
- Back-up blood sample to be drawn for future testing

- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events
- CT/MRI scan (Day 64 only, +/- 5 day window). Same modality as acquired for baseline should be used for follow up scans. Tumor response assessments to be evaluated by the investigator.
- Bone scan (Day 64 only, +/- 5 day window). Follow-up scan to be acquired if performed at baseline.

If patient terminates the treatment early, prior to receiving a fourth dose of ACE-011/placebo, the patient will enter the treatment period follow up. However, if patient terminates ACE-011/placebo early and begins a new chemotherapy regimen, the patient should complete the Day 281/Termination visit procedures and enter the post treatment follow up period.

10.1.6. Treatment Period Follow-up visits : Days 113 and 141 (± 7 days)

Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- 12 lead ECG-including measurement of the QT interval (Day 113 only)
- Serum folate (Day 113 only)
- Coagulation
- Serum chemistry
- Hematology
- Serum erythropoietin marker (Day 141 only)
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test
- Bone biomarkers- OC, BSAP, PINP, CTX, TRACP-5b, and uNTX
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions the collection of serum and urine for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Evaluation of transfusion frequency

- Documentation of concomitant medications
- Evaluation of adverse events
- Females of childbearing potential- collection of menstrual cycle information (date of last menstruation, date of menstrual period onset and duration of menstruation)
- CT/MRI scan (Day 113 only, +/- 5 day window). Same modality as acquired for baseline should be used for follow up scans. Tumor response assessments to be evaluated by the investigator.
- DXA of lumbar spine and hip (Day 113 only)
- Bone scan (Day 113 only, +/- 5 day window). Follow-up scan to be acquired if performed at baseline.

10.1.7. Post Treatment Period Follow-up visits : Days 169, 197, 225, and 253 (± 7 days)

If a CT/MRI and bone scan (if applicable) is performed as per standard of care during the post treatment period follow up, results may be collected within the eCRF to capture the status of the disease. Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Hematology
- Back-up blood sample to be drawn for future testing
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events

10.1.8. Termination Visit: Day 281 (± 7 days)

Day 281 is the final visit. If a patient terminates the study early, the Day 281 procedures should be followed. Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- FACT-Fatigue survey
- 12 lead ECG -including measurement of the QT interval
- Serum iron, total iron binding capacity, transferrin and serum ferritin
- Serum folate
- Coagulation

- Serum chemistry
- Hematology
- Serum erythropoietin marker
- Peripheral blood smear
- Anti-drug antibody test
- Bone biomarkers- OC, BSAP, PINP, CTX, TRACP-5b, and uNTX
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions the collection of serum and urine for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Females of childbearing potential -Urine (dipstick) pregnancy test
- Urinalysis
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events
- DXA of lumbar spine and hip
- Bone scan (Day 281, +/- 5 day window). Follow-up scan to be acquired if performed at baseline.

If a patient has a positive anti-drug antibody result at Day 281/Termination visit, the patient may be asked to return to the clinical site for additional monthly anti-drug antibody testing, 12 months after their last dose.

10.2. Discontinuation of study

The Sponsor may terminate this study, after consultation with the Investigator, at any time for safety or administrative reasons. The Sponsor may terminate the study if the occurrence of serious adverse events (SAEs) or other findings suggest unacceptable risk to the health of the patients.

11. INVESTIGATIONAL PRODUCT MATERIALS AND MANAGEMENT

11.1. Reference Product

The placebo (control agent) to be used in this study will be sterile normal saline (0.9% Sodium Chloride for Injection) administered as a SC injection. Sterile, normal saline will be supplied to the investigational site's pharmacist. The manufacturer's directions for placebo storage and handling are to be followed, as are standard clinical practices for ensuring sterility of the placebo.

11.2. Investigational Product Packaging and Labeling

ACE-011 will be supplied in 2 mL clear glass vials with gray stoppers and red flip-top seals that contain 1 mL of ACE-011. The drug product consists of ACE-011 in PBS at a nominal concentration of approximately 50 mg/mL.

11.3. Investigational Product Storage

ACE-011 is recommended to be stored at ≤ -65℃.

11.4. Investigational Product Preparation

See Investigational Product Preparation Manual.

11.5. Administration

ACE-011/placebo will be administered by subcutaneous injection (SC). Subcutaneous injections will be given in the upper arm and/or thigh. Please refer to Investigational Product Handling and Administration document for further information.

Dose modifications will be determined by the investigator after reference to the dose modification rules listed in Figure 1. In the case of a dose interruption or dose reduction of ACE-011, the investigator or designee will notify the pharmacy staff, who is unblinded, of the treatment decision for appropriate preparation of the study drug to maintain the blind of the study.

11.6. Drug Accountability

Accountability for ACE-011/placebo is the responsibility of the Investigator. Investigational clinical supplies must be received by a designated person at the clinical site and kept in a secured location. The investigational site must maintain accurate records demonstrating dates and amounts of ACE-011/placebo received, to whom it was dispensed (patient-by-patient accounting), and accounts of any ACE-011/placebo accidentally or deliberately destroyed or returned. Unless otherwise notified, all vials of ACE-011/placebo, both used and unused, must be saved for drug accountability. The used vials may be discarded, per the institution's standard

practice, after drug accountability has been completed by the monitor. The Investigator must return all unused vials of ACE-011 to the Sponsor at the end of the study, or the study drug may be destroyed at the clinical site with the permission of the Sponsor. For either scenario, the outcome must be documented on the drug accountability log. The Sponsor will provide direction for the outcome of all unused vials.

11.7. Investigational Product Handling and Disposal

Please refer to the Study Reference Guide, provided under separate cover, for detailed drug handling, administration, and storage instructions.

12. ASSESSMENT OF EFFICACY

Efficacy measurements will include assessments for hematopoietic response which is defined as an increase in hemoglobin of ≥ 0.5 g/dL, ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs, at each dose level. The hematopoietic endpoints will be determined by the monitoring of hematologic laboratory values and RBC transfusion and/or ESA administration. Secondary endpoints associated with assessment of disease progression will be evaluated by CT/MRI scan and bone mineral density by DXA scans, respectively. Bone biomarkers and incidence of skeletal related events (SREs) will also be evaluated for all patients. SREs are defined as pathologic fracture, radiation therapy to bone, surgery to bone, or spinal cord compression. Quality of life assessment (FACT-Fatigue) will be evaluated for each patient.

13. ASSESSMENT OF SAFETY

13.1. Safety Parameters

13.1.1. Adverse Events

All patients will be assessed for safety by monitoring adverse events, clinical laboratory tests, vital signs and physical examinations. AE information will be collected throughout the study. See Section 14 Adverse Events for a full description of AE definitions, AE and SAE reporting procedures, and emergency procedures.

13.1.2. Clinical Laboratory Tests

The following laboratory safety assessments will be drawn at the investigational site's local laboratory according to the laboratory collection recommendations. The samples will then be sent to the central lab for value reporting. Please refer to Lab Reference Manual for further information. Hemoglobin and hematocrit lab tests will be drawn and evaluated by the local laboratory at each visit day throughout the study. The local hemoglobin laboratory value will be used in the evaluation of ACE-011/placebo dose modifications. The sponsor may request additional safety tests be performed based on ongoing data review during the study.

- Iron studies: Serum iron, total iron binding capacity (TIBC), transferrin and serum ferritin on specified days.
- Nutritional tests: Serum folate and vitamin B12
- Coagulation (central lab only): prothrombin time (PT), partial thromboplastin time (PTT), and fibrinogen
- Serum chemistry (central lab only): AST, ALT, lactate dehydrogenase (LDH), total bilirubin, indirect bilirubin, alkaline phosphatase, total protein, albumin, sodium, potassium, chloride, carbon dioxide, calcium, phosphorus, blood urea nitrogen (BUN), creatinine, uric acid, glucose, amylase, and lipase.
- Hematology: complete blood count (CBC) with differential and platelet. CBC includes red blood cells (RBC), white blood cells (WBC), hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), and erythropoietin levels. Reticulocyte percent and peripheral blood smear will be collected on specified days.
- Bone biomarkers: (serum) osteocalcin (OC), procollagen type 1 N-terminal propeptide (P1NP), serum bone specific alkaline phosphatase (BSAP), C-terminal type 1 collagen telopeptide (CTX), tartrate-resistant acid phosphatase isoform-5b (TRACP-5b), and urine N-telopeptide (uNTX).
- Pregnancy test for females of child bearing potential
- Urinalysis: including determination of pH, specific gravity, protein, glucose, ketones, blood, leukocyte esterase and nitrite. Microscopy required only to follow-up abnormal findings.

13.1.3. Other Safety Assessments

- Physical examinations
- Vital signs: weight (kg), heart rate (beats/min), seated blood pressure (mmHg) and oral temperature (°C). Height will be collected at screening only.
- FACT-Fatigue survey
- 12-lead ECG -including measurement of the QT interval
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test
- CT/MRI scan
- DXA
- Bone scans (for those patients with baseline bone metastases)

13.2. Pharmacokinetics

See Appendix 5 for further details regarding PK sampling.

14. ADVERSE EVENT

Adverse Event

An adverse event (AE) is any untoward medical occurrence in a patient administered a pharmaceutical product, which does not necessarily have a causal relationship with the treatment. An AE can be any unfavorable and unintended sign (e.g., including an abnormal laboratory finding), symptom, or disease temporally associated with the use of the study drug, whether or not it is considered to be study drug related. This includes any newly occurring event or previous condition that has increased in severity or frequency since the administration of study drug (treatment-emergent).

Serious Adverse Event

A serious adverse event (SAE) is any AE, occurring at any dose level and regardless of causality that:

- Results in death.
- Is life-threatening: Life-threatening means that the patient was at immediate risk of death from the reaction as it occurred, (i.e., it does not include a reaction which hypothetically might have caused death had it occurred in a more severe form).
- Requires inpatient hospitalization or prolongation of existing hospitalization.
- Results in persistent or significant disability/incapacity.
- Is a congenital anomaly/birth defect.
- Is an important medical event: an important medical event is an event that may not result in death, be life-threatening, or require hospitalization but may be considered an SAE when, based upon appropriate medical judgment, it may jeopardize the patient and may require medical or surgical intervention to prevent one of the outcomes listed in the definitions for SAEs. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.
- A hospitalization for an elective procedure will not be considered a SAE.

Unexpected Adverse Event

An unexpected AE is an AE that is not reported in the Investigator Brochure or in the case of an AE already described, the severity of which is not described in the Investigator Brochure.

14.1. Adverse Event Classification

The following classifications should be used when evaluating the relationship of AEs and SAEs to the investigational drug.

None:	No relationship between the experience and the administration of study drug; related to other etiologies such as concomitant medications or patient's clinical state.
Unlikely:	The current state of knowledge indicates that a relationship is unlikely.
Possibly:	A reaction that follows a plausible temporal sequence from administration of the study drug and follows a known response pattern to the suspected study drug. The reaction might have been produced by the patient's clinical state or other modes of therapy administered to the patient.
Probably:	A reaction that follows a plausible temporal sequence from administration of the study drug and follows a known response pattern to the suspected study drug. The reaction cannot be reasonably explained by the known characteristics of the patient's clinical state or other modes of therapy administered to the patient.
Definitely:	A reaction that follows a plausible temporal sequence from administration of the study drug and follows a known response pattern to the suspected study drug and can be confirmed with a positive re-challenge test or supporting laboratory data.

Severity of AEs will be graded by the investigator using the National Cancer Institute (NCI; US) Common Terminology Criteria for Adverse Events (CTCAE) version 3.0 guidelines (Appendix 3).

14.2. Recording Adverse Events

Patients will be evaluated and questioned generally for AEs during the course of the study, starting from baseline (Day 1). The Medical History CRF should be comprehensive of patient's medical history up to Day 1. All AEs occurring after ACE-011/placebo administration until the Day 281/Termination visit are to be documented on AE CRF. The specific disease or syndrome rather than individual associated signs and symptoms should be identified by the investigator and recorded on the CRF.

Serious Adverse Events (SAEs)

For SAEs, a SAE form must be completed with as much information as possible and submitted in the time frame described in Section 14.3.

When new significant information is obtained as well as when the outcome of an event is known, the investigator should record the information on a new SAE form. If the patient was hospitalized, a copy of the discharge summary must be included as part of the patient medical file. In all instances, the investigator should follow up with patients until the outcome of the SAE is known.

14.3. Reporting Adverse Events

All AEs spontaneously reported by the patient and/or in response to an open question from study personnel or revealed by observation, physical examination or other diagnostic procedures will be recorded on the AE CRF. Any clinically relevant change in laboratory assessments or other clinical findings is considered an AE and must be recorded on the AE CRF. Clinically significant laboratory abnormalities are those that are identified as such by the investigator and/or those that require intervention. All AEs are to be followed until the event resolves or the clinical course is stabilized.

Serious Adverse Events

For SAEs, a SAE form must be completed with as much information as possible and submitted in the time frame described below.

All SAEs that occur during the course of the study (from the signing of the ICF until the last study visit) must be reported by the Investigator to PPD and Sponsor by faxing the SAE form within 24 hours from the point in time when the Investigator becomes aware of the SAE. All SAEs and deaths must be reported whether or not considered causally related to the study drug.

SAE Reporting:

PPD Medical Monitor
PPD Inc.


If there are serious, unexpected AEs associated with the use of the study drug, the Sponsor will notify the appropriate regulatory agency(ies) and all appropriate parties on an expedited basis. It is the responsibility of the Investigator to promptly notify the Institutional Review Board (IRB) of all unexpected SAEs involving risk to human patients.

14.4. Emergency Sponsor Contact

In the event of a medical emergency (i.e., an event that requires immediate attention regarding the treatment of a patient, operation of the clinical study, and/or the use of investigational drug), investigational site personnel must immediately contact the Medical Monitor.

15. STATISTICS

Statistics Analysis Overview

The study will be considered complete with regard to the primary endpoint once all eligible subjects have completed up to 2 months after the last dose of ACE-011. Patients will be randomized to one of three ACE-011 treatment groups (0.1, 0.3 and 0.5 mg/kg) or placebo with a ratio of 2:2:2:1. Patients will be stratified according to administration of either single agent weekly chemotherapy or less frequent chemotherapy regimens.

15.1. Determination of Sample Size

With an anticipated dropout rate of 20%, a sample size of 30 patients per dose level is estimated to achieve at least 80% power with 2 sided significance level of 0.05 to differentiate targeted response rate of p1=0.45 from null hypothesis p0=0.20. An additional 15 patients will be enrolled to the placebo group for safety comparison. A total of 105 patients will be randomized to three ACE-011 dose groups (0.1, 0.3, and 0.5 mg/kg) and placebo with a ratio of 2:2:2:1.

15.2. Analysis Populations

For this study, the following populations will be defined and used in the analysis and presentation of the data.

Modified Intent-to-Treat (MITT) population: The MITT population is defined as all patients randomized who received at least one dose of study drug.

Per-Protocol (PP) population: The PP population is defined as all patients randomized who have confirmed chemotherapy induced anemia, received at least one dose of ACE-011 and on study (treatment) for at least 2 months (57 days).

Safety population: The safety population is defined as all patients who received at least one dose of study drug ACE-011 or placebo.

15.3. Background and Demographic Characteristics

Summary statistics (mean, standard deviation, median, minimum and maximum) will be provided for those variables measured on a continuous scale. The frequency distribution (number and percentage) will be provided for those variables measured on a nominal scale.

15.3.1. Patient Disposition

Patient disposition (analysis population allocation, entered, discontinued, along with primary reason for discontinuation) will be summarized using frequency and percent for each dose group. A summary of patients enrolled by site will be provided.

15.3.2. ACE-011

ACE-011 treatment exposure (total dosage received) will be summarized for each dose group as well as for each cycle for each dose level. Listings for dose adjustment will be provided.

15.3.3. Concomitant Therapy

By-patient listings will be provided for all concomitant medications and therapies taken during the study. All concomitant medications will be coded and categorized by the WHO drug coding system. Usage frequency of each coded concomitant medications will be summarized by each dose group.

15.4. Efficacy Evaluation

15.4.1. Primary Efficacy Analysis

The primary efficacy endpoint is the hematopoietic response rate at each dose level. It is defined as the proportion of patients who have a hematopoietic response of an increase in hemoglobin of ≥ 0.5 g/dL for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs. The ACE-011 treatment period is defined as from the time of first ACE-011 dose to up to 2 months after the last dose of ACE-011 treatment. To be considered as a responder, all hemoglobin measurements during a 28 consecutive day period or longer must have an increase of ≥ 0.5 g/dL from baseline. If RBC transfusions and/or ESAs are required, no hemoglobin measurements within 28 days will be used to determine the hemoglobin response. Primary efficacy analysis will be based on PP population. Primary efficacy analysis will also be conducted on MITT population as supportive to the PP based analysis. Patients who do not receive ACE-011 will be excluded from primary efficacy analysis. Patients who discontinue the ACE-011 treatment prematurely will be counted as non-responder in the calculation of response rate.

A 95% confidence interval (CI) for the response rate for each dose level will be provided to assess whether 95% CI of response rate contains null hypothesis response rate, p0=0.20. The response rate and 95% CI will also be provided for patients with chemotherapy on weekly basis versus those with chemotherapy on a less frequent basis for each dose level as well as overall (all three doses levels analyzed together).

An exploratory analysis with the Kaplan Meier approach will be considered to account for those patients who discontinue the ACE-011 treatment prematurely if the drop out rate is greater than 20%. Exploratory analysis of comparing hematopoietic response rates between active and placebo will be performed using Fisher's exact test. Due to the nature of the study (phase 2 dose determination study), no multiplicity adjustment will made for the multiple comparisons.

15.4.2. Secondary Efficacy Analyses

The secondary efficacy endpoints include:

- Proportion of patients achieving an increase in hemoglobin of ≥ 1 g/dl, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for 28 consecutive days during the treatment period and up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs.
- Duration of hematopoietic response (days) defined as the first time hemoglobin increases at least 0.5 g/dL, ≥ 1 g/dl, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL from baseline to the last time there is hemoglobin ≥ 0.5 g/dL, ≥ 1 g/dl, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL increase from baseline. Duration of response is only calculated for a responder and will be at least 28 days. The duration of response will be at least 28 days is only calculated for a patient who meets the primary efficacy endpoint.
- Time to achieve hematopoietic response (days) defined as time from first dose of ACE-011 to the first time a hemoglobin result at least 0.5 g/dL, ≥ 1 g/dl, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL increase from baseline and maintained for a period of at least 28 consecutive days.
- Proportion of patients receiving red blood cell (RBC) transfusions and/or ESAs in each dose group as well as within each cycle of each ACE-011 dose level.
- Objective tumor response rate for each dose level using RECIST criteria.
- Progression-Free Survival is defined as the time from randomization into the study to the first observation of documented disease progression or death due to any cause. If the subject has not progressed or died at the time of final analysis, PFS will be censored at the time of last tumor assessment.

The statistical analysis for binary endpoints will be similar to the primary efficacy analysis. For time to event endpoints, the Kaplan-Meier method will be used to estimate the survival curve and median time to event. These analyses will be performed for each dose group and all ACE-011 groups combined.

15.5. Safety Evaluation

Safety variables will be tabulated and presented for all patients who receive ACE-011. Adverse events will be coded using the Medical Dictionary for Regulatory Activities (MedDRA). Incidence of treatment-emergent adverse events will be presented by system organ class (SOC) and preferred term. Adverse events will also be presented by severity, and relationship to study drug. Change from baseline in clinical laboratory parameters and vital signs will be summarized across time. Shift tables will be presented for selected laboratory parameters. Physical examination results will be presented in listings.

Data from all patients who receive one or more doses of study drug will be included in the safety analyses. To assess clinical safety, adverse events, vital sign measurements and clinical

laboratory information will be summarized by visit. Adverse events and laboratory results will also be summarized by severity grade (NCI CTCAE v3.0). Descriptive statistics will be generated and shift tables provided as appropriate.

15.6. Pharmacokinetic Evaluation

Listing of individual patient serum ACE-011 concentrations, actual blood sampling times and graphs of concentration vs. time will be prepared for each dose group. Summaries of PK parameters will be summarized by dose group. The trough concentrations will be summarized for all patients who provide pharmacokinetic samples by dose group. Exploratory analyses will be performed to correlate trough concentrations with efficacy, bone biomarker data, and safety data, however these analyses will be data-driven and only conducted if warranted by the data.

15.6.1. Anti-drug Antibody Data

The results of anti-drug and neutralizing antibodies will be presented over time. Exploratory analysis will be performed on the potential effect of anti-drug antibody on ACE-011 PK and drug exposure if anti-drug antibody tests are deemed positive.

15.7. Interim Analysis

There is no planned interim analysis for this study.

15.8. Deviation from Original Analysis Plan

Deviations from the statistical analyses outlined in this protocol will be indicated in the statistical analysis plan; any further modifications will be noted in the final clinical study report.

16. DIRECT ACCESS TO SOURCE DATA/DOCUMENTS

16.1. Study Monitoring

The Clinical Monitor will arrange to visit the Investigator sites at regular intervals during the study. The monitoring visits must be conducted according to the applicable ICH and GCP guidelines to ensure protocol adherence, quality of data, drug accountability, compliance with regulatory requirements, and continued adequacy of the investigational site and its facilities. During these visits, CRFs and other data related to the study will be reviewed and any discrepancies or omissions will be resolved. The Clinical Monitor will be given access to study relevant source documents (including medical records) for purposes of source data verification.

During and/or after completion of the study, quality assurance officers named by the Sponsor or the regulatory authorities may wish to perform on-site audits. The Investigator is expected to cooperate with any audit and provide assistance and documentation (including source data) as requested.

16.2. Audits and Inspections

The Investigators and clinical sites will permit trial related monitoring, audits, IRB review, and regulatory inspections as requested by FDA, the Sponsor or designee, including direct access to source data/documents (i.e., original medical records, laboratory reports, hospital documents, progress reports, signed ICFs, etc.) in addition to CRFs.

17. QUALITY CONTROL AND QUALITY ASSURANCE

17.1. Data Quality Control and Quality Assurance

17.1.1. Investigator responsibility

The investigator is responsible for ensuring the study is conducted according to the protocol, Code of Federal Regulations (CFR), GCP, and any other applicable regulatory requirements. These responsibilities outlined in these documents along with the documentation that a signed informed consent must be obtained prior to a patient participation in the study.

17.1.2. Protocol modifications

The investigator may not modify the protocol without agreement from the Sponsor and prior review or approval by the IRB. Any deviations from the protocol should be documented by the investigator or designee.

17.2. Financing and Insurance

Financing and insurance are addressed in a separate document.

17.3. Confidentiality

To maintain patient privacy, all eCRFs, study drug accountability records, study reports and communications will identify the patient by the assigned patient identification number. The Investigator will grant monitor(s) and auditor(s) from the Sponsor or designee and regulatory authority(ies) access to the patient's original medical records for verification of data gathered on the CRFs and to audit the data collection process. The patient's confidentiality will be maintained and will not be made publicly available to the extent permitted by the applicable laws and regulations.

All information regarding the investigational product supplied by the Sponsor to the Investigator is privileged and confidential information. The Investigator agrees to use this information to accomplish the study and will not use it for other purposes without consent from the Sponsor. It is understood that there is an obligation to provide the Sponsor with complete data obtained during the study. The information obtained from the clinical study will be used towards the development of the investigational product and may be disclosed to regulatory authority(ies), other Investigators, corporate partners, or consultants as required.

17.4. Publication Policy

All information concerning ACE-011 is considered confidential and shall remain the sole property of the Sponsor. The Investigator agrees to use this information only in conducting the study and shall not use it for any other purposes without the Sponsor's written approval. The Investigator agrees not to disclose the Sponsor's confidential information to anyone except to

people involved in the study that need such information to assist in conducting the study, and then only on like terms of confidentiality and nonuse.

It is understood by the Investigator that the information developed from this clinical study will be used by the Sponsor in connection with the development of ACE-011, and therefore may be disclosed as required to regulatory agencies. To allow for the use of the information derived from clinical studies, it is understood that there is an obligation to provide the Sponsor with complete test results and all data developed in the study.

No publication or disclosure of study results will be permitted except as specified in a separate, written, agreement between the Sponsor and the Investigator.

17.5. Protocol Amendments

Protocol amendments that impact patient safety change the scope of the investigation, or affect the scientific quality of the study must be approved by the IRB and submitted to the appropriate regulatory authorities before implementation of such modifications to the study.

In the event that the protocol needs to be modified immediately to eliminate an apparent hazard to a patient, the Sponsor will amend and implement the protocol change and subsequently notify the regulatory authorities and/or the IRB, as appropriate.

18. ETHICS

18.1. Institutional Review Board or Independent Ethics Committee

The Investigator must obtain written IRB approval of the protocol, approval for relevant supporting information and all types of patient recruitment and advertisement and ICF prior to starting the study. The IRB will meet all Food and Drug Administration (FDA) requirements governing IRBs (Code of Federal Regulations, Title 21, Part 56).

The Sponsor or designee must approve the ICF submitted to the investigational site's IRB. All patient recruitment and advertisements must be submitted to the Sponsor or designee prior to submission to the IRB, for review.

18.2. Ethical Conduct of the Study

The Sponsor and the Investigator must comply with all instructions, regulations, and agreements in this protocol and in the applicable International Conference on Harmonisation (ICH) and GCP guidelines, and must also conduct the study in accordance with local regulations.

18.3. Written Informed Consent

18.3.1. Informed Consent

Informed written consent is required from each patient prior to any testing under this protocol, including screening tests and evaluations. The consent form, as specified by the clinical site's IRB, must follow the Protection of Human Patients regulations listed in the Code of Federal Regulations, Title 21, Part 50.

The background of the proposed study and the benefits and risks of the procedures and study must be explained to the patients. It is the responsibility of the Investigator to obtain consent and to provide the patient with a copy of the signed and dated ICF. Confirmation of a patient's informed consent must also be documented in the patient's medical record prior to any testing under this protocol, including screening tests and evaluations.

All ICFs used in this study must be approved by the appropriate IRB and by the Sponsor or designee. The ICF must not be altered without the prior agreement of the relevant IRB and the Sponsor.

18.3.2. Patient Data Protection

Prior to any testing under this protocol, including screening tests and evaluations, patients must authorize the release and use of protected health information (PHI), as required by local law.

The patient will not be identified by name in the CRF or in any study reports. These reports will be used for research purposes only. The Sponsor, its designee, and various government health agencies may inspect the records of this study. Every effort will be made to keep the patient's personal medical data confidential.

19. DATA HANDLING AND RECORDKEEPING

19.1. Case Report Form Completion

CRFs will be completed for each randomized patient. It is the Investigator's responsibility to ensure the accuracy, completeness, and timeliness of the data reported in the patient's CRF. Source documentation supporting the CRF data should indicate the patient's participation in the study and should document the dates and details of study procedures, AEs, and patient status.

Investigators will maintain copies of the CRFs at the clinical site. For patients who discontinue or terminate from the study, the CRFs will be completed as much as possible, and the reason for the discontinuation or termination clearly and concisely specified on the appropriate CRF.

19.2. Retention of Records

The Investigator will maintain all study records according to ICH-GCP and applicable regulatory requirements. Records will be retained for at least 2 years after the last marketing application approval or 2 years after formal discontinuation of the clinical development of the investigational product or according to applicable regulatory requirements. If the Investigator withdraws from the responsibility of keeping the study records, custody must be transferred to a person willing to accept the responsibility. The Sponsor must be notified in writing if a custodial change occurs.

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21. APPENDICES

Appendix 1: Response Evaluation Criteria in Solid Tumors

Response Evaluation Criteria in Solid Tumors (RECIST) v1.1	
Terms	Definition
Measurable disease	The presence of at least one measurable lesion by CT or MRI. If the measurable disease is restricted to a solitary lesion, its neoplastic nature should be confirmed by cytology/histology. All measurements should be taken and recorded in metric notation, using a ruler or calipers. All baseline evaluations should be performed as closely as possible to the beginning of treatment and never more than 4 weeks before the beginning of the treatment.
Measurable lesions	Lesions that can be accurately measured in least one dimension by CT scan with longest diameter $\geq$ 10 mm (CT scan thickness no greater than 5 mm). The same method of assessment and the same technique should be used to characterize each identified and reported lesion at baseline and during follow-up.
Non-measurable lesion	All other lesions, including small lesions (longest diameter <10mm or pathological lymph nodes with P10 to <15mm short axis) as well as truly non-measurable lesions. Lesions considered truly non-measurable include: leptomeningeal disease, ascites, pleural or pericardial effusion, inflammatory breast disease, lymphangitic involvement of skin or lung, abdominal masses/abdominal organomegaly identified by physical exam that is not measurable by reproducible imaging techniques.

RECIST Criteria-Response Evaluation

Evaluation of target lesions	
Complete Response (CR)	Disappearance of all target lesions. Any pathological lymph nodes (whether target or non-target) must have reduction in short axis to <10 mm.
Partial Response (PR):	At least a 30% decrease in the sum of diameters of target lesions, taking as reference the baseline sum diameters.
Stable Disease (SD):	Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum diameters while on study.
Progressive Disease (PD):	At least a 20% increase in the sum of diameters of target lesions, taking as reference the smallest sum on study (this includes the baseline sum if that is the smallest on study). In addition to the relative increase of 20%, the sum must also demonstrate an absolute increase of at least 5 mm. (Note: the appearance of one or more new lesions is also considered progression).

Evaluation of non-target lesions	
Complete Response (CR):	Disappearance of all non-target lesions and normalisation of tumour marker level. All lymph nodes must be non-pathological in size (<10mm short axis).
Non-CR/Non-PD	Persistence of one or more non-target lesion(s) and/or maintenance of tumour marker level above the normal limits.
Progressive Disease (PD):	Unequivocal progression (see comments below) of existing non-target lesions. (Note: the appearance of one or more new lesions is also considered progression).

RECIST Criteria-Response Evaluation cont-

Evaluation of best overall response	The best overall response is the best response recorded from the start of the treatment until disease progression/recurrence (taking as reference for PD the smallest measurements recorded since the treatment started). In general, the patient's best response assignment will depend on the achievement of both measurement and confirmation criteria. Patients with a global deterioration of health status requiring discontinuation of treatment without objective evidence of disease progression at that time should be classified as having "symptomatic deterioration". Every effort should be made to document the objective progression even after discontinuation of treatment. In some circumstances it may be difficult to distinguish residual disease from normal tissue. When the evaluation of complete response depends on this determination, it is recommended that the residual lesion be investigated (fine needle aspirate/biopsy) to confirm the complete response status.

Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, Dancey J, Arbuck S, Gwyther S, Mooney M, Rubinstein L, Shankar L, Dodd L, Kaplan R, Lacombe D, Verweij J.New response evaluation criteria in solid tumours: Revised RECIST guideline (version 1.1) (2009). European Journal of Cancer 45, 228-247.

Appendix 2: ECOG Performance Status

The ECOG scale is used to assess a patient's quality of life in an evaluation by a health professional of the daily activities and how the activities are affected by the disease of the patient.

Grade	ECOG
0	Fully active, able to carry on all pre-disease performance without restriction
1	Restricted in physically strenuous activity but ambulatory and able to carry out work of a light or sedentary nature, e.g., light house work, office work
2	Ambulatory and capable of all selfcare but unable to carry out any work activities. Up and about more than 50% of waking hours
3	Capable of only limited selfcare, confined to bed or chair more than 50% of waking hours
4	Completely disabled. Cannot carry on any selfcare. Totally confined to bed or chair
5	Dead

Oken, M.M., Creech, R.H., Tormey, D.C., Horton, J., Davis, T.E., McFadden, E.T., Carbone, P.P. (1982) Toxicity And Response Criteria Of The Eastern Cooperative Oncology Group. Am J Clin Onco 5:649-655.

Appendix 3: National Cancer Institute (NCI) Common Terminology Criteria for Adverse Events (CTCAE) Version 3.0

See http://ctep.cancer.gov/reporting/ctc.html

Appendix 4: New York Heart Association- Classification of heart failure

Class 1 - Class 1 heart failure - no limitation of activities. No symptoms from ordinary activities.
Class 2 - Class 2 heart failure- mild limitation of activity. Comfortable with rest or mild exertion.
Class 3 - Class 3 heart failure- marked limitation of activity and be comfortable only at rest.
Class 4 –Class 4 heart failure- complete rest, confined to bed or chair; any physical activity brings on discomfort and symptoms occur at rest.

National Heart, Lung, and Blood Institute, National Institutes of Health. New York Heart Association Classification. 2008.

Appendix 5- Pharmacokinetic Sampling

Pharmacokinetics

Blood samples will be collected from approximately one-third of patients at selected study sites for the evaluation of serum concentrations of ACE-011. Pre-dose PK samples will be collected on Day 1.

Blood samples for PK should be collected in a fasted state (defined by no food or drink except water for at least 4 hours prior to the study procedure). The sample should be collected prior to ACE-011/placebo dosing. Chemotherapy regimens scheduled will be administered to the patient as per standard of care. Collection, handling, and shipping procedures for blood samples are provided in the Study Reference Guide.

Schedule of Events

	Screen	Treatment period													Post Treatment Follow up Period				Termination Visit
Placebo Dose period		#1				#2			#3			#4	Follow up						
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	113 (± 7d	141 (± 7d	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
PK Sampling		X				X	X			X			X						
ACE-011/placebo administration		X				X			X			X							
Chemo regimen		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

Protocol Amendment – Summary of Changes

CLINICAL STUDY PROTOCOL

A Phase 2, Double-Blind, Randomized, Placebo-Controlled Study of ACE-011 for the Treatment of Chemotherapy Induced Anemia in Patients with Metastatic Breast Cancer

STUDY DRUG: **ACE-011**

PROTOCOL NUMBER: **A011-08**

SPONSOR: **Acceleron Pharma, Inc.**
128 Sidney Street
Cambridge, MA 02139

PPD

MEDICAL MONITOR: **PPD Inc.**

PPD

PROTOCOL DATE: **13 March 2009**
AMENDMENT #1: **22 April 2009**
AMENDMENT #2: **21 May 2009**

Summary:
Amendment 02 includes the following changes:

- Administrative changes
- Addition of exclusion criteria - Major surgery within 30 days prior to Day 1 (patients must have completely recovered from any previous surgery prior to Day 1).
- Addition of visit day 99 with corresponding study day procedures.
- Additional PK sampling days added to appendix 5.
- Changes to the ECG collection days and procedures.
- Addition of text to clarify AE, SAE, and Concomitant medication collection during the patient participation on the study.
- Addition of text regarding back up blood samples for future testing.

Protocol Location	Change
Section 2. Protocol Synopsis	*Objectives* Clarification of exploratory objective serum bone biomarker "PINP" to "P1NP". Deleted the text "during the study" within exploratory objective #2 (evaluation of quality of life). *Exclusion criteria* Addition of exclusion criteria of "Major surgery within 30 days prior to Day 1 (patients must have completely recovered from any previous surgery prior to Day 1)." *Duration of study* Clarification of "study drug" to "ACE-011/placebo". Additional text added of complete the Day 281/Termination visit procedures" and "following the last dose of ACE-011/placebo." *Criteria for evaluation* Addition of text amending the section regarding the ECG timepoints. Additional clarification added for the timeframe of when lab values and blood pressure would be collected during the study by the following text "during the treatment and treatment follow up period." The following text was deleted "weekly for the first two treatment (dosing) periods and approximately every one to two weeks for subsequent treatment (dosing) periods."
Section 2. Schedule of events Table 7: Schedule of events	Formatting. Day 99 visit day added with the corresponding study day visit procedures. Day 113 visit day window was amended from $\pm$7 days to $\pm$ 3 days. ECG collection days were amended to include visit days 8, 36, and 64. The

	collection of an ECG on Day 29 was deleted. Footnote added to indicate ECGs are to be done in triplicate at each timepoint, at 3 minute intervals (± 1 min). Added clarification that the physical exam, coagulation/serum chemistry, and anti-drug antibody are to be done prior to ACE-011/placebo administration, as applicable. Added footnote assignment during screening and the following text "Confirmatory labs for eligibility criteria must be drawn and resulted within 7 days of Day 1." Deleted back up blood sample for future testing done at screening and Days 169-253. Additional reticulocyte values to be reported on Days 22, 36, and 64. Additional clarification added to indicate that "concomitant medications would be collected up through 90 days from the last dose of ACE-011/placebo." Footnote added to clarify that "AEs will be reported including up to 90 days after the last dose of ACE 011/placebo. Additional AEs noted after 90 days from the last dose of ACE-011/placebo will be reported at the discretion of the investigator. SREs and SAEs will be followed through out the study. If a patient is followed for survival only, SREs and SAEs will be captured up to 90 days from the last dose of ACE-011/placebo." Added text "start and end date of last menstrual period since the last assessment." The following text we deleted "date of last menstruation, date of menstrual period onset and duration of menstruation" Footnote clarification to reference withdrawal criteria in section 8.3 of the protocol.
Section 6. Trial Objectives and Purpose	*Section 6.2: Secondary Objectives* Formatting. *Section 6.3: Exploratory Objectives* Clarified exploratory objective serum bone biomarker "PINP" to "P1NP". Deleted the text "during the study" within exploratory objective #2 (evaluation of quality of life).

Section 7. Investigational Plan	Formatting. Addition of text amending the section regarding the ECG timepoints. Additional clarification added for the timeframe of when lab values and blood pressure would be collected during the study by the following text "during the treatment and treatment follow up period". The following text was deleted "weekly for the first two treatment (dosing) periods and approximately every one to two weeks for subsequent treatment (dosing) periods." Additional text added regarding central and local lab values. Additional text added "Back up (serum) blood samples will be collected for future testing for the evaluation of novel tumor markers, such as endoglin and ALK-1 (activin receptor-like kinase-1). The samples will the stored at the central lab until this evaluation is performed."
Section 8. Selection and Withdrawal of Patients	*Section 8.2.2 Patient exclusion criteria* Addition of exclusion criteria of "Major surgery within 30 days prior to Day 1 (patients must have completely recovered from any previous surgery prior to Day 1)." *Section 8.3 Patient Withdrawal Criteria* Additional clarification added to specify that patients who "withdraw due to ACE-011/placebo toxicity should" enter the treatment follow-up period.
Section 9. Treatment of Patients	Formatting. *Section 9.1.1 General Concomitant Medication Usage* Additional text added "Concomitant medications will be recorded on the eCRF through 90 days from the last dose of ACE-011/placebo." *Section 9.5.2 Selection and Timing of dosing for each patient* Clarification to reference section 8.3 of the protocol for reasons a patient may discontinue study treatment. *Section 9.5.2.2 ACE-011 Dose Modification Rules* Clarification of "The local hemoglobin values and blood pressure values will be evaluated at each study visit and on each ACE-011/placebo dosing day."
Section 10. Study Procedures	Formatting. Clarification of "collection of menstrual cycle information of the start and end date of the last menstruation." was added through out this section. *Section 10.1.2 Screening (within 14 days of Day 1)* Added text "Confirmatory labs for eligibility criteria must be drawn and resulted within 7 days of Day 1." Additional text "performed at two time intervals (approximately one hour

	apart) in triplicate at 3 minute intervals (±1 min).” Deleted back up blood samples for future testing. Serum pregnancy test to be confirmed within 7 days of Day 1. *Section 10.1.3 Initial dosing: Day 1* Deleted text “A patient will be considered randomized once a multi-digit patient identification number has been assigned to the patient through the IVRS.” Text of “and the inclusion/exclusion criteria have been met” was added to clarify when ACE-011/placebo administration may begin. Additional text added to the 12 lead ECG procedure “performed at two time intervals (approximately one hour apart) in triplicate at 3 minute intervals (±1 min)”. *Section 10.1.4 ACE-011/placebo administration: Days 29, 57, and 85 (± 3 days)* Deletion of 12 lead ECG procedure. Deletion of “and Hematocrit” *Section 10.1.5 Additional visits during the Treatment Period: Days 8, 15, 22, 36 (± 1 day), 43, 64 and 71 (± 2 days)* Deletion of 12 lead ECG procedure for Day 15 only. Addition of “12 lead ECG to be performed on Days 8, 36, and 64 only -to be done in triplicate at 3 minute intervals (±1 min).” Clarification of if a patient begins a new chemotherapy regimen; the patient will “be followed for survival only.” *Section 10.1.6 Treatment Period Follow-up visits : Days 99, 113(± 3 days) and 141(± 7 days)* Addition of Day 99 visit. Additional text added to the 12 lead ECG procedure “to be done in triplicate at 3 minute intervals (±1 min)”. Additional text added regarding the procedures to be performed. *Section 10.1.7 Days 169, 197, 225, and 253 (± 7 days)* Additional text added regarding concomitant medication and adverse event follow up. Deleted back up blood samples for future testing. *Section 10.1.8 Termination Visit: Day 281 (± 7 days)* Additional text added to the 12 lead ECG procedure “to be done in triplicate at 3 minute intervals (±1 min)”. Additional text added regarding concomitant medication and adverse event

	follow up.
Section 13. Safety Assessments	Deletion of "oral" from temperature assessment. Additional text added regarding central and local lab values.
Section 14. Adverse Event	*Section 14.2 Recording Adverse Events* Additional text added "AEs...up to 90 days from the last dose of ACE-011/placebo are to be documented on AE CRF. The specific disease or syndrome rather than individual associated signs and symptoms should be identified by the investigator and recorded on the CRF. Skeletal related event information is to be captured and reported for the duration of the patient participation in the study through Day 281/Termination visit" Text added to specify "SAEs will be captured throughout the patient participation in the study through Day 281/Termination visit. SAEs will be captured up to 90 days from the last dose of ACE-011/placebo for those patients who are followed for survival only."
Section 15. Statistics	Formatting. Additional clarification of "study drug" as "ACE-011/placebo".
Section 17. Quality Control and Quality Assurance	Additional clarification of the investigator responsibility.
Appendix 5: PK Sampling	Additional text added regarding PK sampling and the specified days of collection on Day 1 Post 4h, Day 8, Day 15, Day 22, Day 57, Predose Day 85, and Day85 Post 4h.

CLINICAL STUDY PROTOCOL

A Phase 2, Double-Blind, Randomized, Placebo-Controlled Study of ACE-011 for the Treatment of Chemotherapy Induced Anemia in Patients with Metastatic Breast Cancer

STUDY DRUG: **ACE-011**

PROTOCOL NUMBER: **A011-08**

SPONSOR: **Acceleron Pharma, Inc.**
128 Sidney Street
Cambridge, MA 02139

PPD

MEDICAL MONITOR: **PPD Inc.**
PPD

PROTOCOL DATE: **13 March 2009**
AMENDMENT #1 : **22 April 2009**
AMENDMENT #2 : **21 May 2009**

Signature Page

Acceleron Pharma Approval

Signature: PPD _____ **Date:** 22 May 2009

Name (print): _____

Investigator Agreement: I have read the protocol and agree to conduct the study as outlined in the protocol. The study will be conducted in accordance to current United States Food and Drug Administration (FDA) regulations, International Conference of Harmonization (ICH) Guidelines, Good Clinical Practices (GCP), and local ethical and legal requirements.

Signature: _____ **Date:** _____

Name (print): _____

Institution Name and Address:

_____

_____

_____

PROCEDURES IN CASE OF EMERGENCY

Table 1: Emergency Contact Information

Role in Study	Name	Address and Telephone number
PPD Medical Monitor	PPD	PPD Inc. 1800 Perimeter Park Drive, Suite 275 Morrisville, NC 27560-7200 USA PPD
Clinical Trial Manager	PPD	Acceleron Pharma, Inc. 128 Sidney Street Cambridge, MA 02139 USA PPD

2. PROTOCOL SYNOPSIS

Name of Sponsor/Company: Acceleron Pharma, Inc.
128 Sidney Street
Cambridge, MA 02139

Name of Investigational Product: ACE-011

Name of Active Ingredient:
ACE-011 is a fully human fusion protein consisting of the extracellular domain (ECD) of the activin receptor IIA (ActRIIA) linked to the human IgG1 Fc domain.
Mechanism of Action: ACE-011 is a disulfide-linked, glycosylated, dimeric protein. ACE-011 competes with the activin receptor IIA that binds a number of TGF-β superfamily ligands including activin, myostatin (growth differentiation factor [GDF]-8), and GDF-11.

Title of Study: A Phase 2, Double-Blind, Randomized, Placebo-Controlled Study of ACE-011 for the Treatment of Chemotherapy Induced Anemia in Patients with Metastatic Breast Cancer.

Study center(s): approximately 25-35 centers

Phase of development: 2

Objectives

Primary:

1. To evaluate the proportion of patients in each ACE-011 dose regimen who achieve a hematopoietic response during the treatment period including up to 2 months after the last dose of ACE-011 treatment of chemotherapy induced anemia in patients with metastatic breast cancer.

Secondary:

1. To evaluate the safety and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer.
2. To evaluate the percentage of patients who achieve an increase in hemoglobin of ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs.
3. To estimate the duration of the hematopoietic response associated with each ACE-011 dose regimen.
4. To estimate the time to the hematopoietic response associated with each ACE-011 dose regimen.
5. To estimate the proportion of patients receiving red blood cell (RBC) transfusions and/or ESAs within each cycle of ACE-011 with each dose regimen.

6. To estimate the effect of each ACE-011 dose regimen on progression free survival (PFS) and objective response rate (ORR).

Exploratory objectives:

1. To estimate the effect of each ACE-011 dose regimen on serum bone biomarkers (OC, BSAP, P1NP, CTX, TRACP-5b), urine bone biomarker (uNTX), bone mineral density (DXA scans of lumbar spine and hip) and incidence of skeletal related events (SREs).
2. To evaluate the quality of life in relation to the treatment of chemotherapy induced anemia in patients with metastatic breast cancer during the study.

Methodology: This is a Phase 2, double-blind, randomized, placebo-controlled study to evaluate the efficacy, safety, and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer. Patients will be randomized to one of three treatment arms or placebo:

1. ACE-011 0.1 mg/kg subcutaneously (SC) every 28 days for up to 4 doses.
2. ACE-011 0.3 mg/kg SC every 28 days for up to 4 doses.
3. ACE-011 0.5 mg/kg SC every 28 days for up to 4 doses.
4. Placebo SC every 28 days for up to 4 doses.

Number of patients (planned): 105 patients

Diagnosis and main criteria for inclusion

Inclusion Criteria:

1. Women ≥ 18 years of age.
2. Histologically confirmed diagnosis of breast cancer documented by cytology or biopsy.
3. Evidence of metastatic breast cancer with a minimum of one measurable lesion or non measurable lesion per RECIST criteria (Appendix 1).
4. Receiving a multicycle myelosuppressive chemotherapy regimen including one of the following: anthracycline, taxane, gemcitabine or vinorelbine.
5. Planned treatment with the same myelosuppressive chemotherapy regimen for a minimum of 9 weeks after Day 1 of ACE-011/placebo administration.
6. Hemoglobin value between ≥ 6.5 to < 11.0 g/dL (≥ 65 to < 110 g/L).
7. ≥ 30 days must have elapsed (from Day 1) since previous treatment with erythropoiesis-stimulating agent (including concurrent treatment with IV iron) for chemotherapy induced anemia.
8. ≥ 7 days have elapsed (from Day 1) since the last RBC blood transfusion and receipt of ≤ 2 units of blood in the past 30 days.
9. ECOG Performance status of 0 – 2 (Appendix 2).
10. Adequate renal (creatinine ≤ 1.5 x ULN or creatinine clearance ≥ 40 mL/min) and hepatic function (bilirubin ≤ 1.5 x ULN; AST/ALT ≤ 2.5 x ULN).
11. Females of childbearing potential participating in the study are to use adequate birth control measures (i.e. oral contraceptives or two forms of barrier method birth control) during study participation. Females of childbearing potential must have a negative serum pregnancy test within 3 days to ACE-011/placebo administration (Day 1). A female of childbearing potential is a sexually mature woman who has not undergone a hysterectomy or who has not been postmenopausal for at least 24 consecutive months (i.e. who has had menses at some time in the preceding 24 months).
12. Life expectancy of ≥ 6 months.
13. Is willing to adhere to the study visit schedule, understand and comply with all protocol requirements.
14. Understand and sign a written informed consent.

Exclusion Criteria:

1. At the time of screening patients who have any grade ≥ 3 toxicity (as per NCI CTCAE v3.0 (Appendix 3)) (except for the following disease related toxicities: hematological events- anemia, thrombocytopenia or neutropenia or non hematological events- nausea, vomiting, fatigue, or muscle or bone/joint pain).
2. Prior radiation therapy to > 20% of the whole skeleton. Use of palliative radiation if the area being treated is <15% of body surface area with no pelvic radiation or no more than 10% of the bone marrow reserve is radiated is permitted during patient participation in the study, at the discretion of the investigator.
3. Have received more than 5 prior chemotherapy treatment regimens for metastatic breast cancer.
4. Concurrent use of bevacizumab with the chemotherapy regimen during study participation.
5. Clinically significant pulmonary, endocrine, neurologic, gastrointestinal, hepatic or genitourinary disease unrelated to underlying hematologic disorder.
6. Patients with heart failure as classified by the New York Heart Association (NYHA) classification of 3 or higher (Appendix 4).
7. History of thrombosis, deep vein thrombosis (DVT), pulmonary emboli, or embolic stroke.
8. Untreated CNS metastases (exception: CNS metastases treated with whole brain radiotherapy > 6 months prior to randomization).
9. Diagnosis of a myeloid malignancy or known history of myelodysplasia.
10. History of second malignancy within 5 years (except excised and cured basal cell carcinoma, squamous cell carcinoma of the skin or cervical carcinoma in situ).
11. Patients with a recent history (within 14 days of Day 1) of administration of systemic (IV or oral) antibiotics. Patients should be free of infection at least 14 days after the last dose of systemic antibiotic treatment (from Day 1).
12. Uncontrolled hypertension. If hypertension is considered clinically stable, systolic blood pressure (BP) must be < 150 mmHg and diastolic BP must be < 100 mmHg.
13. Known history of hepatitis C antibody (HCV), hepatitis B surface antigen (HBsAg and HB core Ab), or human immunodeficiency virus (HIV) antibody.
14. Deficiency in iron (serum ferritin < 100 ng/mL (< 224.7 pmol/L)), vitamin B_{12}, or folate.
15. History of anemia as a result of inherited hemoglobinopathy such as sickle cell anemia or thalassemia.
16. History of autoimmune or hereditary hemolysis or gastrointestinal bleeding.
17. Patient received treatment with another investigational drug or device within 1 month prior to Day 1, or if the half life of the previous product is known, within 5 times the half life prior to dosing, whichever may be longer.
18. Pregnant or lactating females.
19. History of severe allergic or anaphylactic reactions or hypersensitivity to recombinant proteins or excipients in the investigational product.
20. Major surgery within 30 days prior to Day 1 (patients must have completely recovered from any previous surgery prior to Day 1).

Investigational product, dosage and mode of administration:

ACE-011/placebo will be administered as a SC injection, approximately every 28 days, on days 1, 29, 57, and 85 of the treatment period. Each patient will be randomized to one of three treatment arms or placebo:

Dose	Number of Patients
0.1 mg/kg	30
0.3 mg/kg	30
0.5 mg/kg	30
Placebo	15
Total patients enrolled: 105	

Patients with grade ≥ 3 toxicity related to ACE-011 will be discontinued from ACE-011/ placebo treatment except for the following: Patients with grade 3 hypertension that decreases to grade ≤ 1 within 7 days after treatment with anti-hypertensive therapy may continue on ACE-011/placebo treatment. Dose reduction rules will apply at the subsequent scheduled dosing day(s). Patients with a hemoglobin level above the upper limit of normal at any time during the study after Day 1 will be discontinued.

ACE-011/placebo dose modification rules are based on hemoglobin values and blood pressure measurements during each treatment cycle. ACE-011/placebo dose interruption and dose reduction rules will be implemented for those patients with hemoglobin of ≥ 11 g/dL, an increase of ≥ 2 g/dL/ 28 days, or hypertension ≥ Grade 2 within each cycle (see Figure 1).

Duration of study: Patients will be followed for a minimum of 7 months following their last dose of ACE-011/placebo. Study participation for each patient is approximately 10.5 months (14 day screening period, 3 month treatment period with a 2 month treatment follow up period, and a 5 month post treatment follow-up period). The start date of the chemotherapy regimen used in this protocol to treat metastatic breast cancer will be recorded. If a patient begins a new chemotherapy regimen during the study, the patient will discontinue further ACE-011/placebo treatment, complete the Day 281/Termination visit procedures and be followed for survival only. If a patient has a positive anti-drug antibody result at the Day 281/Termination visit, they may be asked to return to the clinical site for additional monthly repeat anti-drug antibody testing for up to 12 months, following the last dose of ACE-011/placebo. The study is complete when the last patient enrolled has completed the last protocol required follow up visit.

Criteria for evaluation

Efficacy: The primary endpoint is to establish the proportion of patients who have a hematopoietic response of an increase in hemoglobin of ≥ 0.5 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs.

The proportion of patients with an increase in hemoglobin of ≥ 1g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for 28 consecutive days during the treatment period up to

2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs will be evaluated as a secondary efficacy endpoint.

The hematopoietic endpoints will be determined by the monitoring of hematologic laboratory values, RBC transfusion and/or ESAs administration. The evaluation of response will be from the first dose of ACE-011 through 2 months after the last dose (for each patient). Additional endpoints will be evaluated with the assessment of disease progression by CT/MRI and bone scan and bone mineral density by DXA scans. Bone biomarkers will be evaluated in all patients and incidence of SREs will be recorded, as applicable. SREs are defined as pathologic fracture, radiation therapy to bone, surgery to bone, or spinal cord compression.

Safety: Throughout the study, safety will be evaluated by the sponsor, medical monitor and principal investigator. Additionally, the DMC will review safety parameters for patients enrolled in the study. All patients will be assessed for safety by monitoring adverse events, clinical laboratory tests, assessment of quality of life, vital signs and physical examinations on an ongoing basis. ECGs will also be performed during screening, prior to dosing on the first cycle, during the first, second and third cycle, Day 113 (28 days after the last dose of ACE-011) and at Day 281/Termination visit to monitor safety. Blood pressure and lab values will be assessed at each study visit during the treatment and treatment follow up period.

Pharmacokinetics: The blood samples will be collected at selected study sites from approximately one-third of patients for the evaluation of serum concentrations of ACE-011. Pre-dose PK samples will be collected on Day 1. See Appendix 5 for PK sampling schedule.

Statistical methods

Sample size determination: With an anticipated dropout rate of 20%, a sample size of 30 patients per dose level is planned to achieve at least 80% power with 2 sided significance level of 0.05 to differentiate targeted response rate of p1=0.45 from null hypothesis p0=0.20. An additional 15 patients will be enrolled into the placebo group for safety comparison. A total of 105 patients will be randomized to three ACE-011 dose groups or placebo with a ratio of 2:2:2:1.

Efficacy analysis: The primary endpoint is the proportion of patients who have a hematopoietic response of an increase in hemoglobin of ≥ 0.5 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs. The proportion of responders will be determined together with 95% confidence interval.

For binary endpoints, the proportion of responders and 95% confidence interval will be calculated. For time to event endpoints, the Kaplan-Meier method will be used to estimate the survival curve and median time to event. Due to the nature of the study, no multiplicity adjustment will be made for efficacy analysis.

Safety analysis: Data from all patients who receive at least one dose of ACE-011/placebo will be included in the safety analyses. To assess clinical safety, adverse events, vital sign measurements and clinical laboratory information will be summarized. Adverse events and laboratory results will also be summarized by severity grade (NCI CTCAE v3.0). Changes from baseline clinical laboratory values and vital signs will be summarized over time. Descriptive statistics and shift tables will be generated as appropriate.

Schedule of Events

	Screen	Treatment Period														Post Treatment Follow up Period				Term Visit[20]
ACE-011/placebo Dose period		#1				#2			#3			#4	Follow-up							
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	99 (± 3d)	113 (± 3d)	141 (± 7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
Informed consent	X																			
Inclusion/exclusion criteria	X	X[2]																		
Medical history	X																			
Physical examination	X	X[2]				X[2]			X[2]			X[2]		X	X					X
Vital signs[1]	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X					X
FACT-Fatigue	X	X[2]				X[2]			X[2]			X[2]								X
12 lead ECG[3]	X	X	X				X			X				X						X
Serum iron, TIBC, transferrin and serum ferritin	X[8]	X[2]				X[2]			X[2]			X[2]								X
Vitamin B12	X[8]																			
Serum folate	X[8]						X							X						X
Coagulation/ Serum chemistry[4]	X[8]	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X					X
Hematology[5]	X[8]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Serum Erythropoietin marker		X[2]				X[2]			X[2]			X[2]			X					X
LH, FSH, testosterone, progesterone, estradiol	X	X[2]				X[2]			X[2]			X[2]		X	X					
Anti-drug antibody testing [6]		X[2]				X[2]			X[2]			X[2]		X	X					X
Bone biomarker[7]		X[2]				X[2]			X[2]			X[2]		X	X					X
Back up blood sample for future testing		X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X					X
Pregnancy Test	X[8]	X[9]				X[9]			X[9]			X[9]								X
Urinalysis[10]	X	X[2]										X[2]								X
Evaluate transfusion frequency	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X	X	X	X	X	X
Documentation of conmeds	X[11]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X			

Schedule of Events cont.

	Screen	Treatment Period											Post Treatment Follow up Period					Term Visit[20]
ACE-011/Placebo Dose period		#1				#2			#3			#4	Follow-up					
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	99 (± 3d)	113 (± 3d)	141 (±7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
Evaluation of AEs and SAEs[12]	X-SAE only	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X-SRE and SAE only	X-SRE and SAE only	X-SRE and SAE only	X
Evaluation of Menstrual cycle[13]	X	X[2]				X[2]			X[2]			X[2]		X	X					
CT/MRI scan	X[14]									X[15]				X[15]						
Dual-energy X-ray absorptiometry (DXA[16])	X													X						X
Bone scans[17]	X									X				X						X
ACE-011/placebo administration[18]		X				X			X			X								
Chemo regimen[19]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

[1] Vital signs include weight, heart rate, seated systolic and diastolic blood pressure, and temperature will be assessed at each visit Height will be collected at screening only
[2] On dosing days (1, 29, 57 and 85), study procedure to be done prior to dosing of ACE-011/placebo
[3] Screening and Day 1- At two different time intervals (approximately 1 hours apart) triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min), prior to dosing as applicable On all other specified days, one time interval of triplicate (12 lead) ECGs is to be performed at 3 minute intervals (± 1min),,
[4] Coagulation (central lab only) prothrombin time (PT), partial thromboplastin time (PTT), fibrinogen Chemistry (central lab only): AST, ALT, lactate dehydrogenase (LDH), total bilirubin, indirect bilirubin, alkaline phosphatase, total protein, albumin, sodium, potassium, chloride, carbon dioxide, calcium, phosphorus, blood urea nitrogen (BUN), creatinine, uric acid, glucose, amylase, and lipase
[5] Hematology (local lab) hemoglobin and hematocrit On dosing days (1, 29, 57 and 85), local hemoglobin and hematocrit values are to be drawn and resulted prior to dosing to apply dose modification rules for ACE-011/placebo Hematology (central lab only) complete blood count (CBC) with differential and platelet, CBC includes red blood cells (RBC), white blood cells (WBC), hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), Peripheral blood smear will also be done on Days 1, 29, 57, 85, and at the termination visit **Reticulocyte should be done on days 1, 8, 15, 22, 29, 36, 57, 64, 85, and at the termination visit.** On dosing days (1, 29, 57 and 85), local hemoglobin and hematocrit values are to be drawn and resulted prior to dosing to apply dose modification rules for ACE-011/placebo
[6] If a patient has a positive anti-drug antibody result at the Termination visit, they may be asked to return to the clinical site for additional monthly follow-up, for up to 12 months after their last dose for a repeat anti-drug antibody test
[7] Both serum and urine samples will be collected at each visit The urine sample should not be from the first morning urine
[8] Confirmatory labs for eligibility criteria must be drawn and resulted within 7 days of Day 1
[9] For females of child bearing potential only urine (dipstick) pregnancy test to be collected and resulted prior to dosing of ACE-011/placebo
[10] Urinalysis to be performed at screening, prior to administration ACE-011/placebo (Days 1 and 85) and termination visit Assessments (central lab only): pH, specific gravity, protein, glucose, ketones, blood, leukocyte esterase, and nitrite Microscopy required only to follow-up abnormal findings
[11] Screening- any concomitant treatments taken 28 days prior to day 1 including up to 90 days after the last dose of ACE-011/placebo
[12] AEs will be reported from day 1 post dose including up to 90 days after the last dose of ACE-011/placebo Additional AEs noted after 90 days from the last dose of ACE-011/placebo will be reported at the discretion of the investigator SREs and SAEs will be followed through out the study If a patient is followed for survival only, SREs and SAEs will be captured up to 90 days from the last dose of ACE-011/placebo
[13] Females of child bearing potential- collection of menstrual cycle information) start and end date of last menstrual period since the last assessment
[14] A CT/MRI scan acquired within 4 weeks of initiating the current chemotherapy regimen may be used for the screening assessment Head, neck, thoracic, abdominal, and pelvic scans are to be acquired One measurable or non measurable lesion per RECIST is to be identified by the scan Brain (head) CT only to be done to rule out CNS metastases in patients with suspicious clinical symptoms or prior history of CNS metastases
[15] Follow-up CT/MRI scans may be acquired +/- 5 day window for both Day 64 and Day 113 Additional CT/MRI scans acquired during the post treatment follow up period will be performed at the discretion of the investigator
[16] Hip and lumbar spine BMD assessment
[17] Bone scans will be acquired within 4 weeks of initiating the current chemotherapy regimen for those patients who have bone metastases identified Follow up bone scans may be acquired +/- 5 day window for Day 64, Day 113, and Day 281
[18] Patients who discontinue ACE-011/placebo due to ACE-011/placebo related toxicities (section 9 5 2 1) will enter the treatment follow-up period
[19] Chemotherapy regimens will be administered to the patient as per standard of care Continuation of the same chemotherapy regimen may be done through the follow up period of the study at the discretion of the investigator
[20] Patients who terminate treatment early due to disease progression and/or begin a new chemotherapy regimen will complete the day 281/Termination visit procedures and be followed for survival only (7 months from the last dose of ACE-011/placebo) If a patient withdraws from the study (section 8 3), the Day 281/Termination visit procedures are to be completed and the patient will be followed for survival If withdrawal reason is due to ESA administration or ACE-011 related toxicity, the patient should enter the treatment follow up period

3. TABLE OF CONTENTS, LIST OF TABLES AND LIST OF FIGURES

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

4. LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

The following abbreviations and specialist terms are used in this study protocol.

Table 2: Abbreviations and Specialist Terms

Term	Definition
ACTH	Adrenocorticotropic hormone
ActRIIA	Activin receptor IIA
AE	Adverse event
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
AUC	Area under the plasma concentration - time curve
$AUC_{(0\text{-last})}$	AUC from time 0 to time of last quantifiable sample
$AUC_{(0\text{-}\infty)}$	AUC extrapolated to infinity
BMD	Bone mineral density
BP	Blood pressure
BSAP	Bone specific alkaline phosphatase
BUN	Blood urea nitrogen
CBC	Complete Blood Count
CFR	Code of Federal Regulations (US)
CH	Constant domain
CI	Confidence interval
CIA	Chemotherapy induced anemia
CL	Clearance
CL/F	Total clearance
C_{max}	Maximum observed concentration
CNS	Central nervous system
CO_2	Carbon dioxide
CRF	Case report form
CRO	Clinical research organization
CT	Computed tomography
CTCAE	Common Terminology Criteria for Adverse Events
CTX	Serum C-terminal type I collagen telopeptide
DLT	Dose-limiting toxicity
DXA	Dual-energy x-ray absorptiometry
DMC	Data Monitoring Committee
DVT	Deep vein thrombosis
ECD	Extracellular domain
ECG	Electrocardiogram
ECOG	Eastern Cooperative Oncology Group
EDF	Erythroid differentiation factor
ESA	Erythropoiesis-stimulating agents
F	Bioavailability (absolute)
FACT	Functional Assessment of Chronic Illness
FDA	Food and Drug Administration
FSH	Follicle-stimulating hormone
GCP	Good Clinical Practices
GH	Growth hormone

Term	Definition
HBV	Hepatitis B virus
Hct	Hematocrit
HCV	Hepatitis C virus
Hgb	Hemoglobin
HIV	Human immunodeficiency virus
ICF	Informed consent form
ICH	International Conference on Harmonization
IEC	Independent Ethics Committee
IND	Investigational New Drug
IP	Intraperitoneal
IRB	Institutional Review Board
IV	Intravenous
IVRS	Interactive Voice Response System
Kd	Binding coefficient
λz	Elimination rate constant
LDH	Lactate dehydrogenase
LH	Luteinizing hormone
MCH	Mean corpuscular hemoglobin
MCHC	Mean corpuscular hemoglobin concentration
MCV	Mean corpuscular volume
MedDRA	Medical Dictionary for Regulatory Activities
MITT	Modified Intent to Treat (population)
MRI	Magnetic resonance imaging
NOAEL	No adverse events level
NYHA	New York Heart Association
OC	Osteocalcin
ORR	Objective response rate
OVX	Ovariectomized
PD	Progressive disease
PFS	Progression free survival
PHI	Protected health information
P1NP	Serum intact procollagen type I N terminal propeptide
PK	Pharmacokinetic
PP	Per-protocol (population)
PT	Prothrombin time
PTH	Parathyroid hormone
PTT	Partial thromboplastin time
RAP-011	Murine version of ACE-011, ActRIIA mFc
RBC	Red blood cell (count)
RECIST	Response Evaluation Criteria in Solid Tumors
SAE	Serious adverse event
SAP	Statistical analysis plan
SC	Subcutaneous
SERM	Selective estrogen receptor modulator
SHAM	Sham-operated
SPEP	Serum protein electrophoresis
SRE	Skeletal-related event
T1/2	Elimination half-life
TIBC	Total iron binding capacity
Tmax	Time to Cmax

Term	Definition
TRACP-5b	Serum tartrate-resistant acid phosphatase isoform-5b
ULN	Upper limit of normal
Vz/F	Volume of distribution
WBC	White blood cell (count)

5. INTRODUCTION

5.1. Indication and Rationale

This study is designed to evaluate the efficacy, safety, and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer.

Treatment of patients with metastatic breast cancer with myelosuppressive chemotherapy is frequently associated with anemia. Chemotherapy induced anemia (CIA) is a significant problem for patients with cancer, causing fatigue and reduced quality of life (1). Patients with CIA are currently treated with blood transfusion and/or erythropoiesis-stimulating agents (2). However, with these treatment options CIA is still an area of unmet medical need. Erythropoiesis-stimulating agents (ESAs) can successfully mitigate transfusion need in a proportion of these patients, but concerns have emerged regarding apparent negative effects on survival and/or tumor progression in certain patient populations. The murine surrogate to ACE-011, RAP-011, has been tested as a single agent in breast cancer cell lines MDA-MB-231 and MCF-7 and no effect on enhanced proliferation of these cell lines has been observed in vitro. Therefore, treatment with ACE-011 may provide a distinct benefit/risk profile to patients with chemotherapy induced anemia.

In both a Phase 1 single dose and multiple dose study of ACE-011 in postmenopausal women, increases in hemoglobin and hematocrit were observed following ACE-011 treatment and remained elevated over the course of study. The observed hemoglobin and hematocrit effects of ACE-011 were dose and time dependent. Please refer to the Investigator Brochure for further information.

Based on the effect of ACE-011 on hematopoiesis and consistent biological phenomena observed in both non clinical and clinical studies, it is hypothesized that the blockade of ActRIIA receptor signaling impedes the terminal differentiation step in erythropoiesis to allow additional rounds of cell replication before cells enter the final differentiation phase. The result is a substantial increase in mature erythrocytes released into the circulation. Since this proposed mechanism is different to that of known ESAs, ACE-011 may provide a different clinical profile in the treatment of CIA.

This study will provide information on the pharmacodynamics (PD) properties regarding the ability of ACE-011 to increase hemoglobin in patients with CIA.

5.2. Description of ACE-011

ACE-011 (ActRIIA-IgG1) is a fully human fusion protein consisting of the extracellular domain (ECD) of activin receptor IIA (ActRIIA) linked to the human IgG1 Fc domain, which includes the heavy chain hinge and constant domains, CH2 and CH3. ACE-011 avidly binds to activin A with a binding coefficient (Kd) approximately 8.9 pM and prevents its binding to endogenous receptors, thereby inhibiting biological effects of activin.

5.3. Activin Biology

Activin A was initially identified as a gonadal differentiation factor involved in modulating follicle stimulating hormone (FSH) secretion from the pituitary (3). Subsequently, the pleiotropic nature of activin A has become more apparent (4). There is a growing body of data suggesting a role for activin A in bone remodeling, specifically as a negative regulator of bone growth (5). Before the two molecules were shown to be identical (6), activin A was also described as erythroid differentiation factor (EDF), effecting red blood cells in the later stages of maturation (7). The mechanism(s) by which activin A influences erythropoiesis remain under investigation and, in fact, there are data from studies in vitro and in animals that support erythropoiesis-stimulatory (8,9) and -inhibitory effects (10).

At the cellular level, activin A binds initially to the high-affinity Type II receptor (ActRIIA). The ligand-bound ActRIIA then recruits the low-affinity Type I receptor (ActRIA or ALK-4). The receptor heterocomplex, through its cytoplasmic protein kinase activity, then activates the Smad signal cascade to eventually influence nuclear transcriptional factors (11, 12). The competitive binding of activin A in the blood by the ACE-011 soluble fusion protein can result in inhibition of ActRIIA receptor signaling pathway by impeding biological processes attributed to activin A.

Activin has also been attributed to erythroid differentiation and has been reported to have proerythrocytic effects and to induce terminal differentiation of RBCs. Inhibition of activin may lead to increases in proliferation of the erythroid lineage. Based on its pharmacologic effects, ACE-011 (ActRIIA-IgG1) is being developed for the treatment of bone loss associated with various disease states (e.g., osteoporosis and treatment of osteolytic lesions in patients with multiple myeloma), and treatment of anemia associated with a variety of disorders, such as chemotherapy induced anemia.

The extracellular domain sequence of ActRIIA is completely conserved among numerous species including mouse, rat, cynomolgus monkey and humans, thus ACE-011 is active in these animals. However, in order to reduce the potential immunogenicity of the fully human molecule, ACE-011, in mice and to maximize the opportunity to maintain exposures in chronic models, a murine surrogate molecule was constructed by exchanging the human immunoglobulin Fc sequence portion of ACE-011 with its murine IgG2a homolog. The resultant construct is referred to as RAP-011 (ActRIIA-mIgG2aFc).

5.3.1. Pharmacology Studies

RAP-011 was evaluated in a broad range of animal pharmacology studies to assess the effects of inhibition of activin A on the biological processes attributed to that regulatory protein. RAP-011 has been shown to have significant effects on the red cell compartment. RAP-011 treatment of mice at 10, 30 and 50 mg/kg by intraperitoneal injection (IP) twice per week for 3 months resulted in a 16-26% increase in red blood cell (RBC) counts compared to control animals. Rats treated with RAP-011 at 0.3, 3 and 30 mg/kg once per week for 3 months showed RBC count increases of 6-15% over control animals. Finally, in cynomolgus monkeys treated with 10, 30 or

50 mg/kg of ACE-011 twice per month for 3 months there was a 21- 24% increase in RBC counts compared to controls animals. In this study, the RBC count increase was apparent as early as 2 weeks following the initial dose of ACE-011.

While the mechanism(s) underlying the stimulation effect of ACE-011 on erythropoiesis are not yet fully understood, it is hypothesized that a blockade of ActRIIA receptor signaling impedes the terminal differentiation step in erythropoiesis to allow additional rounds of erythroid cell replication before cells enter the terminal differentiation phase. The result is a substantial increase in mature erythrocytes released into circulation.

RAP-011 administered at 10 mg/kg to mice three days prior to administration of paclitaxel at 30 mg/kg was sufficient to prevent decreases in red blood cell parameters seen three days later. Mice receiving paclitaxel alone had decreased hematocrit from 43% to 38% three days later. RAP-011 administered 3 days prior to paclitaxel injection was sufficient to keep the hematocrit above 42% at three days and up to two weeks after paclitaxel administration. Therefore prophylactic treatment with RAP-011 was able to prevent paclitaxel induced anemia in mice.

RAP-011 has also been shown to significantly increase bone mineral density and strength in normal animals and in a variety of animal models of bone loss (13, 14, 15). In a murine ovariectomy (OVX)-induced model of osteopenia, RAP-011 (10 mg/kg IV, twice per week x 12 weeks) increased trabecular BMD > 25% in 6-12 weeks compared to the phosphate buffered saline (PBS)-treated controls ($p<0.002$). RAP-011 treatment was able to reverse the accelerated bone loss associated with ovariectomy. As expected sham-operated (SHAM) mice (i.e., intact ovaries) started the study with a greater trabecular BMD than OVX mice. PBS-treated SHAM mice showed a 10% decrease in trabecular BMD after 12 weeks of treatment compared to baseline, while SHAM-RAP-011 mice had a 32% increase in trabecular BMD over the same period. These data demonstrate that RAP-011 is able to increase trabecular BMD in both the normal state and in a model of established bone loss. The femurs of both the RAP-011 treated OVX and SHAM mice showed statistically significant increases in all measured parameters of bone strength (i.e., maximum load, stiffness, energy, ultimate strength, and elastic modulus). In summary, treatment with RAP-011 produced bone of superior quality consistent with an anabolic agent.

RAP-011 was evaluated in a mouse model of skeletal metastasis using luciferase-tagged, human MDA-MB-231 breast cancer cells (estrogen receptor negative). Mice were pretreated for 2 weeks with RAP-011 (10 mg/kg, biweekly, SC) prior to intracardiac injection of tumor cells. Treatment with RAP-011 was continued for an additional 5 weeks following tumor implantation. A parallel study was conducted to examine the effect of RAP-011 treatment, as described above, on survival of MDA-MB-231 bearing mice.

The data demonstrate that RAP-011 treatment acts to inhibit MDA-MB-231 metastasis to bone and may also play a role in inhibiting tumor growth in soft tissue. Furthermore, in the presence of tumor cells, RAP-011 is able to prevent lytic disease through an anabolic mechanism of bone formation. Lastly, inhibition of tumor growth and metastasis with RAP-011 leads to a greater than 25% survival benefit in this model.

The efficacy of RAP-011 was examined in an orthotopic model of breast cancer using luciferase-tagged human MCF-7 breast cancer cells (estrogen receptor positive). Mice were pretreated for 2 weeks with RAP-011 (10 mg/kg, biweekly, SC) prior to the implantation of tumor cells into the mammary fat pad of female nude mice. Treatment with RAP-011 was continued for an additional 7 weeks following tumor implantation at which time mice were sacrificed and assessed for tumor burden. RAP-011 effectively lowered the tumor burden in mice as detected by bio-luminescence. In addition, a 60% reduction in tumor weight was observed in the RAP-011 treated mice.

The ability of RAP-011 to repair osteolytic lesions caused by metastatic disease was investigated in mice. In this model athymic nude mice received an intratibial injection of MDA-MB-231-Luc cells to mimic bone metastasis. Mice were treated with a chemotherapy regimen of paclitaxel (20 mg/kg, IP, every three days) starting seven days after tumor injection and continuing for the duration of the study. On study day 42 mice with a detectable tumor burden by bioluminescent imaging were divided into two groups and began treatment with RAP-011 (10 mg/kg, SC, biweekly) or vehicle. Prior to dosing, μCT scans of the tumor bearing tibia were conducted to identify osteolytic lesions. RAP-011 treatment continued for four weeks (to study day 70) and a final μCT scan of the tibia was performed. On study day 70 osteolytic lesions remained the same or had progressed further when compared to study day 42 in the vehicle treated mice. While some mice showed progression of osteolytic lesions (most likely related to tumor burden) on study day 70, a majority of mice treated with RAP-011 demonstrated repair of the osteolytic lesions seen on study day 42. Therefore treatment with RAP-011 has the ability to repair osteolytic lesions caused by tumors and after cytoxic chemotherapy with paclitaxel.

5.3.2. Toxicology

ACE-011 has been evaluated for toxicological effects in two species, Sprague-Dawley rats and cynomolgus monkeys. The dose levels ranged from 0.3 to 30 mg/kg in rats and from 1 to 50 mg/kg in monkeys. These dose ranges were designed to support Phase 1a single-dose levels of 0.01 to 3.0 mg/kg IV and Phase 1b multiple doses of 0.1 to 2 mg/kg (monthly, SC). Weekly dosing in animals was designed to provide continuous, but fluctuating serum concentrations of ACE-011, which would be mimicked by a one-month dosing interval in humans.

Initial studies utilized IV dosing to identify target organs while longer-term studies (3 months in rats; 3 and 6 months in monkeys) utilized SC dosing to mimic the intended dosing regimen for patients. The expected pharmacologic effect, increased red blood cells (RBCs), hemoglobin (Hb), hematocrit (HCT) and reticulocytes, was observed in all studies, presumably based on the ability of ACE-011 to inhibit activin. A second expected pharmacologic effect of ACE-011, based on inhibition of activin, was the reversible reduction in sperm production and testicular tubular damage in rats. Since male cynomolgus monkeys were sexually immature in these studies, it was not possible to monitor this effect in this species.

Effects on the adrenal gland were only seen in rats and were more pronounced in female rats. This toxicological observation may not be relevant for predicting effects in humans since adrenal toxicity was not seen in cynomolgus monkeys. Dose-limiting toxicity and no observable adverse effect levels (NOAELs) were primarily based on renal toxicity in both rats and monkeys. There

was some indication that kidney toxicity was an indirect effect, based on the formation of antibody/antigen complexes, but since these studies were not designed to investigate toxicological mechanisms, a definitive cause of renal impairment and renal damage was not determined. Development of antibodies to ACE-011 was noted in all of these studies, which is an expected immune reaction in rats and monkeys dosed with a human protein.

The no observable adverse effect level (NOAELs) from the 3 month SC studies was 3 and 30 mg/kg in rats and monkeys, respectively. Because the kidney findings were observed at all dose levels, the NOAEL from the 6 month monkey study is < 10 mg/kg.

5.4. Summary of Clinical Experience

5.4.1. A011-01: A Phase 1a Study in Healthy Postmenopausal Women

ACE-011 was first studied in a randomized, Phase 1a, single dose, dose escalation study in healthy, post-menopausal females (16). ACE-011 was diluted in normal saline administered as an IV infusion (over approximately 1 hour) or as a SC injection on Day 1. Dose levels were 0.01, 0.03, 0.1, 0.3, 1.0, and 3.0 mg/kg IV and 0.03 and 0.1 mg/kg SC. A total of 48 subjects were enrolled; 5 active and 1 placebo at each of the IV and SC dose levels. All subjects were followed for 4 months following a single dose administration.

The pharmacokinetics of ACE-011 was linear for all IV and SC doses. Across IV doses ranging from 0.1 through 3.0 mg/kg, the mean clearance (CL) ranged from 0.092 to 0.128 mL/h/kg, the volume of distribution ranged from 73.7 to 110 mL/kg, and the mean T1/2 ranged from 23.7 to 31.8 days, with no apparent dependence on dose. After SC administration of 0.03 and 0.1 mg/kg, ACE-011 was completely absorbed, and the mean T1/2 was approximately 30 days, with no apparent dependence on dose.

The most commonly occurring treatment-emergent adverse events (AEs; i.e., those occurring in more than 1 subject in any treatment group) were headache, infusion site reaction, injection site hemorrhage, and toothache. The majority of the injection site reactions and injection site hemorrhages were in the first IV cohort and were related to infiltration of the IV site. The majority of treatment-emergent AEs were mild in severity and were judged to be unrelated to ACE-011. No deaths, serious AEs (SAEs), or AEs leading to discontinuation were reported. Changes in red blood cells (RBCs), hemoglobin, reticulocytes, liver function tests, glucose, uric acid, amylase and lipase occurred in some subjects; however, none of these laboratory results were reported as adverse events. Mild, transient elevations in pancreas enzymes, liver enzymes, or glucose were reported as AEs in five patients. There were no clinically significant changes from baseline in vital signs, physical examination, endocrine function, or electrocardiogram (ECG) data. Preservation of adrenal cortical function was monitored through evaluation of serum electrolytes (sodium and potassium levels) and through cortisol response to adrenocorticotropic hormone (ACTH) stimulation. No clinically significant findings were observed at doses up to 3.0 mg/kg IV, and ACE-011 was well tolerated in healthy, post-menopausal female volunteers in single dose levels up to 0.1 mg/kg SC and 3.0 mg/kg IV, the highest dose levels tested in this study.

5.4.2. A011-02: A Phase 1b Study in Healthy Postmenopausal Women

ACE-011 was studied in a Phase 1b, single-center, randomized, double-blind, placebo-controlled, multi-dose, dose-escalating study to evaluate the safety, tolerability and pharmacodynamics of ACE-011 in healthy postmenopausal women. Four cohorts of 10 subjects each were planned at the following dose levels: 0.1, 0.3, 1.0, and 2.0 mg/kg administered subcutaneously. Within each cohort, subjects were to be randomized to either active or placebo treatment in an 8:2 ratio. Subjects were to receive one SC injection of ACE-011 or placebo every 28 days for a total of 4 doses. All subjects were to be followed for 12 weeks after the last dose.

The treatment phase of the study was terminated early after a dose-limiting pharmacodynamic effect was observed. While on study, one subject in the 1.0 mg/kg cohort experienced a Serious Adverse Event (SAE) of progressive and persistent hypertension that was attributed to a rapid and significant rise in hemoglobin and hematocrit levels. Due to symptoms of headaches, nausea, eye pain, dizziness and vomiting approximately 1 week following the second dose, the subject was hospitalized for monitoring and evaluation of her hypertension. The head MRI, fundoscopy, and ECG performed were reported as normal and headache symptoms resolved following therapeutic phlebotomy. The SAE resolved and the subject was discharged from the hospital the following day. The hypertension was monitored closely and managed initially with antihypertensive medication. The hypertension resolved by the end of the study without need of any antihypertensive medication. The subject received aspirin prophylactically until the end of the study and continued taking ibuprofen as needed for headaches. Further details can be found in the Investigator Brochure.

Based on the magnitude of the hematopoietic response, the Sponsor suspended dose escalation to the 2.0 mg/kg dose level and further dosing in all cohorts as a result of this dose-limiting pharmacodynamic effect at the 1.0 mg/kg dose level. In total, 31 subjects were enrolled and treated. Dose levels of ACE-011 administered included 0.1, 0.3, and 1.0 mg/kg (Cohorts 1, 2, and 3 respectively). All subjects randomized to active treatment in Cohort 1 received all 4 planned doses of ACE-011. Due to early discontinuation of study drug, subjects randomized to active treatment in Cohort 2 received 3 doses of ACE-011, and subjects randomized to active treatment in Cohort 3 received 2 doses of ACE-011. Subjects randomized to placebo treatment received between 1 and 4 doses of study treatment.

In the analysis of the unblinded data, after the administration of the first dose, a dose and time dependent increase in hemoglobin values was observed (see Table 3 below):

Table 3: A011-02: A Phase 1b Study in Healthy Postmenopausal Women: Hemoglobin Evaluation

Evaluation Time Point	Mean Change from Baseline (g/dL)			
	Placebo N=7*	ACE-011 0.1 mg/kg N=8	ACE-011 0.3 mg/kg N=8	ACE-011 1.0 mg/kg N=8
Baseline Mean, pre-Dose 1	13.20	13.11	13.30	12.71
Day 8	0.17	0.68	0.85	1.21
Day 15	-0.27	0.43	0.44	1.75
Day 29	0.27	0.61	1.21	2.68**
Day 36	0.56	0.64	1.89	2.96
Day 43	-0.02	0.89	1.21	2.85
Day 57	0.27	1.28	1.64**	2.09
Day 64	0.53	1.11	2.49	2.21
Day 71	-0.10	1.34	2.09	1.66
Day 85	0.38	1.18**	2.55	1.86
Day 92	0.00***	1.04	1.60***	3.80***
Day 99	-0.20	1.21	3.20***	1.28***
Day 113	-0.02	1.30	1.29	1.04
Day 141	0.30	0.95	0.34	2.30
Day 169	0.20***	0.06		2.00***

*The number of placebo subjects with data decreases over time as a result of the early discontinuation of the study (i.e., there were placebo subjects in each dosing cohort). There were seven placebo subjects with data at baseline, Days 8, 15, and 36; six subjects with data at Days 29, 43, 57, and 85; five subjects with data on Days 71 and 113; four subjects with data on Day 64; three subjects with data on Day 141; two subjects with data on Day 99; and one subject with data on Days 92 and 169.

** Number of doses administered per treatment group: 0.1 mg/kg 4 doses; 0.3 mg/kg 3 doses; 1.0 mg/kg 2 doses. Data beyond this study day are considered follow-up results.

***n=1

No severe or life-threatening events were reported. The incidence of the most common treatment-emergent AEs (i.e., occurring in more than one subject in any treatment group) is presented below (see Table 4). The most notable AEs were those related to increases in hematologic laboratory measures in the 1.0 mg/kg dose group: hematocrit, hemoglobin, and RBC count. AEs of increased hemoglobin and/or hematocrit were reported for seven of the eight subjects in this dose group. These events were reported as mild or moderate elevations; all were considered probably related to study drug treatment. Three of the subjects in the 1.0 mg/kg group with elevated hemoglobin levels underwent phlebotomies and all hemoglobin elevations were resolved by the end of the follow-up period. No erythroid lineage AEs were reported in the 0.1 or 0.3 mg/kg treatment groups.

Table 4: A011-02: Incidence of Most Common Adverse Events

System Organ Class and Preferred Term	Dose Group			
	Placebo N=7 n (%)	0.1 mg/kg N=8 n (%)	0.3 mg/kg N=8 n (%)	1.0 mg/kg N=8 n (%)
GASTROINTESTINAL DISORDERS				
Diarrhea	0 (0.0%)	2 (25.0%)	0 (0.0%)	0 (0.0%)
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS				
Asthenia	0 (0.0%)	1 (12.5%)	2 (25.0%)	0 (0.0%)
Fatigue	2 (28.6%)	0 (0.0%)	1 (12.5%)	2 (25.0%)
INFECTIONS AND INFESTATIONS				
Viral upper respiratory tract infection	0 (0.0%)	4 (50.0%)	0 (0.0%)	0 (0.0%)
INJURY, POISONING AND PROCEDURAL COMPLICATIONS				
Limb injury	0 (0.0%)	1 (12.5%)	0 (0.0%)	2 (25.0%)
INVESTIGATIONS				
Hematocrit increased	0 (0.0%)	0 (0.0%)	0 (0.0%)	6 (75.0%)
Hemoglobin increased	0 (0.0%)	0 (0.0%)	0 (0.0%)	7 (87.5%)
Red blood cell count increased	0 (0.0%)	0 (0.0%)	0 (0.0%)	3 (37.5%)
METABOLISM AND NUTRITION DISORDERS				
Decreased Appetite	1 (14.3%)	0 (0.0%)	2 (25.0%)	0 (0.0%)
MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS				
Arthralgia	2 (28.6%)	0 (0.0%)	2 (25.0%)	0 (0.0%)
Muscle spasms	0 (0.0%)	1 (12.5%)	0 (0.0%)	2 (25.0%)
NERVOUS SYSTEM DISORDERS				
Dizziness	0 (0.0%)	0 (0.0%)	2 (25.0%)	2 (25.0%)
Headache	2 (28.6%)	4 (50.0%)	3 (37.5%)	2 (25.0%)
Paresthesia	1 (14.3%)	0 (0.0%)	0 (0.0%)	3 (37.5%)
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS				
Oropharyngeal pain	2 (28.6%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
VASCULAR DISORDERS				
Hot flush	0 (0.0%)	2 (25.0%)	0 (0.0%)	1 (12.5%)

Note: Table presents AEs reported in more than one subject in any treatment group.

Headache was reported frequently in all treatment groups with no apparent dose response trend, though the reports in the higher dose groups were more often considered possibly related to treatment. Most headaches were reported as mild (one headache was reported as moderate).

Paresthesia and dizziness were reported more frequently in the ACE-011 groups, though the events were generally not considered drug related. Other frequently reported events (e.g. fatigue, upper respiratory infection) did not appear to increase with dose and were generally mild.

Hemoglobin levels for all subjects with elevations had returned to within normal limits by the end of the study.

Adrenal cortical function was monitored through evaluation of serum electrolytes (sodium and potassium levels) and through cortisol response to adrenocorticotropic hormone (ACTH) stimulation. All ACTH simulation test results were normal.

PK results confirmed that the PK of ACE-011 is linear following the first SC doses of all three dose levels tested, 0.1, 0.3, and 1.0 mg/kg. The terminal half life ($T_{1/2}$) of ACE-011 following the last dose in all three dose groups was identical, with mean $T_{1/2}$ being approximately 23 days. The mean CL/F ranged from 3.05 to 3.90 mL/d/kg, the mean Vz/F ranged from 97.47 to 103.03 mL/kg, and the mean $T_{1/2}$ ranged from 20.92 to 23.34 days in all 3 dose levels, with no apparent dependence on dose.

Bone mineral density was assessed by dual-energy X-ray absorptiometry (DXA). Mean change and mean percent change in BMD results from baseline to study end are summarized in the table below. A dose-dependent increase in the BMD of the total hip from baseline to end of study was observed, with a significant and rapid increase of 2.4% in the 1.0 mg/kg dose group, compared to a 0.7% decrease in the placebo group. BMD results for lumbar spine showed slight increases of 0.4% to 1.0% from baseline to study end in all active treatment groups, compared with a 0.5% decrease in the placebo group (see Table 5 below):

Table 5: A011-02: A Phase 1b Study in Healthy Postmenopausal Women: BMD

	Treatment Group							
	Placebo N=7		**ACE-011 0.1 mg/kg N=8**		**ACE-011 0.3 mg/kg N=8**		**ACE-011 1.0 mg/kg N=8**	
	Mean Change	Mean Percent Change	Mean Change	Mean Percent Change	Mean Change	Mean Percent Change	Mean Change	Mean Percent Change
Lumbar Spine (g/cm^2)	-0.0020	-0.5%	0.0099	0.7%	0.0082	1.0%	0.0051	0.4%
Total Hip (g/cm^2)	-0.0062	-0.7%	0.0050	0.6%	0.0075	0.9%	0.0220	2.4%

Number of doses administered and day of last dose per treatment group: 0.1 mg/kg 4 doses (Day 85); 0.3 mg/kg 3 doses (Day 57); 1.0 mg/kg 2 doses (Day 29). Data beyond this study day are considered follow-up results.

5.4.3. A011-04: A Phase 2a Study in Patients with Osteolytic Lesions of Multiple Myeloma

Study A011-04 is an ongoing, non-IND, Phase 2a, multi-center, randomized, multiple-dose study to evaluate the safety, tolerability and efficacy of ACE-011 in patients with osteolytic lesions of multiple myeloma. Safety evaluations include adverse events (AEs), clinical laboratory tests, standard 12-lead electrocardiogram (ECG), vital signs, Eastern Cooperative Oncology Group

(ECOG) performance status and physical examinations. Additionally, the study includes the assessment of biochemical markers of bone formation and resorption, skeletal related events (SREs), bone mineral density (BMD) by duel-energy x-ray absorptiometry (DXA) and bone pain by visual analog scale (VAS).

In this study, patients are randomized in a 4:1 ratio to one of three dose levels of ACE-011 (0.1, 0.3 and 0.5 mg/kg) or placebo, to be administered to patients every 28 days by subcutaneous injection, for up to four doses over a 3-month period. The test article is being evaluated in combination with the anti-myeloma therapy of melphalan (4 mg/m^2 on days 1-7), prednisolone (40 mg/m^2 on days 1-7) and thalidomide (100 mg per day) (MPT). The sites, Sponsor and Sponsor representatives are blinded to treatment assignment.

As of 10 March 2009, 30 patients have been randomized, enrollment is complete and treatment is ongoing. Fifteen patients have received multiple doses of their assigned treatment; 7 patients have received 4 doses, 3 patients have received 3 doses and 5 patients have received 2 doses. Of the remaining 15 patients, 12 patients have received their first dose of ACE-011 or placebo and 3 patients have not yet received their first dose. There have been no ACE-011 related serious adverse events (SAEs) reported. Per the monitored preliminary data on 18 patients presented to the Independent Data Monitoring Committee (DMC) on 9 February 2009, there were 8 Grade 3/severe AEs reported in 4 patients, including neutropenia, platelet count decreased, pain in extremity, back pain and anemia. All Grade 3 AEs were determined to be not related to ACE-011 and the Grade 3 hematologic adverse events were considered either probably or definitely related to the MPT treatment.

Following preliminary analysis of the blinded central laboratory data, increases in hemoglobin values were observed within 28 days after administration of the first dose of ACE-011/placebo. Per data available, 11 out of 30 patients had $\geq$ 1 g/dL increase in hemoglobin within the first 28 days on the study, 4 patients achieved a $\geq$ 2 g/dL increase within their first 28 days on study, while 8 patients achieved a $\geq$ 2 g/dL increase in hemoglobin through Day 85.

Including all visits as of 10 March 2009 through Day 85, 9 out of 30 patients had a dose interruption due to a hemoglobin value of > 13 g/dL according to the dose modification rules defined in the protocol and 1 of the 9 patients, who had a history of hypertension, also had a dose interruption due to Grade 2 hypertension. Three of the 9 patients that required a dose interruption due to a hemoglobin value of > 13 g/dL, as per protocol, had hemoglobin levels above 13 g/dL prior to their first dose. Taken together, these data, suggest a beneficial pharmacodynamic effect of ACE-011 on erythropoiesis in a patient population with cancer chemotherapy induced anemia. No patients have had a dose reduction in ACE-011/placebo treatment or discontinued treatment.

One SAE of prolonged hospitalization due to pneumonia, that was considered not related to ACE-011, was reported in a PPD enrolled in the study. On PPD, four days after the first dose of ACE-011/placebo and the initial doses of melphalan, prednisolone and thalidomide (MPT), the patient presented with elevated temperatures up to 39º C, cough and fatigue. Chest x-ray performed on PPD revealed a pneumonic infiltration in the lower

lobe of the left lung. On PPD hospitalization of the patient was prolonged due to the event. The patient was treated with moxifloxacin, doripenem, meropenem and fluconazole.

On PPD the pneumonic infiltration resolved, as confirmed by chest x-ray. This SAE resolved upon discharge of the patient from the hospital on PPD

The investigator considered the event of pneumonia not related to ACE-011 and possibly related to MPT. The Sponsor agreed with the Investigator's assessment of causality. Pneumonia is considered a labeled event per the current package insert for thalidomide. The SAE was considered serious, related and expected with respect to thalidomide and unrelated to ACE-011.

The A011-04 study has completed enrollment, while treatment, follow-up and data monitoring are ongoing.

5.5. Potential Risks for Human Use

The 1-month safety study in rats suggested that ACE-011 may affect sperm count and motility in a dose-dependent manner, although these findings appeared to be reversible upon drug discontinuation. These reproductive effects are likely a result of the expected biological activity of activin inhibition.

The 1-month safety study in rats also suggested that ACE-011 induces adrenocortical necrosis in rats. At present, the mechanism by which ACE-011 has induced this lesion in rats and the basis for its apparent species specificity are unknown; the finding has not been observed in mice or cynomolgus monkeys at similar dose levels. Adrenal cortical function was monitored in human subjects receiving ACE-011 by the evaluation of serum electrolytes including sodium and potassium levels and by the evaluation of cortisol response to ACTH stimulation in both the Phase 1a single dose and Phase 1b multiple dose healthy volunteer clinical studies. These parameters were evaluated up to doses of 3.0 mg/kg IV and 1.0 mg/kg SC, and no clinically significant perturbations in adrenal function were observed.

Laboratory findings from the Phase 1a single dose study showed elevations in hematology results, pancreas and liver enzymes, and uric acid. The most commonly seen treatment-emergent adverse events in this study were headache, infusion site reaction, injection site hemorrhage, and toothache.

Based on data from the Phase 1b study, the most notable AEs were increases in hematologic laboratory measures in the 1.0 mg/kg dose group: hematocrit, hemoglobin, and RBC count. AEs of increased hemoglobin and/or hematocrit were reported for seven of the eight subjects in this dose group. These events were reported as mild or moderate elevations; all were considered probably related to study drug treatment. No erythroid lineage AEs were reported in the 0.1 or 0.3 mg/kg treatment groups. Hematologic parameters will be monitored carefully in this study.

Headache was reported frequently in all treatment groups with no apparent dose response trend, though the reports in the higher dose groups were more often considered possibly related to treatment. Most headaches were reported as mild (one headache was reported as moderate). Paresthesia and dizziness were reported more frequently in the ACE-011 groups, though the

events were generally not considered drug related. Other frequently reported events (e.g. fatigue, upper respiratory infection) did not appear to increase with dose and were generally mild.

In the Phase 1b multiple-dose healthy volunteer study, some subjects treated with ACE-011 showed elevations in blood pressure. One subject who had received two doses of ACE-011 at the 1.0 mg/kg dose level had a serious adverse event. The patient was hospitalized for further evaluation of increases in blood pressure and symptoms that may have been caused by the increase in blood pressure. The symptoms that the subject experienced included headache, dizziness, nausea, vomiting, and elevated hematology values (increases in the number of red blood cells). This serious adverse event was judged to be probably related to the study drug and resolved following discharge from hospital the following day. Throughout the course of the follow up period hematologic parameters were monitored frequently until the hemoglobin and hematocrit levels returned to within normal limits. Further details are outlined in the Investigator Brochure.

As with all biologics there is the potential for anti-drug antibodies that can be associated with increased drug clearance and hypersensitivity reactions. An immune response to ACE-011 has been seen in monkeys that received the drug for 1 month and longer. Some of those animals developed kidney inflammation possibly related to that immune response. Anti-drug antibody formation will be assessed in this study to examine the immunogenicity of ACE-011, and to monitor reversibility of any AEs over time.

Please refer to the Investigator Brochure for more information.

6. TRIAL OBJECTIVES AND PURPOSE

6.1. Primary objective

1. To evaluate the proportion of patients in each ACE-011 dose regimen who achieve a hematopoietic response during the treatment period including up to 2 months after the last dose of ACE-011 treatment of chemotherapy induced anemia in patients with metastatic breast cancer.

6.2. Secondary objectives

1. To evaluate the safety and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer.
2. To evaluate the percentage of patients who achieve an increase in hemoglobin of ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for a consecutive 28 day period during the treatment period including up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs.
3. To estimate the duration of the hematopoietic response associated with each ACE-011 dose regimen.
4. To estimate the time to the hematopoietic response associated with each ACE-011 dose regimen.
5. To estimate the proportion of patients receiving red blood cell (RBC) transfusions and/or ESAs within each cycle of ACE-011 with each dose regimen.
6. To estimate the effect of each ACE-011 dose regimen on progression free survival (PFS) and objective response rate (ORR).

6.3. Exploratory objectives

1. To estimate the effect of each ACE-011 dose regimen on serum bone biomarkers (OC, BSAP, P1NP, CTX, TRACP-5b), urine bone biomarker (uNTX), bone mineral density (DXA scans of lumbar spine and hip) and incidence of skeletal related events (SREs).
2. To evaluate the quality of life in relation to the treatment of chemotherapy induced anemia in patients with metastatic breast cancer during the study.

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7. INVESTIGATIONAL PLAN

7.1. Overall Study Design and Plan

This is a Phase 2, double-blind, randomized, placebo-controlled study to evaluate the efficacy, safety, and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer. A total of 105 patients will be enrolled.

The Data Monitoring Committee (DMC) is responsible for reviewing safety. The DMC will be comprised of a minimum of 3 members. The DMC will meet regularly during the study to review serious adverse events (SAEs), adverse events (AEs), laboratory results, and vital signs. Safety data will be reviewed throughout the study by the DMC, Medical Monitor and the Investigator. DMC responsibilities, membership, meeting frequencies and procedures will be outlined in the DMC charter.

Patients will be evaluated for study inclusion/exclusion criteria by the Investigator. Patients who meet the study entry criteria will be enrolled within 14 days of the screening visit. Central lab values and local lab values for hemoglobin will be utilized for the evaluation of patient eligibility. Central labs will also be used in the evaluation of patient safety throughout the study. Local lab values for hemoglobin and hematocrit will be collected and reported from screening through the Day 281/Termination visit. Three cohorts of 30 patients each are planned at the following doses of ACE-011: 0.1 mg/kg, 0.3 mg/kg and 0.5 mg/kg. Fifteen patients will be enrolled into the placebo cohort (as shown in Table 6).

Table 6: Study Design

Dose	Number of Patients
0.1 mg/kg	30
0.3 mg/kg	30
0.5 mg/kg	30
Placebo	15
Total patients enrolled: 105	

Each eligible patient will be randomized to one of the four cohorts to receive a dose administered as a subcutaneous injection every 28 days for up to 4 doses. Dosing will be administered on days 1, 29, 57, and 85. Concurrent treatment with a myelosuppressive chemotherapy regimen for metastatic breast cancer will also be administered per standard of care at the site. Patients will be stratified according to administration of either single agent weekly chemotherapy or less frequent chemotherapy regimens.

Blood samples will be collected from approximately one-third of patients participating in the study at selected study sites for the evaluation of serum concentrations of ACE-011. Pre-dose PK samples will be collected as specified in Appendix 5 (PK sampling schedule.)

Patients will be assessed for safety by monitoring adverse events, clinical laboratory tests, assessment of quality of life (FACT- Fatigue (17)), vital signs and physical examinations on an ongoing basis. ECGs will be performed during screening, prior to dosing on the first cycle, during the first, second and third cycle, Day 113 (28 days after the last dose of ACE-011) and at Day 281/ Termination visit to monitor safety. Blood pressure and lab values will be assessed at each study visit during the treatment and treatment follow up period.

Local laboratory hemoglobin values and blood pressure values will be evaluated prior to dosing per the dose modification criteria, as applicable.

Patients will be followed for a minimum of 7 months following their last dose of ACE-011/placebo. Study participation for each patient is approximately 10.5 months (14 day screening period, 3 month treatment period with a 2 month treatment follow up period, and a 5 month post treatment follow-up period). If a patient discontinues the study due to progression of disease (metastatic breast cancer) and/or begins another chemotherapy regimen, the patient will discontinue ACE-011/placebo treatment, complete the Day 281/Termination visit procedures, and be followed for survival only (a minimum 7 months from the last dose of ACE-011/placebo).

Anti-drug antibody testing will be performed at Day 281/Termination visit and if positive, the patient may be asked to return to the clinical site for additional monthly anti-drug antibody testing for up to 12 months, after their last dose.

Back up (serum) blood samples will be collected for future testing for the evaluation of novel tumor markers, such as endoglin and ALK-1 (activin receptor-like kinase-1). The samples will the stored at the central lab until this evaluation is performed.

Table 7: Schedule of Events

Schedule of Events																				
	Screen	Treatment Period														Post Treatment Follow up Period				Term Visit[20]
ACE-011/placebo Dose period		#1				#2			#3			#4	Follow-up							
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	99 (± 3d)	113 (± 3d)	141 (± 7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
Informed consent	X																			
Inclusion/exclusion criteria	X	X[2]																		
Medical history	X																			
Physical examination	X	X[2]				X[2]			X[2]			X[2]		X	X					X
Vital signs[1]	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X					X
FACT-Fatigue	X	X[2]				X[2]			X[2]			X[2]								X
12 lead ECG[3]	X	X	X				X			X				X						X
Serum iron, TIBC, transferrin and serum ferritin	X[8]	X[2]				X[2]			X[2]			X[2]								X
Vitamin B12	X[8]																			
Serum folate	X[8]						X							X						X
Coagulation/ Serum chemistry[4]	X[8]	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X					X
Hematology[5]	X[8]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Serum Erythropoietin marker		X[2]				X[2]			X[2]			X[2]			X					X
LH, FSH, testosterone, progesterone, estradiol	X	X[2]				X[2]			X[2]			X[2]		X	X					
Anti-drug antibody testing [6]		X[2]				X[2]			X[2]			X[2]		X	X					X
Bone biomarker[7]		X[2]				X[2]			X[2]			X[2]		X	X					X
Back up blood sample for future testing		X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X					X
Pregnancy Test	X[8]	X[9]				X[9]			X[9]			X[9]								X
Urinalysis[10]	X	X[2]										X[2]								X
Evaluate transfusion frequency	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X	X	X	X	X	X
Documentation of conmeds	X[11]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X			

Schedule of Events cont.

	Screen	Treatment Period														Post Treatment Follow up Period				Term Visit[20]
ACE-011/Placebo Dose period		#1				#2			#3			#4	Follow-up							
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	99 (± 3d)	113 (± 3d)	141 (±7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
Evaluation of AEs and SAEs[12]	X-SAE only	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X-SRE and SAE only	X-SRE and SAE only	X-SRE and SAE only	X
Evaluation of Menstrual cycle[13]	X	X[2]				X[2]			X[2]			X[2]		X	X					
CT/MRI scan	X[14]									X[15]				X[15]						
Dual-energy X-ray absorptiometry (DXA[16])	X													X						X
Bone scans[17]	X									X				X						X
ACE-011/placebo administration[18]		X				X			X			X								
Chemo regimen[19]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

[1] Vital signs include weight, heart rate, seated systolic and diastolic blood pressure, and temperature will be assessed at each visit Height will be collected at screening only
[2] On dosing days (1, 29, 57 and 85), study procedure to be done prior to dosing of ACE-011/placebo
[3] Screening and Day 1- At two different time intervals (approximately 1 hours apart) triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min), prior to dosing as applicable On all other specified days, one time interval of triplicate (12 lead) ECGs is to be performed at 3 minute intervals (± 1min),
[4] Coagulation (central lab only) prothrombin time (PT), partial thromboplastin time (PTT), fibrinogen Chemistry (central lab only): AST, ALT, lactate dehydrogenase (LDH), total bilirubin, indirect bilirubin, alkaline phosphatase, total protein, albumin, sodium, potassium, chloride, carbon dioxide, calcium, phosphorus, blood urea nitrogen (BUN), creatinine, uric acid, glucose, amylase, and lipase
[5] Hematology (local lab) hemoglobin and hematocrit On dosing days (1, 29, 57 and 85), local hemoglobin and hematocrit values are to be drawn and resulted prior to dosing to apply dose modification rules for ACE-011/placebo Hematology (central lab only) complete blood count (CBC) with differential and platelet, CBC includes red blood cells (RBC), white blood cells (WBC), hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), Peripheral blood smear will also be done on Days 1, 29, 57, 85, and at the termination visit **Reticulocyte should be done on days 1, 8, 15, 22, 29, 36, 57, 64, 85, and at the termination visit.** On dosing days (1, 29, 57 and 85), local hemoglobin and hematocrit values are to be drawn and resulted prior to dosing to apply dose modification rules for ACE-011/placebo
[6] If a patient has a positive anti-drug antibody result at the Termination visit, they may be asked to return to the clinical site for additional monthly follow-up, for up to 12 months after their last dose for a repeat anti-drug antibody test
[7] Both serum and urine samples will be collected at each visit The urine sample should not be from the first morning urine
[8] Confirmatory labs for eligibility criteria must be drawn and resulted within 7 days of Day 1
[9] For females of child bearing potential only urine (dipstick) pregnancy test to be collected and resulted prior to dosing of ACE-011/placebo
[10] Urinalysis to be performed at screening, prior to administration ACE-011/placebo (Days 1 and 85) and termination visit Assessments (central lab only): pH, specific gravity, protein, glucose, ketones, blood, leukocyte esterase, and nitrite Microscopy required only to follow-up abnormal findings
[11] Screening- any concomitant treatments taken 28 days prior to day 1 including up to 90 days after the last dose of ACE-011/placebo
[12] AEs will be reported from day 1 post dose including up to 90 days after the last dose of ACE-011/placebo Additional AEs noted after 90 days from the last dose of ACE-011/placebo will be reported at the discretion of the investigator SREs and SAEs will be followed through out the study If a patient is followed for survival only, SREs and SAEs will be captured up to 90 days from the last dose of ACE-011/placebo
[13] Females of child bearing potential- collection of menstrual cycle information start and end date pf last menstrual period since the last assessment
[14] A CT/MRI scan acquired within 4 weeks of initiating the current chemotherapy regimen may be used for the screening assessment Head, neck, thoracic, abdominal, and pelvic scans are to be acquired One measurable or non measurable lesion per RECIST is to be identified by the scan Brain (head) CT only to be done to rule out CNS metastases in patients with suspicious clinical symptoms or prior history of CNS metastases
[15] Follow-up CT/MRI scans may be acquired +/- 5 day window for both Day 64 and Day 113 Additional CT/MRI scans acquired during the post treatment follow up period will be performed at the discretion of the investigator
[16] Hip and lumbar spine BMD assessment
[17] Bone scans will be acquired within 4 weeks of initiating the current chemotherapy regimen for those patients who have bone metastases identified Follow up bone scans may be acquired +/- 5 day window for Day 64, Day 113, and Day 281
[18] Patients who discontinue ACE-011/placebo due to ACE-011/placebo related toxicities (section 9 5 2 1) will enter the treatment follow-up period
[19] Chemotherapy regimens will be administered to the patient as per standard of care Continuation of the same chemotherapy regimen may be done through the follow up period of the study at the discretion of the investigator
[20] Patients who terminate treatment early due to disease progression and/or begin a new chemotherapy regimen will complete the day 281/Termination visit procedures and be followed for survival only (7 months from the last dose of ACE-011/placebo) If a patient withdraws from the study (section 8 3), the Day 281/Termination visit procedures are to be completed and the patient will be followed for survival If withdrawal reason is due to ESA administration or ACE-011 related toxicity, the patient should enter the treatment follow up period

8. SELECTION AND WITHDRAWAL OF PATIENTS

8.1. Number of Patients

The study will include three dose levels of ACE-011 at 0.1 mg/kg, 0.3 mg/kg and 0.5 mg/kg and a placebo group. A total of 105 will be treated with ACE-011 or placebo.

8.2. Entry Criteria

8.2.1. Patient Inclusion Criteria

1. Women ≥ 18 years of age.
2. Histologically confirmed diagnosis of breast cancer documented by cytology or biopsy.
3. Evidence of metastatic breast cancer with a minimum of one measurable lesion or non measurable lesion per RECIST criteria (Appendix 1).
4. Receiving a multicycle myelosuppressive chemotherapy regimen including one of the following: anthracycline, taxane, gemcitabine or vinorelbine.
5. Planned treatment with the same myelosuppressive chemotherapy regimen for a minimum of 9 weeks after Day 1 of ACE-011/placebo administration.
6. Hemoglobin value between ≥ 6.5 to < 11.0 g/dL (≥ 65 to < 110 g/L).
7. ≥ 30 days must have elapsed (from Day 1) since previous treatment with erythropoiesis-stimulating agent (including concurrent treatment with IV iron) for chemotherapy induced anemia.
8. ≥ 7 days have elapsed (from Day 1) since the last RBC blood transfusion and receipt of ≤ 2 units of blood in the past 30 days.
9. ECOG Performance status of 0 – 2 (Appendix 2).
10. Adequate renal (creatinine ≤ 1.5 x ULN or creatinine clearance ≥ 40 mL/min) and hepatic function (bilirubin ≤ 1.5 x ULN; AST/ALT ≤ 2.5 x ULN).
11. Females of childbearing potential participating in the study are to use adequate birth control measures (i.e. oral contraceptives or two forms of barrier method birth control) during study participation. Females of childbearing potential must have a negative serum pregnancy test within 3 days to ACE-011/placebo administration (Day 1). A female of childbearing potential is a sexually mature woman who has not undergone a hysterectomy or who has not been postmenopausal for at least 24 consecutive months (i.e. who has had menses at some time in the preceding 24 months).
12. Life expectancy of ≥ 6 months.
13. Is willing to adhere to the study visit schedule, understand and comply with all protocol requirements.

14. Understand and sign a written informed consent.

8.2.2. Patient Exclusion Criteria

1. At the time of screening patients who have any grade ≥ 3 toxicity (as per NCI CTCAE v3.0 (Appendix 3)) (except for the following disease related toxicities: hematological events- anemia, thrombocytopenia or neutropenia or non hematological events- nausea, vomiting, fatigue, or muscle or bone/joint pain).
2. Prior radiation therapy to > 20% of the whole skeleton. Use of palliative radiation if the area being treated is <15% of body surface area with no pelvic radiation or no more than 10% of the bone marrow reserve is radiated is permitted during patient participation in the study, at the discretion of the investigator.
3. Have received more than 5 prior chemotherapy treatment regimens for metastatic breast cancer.
4. Concurrent use of bevacizumab with the chemotherapy regimen during study participation.
5. Clinically significant pulmonary, endocrine, neurologic, gastrointestinal, hepatic or genitourinary disease unrelated to underlying hematologic disorder.
6. Patients with heart failure as classified by the New York Heart Association (NYHA) classification of 3 or higher (Appendix 4).
7. History of thrombosis, deep vein thrombosis (DVT), pulmonary emboli, or embolic stroke.
8. Untreated CNS metastases (exception: CNS metastases treated with whole brain radiotherapy > 6 months prior to randomization).
9. Diagnosis of a myeloid malignancy or known history of myelodysplasia.
10. History of second malignancy within 5 years (except excised and cured basal cell carcinoma, squamous cell carcinoma of the skin or cervical carcinoma in situ).
11. Patients with a recent history (within 14 days of Day 1) of administration of systemic (IV or oral) antibiotics. Patients should be free of infection at least 14 days after the last dose of systemic antibiotic treatment (from Day 1).
12. Uncontrolled hypertension. If hypertension is considered clinically stable, systolic blood pressure (BP) must be < 150 mmHg and diastolic BP must be < 100 mmHg.
13. Known history of hepatitis C antibody (HCV), hepatitis B surface antigen (HBsAg and HB core Ab), or human immunodeficiency virus (HIV) antibody.
14. Deficiency in iron (serum ferritin < 100 ng/mL (< 224.7 pmol/L)), vitamin B_{12}, or folate.
15. History of anemia as a result of inherited hemoglobinopathy such as sickle cell anemia or thalassemia.
16. History of autoimmune or hereditary hemolysis or gastrointestinal bleeding.
17. Patient received treatment with another investigational drug or device within 1 month prior to Day 1, or if the half life of the previous product is known, within 5 times the half life prior to dosing, whichever may be longer.

18. Pregnant or lactating females.
19. History of severe allergic or anaphylactic reactions or hypersensitivity to recombinant proteins or excipients in the investigational product.
20. Major surgery within 30 days prior to Day 1 (patients must have completely recovered from any previous surgery prior to Day 1).

8.3. Patient Withdrawal Criteria

Patients will be informed that they have the right to withdraw from the study at any time for any reason without prejudice to their medical care.

Patients must be withdrawn from the study for any of the following reasons:

- Patient request
- Patient is unwilling or unable to comply with the protocol
- ESA administration during the treatment period
- Medical reason, such as cancer or treatment related toxicity, or at the discretion of the Investigator and/or the Medical Monitor(s)

The reasons for withdrawal must be recorded in the patient's case report form (CRF). The Investigator must notify the Medical Monitor and the contract research organization (CRO) immediately when a patient has been discontinued/withdrawn due to an AE. All patients who are withdrawn from the study should complete the tests and evaluations scheduled for Day 281/Termination at the time of withdrawal. Discontinued/withdrawn patients should be followed for survival only for 7 months after the last dose of ACE-011/placebo. Patients who withdraw due to ESA administration during the treatment period or due to ACE-011 related toxicity should enter the treatment follow-up period.

9. TREATMENT OF PATIENTS

9.1. Concomitant Medications

9.1.1. General Concomitant Medication Usage

During screening, and during the study, patients may take only stable doses of medications for chronic conditions that are not specifically excluded by the protocol (see Sections 8.2.1 Patient Inclusion Criteria and 8.2.2 Patient Exclusion Criteria). The Medical Monitor should be consulted by the Investigator if there are any concerns about what constitutes a stable dose or what is a chronic condition. Concomitant medications will be recorded in the CRF through 90 days from the last dose of ACE-011/placebo.

9.1.2. Concomitant treatment for anemia

Concurrent therapy with a new prescription medication related to treatment of anemia during the course of the study must first be discussed with the Investigator and Medical Monitor prior to administration, unless appropriate medical care necessitates that medication should begin before the Medical Monitor can be consulted.

9.1.2.1. Iron supplementation

Iron supplementation within 7 days of Day 1 (besides an oral general multivitamin with iron) will not be allowed. If a patient becomes iron replete during study treatment (ferritin < 100 ng/mL (< 224.7 pmol/L) or transferrin < 20%), treatment with iron supplementation is at the discretion of the investigator.

9.1.2.2. Erythropoiesis-stimulating agents (ESAs)

If concurrent treatment with erythropoiesis-stimulating agent is required in the opinion of the investigator, the erythropoiesis-stimulating agent label instructions are to be followed. The patient is to discontinue further treatment with ACE-011/placebo and enter the treatment follow-up period of the study. See Table 7 for Schedule of Events.

9.1.2.3. RBC Transfusions

Concurrent treatment for chemotherapy induced anemia with blood transfusions is recommended when hemoglobin value is < 8 g/dL or at investigator discretion if the hemoglobin value is above 8 g/dL and associated with anemia symptoms such as hemodynamic or pulmonary compromise that require treatment. If a transfusion is given to a patient during the treatment period, ACE-011/placebo should be administered no sooner than 7 days from the date of the transfusion. After the transfusion, the hemoglobin value should be assessed no later than 7 days from the date of the transfusion. On the day of ACE-011/placebo administration, the hemoglobin value and blood pressure will be assessed. See Figure 1 for ACE-011/placebo dose modification rules.

9.2. Chemotherapy

Chemotherapy treatment for metastatic breast cancer is to be given as per standard of care at the site. ACE-011/placebo administration should be given prior to chemotherapy administration on ACE-011/placebo administration days. Continuation of treatment with the same chemotherapy regimen through the treatment or post-treatment follow-up period of the study is at the discretion of the investigator.

9.3. Treatment Compliance

Each dose of ACE-011/placebo will be administered as a SC injection at the clinical site by the study staff and will be documented in the study record. Monitoring for patient compliance with the treatment regimen is therefore unnecessary.

9.4. Randomization

Patients will be randomized to limit scientific bias within the study. Randomization assignments will be generated through a computerized system, provided by an Interactive Web/Voice Response System (IWRS/IVRS). Patients will be stratified according to frequency of the planned chemotherapy regimen (weekly chemotherapy vs. less frequent chemotherapy administration).

9.4.1. Blinding

The Investigators, patients, and sponsor or representative will remain blinded to the treatment arm assignment of each patient. The unblinded statistician will remain uninvolved in the study conduct until database lock and unblinding of the data has occurred.

Safety parameters will be reviewed on an ongoing basis throughout the study including adverse events, serious adverse events, laboratory listings, and vital signs. If substantial toxicity trends are observed in one treatment group versus another, the DMC may request unblinding the treatment code assignment schema.

9.4.2. Unblinding

In the event of a medical emergency for an individual patient in which knowledge of the study medication is critical to the patient's medical management, the investigator may break the blind for that patient. However, prior to breaking the blind, every effort must be made by the investigator to first discuss the need to break the blind with the sponsor or sponsor representative. Further, it must be determined by the investigator that breaking the treatment blind is necessary information for the medical management of that patient. If the blind is broken, the patient must withdraw from the study by completing the Day 281/Termination visit procedures and be followed for survival only.

9.5. Treatments Administered

9.5.1. Selection of Doses in the Study

Single dose administration of ACE-011 up to 3.0 mg/kg was previously demonstrated to be safe and to have durable effects on markers of bone formation and resorption as well as red blood cells following a single IV administration. A dose-limiting pharmacodynamic effect of ACE-011 with increases in hemoglobin, hematocrit and RBCs was established in healthy post-menopausal women at the 1.0 mg/kg dose level following multiple SC administration. This study will confirm the safety in following repeated SC administration of ACE-011 and further evaluate the potential efficacy in patients with chemotherapy induced anemia. The safety and preliminary efficacy of ACE-011 following multiple doses up to 0.5 mg/kg will be assessed using a parallel randomization design.

9.5.2. Selection and Timing of Dosing for Each Patient

Patients will be enrolled and receive their assigned dose of ACE-011/placebo every 28 days (i.e. on Days 1, 29, 57, and 85). Patients will be receiving chemotherapy, concurrently for metastatic breast cancer. After completion of ACE-011/placebo treatment, patients will return to the site monthly for 3 follow-up assessments (i.e., Days 99, 113 and 141). Subsequently, the patient will return to the site for 4 post treatment follow-up assessments (i.e. Days 169, 197, 225, and 253). The patient will return to the site for a termination visit approximately 1 month after the post treatment follow-up period (Day 281). Patients will be discontinued from the ACE-011/placebo treatment for reasons listed in section 8.3 of the protocol, for unacceptable toxicity, if the chemotherapy regimen is discontinued or for disease progression that requires the initiation of another chemotherapy treatment.

9.5.2.1. ACE-011 Treatment Related Toxicity

Patients with grade ≥3 toxicity related to ACE-011 will be discontinued from ACE-011/placebo treatment except for the following. Patients with grade 3 hypertension that decreases to grade ≤ 1 within 7 days after treatment with anti-hypertensive therapy may continue on ACE-011/placebo treatment. Dose reduction rules will apply at the subsequent scheduled dosing day(s).

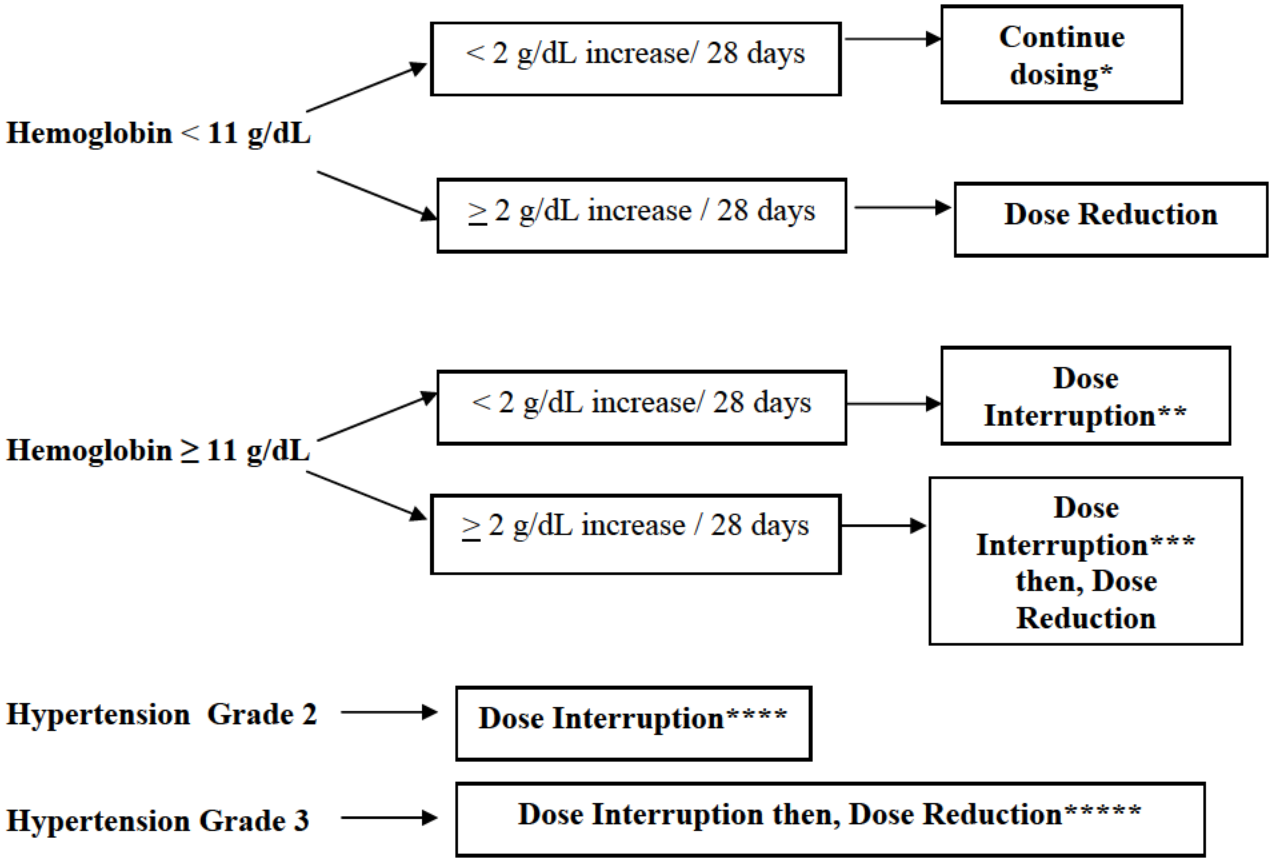
Patients with a hemoglobin level above the upper limit of normal at any time during the study after Day 1 will be discontinued. Patients who discontinue treatment with ACE-011/placebo will enter the treatment follow-up period.

9.5.2.2. ACE-011 Dose Modification Rules

Throughout the study blood pressure and hemoglobin values will be evaluated for patient safety. The local hemoglobin values and blood pressure values will be evaluated at each study visit and on each ACE-011/placebo dosing day. The hemoglobin values from the previous 28 days between each ACE-011/placebo dosing day and blood pressure will be reviewed and evaluated

for dose modifications. The parameters below outline rules for dosing modifications (Figure 1). Table 8 outlines the dose reduction levels of ACE-011.

Figure 1: ACE-011/Placebo Dose Modification Rules (after initial dose of ACE-011/placebo)



* Patients who have received a blood transfusion in the past 28 days should continue with dosing at the same ACE-011/placebo dosing level if the transfusion was given no sooner than 7 days from the dosing day and the hemoglobin level is < 11 g/dL and hypertension ≤ Grade 1 on the day of dosing

** ACE-011/placebo should be held until the following scheduled treatment visit

*** ACE-011/placebo should be held and at the following scheduled treatment visit, a dose reduction of ACE-011/placebo will be administered based on the evaluation of the hemoglobin and blood pressure at that time

**** ACE-011/placebo should be held until resolution to ≤ Grade 1 (NCI CTCAE v3 0) and may then be resumed at the following scheduled treatment visit.

***** ACE-011/placebo should be held until resolution to ≤Grade 1 (NCI CTCAE v3 0) within 7 days after treatment with anti- hypertensive therapy and may continue on a reduced dose of ACE-011/placebo treatment for the subsequent scheduled dosing day

Table 8: ACE-011/Placebo Dose Reduction Levels

When required per the dose modification rules above (Figure 1), ACE-011/placebo dose(s) should be reduced as follows for dose 2 (Day 29), dose 3 (Day 57) and/or dose 4 (Day 85):

Dose	Dose reduction #1	Dose Reduction #2	Dose Reduction #3
Placebo	N/A	N/A	N/A
0.1 mg/kg	0.05 mg/kg	0.03 mg/kg	0.01 mg/kg
0.3 mg/kg	0.1 mg/kg	0.05 mg/kg	0.03 mg/kg
0.5 mg/kg	0.3 mg/kg	0.1 mg/kg	0.05 mg/kg

Blood pressure and hemoglobin values must be evaluated at each dosing day for consideration of administration of ACE-011/placebo.

The dose level for the initial dose reduction will be maintained unless subsequent dose reductions need to be administered, upon evaluation of the dose modifications listed above. A patient may have up to 3 dose reductions in the study.

Dose reduction steps and the administration of the reduced dose(s) will be conducted in a manner that will preserve the blinded status of the original treatment group of each patient. Patients in the placebo group who are designated by the investigator to undergo a dose reduction will continue to receive placebo.

10. STUDY PROCEDURES AND SCHEDULE

10.1.1. Written Informed consent

Patients will be required to sign an Institutional Review Board/Institutional Ethics Committee (IRB/IEC) approved informed consent form (ICF) prior to any study related procedures, including screening evaluations.

Screen failure information will be maintained, including but not limited to, reason for failure.

10.1.2. Screening (Within 14 days of dosing)

The following will be collected within 14 days prior to the initial dosing Day 1. Confirmatory labs for eligibility criteria must be drawn and resulted within 7 days of Day 1.:

- Informed consent
- Assessment of inclusion/exclusion criteria
- Complete Medical History and Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), temperature (°C) and height.
- FACT-Fatigue survey
- 12 lead Electrocardiogram (ECG) performed at two time intervals (approximately one hour apart) in triplicate at 3 minute intervals (±1 min)
- Serum iron, total iron binding capacity (TIBC), transferrin and serum ferritin
- Vitamin B12 and serum folate levels
- Coagulation
- Serum chemistry
- Hematology
- LH, FSH, testosterone, progesterone, and estradiol
- Serum Pregnancy Test for females of childbearing potential- to be assessed within 7 days of Day 1.
- Urinalysis
- Evaluation of transfusion frequency (including history of transfusion up to 8 weeks prior to Day 1)
- Documentation of concomitant medications (any treatments taken 28 days prior to Day 1)

- Females of childbearing potential- collection of menstrual cycle information of the start and end date of the last menstruation.
- CT/MRI scan: Head, neck, thoracic, abdominal, and pelvic scans are to be acquired. A CT/MRI scan acquired within 4 weeks of initiating the current chemotherapy regimen may be used for the screening assessment. One measurable or non-measurable lesion per RECIST is to be identified by the scan.
    - Brain (head) CT only to be done to rule out CNS metastases in patients with suspicious clinical symptoms or prior history of CNS metastases.
- Dual-energy X-ray absorptiometry (DXA) of lumbar spine and hip
- Bone scans (only for patients with bone metastases) acquired within 4 weeks of initiating the current chemotherapy regimen may be used for the screening assessment.

10.1.3. Initial Dosing: Day 1

Patients will be dosed on Day 1. Results from screening evaluations must be reviewed prior to randomization to confirm patient eligibility. A patient will be considered randomized once a multi-digit patient identification number has been assigned to the patient through the IVRS/IWRS. After assignment of the patient identification number and the inclusion/exclusion criteria have been met, ACE-011/placebo administration may begin.

Chemotherapy regimen will be administered per standard of care at the site. The following tests must be performed prior to ACE-011/placebo administration:

- Confirm eligibility of patient by inclusion/exclusion criteria. The local hemoglobin value should be drawn prior to ACE-011/placebo administration.
- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- FACT-Fatigue survey
- 12 lead Electrocardiogram (ECG) performed at two time intervals (approximately one hour apart) in triplicate at 3 minute interval (±1 min)
- Serum iron, total iron binding capacity, transferrin and serum ferritin
- Coagulation
- Chemistry
- Hematology
- Serum erythropoietin marker

- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test
- Bone biomarkers- osteocalcin (OC), procollagen type 1 N-terminal propeptide (P1NP), serum bone specific alkaline phosphatase (BSAP), C-terminal type 1 collagen telopeptide (CTX), tartrate-resistant acid phosphatase isoform-5b (TRACP-5b), and urine N-telopeptide (uNTX).
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions the on collection of serum and urine for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Females of childbearing potential- Urine (dipstick) pregnancy test to be collected and resulted prior to ACE-011/placebo dosing
- Urinalysis
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events (after ACE-011/placebo administration)
- Females of childbearing potential-collection of menstrual cycle information of the start and end date of the last menstruation.
- Patients will be administered ACE-011/placebo before the chemotherapy regimen begins during the study visits.

After ACE-011/placebo and chemotherapy are administered, the patient may leave the clinic, based on the clinical judgment of the staff.

10.1.4. ACE-011/placebo administration: Days 29, 57, and 85 (± 3 days)

Chemotherapy regimen will be administered per standard of care at the site. ACE-011/placebo will be administered approximately every 28 days. The following procedures must be performed prior to ACE-011/placebo administration:

- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- FACT-Fatigue survey
- Serum iron, total iron binding capacity, transferrin and serum ferritin
- Coagulation
- Serum chemistry

- Hematology
- Serum erythropoietin marker
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test
- Bone biomarkers- OC, BSAP, P1NP, CTX, TRACP-5b, and uNTX
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions the collection of serum and urine for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Females of childbearing potential -Urine (dipstick) pregnancy test to be collected and resulted prior to ACE-011/placebo dosing
- Urinalysis (Day 85 only)
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events
- Females of childbearing potential- collection of menstrual cycle information of the start and end date of the last menstruation.

Hemoglobin (HgB) and blood pressure (BP) measurement must be assessed prior to ACE-011/placebo dose administration by review of local laboratory values.

10.1.5. Additional visits during Treatment Period: Days 8, 15, 22, 36 (± 1 day), 43, 64 and 71 (± 2 days)

Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- 12 lead ECG (Days 8, 36, and 64 only- to be done in triplicate at 3 minute intervals (±1 min)
- Serum folate (Day 36 only)
- Coagulation
- Serum chemistry

- Hematology
- Back-up blood sample to be drawn for future testing
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events
- CT/MRI scan (Day 64 only, +/- 5 day window). Same modality as acquired for baseline should be used for follow up scans. Tumor response assessments to be evaluated by the investigator.
- Bone scan (Day 64 only, +/- 5 day window). Follow-up scan to be acquired if performed at baseline.

If patient terminates the treatment period early, prior to the Day 85 of ACE-011/placebo, the patient will enter the treatment period follow up. However, if patient terminates ACE-011/placebo early and begins a new chemotherapy regimen, the patient should complete the Day 281/Termination visit procedures and be followed for survival only.

10.1.6. Treatment Period Follow-up visits : Days 99, 113 (± 3 days) and 141 (± 7 days)

Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Physical Examination (Days 113 and 141 only)
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- 12 lead ECG-including (Day 113 only- to be done in triplicate at 3 minute intervals (±1 min)
- Serum folate (Day 113 only)
- Coagulation
- Serum chemistry
- Hematology
- Serum erythropoietin marker (Day 141 only)
- LH, FSH, testosterone, progesterone, and estradiol (Days 113 and 141 only)
- Anti-drug antibody test (Days 113 and 141 only)

- Bone biomarkers- OC, BSAP, P1NP, CTX, TRACP-5b, and uNTX (Days 113 and 141 only)
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions on the collection of serum and urine for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events
- Females of childbearing potential- collection of menstrual cycle information of the start and end date of the last menstruation. (Days 113 and 141 only)
- CT/MRI scan (Day 113 only, +/- 5 day window). Same modality as acquired for baseline should be used for follow up scans. Tumor response assessments to be evaluated by the investigator.
- DXA of lumbar spine and hip (Day 113 only)
- Bone scan (Day 113 only, +/- 5 day window). Follow-up scan to be acquired if performed at baseline.

10.1.7. Post Treatment Period Follow-up visits : Days 169, 197, 225, and 253 (± 7 days)

If a CT/MRI and bone scan (if applicable) is performed as per standard of care during the post treatment period follow up, results may be collected within the CRF to capture the status of the disease. Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Hematology
- Evaluation of transfusion frequency
- Documentation of concomitant medications (up to 90 days from the last dose of ACE-011/placebo)
- Evaluation of adverse events (up to 90 days from the last dose of ACE-011/placebo)

10.1.8. Termination Visit: Day 281 (± 7 days)

Day 281 is the final visit. If a patient terminates the study early, the Day 281 procedures should be followed. Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- FACT-Fatigue survey
- 12 lead ECG (in triplicate at 3 minute intervals (±1 min)
- Serum iron, total iron binding capacity, transferrin and serum ferritin
- Serum folate
- Coagulation
- Serum chemistry
- Hematology
- Serum erythropoietin marker
- Anti-drug antibody test
- Bone biomarkers- OC, BSAP, P1NP, CTX, TRACP-5b, and uNTX
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions on the collection of serum and urine for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Females of childbearing potential -Urine (dipstick) pregnancy test
- Urinalysis
- Evaluation of transfusion frequency
- Documentation of concomitant medications (up to 90 days from the last dose of ACE-011/placebo)
- Evaluation of adverse events (up to 90 days from the last dose of ACE-011/placebo)
- DXA of lumbar spine and hip
- Bone scan (Day 281, +/- 5 day window). Follow-up scan to be acquired if performed at baseline.

If a patient has a positive anti-drug antibody result at Day 281/Termination visit, the patient may be asked to return to the clinical site for additional monthly anti-drug antibody testing, up to 12 months after their last dose.

10.2. Discontinuation of study

The Sponsor may terminate this study, after consultation with the Investigator, at any time for safety or administrative reasons. The Sponsor may terminate the study if the occurrence of serious adverse events (SAEs) or other findings suggest unacceptable risk to the health of the patients.

11. INVESTIGATIONAL PRODUCT MATERIALS AND MANAGEMENT

11.1. Reference Product

The placebo (control agent) to be used in this study will be sterile normal saline (0.9% Sodium Chloride for Injection) administered as a SC injection. Sterile, normal saline will be supplied by the investigational site's pharmacy. The manufacturer's directions for placebo storage and handling are to be followed, as are standard clinical practices for ensuring sterility of the placebo.

11.2. Investigational Product Packaging and Labeling

ACE-011 will be supplied in 2 mL clear glass vials with gray stoppers and red flip-top seals that contain 1 mL of ACE-011. The drug product consists of ACE-011 in PBS at a nominal concentration of approximately 50 mg/mL.

11.3. Investigational Product Storage

ACE-011 is recommended to be stored at ≤ -65℃.

11.4. Investigational Product Preparation

See Investigational Product Preparation Manual.

11.5. Administration

ACE-011/placebo will be administered by subcutaneous injection (SC). Subcutaneous injections will be given in the upper arm and/or thigh. Please refer to Investigational Product Handling and Administration document for further information.

Dose modifications will be determined by the investigator after reference to the dose modification rules listed in Figure 1. In the case of a dose interruption or dose reduction of ACE-011, the investigator or designee will notify the pharmacy staff, who is unblinded, of the treatment decision for appropriate preparation of the study drug to maintain the blind of the study.

11.6. Drug Accountability

Accountability for ACE-011/placebo is the responsibility of the Investigator. Investigational clinical supplies must be received by a designated person at the clinical site and kept in a secured location. The investigational site must maintain accurate records demonstrating dates and amounts of ACE-011/placebo received, to whom it was dispensed (patient-by-patient accounting), and accounts of any ACE-011/placebo accidentally or deliberately destroyed or returned. Unless otherwise notified, all vials of ACE-011/placebo, both used and unused, must be saved for drug accountability. The used vials may be discarded, per the institution's standard

practice, after drug accountability has been completed by the monitor. The Investigator must return all unused vials of ACE-011 to the Sponsor at the end of the study, or the ACE-011 may be destroyed at the clinical site with the permission of the Sponsor. For either scenario, the outcome must be documented on the drug accountability log. The Sponsor will provide direction for the outcome of all unused vials.

11.7. Investigational Product Handling and Disposal

Please refer to the Study Reference Guide, provided under separate cover, for detailed drug handling, administration, and storage instructions.

12. ASSESSMENT OF EFFICACY

Efficacy measurements will include assessments for hematopoietic response which is defined as an increase in hemoglobin of ≥ 0.5 g/dL, ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs, at each dose level. The hematopoietic endpoints will be determined by the monitoring of hematologic laboratory values and RBC transfusion and/or ESA administration. Secondary endpoints associated with assessment of disease progression will be evaluated by CT/MRI scan and bone mineral density by DXA scans, respectively. Bone biomarkers and incidence of skeletal related events (SREs) will also be evaluated for all patients. SREs are defined as pathologic fracture, radiation therapy to bone, surgery to bone, or spinal cord compression. Quality of life assessment (FACT-Fatigue) will be evaluated for each patient.

13. ASSESSMENT OF SAFETY

13.1. Safety Parameters

13.1.1. Adverse Events

All patients will be assessed for safety by monitoring adverse events, clinical laboratory tests, vital signs and physical examinations. AE information will be collected throughout the study. See Section 14 Adverse Events for a full description of AE definitions, AE and SAE reporting procedures, and emergency procedures.

13.1.2. Clinical Laboratory Tests

The following laboratory safety assessments will be drawn at the investigational site's local laboratory according to the laboratory collection recommendations. The samples will then be sent to the central lab for value reporting. Please refer to Lab Reference Manual for further information. Hemoglobin and hematocrit lab tests will be drawn and evaluated by the local laboratory at screening and at each visit day throughout the study. The local hemoglobin laboratory value will be used in the evaluation of ACE-011/placebo dose modifications. The sponsor may request additional safety tests be performed based on ongoing data review during the study.

- Iron studies: Serum iron, total iron binding capacity (TIBC), transferrin and serum ferritin on specified days.
- Nutritional tests: Serum folate and vitamin B12
- Coagulation (central lab only): prothrombin time (PT), partial thromboplastin time (PTT), and fibrinogen
- Serum chemistry (central lab only): AST, ALT, lactate dehydrogenase (LDH), total bilirubin, indirect bilirubin, alkaline phosphatase, total protein, albumin, sodium, potassium, chloride, carbon dioxide, calcium, phosphorus, blood urea nitrogen (BUN), creatinine, uric acid, glucose, amylase, and lipase.
- Hematology: complete blood count (CBC) with differential and platelet. CBC includes red blood cells (RBC), white blood cells (WBC), hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC). Reticulocyte percent, erythropoietin levels and peripheral blood smear will be collected on specified days.
- Bone biomarkers: (serum) osteocalcin (OC), procollagen type 1 N-terminal propeptide (P1NP), serum bone specific alkaline phosphatase (BSAP), C-terminal type 1 collagen telopeptide (CTX), tartrate-resistant acid phosphatase isoform-5b (TRACP-5b), and urine N-telopeptide (uNTX).
- Pregnancy test for females of child bearing potential

- Urinalysis (central lab only): including determination of pH, specific gravity, protein, glucose, ketones, blood, leukocyte esterase and nitrite. Microscopy required only to follow-up abnormal findings.

13.1.3. Other Safety Assessments

- Physical examinations
- Vital signs: weight (kg), heart rate (beats/min), seated blood pressure (mmHg) and temperature (°C). Height will be collected at screening only.
- FACT-Fatigue survey
- 12-lead ECG (done in triplicate)
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test
- CT/MRI scan
- DXA
- Bone scans (for those patients with baseline bone metastases)

13.2. Pharmacokinetics

See Appendix 5 for further details regarding PK sampling.

14. ADVERSE EVENT

Adverse Event

An adverse event (AE) is any untoward medical occurrence in a patient administered a pharmaceutical product, which does not necessarily have a causal relationship with the treatment. An AE can be any unfavorable and unintended sign (e.g., including an abnormal laboratory finding), symptom, or disease temporally associated with the use of the study drug, whether or not it is considered to be study drug related. This includes any newly occurring event or previous condition that has increased in severity or frequency since the administration of study drug (treatment-emergent).

Serious Adverse Event

A serious adverse event (SAE) is any AE, occurring at any dose level and regardless of causality that:

- Results in death.
- Is life-threatening: Life-threatening means that the patient was at immediate risk of death from the reaction as it occurred, (i.e., it does not include a reaction which hypothetically might have caused death had it occurred in a more severe form).
- Requires inpatient hospitalization or prolongation of existing hospitalization.
- Results in persistent or significant disability/incapacity.
- Is a congenital anomaly/birth defect.
- Is an important medical event: an important medical event is an event that may not result in death, be life-threatening, or require hospitalization but may be considered an SAE when, based upon appropriate medical judgment, it may jeopardize the patient and may require medical or surgical intervention to prevent one of the outcomes listed in the definitions for SAEs. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.
- A hospitalization for an elective procedure will not be considered a SAE.

SAEs will be captured throughout the patient participation in the study through Day 281/Termination visit. SAEs will be captured up to 90 days from the last dose of ACE-011/placebo for those patients who are followed for survival only.

Unexpected Adverse Event

An unexpected AE is an AE that is not reported in the Investigator Brochure or in the case of an AE already described, the severity of which is not described in the Investigator Brochure.

14.1. Adverse Event Classification

The following classifications should be used when evaluating the relationship of AEs and SAEs to the investigational drug (study drug).

None:	No relationship between the experience and the administration of study drug; related to other etiologies such as concomitant medications or patient's clinical state.
Unlikely:	The current state of knowledge indicates that a relationship is unlikely.
Possibly:	A reaction that follows a plausible temporal sequence from administration of the study drug and follows a known response pattern to the suspected study drug. The reaction might have been produced by the patient's clinical state or other modes of therapy administered to the patient.
Probably:	A reaction that follows a plausible temporal sequence from administration of the study drug and follows a known response pattern to the suspected study drug. The reaction cannot be reasonably explained by the known characteristics of the patient's clinical state or other modes of therapy administered to the patient.
Definitely:	A reaction that follows a plausible temporal sequence from administration of the study drug and follows a known response pattern to the suspected study drug and can be confirmed with a positive re-challenge test or supporting laboratory data.

Severity of AEs will be graded by the investigator using the National Cancer Institute (NCI; US) Common Terminology Criteria for Adverse Events (CTCAE) version 3.0 guidelines (Appendix 3).

14.2. Recording Adverse Events

Patients will be evaluated and questioned generally for AEs during the course of the study, starting from baseline (Day 1). The Medical History CRF should be comprehensive of patient's medical history up to Day 1. All AEs occurring after ACE-011/placebo administration up to 90 days from the last dose of ACE-011/placebo are to be documented on AE CRF. The specific disease or syndrome rather than individual associated signs and symptoms should be identified by the investigator and recorded on the CRF.

Skeletal related event information is to be captured and reported for the duration of the patient participation in the study through Day 281/Terminiation visit. SREs will be captured up to 90 days from the last dose of ACE-011/placebo for those patients who are followed for survival only.

Serious Adverse Events (SAEs)

For SAEs, a SAE form must be completed with as much information as possible and submitted in the time frame described in Section 14.3.

When new significant information is obtained as well as when the outcome of an event is known, the investigator should record the information on a new SAE form. If the patient was hospitalized, a copy of the discharge summary must be included as part of the patient medical file. In all instances, the investigator should follow up with patients until the outcome of the SAE is known.

14.3. Reporting Adverse Events

All AEs spontaneously reported by the patient and/or in response to an open question from study personnel or revealed by observation, physical examination or other diagnostic procedures will be recorded on the AE CRF. Any clinically relevant change in laboratory assessments or other clinical findings is considered an AE and must be recorded on the AE CRF. Clinically significant laboratory abnormalities are those that are identified as such by the investigator and/or those that require intervention. All AEs are to be followed until the event resolves or the clinical course is stabilized.

Serious Adverse Events

For SAEs, a SAE form must be completed with as much information as possible and submitted in the time frame described below.

All SAEs that occur during the course of the study (from the signing of the ICF and throughout patient participation in the study procedures) must be reported by the Investigator to PPD and Sponsor by faxing the SAE form within 24 hours from the point in time when the Investigator becomes aware of the SAE. All SAEs and deaths must be reported whether or not considered causally related to the study drug.

SAE Reporting:

PPD Medical Monitor
PPD Inc.

If there are serious, unexpected AEs associated with the use of the study drug, the Sponsor will notify the appropriate regulatory agency(ies) and all appropriate parties on an expedited basis. It is the responsibility of the Investigator to promptly notify the Institutional Review Board (IRB) of all unexpected SAEs involving risk to human patients.

14.4. Emergency Sponsor Contact

In the event of a medical emergency (i.e., an event that requires immediate attention regarding the treatment of a patient, operation of the clinical study, and/or the use of investigational drug), investigational site personnel must immediately contact the Medical Monitor.

15. STATISTICS

Statistics Analysis Overview

The study will be considered complete with regard to the primary endpoint once all eligible subjects have completed up to 2 months after the last dose of ACE-011. Patients will be randomized to one of three ACE-011 treatment groups (0.1, 0.3 and 0.5 mg/kg) or placebo with a ratio of 2:2:2:1. Patients will be stratified according to administration of either single agent weekly chemotherapy or less frequent chemotherapy regimens.

15.1. Determination of Sample Size

With an anticipated dropout rate of 20%, a sample size of 30 patients per dose level is estimated to achieve at least 80% power with 2 sided significance level of 0.05 to differentiate targeted response rate of p1=0.45 from null hypothesis p0=0.20. An additional 15 patients will be enrolled to the placebo group for safety comparison. A total of 105 patients will be randomized to three ACE-011 dose groups (0.1, 0.3, and 0.5 mg/kg) and placebo with a ratio of 2:2:2:1.

15.2. Analysis Populations

For this study, the following populations will be defined and used in the analysis and presentation of the data.

Modified Intent-to-Treat (MITT) population: The MITT population is defined as all patients randomized who received at least one dose of ACE-011/placebo.

Per-Protocol (PP) population: The PP population is defined as all patients randomized who have confirmed chemotherapy induced anemia, received at least one dose of ACE-011 and on study (treatment) for at least 2 months (57 days).

Safety population: The safety population is defined as all patients who received at least one dose of ACE-011 or placebo.

15.3. Background and Demographic Characteristics

Summary statistics (mean, standard deviation, median, minimum and maximum) will be provided for those variables measured on a continuous scale. The frequency distribution (number and percentage) will be provided for those variables measured on a nominal scale.

15.3.1. Patient Disposition

Patient disposition (analysis population allocation, entered, discontinued, along with primary reason for discontinuation) will be summarized using frequency and percent for each dose group. A summary of patients enrolled by site will be provided.

15.3.2. ACE-011

ACE-011 treatment exposure (total dosage received) will be summarized for each dose group as well as for each cycle for each dose level. Listings for dose adjustment will be provided.

15.3.3. Concomitant Therapy

By-patient listings will be provided for all concomitant medications and therapies taken during the study. All concomitant medications will be coded and categorized by the WHO drug coding system. Usage frequency of each coded concomitant medications will be summarized by each dose group.

15.4. Efficacy Evaluation

15.4.1. Primary Efficacy Analysis

The primary efficacy endpoint is the hematopoietic response rate at each dose level. It is defined as the proportion of patients who have a hematopoietic response of an increase in hemoglobin of ≥ 0.5 g/dL for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs. The ACE-011 treatment period is defined as from the time of first ACE-011 dose to up to 2 months after the last dose of ACE-011 treatment. To be considered as a responder, all hemoglobin measurements during a 28 consecutive day period or longer must have an increase of ≥ 0.5 g/dL from baseline. If RBC transfusions and/or ESAs are required, no hemoglobin measurements within 28 days will be used to determine the hemoglobin response. Primary efficacy analysis will be based on PP population. Primary efficacy analysis will also be conducted on MITT population as supportive to the PP based analysis. Patients who do not receive ACE-011 will be excluded from primary efficacy analysis. Patients who discontinue the ACE-011 treatment prematurely will be counted as non-responder in the calculation of response rate.

A 95% confidence interval (CI) for the response rate for each dose level will be provided to assess whether 95% CI of response rate contains null hypothesis response rate, p0=0.20. The response rate and 95% CI will also be provided for patients with chemotherapy on weekly basis versus those with chemotherapy on a less frequent basis for each dose level as well as overall (all three doses levels analyzed together).

An exploratory analysis with the Kaplan Meier approach will be considered to account for those patients who discontinue the ACE-011 treatment prematurely if the drop out rate is greater than 20%. Exploratory analysis of comparing hematopoietic response rates between active and placebo will be performed using Fisher's exact test. Due to the nature of the study (phase 2 dose determination study), no multiplicity adjustment will made for the multiple comparisons.

15.4.2. Secondary Efficacy Analyses

The secondary efficacy endpoints include:

- Proportion of patients achieving an increase in hemoglobin of ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for 28 consecutive days during the treatment period and up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs.
- Duration of hematopoietic response (days) defined as the first time hemoglobin increases at least 0.5 g/dL, ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL from baseline to the last time there is hemoglobin ≥ 0.5 g/dL, ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL increase from baseline. Duration of response is only calculated for a responder and will be at least 28 days. The duration of response will be at least 28 days is only calculated for a patient who meets the primary efficacy endpoint.
- Time to achieve hematopoietic response (days) defined as time from first dose of ACE-011 to the first time a hemoglobin result at least 0.5 g/dL, ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL increase from baseline and maintained for a period of at least 28 consecutive days.
- Proportion of patients receiving red blood cell (RBC) transfusions and/or ESAs in each dose group as well as within each cycle of each ACE-011 dose level.
- Objective tumor response rate for each dose level using RECIST criteria.
- Progression-Free Survival is defined as the time from randomization into the study to the first observation of documented disease progression or death due to any cause. If the subject has not progressed or died at the time of final analysis, PFS will be censored at the time of last tumor assessment.

The statistical analysis for binary endpoints will be similar to the primary efficacy analysis. For time to event endpoints, the Kaplan-Meier method will be used to estimate the survival curve and median time to event. These analyses will be performed for each dose group and all ACE-011 groups combined.

15.5. Safety Evaluation

Safety variables will be tabulated and presented for all patients who receive ACE-011. Adverse events will be coded using the Medical Dictionary for Regulatory Activities (MedDRA). Incidence of treatment-emergent adverse events will be presented by system organ class (SOC) and preferred term. Adverse events will also be presented by severity, and relationship to study drug. Change from baseline in clinical laboratory parameters and vital signs will be summarized across time. Shift tables will be presented for selected laboratory parameters. Physical examination results will be presented in listings.

Data from all patients who receive one or more doses of ACE-011/placebo will be included in the safety analyses. To assess clinical safety, adverse events, vital sign measurements and clinical laboratory information will be summarized by visit. Adverse events and laboratory results will also be summarized by severity grade (NCI CTCAE v3.0). Descriptive statistics will be generated and shift tables provided as appropriate.

15.6. Pharmacokinetic Evaluation

Listing of individual patient serum ACE-011 concentrations, actual blood sampling times and graphs of concentration vs. time will be prepared for each dose group. Summaries of PK parameters will be summarized by dose group. The trough concentrations will be summarized for all patients who provide pharmacokinetic samples by dose group. Exploratory analyses will be performed to correlate trough concentrations with efficacy, bone biomarker data, and safety data, however these analyses will be data-driven and only conducted if warranted by the data.

15.6.1. Anti-drug Antibody Data

The results of anti-drug and neutralizing antibodies will be presented over time. Exploratory analysis will be performed on the potential effect of anti-drug antibody on ACE-011 PK and drug exposure if anti-drug antibody tests are deemed positive.

15.7. Interim Analysis

There is no planned interim analysis for this study.

15.8. Deviation from Original Analysis Plan

Deviations from the statistical analyses outlined in this protocol will be indicated in the statistical analysis plan; any further modifications will be noted in the final clinical study report.

16. DIRECT ACCESS TO SOURCE DATA/DOCUMENTS

16.1. Study Monitoring

The Clinical Monitor will arrange to visit the Investigator sites at regular intervals during the study. The monitoring visits must be conducted according to the applicable ICH and GCP guidelines to ensure protocol adherence, quality of data, drug accountability, compliance with regulatory requirements, and continued adequacy of the investigational site and its facilities. During these visits, CRFs and other data related to the study will be reviewed and any discrepancies or omissions will be resolved. The Clinical Monitor will be given access to study relevant source documents (including medical records) for purposes of source data verification.

During and/or after completion of the study, quality assurance officers named by the Sponsor or the regulatory authorities may wish to perform on-site audits. The Investigator is expected to cooperate with any audit and provide assistance and documentation (including source data) as requested.

16.2. Audits and Inspections

The Investigators and clinical sites will permit trial related monitoring, audits, IRB review, and regulatory inspections as requested by FDA, the Sponsor or designee, including direct access to source data/documents (i.e., original medical records, laboratory reports, hospital documents, progress reports, signed ICFs, etc.) in addition to CRFs.

17. QUALITY CONTROL AND QUALITY ASSURANCE

17.1. Data Quality Control and Quality Assurance

17.1.1. Investigator responsibility

The investigator is responsible for ensuring the study is conducted according to the protocol, Code of Federal Regulations (CFR), GCP, and any other applicable regulatory requirements. The investigator responsibilities are outlined in these documents along with ensuring that documentation of a signed informed consent, is obtained prior to patient participation in the study.

17.1.2. Protocol modifications

The investigator may not modify the protocol without agreement from the Sponsor and prior review or approval by the IRB. Any deviations from the protocol should be documented by the investigator or designee.

17.2. Financing and Insurance

Financing and insurance are addressed in a separate document.

17.3. Confidentiality

To maintain patient privacy, all CRFs, study drug accountability records, study reports and communications will identify the patient by the assigned patient identification number. The Investigator will grant monitor(s) and auditor(s) from the Sponsor or designee and regulatory authority(ies) access to the patient's original medical records for verification of data gathered on the CRFs and to audit the data collection process. The patient's confidentiality will be maintained and will not be made publicly available to the extent permitted by the applicable laws and regulations.

All information regarding the investigational product supplied by the Sponsor to the Investigator is privileged and confidential information. The Investigator agrees to use this information to accomplish the study and will not use it for other purposes without consent from the Sponsor. It is understood that there is an obligation to provide the Sponsor with complete data obtained during the study. The information obtained from the clinical study will be used towards the development of the investigational product and may be disclosed to regulatory authority(ies), other Investigators, corporate partners, or consultants as required.

17.4. Publication Policy

All information concerning ACE-011 is considered confidential and shall remain the sole property of the Sponsor. The Investigator agrees to use this information only in conducting the study and shall not use it for any other purposes without the Sponsor's written approval. The Investigator agrees not to disclose the Sponsor's confidential information to anyone except to

people involved in the study that need such information to assist in conducting the study, and then only on like terms of confidentiality and nonuse.

It is understood by the Investigator that the information developed from this clinical study will be used by the Sponsor in connection with the development of ACE-011, and therefore may be disclosed as required to regulatory agencies. To allow for the use of the information derived from clinical studies, it is understood that there is an obligation to provide the Sponsor with complete test results and all data developed in the study.

No publication or disclosure of study results will be permitted except as specified in a separate, written, agreement between the Sponsor and the Investigator.

17.5. Protocol Amendments

Protocol amendments that impact patient safety change the scope of the investigation, or affect the scientific quality of the study must be approved by the IRB and submitted to the appropriate regulatory authorities before implementation of such modifications to the study.

In the event that the protocol needs to be modified immediately to eliminate an apparent hazard to a patient, the Sponsor will amend and implement the protocol change and subsequently notify the regulatory authorities and/or the IRB, as appropriate.

18. ETHICS

18.1. Institutional Review Board or Independent Ethics Committee

The Investigator must obtain written IRB approval of the protocol, approval for relevant supporting information and all types of patient recruitment and advertisement and ICF prior to starting the study. The IRB will meet all Food and Drug Administration (FDA) requirements governing IRBs (Code of Federal Regulations, Title 21, Part 56).

The Sponsor or designee must approve the ICF submitted to the investigational site's IRB. All patient recruitment and advertisements must be submitted to the Sponsor or designee prior to submission to the IRB, for review.

18.2. Ethical Conduct of the Study

The Sponsor and the Investigator must comply with all instructions, regulations, and agreements in this protocol and in the applicable International Conference on Harmonisation (ICH) and GCP guidelines, and must also conduct the study in accordance with local regulations.

18.3. Written Informed Consent

18.3.1. Informed Consent

Informed written consent is required from each patient prior to any testing under this protocol, including screening tests and evaluations. The consent form, as specified by the clinical site's IRB, must follow the Protection of Human Patients regulations listed in the Code of Federal Regulations, Title 21, Part 50.

The background of the proposed study and the benefits and risks of the procedures and study must be explained to the patients. It is the responsibility of the Investigator to obtain consent and to provide the patient with a copy of the signed and dated ICF. Confirmation of a patient's informed consent must also be documented in the patient's medical record prior to any testing under this protocol, including screening tests and evaluations.

All ICFs used in this study must be approved by the appropriate IRB and by the Sponsor or designee. The ICF must not be altered without the prior agreement of the relevant IRB and the Sponsor.

18.3.2. Patient Data Protection

Prior to any testing under this protocol, including screening tests and evaluations, patients must authorize the release and use of protected health information (PHI), as required by local law.

The patient will not be identified by name in the CRF or in any study reports. These reports will be used for research purposes only. The Sponsor, its designee, and various government health agencies may inspect the records of this study. Every effort will be made to keep the patient's personal medical data confidential.

19. DATA HANDLING AND RECORDKEEPING

19.1. Case Report Form Completion

CRFs will be completed for each randomized patient. It is the Investigator's responsibility to ensure the accuracy, completeness, and timeliness of the data reported in the patient's CRF. Source documentation supporting the CRF data should indicate the patient's participation in the study and should document the dates and details of study procedures, AEs, and patient status.

Investigators will maintain copies of the CRFs at the clinical site. For patients who discontinue or terminate from the study, the CRFs will be completed as much as possible, and the reason for the discontinuation or termination clearly and concisely specified on the appropriate CRF.

19.2. Retention of Records

The Investigator will maintain all study records according to ICH-GCP and applicable regulatory requirements. Records will be retained for at least 2 years after the last marketing application approval or 2 years after formal discontinuation of the clinical development of the investigational product or according to applicable regulatory requirements. If the Investigator withdraws from the responsibility of keeping the study records, custody must be transferred to a person willing to accept the responsibility. The Sponsor must be notified in writing if a custodial change occurs.

20. REFERENCES

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21. APPENDICES

Appendix 1: Response Evaluation Criteria in Solid Tumors

Response Evaluation Criteria in Solid Tumors (RECIST) v1.1	
Terms	Definition
Measurable disease	The presence of at least one measurable lesion by CT or MRI. If the measurable disease is restricted to a solitary lesion, its neoplastic nature should be confirmed by cytology/histology. All measurements should be taken and recorded in metric notation, using a ruler or calipers. All baseline evaluations should be performed as closely as possible to the beginning of treatment and never more than 4 weeks before the beginning of the treatment.
Measurable lesions	Lesions that can be accurately measured in least one dimension by CT scan with longest diameter $\geq$ 10 mm (CT scan thickness no greater than 5 mm). The same method of assessment and the same technique should be used to characterize each identified and reported lesion at baseline and during follow-up.
Non-measurable lesion	All other lesions, including small lesions (longest diameter <10mm or pathological lymph nodes with P10 to <15mm short axis) as well as truly non-measurable lesions. Lesions considered truly non-measurable include: leptomeningeal disease, ascites, pleural or pericardial effusion, inflammatory breast disease, lymphangitic involvement of skin or lung, abdominal masses/abdominal organomegaly identified by physical exam that is not measurable by reproducible imaging techniques.

RECIST Criteria-Response Evaluation

Evaluation of target lesions	
Complete Response (CR)	Disappearance of all target lesions. Any pathological lymph nodes (whether target or non-target) must have reduction in short axis to <10 mm.
Partial Response (PR):	At least a 30% decrease in the sum of diameters of target lesions, taking as reference the baseline sum diameters.
Stable Disease (SD):	Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum diameters while on study.
Progressive Disease (PD):	At least a 20% increase in the sum of diameters of target lesions, taking as reference the smallest sum on study (this includes the baseline sum if that is the smallest on study). In addition to the relative increase of 20%, the sum must also demonstrate an absolute increase of at least 5 mm. (Note: the appearance of one or more new lesions is also considered progression).

Evaluation of non-target lesions	
Complete Response (CR):	Disappearance of all non-target lesions and normalisation of tumour marker level. All lymph nodes must be non-pathological in size (<10mm short axis).
Non-CR/Non-PD	Persistence of one or more non-target lesion(s) and/or maintenance of tumour marker level above the normal limits.
Progressive Disease (PD):	Unequivocal progression (see comments below) of existing non-target lesions. (Note: the appearance of one or more new lesions is also considered progression).

Evaluation of best overall response	The best overall response is the best response recorded from the start of the treatment until disease progression/recurrence (taking as reference for PD the smallest measurements recorded since the treatment started). In general, the patient's best response assignment will depend on the achievement of both measurement and confirmation criteria. Patients with a global deterioration of health status requiring discontinuation of treatment without objective evidence of disease progression at that time should be classified as having "symptomatic deterioration". Every effort should be made to document the objective progression even after discontinuation of treatment. In some circumstances it may be difficult to distinguish residual disease from normal tissue. When the evaluation of complete response depends on this determination, it is recommended that the residual lesion be investigated (fine needle aspirate/biopsy) to confirm the complete response status.

Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, Dancey J, Arbuck S, Gwyther S, Mooney M, Rubinstein L, Shankar L, Dodd L, Kaplan R, Lacombe D, Verweij J.New response evaluation criteria in solid tumours: Revised RECIST guideline (version 1.1) (2009). European Journal of Cancer 45, 228-247.

Appendix 2: ECOG Performance Status

The ECOG scale is used to assess a patient's quality of life in an evaluation by a health professional of the daily activities and how the activities are affected by the disease of the patient.

Oken, M.M., Creech, R.H., Tormey, D.C., Horton, J., Davis, T.E., McFadden, E.T., Carbone, P.P. (1982) Toxicity And Response

Grade	ECOG
0	Fully active, able to carry on all pre-disease performance without restriction
1	Restricted in physically strenuous activity but ambulatory and able to carry out work of a light or sedentary nature, e.g., light house work, office work
2	Ambulatory and capable of all selfcare but unable to carry out any work activities. Up and about more than 50% of waking hours
3	Capable of only limited selfcare, confined to bed or chair more than 50% of waking hours
4	Completely disabled. Cannot carry on any selfcare. Totally confined to bed or chair
5	Dead

Criteria Of The Eastern Cooperative Oncology Group. Am J Clin Onco 5:649-655.

Appendix 3: National Cancer Institute (NCI) Common Terminology Criteria for Adverse Events (CTCAE) Version 3.0

See http://ctep.cancer.gov/reporting/ctc.html

Appendix 4: New York Heart Association- Classification of heart failure

Class 1 - Class 1 heart failure - no limitation of activities. No symptoms from ordinary activities.
Class 2 - Class 2 heart failure- mild limitation of activity. Comfortable with rest or mild exertion.
Class 3 - Class 3 heart failure- marked limitation of activity and be comfortable only at rest.
Class 4 –Class 4 heart failure- complete rest, confined to bed or chair; any physical activity brings on discomfort and symptoms occur at rest.

National Heart, Lung, and Blood Institute, National Institutes of Health. New York Heart Association Classification. 2008.

Appendix 5- Pharmacokinetic Sampling

Pharmacokinetics

Blood samples will be collected from approximately one-third of patients at selected study sites for the evaluation of serum concentrations of ACE-011. Pre-dose PK samples will be collected on Day 1 and Day 85.

Blood samples for PK should be collected in a fasted state (defined by no food or drink except water for at least 4 hours prior to the study procedure) with the exception of the Day 1 post 4 hour dose and the Day 85 post 4 hour dose. Pre-dose samples should be collected prior to ACE-011/placebo dosing (Day 1, 29, 57, and 85). The post 4 hour collection (± 10 minute window) on Day 1 and Day 85 is calculated to be 4 hours post the administration of ACE-011/placebo. Chemotherapy regimens scheduled will be administered to the patient as per standard of care. Collection, handling, and shipping procedures for blood samples are provided in the Study Reference Guide.

Schedule of Events cont.

	Screen	Treatment Period														Post Treatment Follow up Period				Term Visit[20]
ACE-011/Placebo Dose period		#1				#2			#3			#4	Follow-up							
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	99 (± 3d)	113 (± 3d)	141 (±7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
PK Sampling		X X- Post 4 hours	X	X	X	X	X		X	X		X X-Post 4 hours		X						
ACE-011/Placebo administration		X				X			X			X								
Chemo regimen		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

Protocol Amendment – Summary of Changes

CLINICAL STUDY PROTOCOL

A Phase 2, Double-Blind, Randomized, Placebo-Controlled Study of ACE-011 for the Treatment of Chemotherapy Induced Anemia in Patients with Metastatic Breast Cancer

STUDY DRUG: **ACE-011**

PROTOCOL NUMBER: **A011-08**

SPONSOR: **Acceleron Pharma, Inc.**
128 Sidney Street
Cambridge, MA 02139

PPD

MEDICAL MONITOR: **PPD Inc.**

PPD

PROTOCOL DATE: **13 March 2009**
AMENDMENT #1: **22 April 2009**
AMENDMENT #2: **21 May 2009**
AMENDMENT #3: **07 October 2009**

Summary:
Amendment 03 includes the following changes:

- Administrative changes
- Deletion of exclusion criteria- Concurrent use of bevacizumab with the chemotherapy regimen during study participation.
- Addition of SAE information from previous clinical studies
- Deletion of bone scan timepoints
- Amend CT/MRI scan and DXA scan information
    - CT/MRI- screening- Scans of the anatomical areas of head (if applicable), neck, thoracic, abdominal, and pelvic are to be available for evaluation. If one of the anatomical areas listed has not be acquired, prior to screening, then a CT/MRI must be done, during screening for this study, to ensure a scan of the head (if applicable), neck, thoracic, abdominal, and pelvic is available.
    - DXA- screening- Refer to Imaging Acquisition Guidelines for guidelines regarding washout period for contrast (if used) for radiological images, prior to collection of DXA
- Amend Progression Free Survival statistical analysis evaluation
- Primary endpoint hematopoietic value was changed from $\geq$ 0.5 g/dL to $\geq$ 1.0 g/dL
- Addition of section for "rescreening" patients

Protocol Location	Change
Section 2. Protocol Synopsis	*Objectives* o Added additional language defining the primary endpoint. o Deletion of $\geq$ 1g/dL increase variable from the second secondary endpoint. *Inclusion criteria* o Added "cytologically" and removed "documented by cytology or biopsy" from inclusion criteria "Histologically confirmed diagnosis of breast cancer documented by cytology or biopsy." o Added version number to RECIST text with the inclusion criteria. o Added "anticipated" instead of "planned" for treatment with a myelosuppressive chemotherapy criteria. *Exclusion criteria* o Deletion of "Concurrent use of bevacizumab with the chemotherapy regimen during study participation. o Addition of text "active gastrointestinal bleed (within the last 12 months as compared to Day 1)

	Criteria for evaluation o Amended the primary endpoint language defining the hematopoietic value from ≥ 0.5 g/dL to ≥ 1.0 g/dL. o Amended the secondary endpoint language based on the changes to the primary endpoint. o *Statistical methods- Efficacy* o Amended the primary endpoint language defining the hematopoietic value from ≥ 0.5 g/dL to ≥ 1.0 g/dL.
Section 2. Schedule of events Table 7: Schedule of events	o Formatting. o Deletion of timepoints for acquiring a bone scan on days 64 and 113. o Addition of the following text "Refer to Imaging Acquisition Guidelines for o guidelines regarding washout period for contrast (if used) for radiological o images, prior to collection of DXA" to the footnote for DXA scans. o Amended language regarding the baseline CT/MRI scans from 4 weeks to 6 weeks. o Addition of the following text "Follow up bone scan, **when the baseline bone scan was abnormal."** to the footnote for acquiring follow up bone scans. o Addition of text "All study visit day dates (inclusive of windows) are calculated from Day 1(day of initial ACE-011/placebo administration).
Section 5: Introduction	*Section 5.4 Clinical Experience* o Additional text added to update the SAE information for the A011-04 study.
Section 6. Trial Objectives and Purpose	*Section 6.1: Primary Objective* o Amended the primary endpoint language defining the hematopoietic value from ≥ 0.5 g/dL to ≥ 1.0 g/dL. *Section 6.2: Secondary Objectives* o Amended the secondary endpoint language based on the changes to the primary endpoint.

Section 7. Investigational Plan	o Additional text added regarding central and local lab values.
Section 8. Selection	*Section 8.2.1 Patient inclusion criteria* o Added "cytologically" and removed "documented by cytology or biopsy" from inclusion criteria "Histologically confirmed diagnosis of breast cancer documented by cytology or biopsy." o Added version number to RECIST text with the inclusion criteria. o Added "anticipated" instead of "planned" for treatment with a myelosuppressive chemotherapy criteria. *Section 8.2.2 Patient exclusion criteria* o Deletion of "Concurrent use of bevacizumab with the chemotherapy regimen during study participation. o Addition of text "active gastrointestinal bleed (within the last 12 months as compared to Day 1)
Section 9. Treatment of Patients	o Formatting. o Addition of "Rescreening" section. o Table 8 Amended.
Section 10. Study Procedures	Formatting. *Section 10.1.2 Screening (within 14 days of Day 1)* o *Amended the following language (in bold):* A CT/MRI scan acquired within **6** weeks of initiating the current chemotherapy regimen may be used for the screening assessment. One measurable or non-measurable lesion per RECIST v**1.1** is to be identified by the scan. o Added text – "Scans of the anatomical areas of head (if applicable), neck, thoracic, abdominal, and pelvic are to be available for evaluation. If one of the anatomical areas listed has not be acquired, prior to screening, then a CT/MRI must be done, during screening for this study, to ensure a scan of the head (if applicable), neck, thoracic, abdominal, and pelvic is available." o Added text- "Refer to Imaging Acquisition Guidelines for guidelines regarding washout period for contrast (if used) for radiological images, prior to acquiring DXA scan." *Section 10.1.4 ACE-011/placebo administration: Days 29, 57, and 85 (± 3 days)* o Added text- "Local Hgb must be done within 1 day prior to the ACE-011/placebo dosing day."

	Section 10.1.5 Additional visits during the Treatment Period: Days 8, 15, 22, 36 (± 1 day), 43, 64 and 71 (± 2 days) o Added text- *"*anatomical areas" to the following section: CT/MRI scan (Day 64 only, +/- 5 day window). Same modality and anatomical areas as acquired prior to enrollment should be used for follow up scans. Tumor response assessments to be evaluated by the investigator. o Deletion of bone scan text. *Section 10.1.6 Treatment Period Follow-up visits : Days 99, 113(± 3 days) and 141(± 7 days)* o Added text- *"*anatomical areas" to the following section: CT/MRI scan (Day 113 only, +/- 5 day window). Same modality and anatomical areas as acquired prior to enrollment should be used for follow up scans. Tumor response assessments to be evaluated by the investigator. o Deletion of text for bone scan. o Added text- "Refer to Imaging Acquisition Guidelines for guidelines regarding washout period for contrast (if used) for radiological images, prior to acquiring DXA scan." *Section 10.1.8 Termination Visit: Day 281 (± 7 days)* o Added text- "Refer to Imaging Acquisition Guidelines for guidelines regarding washout period for contrast (if used) for radiological images, prior to acquiring DXA scan." o Added text- "and the scan was abnormal" to the following: Bone scan (Day 281, +/- 5 day window). Follow-up scan to be acquired if performed at baseline.

Section 11. Investigational Product Materials and Management	o Added text: ACE-011 Handling and Administration Guidelines through out the section.
Section 14. Adverse Event	o Formatting. o Amended text "study drug" to investigational drug" throughout the section.
Section 15. Statistics	*Section 15.4.1 Primary Efficacy Analysis* o Amended the primary endpoint language defining the hematopoietic value from $\geq$ 0.5 g/dL to $\geq$ 1.0 g/dL. o Added text "with no sufficient hemoglobin measurement" *Section 15.4.2 Secondary Efficacy Analysis* o Amended the secondary endpoint language based on the changes to the primary endpoint. o Added the following language to the Progression Free Survival section- "the start date of current chemotherapy regimen"

CLINICAL STUDY PROTOCOL

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AMENDMENT #2 : **21 May 2009**
AMENDMENT #3 : **07 October 2009**

Signature Page

Acceleron Pharma Approval

Signature: PPD **Date:** 9 Oct 2007

Name (print): _____

Investigator Agreement: I have read the protocol and agree to conduct the study as outlined in the protocol. The study will be conducted in accordance to current United States Food and Drug Administration (FDA) regulations, International Conference of Harmonization (ICH) Guidelines, Good Clinical Practices (GCP), and local ethical and legal requirements.

Signature: _____ **Date:** _____

Name (print): _____

Institution Name and Address:

_____

_____

_____

PROCEDURES IN CASE OF EMERGENCY

Table 1: Emergency Contact Information

Role in Study	Name	Address and Telephone number
PPD Medical Monitor	PPD	PPD Inc. 1800 Perimeter Park Drive, Suite 275 Morrisville, NC 27560-7200 USA PPD
Clinical Trial Manager	PPD	Acceleron Pharma, Inc. 128 Sidney Street Cambridge, MA 02139 USA PPD

2. PROTOCOL SYNOPSIS

Name of Sponsor/Company: Acceleron Pharma, Inc.
128 Sidney Street
Cambridge, MA 02139

Name of Investigational Product: ACE-011

Name of Active Ingredient:
ACE-011 is a fully human fusion protein consisting of the extracellular domain (ECD) of the activin receptor IIA (ActRIIA) linked to the human IgG1 Fc domain.
Mechanism of Action: ACE-011 is a disulfide-linked, glycosylated, dimeric protein. ACE-011 competes with the activin receptor IIA that binds a number of TGF-β superfamily ligands including activin, myostatin (growth differentiation factor [GDF]-8), and GDF-11.

Title of Study: A Phase 2, Double-Blind, Randomized, Placebo-Controlled Study of ACE-011 for the Treatment of Chemotherapy Induced Anemia in Patients with Metastatic Breast Cancer.

Study center(s): approximately 25-35 centers

Phase of development: 2

Objectives

Primary:

1. To evaluate the proportion of patients in each ACE-011 dose regimen who achieve a hematopoietic response during the treatment period including up to 2 months after the last dose of ACE-011 treatment of chemotherapy induced anemia in patients with metastatic breast cancer. Hematopoietic response is defined as an increase in hemoglobin of ≥ 1.0 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs.

Secondary:

1. To evaluate the safety and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer.
2. To evaluate the percentage of patients who achieve an increase in hemoglobin of, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs.
3. To estimate the duration of the hematopoietic response associated with each ACE-011 dose regimen.
4. To estimate the time to the hematopoietic response associated with each ACE-011 dose regimen.

5. To estimate the proportion of patients receiving red blood cell (RBC) transfusions and/or ESAs within each cycle of ACE-011 with each dose regimen.
6. To estimate the effect of each ACE-011 dose regimen on progression free survival (PFS) and objective response rate (ORR).

Exploratory objectives:

1. To estimate the effect of each ACE-011 dose regimen on serum bone biomarkers (OC, BSAP, P1NP, CTX, TRACP-5b), urine bone biomarker (uNTX), bone mineral density (DXA scans of lumbar spine and hip) and incidence of skeletal related events (SREs).
2. To evaluate the quality of life in relation to the treatment of chemotherapy induced anemia in patients with metastatic breast cancer during the study.

Methodology: This is a Phase 2, double-blind, randomized, placebo-controlled study to evaluate the efficacy, safety, and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer. Patients will be randomized to one of three treatment arms or placebo:

1. ACE-011 0.1 mg/kg subcutaneously (SC) every 28 days for up to 4 doses.
2. ACE-011 0.3 mg/kg SC every 28 days for up to 4 doses.
3. ACE-011 0.5 mg/kg SC every 28 days for up to 4 doses.
4. Placebo SC every 28 days for up to 4 doses.

Number of patients (planned): 105 patients

Diagnosis and main criteria for inclusion

Inclusion Criteria:

1. Women ≥ 18 years of age.
2. Histologically or cytologically confirmed diagnosis of breast cancer.
3. Evidence of metastatic breast cancer with a minimum of one measurable lesion or non measurable lesion per RECIST 1.1 criteria (Appendix 1).
4. Receiving a multicycle myelosuppressive chemotherapy regimen including one of the following: anthracycline, taxane, gemcitabine or vinorelbine.
5. Anticipated treatment with the same myelosuppressive chemotherapy regimen for a minimum of 9 weeks after Day 1 of ACE-011/placebo administration.
6. Hemoglobin value between ≥ 6.5 to < 11.0 g/dL (≥ 65 to < 110 g/L).
7. ≥ 30 days must have elapsed (from Day 1) since previous treatment with erythropoiesis-stimulating agent (including concurrent treatment with IV iron) for chemotherapy induced anemia.
8. ≥ 7 days have elapsed (from Day 1) since the last RBC blood transfusion and receipt of ≤ 2 units of blood in the past 30 days.
9. ECOG Performance status of 0 – 2 (Appendix 2).
10. Adequate renal (creatinine ≤ 1.5 x ULN or creatinine clearance ≥ 40 mL/min) and hepatic function (bilirubin ≤ 1.5 x ULN; AST/ALT ≤ 2.5 x ULN).
11. Females of childbearing potential participating in the study are to use adequate birth control measures (i.e. oral contraceptives or two forms of barrier method birth control) during study participation. Females of childbearing potential must have a negative serum pregnancy test within 7 days to ACE-011/placebo administration (Day 1). A female of childbearing potential is a sexually mature woman who has not undergone a hysterectomy or who has not been postmenopausal for at least 24 consecutive months (i.e. who has had menses at some time in the preceding 24 months).
12. Life expectancy of ≥ 6 months.
13. Is willing to adhere to the study visit schedule, understand and comply with all protocol requirements.
14. Understand and sign a written informed consent.

Exclusion Criteria:

1. At the time of screening patients who have any grade ≥ 3 toxicity (as per NCI CTCAE v3.0 (Appendix 3)) (except for the following disease related toxicities: hematological events- anemia, thrombocytopenia or neutropenia or non hematological events- nausea, vomiting, fatigue, or muscle or bone/joint pain).
2. Prior radiation therapy to > 20% of the whole skeleton. Use of palliative radiation if the area being treated is <15% of body surface area with no pelvic radiation or no more than 10% of the bone marrow reserve is radiated is permitted during patient participation in the study, at the discretion of the investigator.
3. Have received more than 5 prior chemotherapy treatment regimens for metastatic breast cancer.
4. Clinically significant pulmonary, endocrine, neurologic, gastrointestinal, hepatic or genitourinary disease unrelated to underlying hematologic disorder.
5. Patients with heart failure as classified by the New York Heart Association (NYHA) classification of 3 or higher (Appendix 4).
6. History of thrombosis, deep vein thrombosis (DVT), pulmonary emboli, or embolic stroke.
7. Untreated CNS metastases (exception: CNS metastases treated with whole brain radiotherapy > 6 months prior to randomization).
8. Diagnosis of a myeloid malignancy or known history of myelodysplasia.
9. History of second malignancy within 5 years (except excised and cured basal cell carcinoma, squamous cell carcinoma of the skin or cervical carcinoma in situ).
10. Patients with a recent history (within 14 days of Day 1) of administration of systemic (IV or oral) antibiotics. Patients should be free of infection at least 14 days after the last dose of systemic antibiotic treatment (from Day 1).
11. Uncontrolled hypertension. If hypertension is considered clinically stable, systolic blood pressure (BP) must be < 150 mmHg and diastolic BP must be < 100 mmHg.
12. Known history of hepatitis C antibody (HCV), hepatitis B surface antigen (HBsAg and HB core Ab), or human immunodeficiency virus (HIV) antibody.
13. Deficiency in iron (serum ferritin < 100 ng/mL (< 224.7 pmol/L)), vitamin B_{12}, or folate.
14. History of anemia as a result of inherited hemoglobinopathy such as sickle cell anemia or thalassemia.
15. History of autoimmune or hereditary hemolysis or active gastrointestinal bleeding (within the previous 12 months as compared to Day 1).
16. Patient received treatment with another investigational drug or device within 1 month prior to Day 1, or if the half life of the previous product is known, within 5 times the half life prior to dosing, whichever may be longer.
17. Pregnant or lactating females.
18. History of severe allergic or anaphylactic reactions or hypersensitivity to recombinant proteins or excipients in the investigational product.
19. Major surgery within 30 days prior to Day 1 (patients must have completely recovered from any previous surgery prior to Day 1).

Investigational product, dosage and mode of administration:

ACE-011/placebo will be administered as a SC injection, approximately every 28 days, on days 1, 29, 57, and 85 of the treatment period. Each patient will be randomized to one of three treatment arms or placebo:

Dose	Number of Patients
0.1 mg/kg	30
0.3 mg/kg	30
0.5 mg/kg	30
Placebo	15
Total patients enrolled: 105	

Patients with grade ≥ 3 toxicity related to ACE-011 will be discontinued from ACE-011/ placebo treatment except for the following: Patients with grade 3 hypertension that decreases to grade ≤ 1 within 7 days after treatment with anti-hypertensive therapy may continue on ACE-011/placebo treatment. Dose reduction rules will apply at the subsequent scheduled dosing day(s). Patients with a hemoglobin level above the upper limit of normal at any time during the study after Day 1 will be discontinued.

ACE-011/placebo dose modification rules are based on hemoglobin values and blood pressure measurements during each treatment cycle. ACE-011/placebo dose interruption and dose reduction rules will be implemented for those patients with hemoglobin of ≥ 11 g/dL, an increase of ≥ 2 g/dL/ 28 days, or hypertension ≥ Grade 2 within each cycle (see Figure 1).

Duration of study: Patients will be followed for a minimum of 7 months following their last dose of ACE-011/placebo. Study participation for each patient is approximately 10.5 months (14 day screening period, 3 month treatment period with a 2 month treatment follow up period, and a 5 month post treatment follow-up period). The start date of the chemotherapy regimen used in this protocol to treat metastatic breast cancer will be recorded. If a patient begins a new chemotherapy regimen during the study, the patient will discontinue further ACE-011/placebo treatment, complete the Day 281/Termination visit procedures and be followed for survival only. If a patient has a positive anti-drug antibody result at the Day 281/Termination visit, they may be asked to return to the clinical site for additional monthly repeat anti-drug antibody testing for up to 12 months, following the last dose of ACE-011/placebo. The study is complete when the last patient enrolled has completed the last protocol required follow up visit.

Criteria for evaluation

Efficacy: The primary endpoint is to establish the proportion of patients who have a hematopoietic response of an increase in hemoglobin of ≥ 1.0 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs.

The proportion of patients with an increase in hemoglobin of ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for 28 consecutive days during the treatment period up to 2 months

after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs will be evaluated as a secondary efficacy endpoint.

The hematopoietic endpoints will be determined by the monitoring of hematologic laboratory values, RBC transfusion and/or ESAs administration. The evaluation of response will be from the first dose of ACE-011 through 2 months after the last dose (for each patient). Additional endpoints will be evaluated with the assessment of disease progression by CT/MRI and bone scan and bone mineral density by DXA scans. Bone biomarkers will be evaluated in all patients and incidence of SREs will be recorded, as applicable. SREs are defined as pathologic fracture, radiation therapy to bone, surgery to bone, or spinal cord compression.

Safety: Throughout the study, safety will be evaluated by the sponsor, medical monitor and principal investigator. Additionally, the DMC will review safety parameters for patients enrolled in the study. All patients will be assessed for safety by monitoring adverse events, clinical laboratory tests, assessment of quality of life, vital signs and physical examinations on an ongoing basis. ECGs will also be performed during screening, prior to dosing on the first cycle, during the first, second and third cycle, Day 113 (28 days after the last dose of ACE-011) and at Day 281/Termination visit to monitor safety. Blood pressure and lab values will be assessed at each study visit during the treatment and treatment follow up period.

Pharmacokinetics: The blood samples will be collected at selected study sites from approximately one-third of patients for the evaluation of serum concentrations of ACE-011. Pre-dose PK samples will be collected on Day 1. See Appendix 5 for PK sampling schedule.

Statistical methods

Sample size determination: With an anticipated dropout rate of 20%, a sample size of 30 patients per dose level is planned to achieve at least 80% power with 2 sided significance level of 0.05 to differentiate targeted response rate of p1=0.45 from null hypothesis p0=0.20. An additional 15 patients will be enrolled into the placebo group for safety comparison. A total of 105 patients will be randomized to three ACE-011 dose groups or placebo with a ratio of 2:2:2:1.

Efficacy analysis: The primary endpoint is the proportion of patients who have a hematopoietic response of an increase in hemoglobin of ≥ 1.0 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs. The proportion of responders will be determined together with 95% confidence interval.

For binary endpoints, the proportion of responders and 95% confidence interval will be calculated. For time to event endpoints, the Kaplan-Meier method will be used to estimate the survival curve and median time to event. Due to the nature of the study, no multiplicity adjustment will be made for efficacy analysis.

Safety analysis: Data from all patients who receive at least one dose of ACE-011/placebo will be included in the safety analyses. To assess clinical safety, adverse events, vital sign measurements and clinical laboratory information will be summarized. Adverse events and laboratory results will also be summarized by severity grade (NCI CTCAE v3.0). Changes from baseline clinical laboratory values and vital signs will be summarized over time. Descriptive statistics and shift tables will be generated as appropriate.

Schedule of Events [21]

	Screen	Treatment Period														Post Treatment Follow up Period				Term Visit[20]
ACE-011/placebo Dose period		#1				#2			#3			#4	Follow-up							
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	99 (± 3d)	113 (± 3d)	141 (± 7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
Informed consent	X																			
Inclusion/exclusion criteria	X	X[2]																		
Medical history	X																			
Physical examination	X	X[2]				X[2]			X[2]			X[2]		X	X					X
Vital signs[1]	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X					X
FACT-Fatigue	X	X[2]				X[2]			X[2]			X[2]								X
12 lead ECG[3]	X	X	X				X			X				X						X
Serum iron, TIBC, transferrin and serum ferritin	X[8]	X[2]				X[2]			X[2]			X[2]								X
Vitamin B12	X[8]																			
Serum folate	X[8]						X							X						X
Coagulation/ Serum chemistry[4]	X[8]	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X					X
Hematology[5]	X[8]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Serum Erythropoietin marker		X[2]				X[2]			X[2]			X[2]			X					X
LH, FSH, testosterone, progesterone, estradiol	X	X[2]				X[2]			X[2]			X[2]		X	X					
Anti-drug antibody testing [6]		X[2]				X[2]			X[2]			X[2]		X	X					X
Bone biomarker[7]		X[2]				X[2]			X[2]			X[2]		X	X					X
Back up blood sample for future testing		X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X					X
Pregnancy Test	X[8]	X[9]				X[9]			X[9]			X[9]								X
Urinalysis[10]	X	X[2]										X[2]								X
Evaluate transfusion frequency	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X	X	X	X	X	X
Documentation of conmeds	X[11]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X			

Schedule of Events cont.

	Screen	Treatment Period														Post Treatment Follow up Period				Term Visit[20]
ACE-011/Placebo Dose period		#1				#2			#3			#4	Follow-up							
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	99 (± 3d)	113 (± 3d)	141 (±7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
Evaluation of AEs and SAEs[12]	X-SAE only	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X-SRE and SAE only	X-SRE and SAE only	X-SRE and SAE only	X
Evaluation of Menstrual cycle[13]	X	X[2]				X[2]			X[2]			X[2]		X	X					
CT/MRI scan	X[14]									X[15]				X[15]						
Dual-energy X-ray absorptiometry (DXA[16])	X													X						X
Bone scans[17]	X																			X
ACE-011/placebo administration[18]		X				X			X			X								
Chemo regimen[19]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

[1] Vital signs include weight, heart rate, seated systolic and diastolic blood pressure, and temperature will be assessed at each visit Height will be collected at screening only

[2] On dosing days (1, 29, 57 and 85), study procedure to be done prior to dosing of ACE-011/placebo

[3] Screening and Day 1- At two different time intervals (approximately 1 hours apart) triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min), prior to dosing as applicable On all other specified days, one time interval of triplicate (12 lead) ECGs is to be performed at 3 minute intervals (± 1min),,

[4] Coagulation (central lab only) prothrombin time (PT), partial thromboplastin time (PTT), fibrinogen Chemistry (central lab only): AST, ALT, lactate dehydrogenase (LDH), total bilirubin, indirect bilirubin, alkaline phosphatase, total protein, albumin, sodium, potassium, chloride, carbon dioxide, calcium, phosphorus, blood urea nitrogen (BUN), creatinine, uric acid, glucose, amylase, and lipase

[5] Hematology (local lab) hemoglobin and hematocrit On dosing days (1, 29, 57 and 85), local hemoglobin and hematocrit values are to be drawn and resulted (up to 1 day) prior to dosing to apply dose modification rules for ACE-011/placebo Hematology (central lab only) complete blood count (CBC) with differential and platelet, CBC includes red blood cells (RBC), white blood cells (WBC), hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), Peripheral blood smear will also be done on Days 1, 29, 57, 85, and at the termination visit **Reticulocyte should be done on days 1, 8, 15, 22, 29, 36, 57, 64, 85, and at the termination visit.**

[6] If a patient has a positive anti-drug antibody result at the Termination visit, they may be asked to return to the clinical site for additional monthly follow-up, for up to 12 months after their last dose for a repeat anti-drug antibody test

[7] Both serum and urine samples will be collected at each visit The urine sample should not be from the first morning urine

[8] Labs may be drawn within 1 day prior to Day 1 Confirmatory labs for eligibility criteria must be drawn and resulted within 7 days of Day 1

[9] For females of child bearing potential only urine (dipstick) pregnancy test to be collected and resulted prior to dosing of ACE-011/placebo

[10] Urinalysis to be performed at screening, prior to administration ACE-011/placebo (Days 1 and 85) and termination visit Assessments (central lab only): pH, specific gravity, protein, glucose, ketones, blood, leukocyte esterase, and nitrite Microscopy required only to follow-up abnormal findings

[11] Screening- any concomitant treatments taken 28 days prior to day 1 including up to 90 days after the last dose of ACE-011/placebo

[12] AEs will be reported from day 1 post dose including up to 90 days after the last dose of ACE-011/placebo Additional AEs noted after 90 days from the last dose of ACE-011/placebo will be reported at the discretion of the investigator SREs and SAEs will be followed through out the study If a patient is followed for survival only, SREs and SAEs will be captured up to 90 days from the last dose of ACE-011/placebo

[13] Females of child bearing potential- collection of menstrual cycle information) start and end date of last menstrual period since the last assessment

[14] A CT/MRI scan acquired within 6 weeks of initiating the current chemotherapy regimen may be used for the screening assessment Head, neck, thoracic, abdominal, and pelvic scans are to be acquired One measurable or non measurable lesion per RECIST is to be identified by the scan Brain (head) CT only to be done to rule out CNS metastases in patients with suspicious clinical symptoms or prior history of CNS metastases

[15] Follow-up CT/MRI scans may be acquired +/- 5 day window for both Day 64 and Day 113 Additional CT/MRI scans acquired during the post treatment follow up period will be performed at the discretion of the investigator

[16] Hip and lumbar spine BMD assessment Refer to Imaging Acquisition Guidelines for guidelines regarding washout period for contrast (if used) for radiological images, prior to collection of DXA

[17] Bone scans will be acquired within 4 weeks of initiating the current chemotherapy regimen for those patients who have bone metastases identified Follow up bone scan, when the baseline bone scan was abnormal may be acquired +/- 5 day window for Day 281

[18] All study visit day dates (inclusive of windows) are calculated from Day 1(day of initial ACE-011/placebo administration) Patients who discontinue ACE-011/placebo due to ACE-011/placebo related toxicities (section 9 6 2 1) will enter the treatment follow-up period

[19] Chemotherapy regimens will be administered to the patient as per standard of care Continuation of the same chemotherapy regimen may be done through the follow up period of the study at the discretion of the investigator

[20] Patients who terminate treatment early due to disease progression and/or begin a new chemotherapy regimen will complete the Day 281/Termination visit procedures and be followed for survival only (7 months from the last dose of ACE-011/placebo) If a patient withdraws from the study (section 8 3), the Day 281/Termination visit procedures are to be completed and the patient will be followed for survival If withdrawal reason is due to ESA administration or ACE-011 related toxicity, the patient should enter the treatment follow up period

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4. LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

The following abbreviations and specialist terms are used in this study protocol.

Table 2: Abbreviations and Specialist Terms

Term	Definition
ACTH	Adrenocorticotropic hormone
ActRIIA	Activin receptor IIA
AE	Adverse event
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
AUC	Area under the plasma concentration - time curve
$AUC_{(0\text{-last})}$	AUC from time 0 to time of last quantifiable sample
$AUC_{(0\text{-}\infty)}$	AUC extrapolated to infinity
BMD	Bone mineral density
BP	Blood pressure
BSAP	Bone specific alkaline phosphatase
BUN	Blood urea nitrogen
CBC	Complete Blood Count
CFR	Code of Federal Regulations (US)
CH	Constant domain
CI	Confidence interval
CIA	Chemotherapy induced anemia
CL	Clearance
CL/F	Total clearance
C_{max}	Maximum observed concentration
CNS	Central nervous system
CO_2	Carbon dioxide
CRF	Case report form
CRO	Clinical research organization
CT	Computed tomography
CTCAE	Common Terminology Criteria for Adverse Events
CTX	Serum C-terminal type I collagen telopeptide
DLT	Dose-limiting toxicity
DXA	Dual-energy x-ray absorptiometry
DMC	Data Monitoring Committee
DVT	Deep vein thrombosis
ECD	Extracellular domain
ECG	Electrocardiogram
ECOG	Eastern Cooperative Oncology Group
EDF	Erythroid differentiation factor
ESA	Erythropoiesis-stimulating agents
F	Bioavailability (absolute)
FACT	Functional Assessment of Chronic Illness
FDA	Food and Drug Administration
FSH	Follicle-stimulating hormone
GCP	Good Clinical Practices
GH	Growth hormone

Term	Definition
HBV	Hepatitis B virus
Hct	Hematocrit
HCV	Hepatitis C virus
Hgb	Hemoglobin
HIV	Human immunodeficiency virus
ICF	Informed consent form
ICH	International Conference on Harmonization
IEC	Independent Ethics Committee
IND	Investigational New Drug
IP	Intraperitoneal
IRB	Institutional Review Board
IV	Intravenous
IVRS	Interactive Voice Response System
Kd	Binding coefficient
λz	Elimination rate constant
LDH	Lactate dehydrogenase
LH	Luteinizing hormone
MCH	Mean corpuscular hemoglobin
MCHC	Mean corpuscular hemoglobin concentration
MCV	Mean corpuscular volume
MedDRA	Medical Dictionary for Regulatory Activities
MITT	Modified Intent to Treat (population)
MRI	Magnetic resonance imaging
NOAEL	No adverse events level
NYHA	New York Heart Association
OC	Osteocalcin
ORR	Objective response rate
OVX	Ovariectomized
PD	Progressive disease
PFS	Progression free survival
PHI	Protected health information
P1NP	Serum intact procollagen type I N terminal propeptide
PK	Pharmacokinetic
PP	Per-protocol (population)
PT	Prothrombin time
PTH	Parathyroid hormone
PTT	Partial thromboplastin time
RAP-011	Murine version of ACE-011, ActRIIA mFc
RBC	Red blood cell (count)
RECIST	Response Evaluation Criteria in Solid Tumors
SAE	Serious adverse event
SAP	Statistical analysis plan
SC	Subcutaneous
SERM	Selective estrogen receptor modulator
SHAM	Sham-operated
SPEP	Serum protein electrophoresis
SRE	Skeletal-related event
T1/2	Elimination half-life
TIBC	Total iron binding capacity
Tmax	Time to Cmax

Term	Definition
TRACP-5b	Serum tartrate-resistant acid phosphatase isoform-5b
ULN	Upper limit of normal
Vz/F	Volume of distribution
WBC	White blood cell (count)

5. INTRODUCTION

5.1. Indication and Rationale

This study is designed to evaluate the efficacy, safety, and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer.

Treatment of patients with metastatic breast cancer with myelosuppressive chemotherapy is frequently associated with anemia. Chemotherapy induced anemia (CIA) is a significant problem for patients with cancer, causing fatigue and reduced quality of life (1). Patients with CIA are currently treated with blood transfusion and/or erythropoiesis-stimulating agents (2). However, with these treatment options CIA is still an area of unmet medical need. Erythropoiesis-stimulating agents (ESAs) can successfully mitigate transfusion need in a proportion of these patients, but concerns have emerged regarding apparent negative effects on survival and/or tumor progression in certain patient populations. The murine surrogate to ACE-011, RAP-011, has been tested as a single agent in breast cancer cell lines MDA-MB-231 and MCF-7 and no effect on enhanced proliferation of these cell lines has been observed in vitro. Therefore, treatment with ACE-011 may provide a distinct benefit/risk profile to patients with chemotherapy induced anemia.

In both a Phase 1 single dose and multiple dose study of ACE-011 in postmenopausal women, increases in hemoglobin and hematocrit were observed following ACE-011 treatment and remained elevated over the course of study. The observed hemoglobin and hematocrit effects of ACE-011 were dose and time dependent. Please refer to the Investigator Brochure for further information.

Based on the effect of ACE-011 on hematopoiesis and consistent biological phenomena observed in both non clinical and clinical studies, it is hypothesized that the blockade of ActRIIA receptor signaling impedes the terminal differentiation step in erythropoiesis to allow additional rounds of cell replication before cells enter the final differentiation phase. The result is a substantial increase in mature erythrocytes released into the circulation. Since this proposed mechanism is different to that of known ESAs, ACE-011 may provide a different clinical profile in the treatment of CIA.

This study will provide information on the pharmacodynamics (PD) properties regarding the ability of ACE-011 to increase hemoglobin in patients with CIA.

5.2. Description of ACE-011

ACE-011 (ActRIIA-IgG1) is a fully human fusion protein consisting of the extracellular domain (ECD) of activin receptor IIA (ActRIIA) linked to the human IgG1 Fc domain, which includes the heavy chain hinge and constant domains, CH2 and CH3. ACE-011 avidly binds to activin A with a binding coefficient (Kd) approximately 8.9 pM and prevents its binding to endogenous receptors, thereby inhibiting biological effects of activin.

5.3. Activin Biology

Activin A was initially identified as a gonadal differentiation factor involved in modulating follicle stimulating hormone (FSH) secretion from the pituitary (3). Subsequently, the pleiotropic nature of activin A has become more apparent (4). There is a growing body of data suggesting a role for activin A in bone remodeling, specifically as a negative regulator of bone growth (5). Before the two molecules were shown to be identical (6), activin A was also described as erythroid differentiation factor (EDF), effecting red blood cells in the later stages of maturation (7). The mechanism(s) by which activin A influences erythropoiesis remain under investigation and, in fact, there are data from studies in vitro and in animals that support erythropoiesis-stimulatory (8,9) and -inhibitory effects (10).

At the cellular level, activin A binds initially to the high-affinity Type II receptor (ActRIIA). The ligand-bound ActRIIA then recruits the low-affinity Type I receptor (ActRIA or ALK-4). The receptor heterocomplex, through its cytoplasmic protein kinase activity, then activates the Smad signal cascade to eventually influence nuclear transcriptional factors (11, 12). The competitive binding of activin A in the blood by the ACE-011 soluble fusion protein can result in inhibition of ActRIIA receptor signaling pathway by impeding biological processes attributed to activin A.

Activin has also been attributed to erythroid differentiation and has been reported to have proerythrocytic effects and to induce terminal differentiation of RBCs. Inhibition of activin may lead to increases in proliferation of the erythroid lineage. Based on its pharmacologic effects, ACE-011 (ActRIIA-IgG1) is being developed for the treatment of bone loss associated with various disease states (e.g., osteoporosis and treatment of osteolytic lesions in patients with multiple myeloma), and treatment of anemia associated with a variety of disorders, such as chemotherapy induced anemia.

The extracellular domain sequence of ActRIIA is completely conserved among numerous species including mouse, rat, cynomolgus monkey and humans, thus ACE-011 is active in these animals. However, in order to reduce the potential immunogenicity of the fully human molecule, ACE-011, in mice and to maximize the opportunity to maintain exposures in chronic models, a murine surrogate molecule was constructed by exchanging the human immunoglobulin Fc sequence portion of ACE-011 with its murine IgG2a homolog. The resultant construct is referred to as RAP-011 (ActRIIA-mIgG2aFc).

5.3.1. Pharmacology Studies

RAP-011 was evaluated in a broad range of animal pharmacology studies to assess the effects of inhibition of activin A on the biological processes attributed to that regulatory protein. RAP-011 has been shown to have significant effects on the red cell compartment. RAP-011 treatment of mice at 10, 30 and 50 mg/kg by intraperitoneal injection (IP) twice per week for 3 months resulted in a 16-26% increase in red blood cell (RBC) counts compared to control animals. Rats treated with RAP-011 at 0.3, 3 and 30 mg/kg once per week for 3 months showed RBC count increases of 6-15% over control animals. Finally, in cynomolgus monkeys treated with 10, 30 or

50 mg/kg of ACE-011 twice per month for 3 months there was a 21- 24% increase in RBC counts compared to controls animals. In this study, the RBC count increase was apparent as early as 2 weeks following the initial dose of ACE-011.

While the mechanism(s) underlying the stimulation effect of ACE-011 on erythropoiesis are not yet fully understood, it is hypothesized that a blockade of ActRIIA receptor signaling impedes the terminal differentiation step in erythropoiesis to allow additional rounds of erythroid cell replication before cells enter the terminal differentiation phase. The result is a substantial increase in mature erythrocytes released into circulation.

RAP-011 administered at 10 mg/kg to mice three days prior to administration of paclitaxel at 30 mg/kg was sufficient to prevent decreases in red blood cell parameters seen three days later. Mice receiving paclitaxel alone had decreased hematocrit from 43% to 38% three days later. RAP-011 administered 3 days prior to paclitaxel injection was sufficient to keep the hematocrit above 42% at three days and up to two weeks after paclitaxel administration. Therefore prophylactic treatment with RAP-011 was able to prevent paclitaxel induced anemia in mice.

RAP-011 has also been shown to significantly increase bone mineral density and strength in normal animals and in a variety of animal models of bone loss (13, 14, 15). In a murine ovariectomy (OVX)-induced model of osteopenia, RAP-011 (10 mg/kg IV, twice per week x 12 weeks) increased trabecular BMD > 25% in 6-12 weeks compared to the phosphate buffered saline (PBS)-treated controls ($p<0.002$). RAP-011 treatment was able to reverse the accelerated bone loss associated with ovariectomy. As expected sham-operated (SHAM) mice (i.e., intact ovaries) started the study with a greater trabecular BMD than OVX mice. PBS-treated SHAM mice showed a 10% decrease in trabecular BMD after 12 weeks of treatment compared to baseline, while SHAM-RAP-011 mice had a 32% increase in trabecular BMD over the same period. These data demonstrate that RAP-011 is able to increase trabecular BMD in both the normal state and in a model of established bone loss. The femurs of both the RAP-011 treated OVX and SHAM mice showed statistically significant increases in all measured parameters of bone strength (i.e., maximum load, stiffness, energy, ultimate strength, and elastic modulus). In summary, treatment with RAP-011 produced bone of superior quality consistent with an anabolic agent.

RAP-011 was evaluated in a mouse model of skeletal metastasis using luciferase-tagged, human MDA-MB-231 breast cancer cells (estrogen receptor negative). Mice were pretreated for 2 weeks with RAP-011 (10 mg/kg, biweekly, SC) prior to intracardiac injection of tumor cells. Treatment with RAP-011 was continued for an additional 5 weeks following tumor implantation. A parallel study was conducted to examine the effect of RAP-011 treatment, as described above, on survival of MDA-MB-231 bearing mice.

The data demonstrate that RAP-011 treatment acts to inhibit MDA-MB-231 metastasis to bone and may also play a role in inhibiting tumor growth in soft tissue. Furthermore, in the presence of tumor cells, RAP-011 is able to prevent lytic disease through an anabolic mechanism of bone formation. Lastly, inhibition of tumor growth and metastasis with RAP-011 leads to a greater than 25% survival benefit in this model.

The efficacy of RAP-011 was examined in an orthotopic model of breast cancer using luciferase-tagged human MCF-7 breast cancer cells (estrogen receptor positive). Mice were pretreated for 2 weeks with RAP-011 (10 mg/kg, biweekly, SC) prior to the implantation of tumor cells into the mammary fat pad of female nude mice. Treatment with RAP-011 was continued for an additional 7 weeks following tumor implantation at which time mice were sacrificed and assessed for tumor burden. RAP-011 effectively lowered the tumor burden in mice as detected by bio-luminescence. In addition, a 60% reduction in tumor weight was observed in the RAP-011 treated mice.

The ability of RAP-011 to repair osteolytic lesions caused by metastatic disease was investigated in mice. In this model athymic nude mice received an intratibial injection of MDA-MB-231-Luc cells to mimic bone metastasis. Mice were treated with a chemotherapy regimen of paclitaxel (20 mg/kg, IP, every three days) starting seven days after tumor injection and continuing for the duration of the study. On study day 42 mice with a detectable tumor burden by bioluminescent imaging were divided into two groups and began treatment with RAP-011 (10 mg/kg, SC, biweekly) or vehicle. Prior to dosing, μCT scans of the tumor bearing tibia were conducted to identify osteolytic lesions. RAP-011 treatment continued for four weeks (to study day 70) and a final μCT scan of the tibia was performed. On study day 70 osteolytic lesions remained the same or had progressed further when compared to study day 42 in the vehicle treated mice. While some mice showed progression of osteolytic lesions (most likely related to tumor burden) on study day 70, a majority of mice treated with RAP-011 demonstrated repair of the osteolytic lesions seen on study day 42. Therefore treatment with RAP-011 has the ability to repair osteolytic lesions caused by tumors and after cytoxic chemotherapy with paclitaxel.

5.3.2. Toxicology

ACE-011 has been evaluated for toxicological effects in two species, Sprague-Dawley rats and cynomolgus monkeys. The dose levels ranged from 0.3 to 30 mg/kg in rats and from 1 to 50 mg/kg in monkeys. These dose ranges were designed to support Phase 1a single-dose levels of 0.01 to 3.0 mg/kg IV and Phase 1b multiple doses of 0.1 to 2 mg/kg (monthly, SC). Weekly dosing in animals was designed to provide continuous, but fluctuating serum concentrations of ACE-011, which would be mimicked by a one-month dosing interval in humans.

Initial studies utilized IV dosing to identify target organs while longer-term studies (3 months in rats; 3 and 6 months in monkeys) utilized SC dosing to mimic the intended dosing regimen for patients. The expected pharmacologic effect, increased red blood cells (RBCs), hemoglobin (Hb), hematocrit (HCT) and reticulocytes, was observed in all studies, presumably based on the ability of ACE-011 to inhibit activin. A second expected pharmacologic effect of ACE-011, based on inhibition of activin, was the reversible reduction in sperm production and testicular tubular damage in rats. Since male cynomolgus monkeys were sexually immature in these studies, it was not possible to monitor this effect in this species.

Effects on the adrenal gland were only seen in rats and were more pronounced in female rats. This toxicological observation may not be relevant for predicting effects in humans since adrenal toxicity was not seen in cynomolgus monkeys. Dose-limiting toxicity and no observable adverse effect levels (NOAELs) were primarily based on renal toxicity in both rats and monkeys. There

was some indication that kidney toxicity was an indirect effect, based on the formation of antibody/antigen complexes, but since these studies were not designed to investigate toxicological mechanisms, a definitive cause of renal impairment and renal damage was not determined. Development of antibodies to ACE-011 was noted in all of these studies, which is an expected immune reaction in rats and monkeys dosed with a human protein.

The no observable adverse effect level (NOAELs) from the 3 month SC studies was 3 and 30 mg/kg in rats and monkeys, respectively. Because the kidney findings were observed at all dose levels, the NOAEL from the 6 month monkey study is < 10 mg/kg.

5.4. Summary of Clinical Experience

5.4.1. A011-01: A Phase 1a Study in Healthy Postmenopausal Women

ACE-011 was first studied in a randomized, Phase 1a, single dose, dose escalation study in healthy, post-menopausal females (16). ACE-011 was diluted in normal saline administered as an IV infusion (over approximately 1 hour) or as a SC injection on Day 1. Dose levels were 0.01, 0.03, 0.1, 0.3, 1.0, and 3.0 mg/kg IV and 0.03 and 0.1 mg/kg SC. A total of 48 subjects were enrolled; 5 active and 1 placebo at each of the IV and SC dose levels. All subjects were followed for 4 months following a single dose administration.

The pharmacokinetics of ACE-011 was linear for all IV and SC doses. Across IV doses ranging from 0.1 through 3.0 mg/kg, the mean clearance (CL) ranged from 0.092 to 0.128 mL/h/kg, the volume of distribution ranged from 73.7 to 110 mL/kg, and the mean T1/2 ranged from 23.7 to 31.8 days, with no apparent dependence on dose. After SC administration of 0.03 and 0.1 mg/kg, ACE-011 was completely absorbed, and the mean T1/2 was approximately 30 days, with no apparent dependence on dose.

The most commonly occurring treatment-emergent adverse events (AEs; i.e., those occurring in more than 1 subject in any treatment group) were headache, infusion site reaction, injection site hemorrhage, and toothache. The majority of the injection site reactions and injection site hemorrhages were in the first IV cohort and were related to infiltration of the IV site. The majority of treatment-emergent AEs were mild in severity and were judged to be unrelated to ACE-011. No deaths, serious AEs (SAEs), or AEs leading to discontinuation were reported. Changes in red blood cells (RBCs), hemoglobin, reticulocytes, liver function tests, glucose, uric acid, amylase and lipase occurred in some subjects; however, none of these laboratory results were reported as adverse events. Mild, transient elevations in pancreas enzymes, liver enzymes, or glucose were reported as AEs in five patients. There were no clinically significant changes from baseline in vital signs, physical examination, endocrine function, or electrocardiogram (ECG) data. Preservation of adrenal cortical function was monitored through evaluation of serum electrolytes (sodium and potassium levels) and through cortisol response to adrenocorticotropic hormone (ACTH) stimulation. No clinically significant findings were observed at doses up to 3.0 mg/kg IV, and ACE-011 was well tolerated in healthy, post-menopausal female volunteers in single dose levels up to 0.1 mg/kg SC and 3.0 mg/kg IV, the highest dose levels tested in this study.

5.4.2. A011-02: A Phase 1b Study in Healthy Postmenopausal Women

ACE-011 was studied in a Phase 1b, single-center, randomized, double-blind, placebo-controlled, multi-dose, dose-escalating study to evaluate the safety, tolerability and pharmacodynamics of ACE-011 in healthy postmenopausal women. Four cohorts of 10 subjects each were planned at the following dose levels: 0.1, 0.3, 1.0, and 2.0 mg/kg administered subcutaneously. Within each cohort, subjects were to be randomized to either active or placebo treatment in an 8:2 ratio. Subjects were to receive one SC injection of ACE-011 or placebo every 28 days for a total of 4 doses. All subjects were to be followed for 12 weeks after the last dose.

The treatment phase of the study was terminated early after a dose-limiting pharmacodynamic effect was observed. While on study, one subject in the 1.0 mg/kg cohort experienced a Serious Adverse Event (SAE) of progressive and persistent hypertension that was attributed to a rapid and significant rise in hemoglobin and hematocrit levels. Due to symptoms of headaches, nausea, eye pain, dizziness and vomiting approximately 1 week following the second dose, the subject was hospitalized for monitoring and evaluation of her hypertension. The head MRI, fundoscopy, and ECG performed were reported as normal and headache symptoms resolved following therapeutic phlebotomy. The SAE resolved and the subject was discharged from the hospital the following day. The hypertension was monitored closely and managed initially with antihypertensive medication. The hypertension resolved by the end of the study without need of any antihypertensive medication. The subject received aspirin prophylactically until the end of the study and continued taking ibuprofen as needed for headaches. Further details can be found in the Investigator Brochure.

Based on the magnitude of the hematopoietic response, the Sponsor suspended dose escalation to the 2.0 mg/kg dose level and further dosing in all cohorts as a result of this dose-limiting pharmacodynamic effect at the 1.0 mg/kg dose level. In total, 31 subjects were enrolled and treated. Dose levels of ACE-011 administered included 0.1, 0.3, and 1.0 mg/kg (Cohorts 1, 2, and 3 respectively). All subjects randomized to active treatment in Cohort 1 received all 4 planned doses of ACE-011. Due to early discontinuation of study drug, subjects randomized to active treatment in Cohort 2 received 3 doses of ACE-011, and subjects randomized to active treatment in Cohort 3 received 2 doses of ACE-011. Subjects randomized to placebo treatment received between 1 and 4 doses of study treatment.

In the analysis of the unblinded data, after the administration of the first dose, a dose and time dependent increase in hemoglobin values was observed (see Table 3 below):

Table 3: A011-02: A Phase 1b Study in Healthy Postmenopausal Women: Hemoglobin Evaluation

Evaluation Time Point	Mean Change from Baseline (g/dL)			
	Placebo N=7*	ACE-011 0.1 mg/kg N=8	ACE-011 0.3 mg/kg N=8	ACE-011 1.0 mg/kg N=8
Baseline Mean, pre-Dose 1	13.20	13.11	13.30	12.71
Day 8	0.17	0.68	0.85	1.21
Day 15	-0.27	0.43	0.44	1.75
Day 29	0.27	0.61	1.21	2.68**
Day 36	0.56	0.64	1.89	2.96
Day 43	-0.02	0.89	1.21	2.85
Day 57	0.27	1.28	1.64**	2.09
Day 64	0.53	1.11	2.49	2.21
Day 71	-0.10	1.34	2.09	1.66
Day 85	0.38	1.18**	2.55	1.86
Day 92	0.00***	1.04	1.60***	3.80***
Day 99	-0.20	1.21	3.20***	1.28***
Day 113	-0.02	1.30	1.29	1.04
Day 141	0.30	0.95	0.34	2.30
Day 169	0.20***	0.06		2.00***

*The number of placebo subjects with data decreases over time as a result of the early discontinuation of the study (i.e., there were placebo subjects in each dosing cohort). There were seven placebo subjects with data at baseline, Days 8, 15, and 36; six subjects with data at Days 29, 43, 57, and 85; five subjects with data on Days 71 and 113; four subjects with data on Day 64; three subjects with data on Day 141; two subjects with data on Day 99; and one subject with data on Days 92 and 169.

** Number of doses administered per treatment group: 0.1 mg/kg 4 doses; 0.3 mg/kg 3 doses; 1.0 mg/kg 2 doses. Data beyond this study day are considered follow-up results.

***n=1

No severe or life-threatening events were reported. The incidence of the most common treatment-emergent AEs (i.e., occurring in more than one subject in any treatment group) is presented below (see Table 4). The most notable AEs were those related to increases in hematologic laboratory measures in the 1.0 mg/kg dose group: hematocrit, hemoglobin, and RBC count. AEs of increased hemoglobin and/or hematocrit were reported for seven of the eight subjects in this dose group. These events were reported as mild or moderate elevations; all were considered probably related to study drug treatment. Three of the subjects in the 1.0 mg/kg group with elevated hemoglobin levels underwent phlebotomies and all hemoglobin elevations were resolved by the end of the follow-up period. No erythroid lineage AEs were reported in the 0.1 or 0.3 mg/kg treatment groups.

Table 4: A011-02: Incidence of Most Common Adverse Events

System Organ Class and Preferred Term	Dose Group			
	Placebo N=7 n (%)	0.1 mg/kg N=8 n (%)	0.3 mg/kg N=8 n (%)	1.0 mg/kg N=8 n (%)
GASTROINTESTINAL DISORDERS				
Diarrhea	0 (0.0%)	2 (25.0%)	0 (0.0%)	0 (0.0%)
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS				
Asthenia	0 (0.0%)	1 (12.5%)	2 (25.0%)	0 (0.0%)
Fatigue	2 (28.6%)	0 (0.0%)	1 (12.5%)	2 (25.0%)
INFECTIONS AND INFESTATIONS				
Viral upper respiratory tract infection	0 (0.0%)	4 (50.0%)	0 (0.0%)	0 (0.0%)
INJURY, POISONING AND PROCEDURAL COMPLICATIONS				
Limb injury	0 (0.0%)	1 (12.5%)	0 (0.0%)	2 (25.0%)
INVESTIGATIONS				
Hematocrit increased	0 (0.0%)	0 (0.0%)	0 (0.0%)	6 (75.0%)
Hemoglobin increased	0 (0.0%)	0 (0.0%)	0 (0.0%)	7 (87.5%)
Red blood cell count increased	0 (0.0%)	0 (0.0%)	0 (0.0%)	3 (37.5%)
METABOLISM AND NUTRITION DISORDERS				
Decreased Appetite	1 (14.3%)	0 (0.0%)	2 (25.0%)	0 (0.0%)
MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS				
Arthralgia	2 (28.6%)	0 (0.0%)	2 (25.0%)	0 (0.0%)
Muscle spasms	0 (0.0%)	1 (12.5%)	0 (0.0%)	2 (25.0%)
NERVOUS SYSTEM DISORDERS				
Dizziness	0 (0.0%)	0 (0.0%)	2 (25.0%)	2 (25.0%)
Headache	2 (28.6%)	4 (50.0%)	3 (37.5%)	2 (25.0%)
Paresthesia	1 (14.3%)	0 (0.0%)	0 (0.0%)	3 (37.5%)
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS				
Oropharyngeal pain	2 (28.6%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
VASCULAR DISORDERS				
Hot flush	0 (0.0%)	2 (25.0%)	0 (0.0%)	1 (12.5%)

Note: Table presents AEs reported in more than one subject in any treatment group.

Headache was reported frequently in all treatment groups with no apparent dose response trend, though the reports in the higher dose groups were more often considered possibly related to treatment. Most headaches were reported as mild (one headache was reported as moderate).

Paresthesia and dizziness were reported more frequently in the ACE-011 groups, though the events were generally not considered drug related. Other frequently reported events (e.g. fatigue, upper respiratory infection) did not appear to increase with dose and were generally mild.

Hemoglobin levels for all subjects with elevations had returned to within normal limits by the end of the study.

Adrenal cortical function was monitored through evaluation of serum electrolytes (sodium and potassium levels) and through cortisol response to adrenocorticotropic hormone (ACTH) stimulation. All ACTH simulation test results were normal.

PK results confirmed that the PK of ACE-011 is linear following the first SC doses of all three dose levels tested, 0.1, 0.3, and 1.0 mg/kg. The terminal half life ($T_{1/2}$) of ACE-011 following the last dose in all three dose groups was identical, with mean $T_{1/2}$ being approximately 23 days. The mean CL/F ranged from 3.05 to 3.90 mL/d/kg, the mean Vz/F ranged from 97.47 to 103.03 mL/kg, and the mean $T_{1/2}$ ranged from 20.92 to 23.34 days in all 3 dose levels, with no apparent dependence on dose.

Bone mineral density was assessed by dual-energy X-ray absorptiometry (DXA). Mean change and mean percent change in BMD results from baseline to study end are summarized in the table below. A dose-dependent increase in the BMD of the total hip from baseline to end of study was observed, with a significant and rapid increase of 2.4% in the 1.0 mg/kg dose group, compared to a 0.7% decrease in the placebo group. BMD results for lumbar spine showed slight increases of 0.4% to 1.0% from baseline to study end in all active treatment groups, compared with a 0.5% decrease in the placebo group (see Table 5 below):

Table 5: A011-02: A Phase 1b Study in Healthy Postmenopausal Women: BMD

	Treatment Group							
	Placebo N=7		**ACE-011 0.1 mg/kg N=8**		**ACE-011 0.3 mg/kg N=8**		**ACE-011 1.0 mg/kg N=8**	
	Mean Change	Mean Percent Change	Mean Change	Mean Percent Change	Mean Change	Mean Percent Change	Mean Change	Mean Percent Change
Lumbar Spine (g/cm^2)	-0.0020	-0.5%	0.0099	0.7%	0.0082	1.0%	0.0051	0.4%
Total Hip (g/cm^2)	-0.0062	-0.7%	0.0050	0.6%	0.0075	0.9%	0.0220	2.4%

Number of doses administered and day of last dose per treatment group: 0.1 mg/kg 4 doses (Day 85); 0.3 mg/kg 3 doses (Day 57); 1.0 mg/kg 2 doses (Day 29). Data beyond this study day are considered follow-up results.

5.4.3. A011-04: A Phase 2a Study in Patients with Osteolytic Lesions of Multiple Myeloma

Study A011-04 is an ongoing, non-IND, Phase 2a, multi-center, randomized, multiple-dose study to evaluate the safety, tolerability and efficacy of ACE-011 in patients with osteolytic lesions of multiple myeloma. Safety evaluations include adverse events (AEs), clinical laboratory tests, standard 12-lead electrocardiogram (ECG), vital signs, Eastern Cooperative Oncology Group

(ECOG) performance status and physical examinations. Additionally, the study includes the assessment of biochemical markers of bone formation and resorption, skeletal related events (SREs), bone mineral density (BMD) by duel-energy x-ray absorptiometry (DXA) and bone pain by visual analog scale (VAS).

In this study, patients are randomized in a 4:1 ratio to one of three dose levels of ACE-011 (0.1, 0.3 and 0.5 mg/kg) or placebo, to be administered to patients every 28 days by subcutaneous injection, for up to four doses over a 3-month period. The test article is being evaluated in combination with the anti-myeloma therapy of melphalan (4 mg/m^2 on days 1-7), prednisolone (40 mg/m^2 on days 1-7) and thalidomide (100 mg per day) (MPT). The sites, Sponsor and Sponsor representatives are blinded to treatment assignment.

As of 10 March 2009, 30 patients have been randomized, enrollment is complete and treatment is ongoing. Fifteen patients have received multiple doses of their assigned treatment; 7 patients have received 4 doses, 3 patients have received 3 doses and 5 patients have received 2 doses. Of the remaining 15 patients, 12 patients have received their first dose of ACE-011 or placebo and 3 patients have not yet received their first dose. There have been no ACE-011 related serious adverse events (SAEs) reported. Per the monitored preliminary data on 18 patients presented to the Independent Data Monitoring Committee (DMC) on 9 February 2009, there were 8 Grade 3/severe AEs reported in 4 patients, including neutropenia, platelet count decreased, pain in extremity, back pain and anemia. All Grade 3 AEs were determined to be not related to ACE-011 and the Grade 3 hematologic adverse events were considered either probably or definitely related to the MPT treatment.

Following preliminary analysis of the blinded central laboratory data, increases in hemoglobin values were observed within 28 days after administration of the first dose of ACE-011/placebo. Per data available, 11 out of 30 patients had $\geq$ 1 g/dL increase in hemoglobin within the first 28 days on the study, 4 patients achieved a $\geq$ 2 g/dL increase within their first 28 days on study, while 8 patients achieved a $\geq$ 2 g/dL increase in hemoglobin through Day 85.

Including all visits as of 10 March 2009 through Day 85, 9 out of 30 patients had a dose interruption due to a hemoglobin value of > 13 g/dL according to the dose modification rules defined in the protocol and 1 of the 9 patients, who had a history of hypertension, also had a dose interruption due to Grade 2 hypertension. Three of the 9 patients that required a dose interruption due to a hemoglobin value of > 13 g/dL, as per protocol, had hemoglobin levels above 13 g/dL prior to their first dose. Taken together, these data, suggest a beneficial pharmacodynamic effect of ACE-011 on erythropoiesis in a patient population with cancer chemotherapy induced anemia. No patients have had a dose reduction in ACE-011/placebo treatment or discontinued treatment.

One (1) subject died on PPD due to sudden death (including progression of disease). PPD The subject had received the first dose of ACE-011/placebo and started cycle 1 MPT on PPD The last study dose was administered on PPD On PPD the patient developed grade 2 blood pressure increase, which resolved with the patient's blood pressure medication. The following day (PPD), the patient's blood pressure in

the morning was back to baseline. The patient then died on PPD, 18 days after last study dose, due to the described sudden death. No autopsy was performed. The Investigator assessed event causality as possibly related to ACE-011/placebo and probably related to MPT.

One SAE of atrial fibrillation with syncope occurred resulting in study discontinuation. This was in a patient with prior history of Chronic Atrial Fibrillation. On further review this patient's inclusion into the study was a protocol violation. The Investigator assessed event causality as unrelated to ACE-011/placebo and possibly related to MPT.

One SAE of prolonged hospitalization due to pneumonia, that was considered not related to ACE-011, was reported in a PPD enrolled in the study. On PPD four days after the first dose of ACE-011/placebo and the initial doses of melphalan, prednisolone and thalidomide (MPT), the patient presented with elevated temperatures up to 39° C, cough and fatigue. Chest x-ray performed on PPD revealed a pneumonic infiltration in the lowerlobe of the left lung. On PPD, hospitalization of the patient was prolonged due to the event. The patient was treated with moxifloxacin, doripenem, meropenem and fluconazole.

On PPD, the pneumonic infiltration resolved, as confirmed by chest x-ray. This SAE resolved upon discharge of the patient from the hospital on PPD Pneumonia is considered a labeled event per the current package insert for thalidomide. The Investigator considered the event of pneumonia unrelated to ACE-011 and possibly related to MPT.

An SAE of pain was reported in a PPD and was found to have a pathological fracture of the right upper femoral bone approximately 2.5 months after last dose of ACE-011/placebo, approximately 5 weeks after last dose of Melphalan-Prednisolone and one day after the last dose of Thalidomide. Radiologic evaluations demonstrated progressive bone tissue destruction at presentation. The subject was casted, and then underwent coxal bandaging. The event resolved with sequelae. The investigator considered the events of pain in the right leg and pathological fracture of the upper third of the right femur unrelated to ACE-011/placebo and unrelated to Thalidomide (MPT).

The A011-04 study has completed enrollment and data monitoring is ongoing.

5.5. Potential Risks for Human Use

The 1-month safety study in rats suggested that ACE-011 may affect sperm count and motility in a dose-dependent manner, although these findings appeared to be reversible upon drug discontinuation. These reproductive effects are likely a result of the expected biological activity of activin inhibition.

The 1-month safety study in rats also suggested that ACE-011 induces adrenocortical necrosis in rats. At present, the mechanism by which ACE-011 has induced this lesion in rats and the basis for its apparent species specificity are unknown; the finding has not been observed in mice or cynomolgus monkeys at similar dose levels. Adrenal cortical function was monitored in human subjects receiving ACE-011 by the evaluation of serum electrolytes including sodium and potassium levels and by the evaluation of cortisol response to ACTH stimulation in both the

Phase 1a single dose and Phase 1b multiple dose healthy volunteer clinical studies. These parameters were evaluated up to doses of 3.0 mg/kg IV and 1.0 mg/kg SC, and no clinically significant perturbations in adrenal function were observed.

Laboratory findings from the Phase 1a single dose study showed elevations in hematology results, pancreas and liver enzymes, and uric acid. The most commonly seen treatment-emergent adverse events in this study were headache, infusion site reaction, injection site hemorrhage, and toothache.

Based on data from the Phase 1b study, the most notable AEs were increases in hematologic laboratory measures in the 1.0 mg/kg dose group: hematocrit, hemoglobin, and RBC count. AEs of increased hemoglobin and/or hematocrit were reported for seven of the eight subjects in this dose group. These events were reported as mild or moderate elevations; all were considered probably related to study drug treatment. No erythroid lineage AEs were reported in the 0.1 or 0.3 mg/kg treatment groups. Hematologic parameters will be monitored carefully in this study.

Headache was reported frequently in all treatment groups with no apparent dose response trend, though the reports in the higher dose groups were more often considered possibly related to treatment. Most headaches were reported as mild (one headache was reported as moderate). Paresthesia and dizziness were reported more frequently in the ACE-011 groups, though the events were generally not considered drug related. Other frequently reported events (e.g. fatigue, upper respiratory infection) did not appear to increase with dose and were generally mild.

In the Phase 1b multiple-dose healthy volunteer study, some subjects treated with ACE-011 showed elevations in blood pressure. One subject who had received two doses of ACE-011 at the 1.0 mg/kg dose level had a serious adverse event. The patient was hospitalized for further evaluation of increases in blood pressure and symptoms that may have been caused by the increase in blood pressure. The symptoms that the subject experienced included headache, dizziness, nausea, vomiting, and elevated hematology values (increases in the number of red blood cells). This serious adverse event was judged to be probably related to the study drug and resolved following discharge from hospital the following day. Throughout the course of the follow up period hematologic parameters were monitored frequently until the hemoglobin and hematocrit levels returned to within normal limits. Further details are outlined in the Investigator Brochure.

As with all biologics there is the potential for anti-drug antibodies that can be associated with increased drug clearance and hypersensitivity reactions. An immune response to ACE-011 has been seen in monkeys that received the drug for 1 month and longer. Some of those animals developed kidney inflammation possibly related to that immune response. Anti-drug antibody formation will be assessed in this study to examine the immunogenicity of ACE-011, and to monitor reversibility of any AEs over time.

Please refer to the Investigator Brochure for more information.

6. TRIAL OBJECTIVES AND PURPOSE

6.1. Primary objective

1. To evaluate the proportion of patients in each ACE-011 dose regimen who achieve a hematopoietic response during the treatment period including up to 2 months after the last dose of ACE-011 treatment of chemotherapy induced anemia in patients with metastatic breast cancer. Hematopoietic response is defined as an increase in hemoglobin of ≥ 1.0 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs.

6.2. Secondary objectives

1. To evaluate the safety and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer.
2. To evaluate the percentage of patients who achieve an increase in hemoglobin of ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for a consecutive 28 day period during the treatment period including up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs.
3. To estimate the duration of the hematopoietic response associated with each ACE-011 dose regimen.
4. To estimate the time to the hematopoietic response associated with each ACE-011 dose regimen.
5. To estimate the proportion of patients receiving red blood cell (RBC) transfusions and/or ESAs within each cycle of ACE-011 with each dose regimen.
6. To estimate the effect of each ACE-011 dose regimen on progression free survival (PFS) and objective response rate (ORR).

6.3. Exploratory objectives

1. To estimate the effect of each ACE-011 dose regimen on serum bone biomarkers (OC, BSAP, P1NP, CTX, TRACP-5b), urine bone biomarker (uNTX), bone mineral density (DXA scans of lumbar spine and hip) and incidence of skeletal related events (SREs).
2. To evaluate the quality of life in relation to the treatment of chemotherapy induced anemia in patients with metastatic breast cancer during the study.

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7. INVESTIGATIONAL PLAN

7.1. Overall Study Design and Plan

This is a Phase 2, double-blind, randomized, placebo-controlled study to evaluate the efficacy, safety, and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer. A total of 105 patients will be enrolled.

The Data Monitoring Committee (DMC) is responsible for reviewing safety. The DMC will be comprised of a minimum of 3 members. The DMC will meet regularly during the study to review serious adverse events (SAEs), adverse events (AEs), laboratory results, and vital signs. Safety data will be reviewed throughout the study by the DMC, Medical Monitor and the Investigator. DMC responsibilities, membership, meeting frequencies and procedures will be outlined in the DMC charter.

Patients will be evaluated for study inclusion/exclusion criteria by the Investigator. Patients who meet the study entry criteria will be enrolled within 14 days of the screening visit. Local lab values for hemoglobin and central lab values will be utilized for the evaluation of patient eligibility. Central labs will also be used in the evaluation of patient safety throughout the study. Local lab values for hemoglobin and hematocrit will be collected and reported from screening (for eligibility) through the Day 281/Termination visit. Three cohorts of 30 patients each are planned at the following doses of ACE-011: 0.1 mg/kg, 0.3 mg/kg and 0.5 mg/kg. Fifteen patients will be enrolled into the placebo cohort (as shown in Table 6).

Table 6: Study Design

Dose	Number of Patients
0.1 mg/kg	30
0.3 mg/kg	30
0.5 mg/kg	30
Placebo	15
Total patients enrolled: 105	

Each eligible patient will be randomized to one of the four cohorts to receive a dose administered as a subcutaneous injection every 28 days for up to 4 doses. Dosing will be administered on days 1, 29, 57, and 85. Concurrent treatment with a myelosuppressive chemotherapy regimen for metastatic breast cancer will also be administered per standard of care at the site. Patients will be stratified according to administration of either single agent weekly chemotherapy or less frequent chemotherapy regimens.

Blood samples will be collected from approximately one-third of patients participating in the study at selected study sites for the evaluation of serum concentrations of ACE-011. Pre-dose PK samples will be collected as specified in Appendix 5 (PK sampling schedule.)

Patients will be assessed for safety by monitoring adverse events, clinical laboratory tests, assessment of quality of life (FACT- Fatigue (17)), vital signs and physical examinations on an ongoing basis. ECGs will be performed during screening, prior to dosing on the first cycle, during the first, second and third cycle, Day 113 (28 days after the last dose of ACE-011) and at Day 281/ Termination visit to monitor safety. Blood pressure and lab values will be assessed at each study visit during the treatment and treatment follow up period.

Local laboratory hemoglobin values and blood pressure values will be evaluated prior to dosing per the dose modification criteria, as applicable.

Patients will be followed for a minimum of 7 months following their last dose of ACE-011/placebo. Study participation for each patient is approximately 10.5 months (14 day screening period, 3 month treatment period with a 2 month treatment follow up period, and a 5 month post treatment follow-up period). If a patient discontinues the study due to progression of disease (metastatic breast cancer) and/or begins another chemotherapy regimen, the patient will discontinue ACE-011/placebo treatment, complete the Day 281/Termination visit procedures, and be followed for survival only (a minimum 7 months from the last dose of ACE-011/placebo).

Anti-drug antibody testing will be performed at Day 281/Termination visit and if positive, the patient may be asked to return to the clinical site for additional monthly anti-drug antibody testing for up to 12 months, after their last dose.

Back up (serum) blood samples will be collected for future testing for the evaluation of novel tumor markers, such as endoglin and ALK-1 (activin receptor-like kinase-1). The samples will the stored at the central lab until this evaluation is performed.

Table 7: Schedule of Events

Schedule of Events																				
	Screen	Treatment Period														Post Treatment Follow up Period				Term Visit[20]
ACE-011/placebo Dose period		#1				#2			#3			#4	Follow-up							
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	99 (± 3d)	113 (± 3d)	141 (± 7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
Informed consent	X																			
Inclusion/exclusion criteria	X	X[2]																		
Medical history	X																			
Physical examination	X	X[2]				X[2]			X[2]			X[2]		X	X					X
Vital signs[1]	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X					X
FACT-Fatigue	X	X[2]				X[2]			X[2]			X[2]								X
12 lead ECG[3]	X	X	X				X			X				X						X
Serum iron, TIBC, transferrin and serum ferritin	X[8]	X[2]				X[2]			X[2]			X[2]								X
Vitamin B12	X[8]																			
Serum folate	X[8]						X							X						X
Coagulation/ Serum chemistry[4]	X[8]	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X					X
Hematology[5]	X[8]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Serum Erythropoietin marker		X[2]				X[2]			X[2]			X[2]			X					X
LH, FSH, testosterone, progesterone, estradiol	X	X[2]				X[2]			X[2]			X[2]		X	X					
Anti-drug antibody testing [6]		X[2]				X[2]			X[2]			X[2]		X	X					X
Bone biomarker[7]		X[2]				X[2]			X[2]			X[2]		X	X					X
Back up blood sample for future testing		X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X					X
Pregnancy Test	X[8]	X[9]				X[9]			X[9]			X[9]								X
Urinalysis[10]	X	X[2]										X[2]								X
Evaluate transfusion frequency	X	X[2]	X	X	X	X[2]	X	X	X[2]	X	X	X[2]	X	X	X	X	X	X	X	X
Documentation of conmeds	X[11]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X			

Schedule of Events cont.

	Screen	Treatment Period														Post Treatment Follow up Period				Term Visit[20]
ACE-011/Placebo Dose period		#1				#2			#3			#4	Follow-up							
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	99 (± 3d)	113 (± 3d)	141 (±7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
Evaluation of AEs and SAEs[12]	X-SAE only	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X-SRE and SAE only	X-SRE and SAE only	X-SRE and SAE only	X
Evaluation of Menstrual cycle[13]	X	X[2]				X[2]			X[2]			X[2]		X	X					
CT/MRI scan	X[14]									X[15]				X[15]						
Dual-energy X-ray absorptiometry (DXA[16])	X													X						X
Bone scans[17]	X																			X
ACE-011/placebo administration[18]		X				X			X			X								
Chemo regimen[19]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

[1] Vital signs include weight, heart rate, seated systolic and diastolic blood pressure, and temperature will be assessed at each visit Height will be collected at screening only

[2] On dosing days (1, 29, 57 and 85), study procedure to be done prior to dosing of ACE-011/placebo

[3] Screening and Day 1- At two different time intervals (approximately 1 hours apart) triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min), prior to dosing as applicable On all other specified days, one time interval of triplicate (12 lead) ECGs is to be performed at 3 minute intervals (± 1min),

[4] Coagulation (central lab only) prothrombin time (PT), partial thromboplastin time (PTT), fibrinogen Chemistry (central lab only): AST, ALT, lactate dehydrogenase (LDH), total bilirubin, indirect bilirubin, alkaline phosphatase, total protein, albumin, sodium, potassium, chloride, carbon dioxide, calcium, phosphorus, blood urea nitrogen (BUN), creatinine, uric acid, glucose, amylase, and lipase

[5] Hematology (local lab) hemoglobin and hematocrit On dosing days (1, 29, 57 and 85), local hemoglobin and hematocrit values are to be drawn and resulted (up to 1 day) prior to dosing to apply dose modification rules for ACE-011/placebo Hematology (central lab only) complete blood count (CBC) with differential and platelet, CBC includes red blood cells (RBC), white blood cells (WBC), hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), Peripheral blood smear will also be done on Days 1, 29, 57, 85, and at the termination visit **Reticulocyte should be done on days 1, 8, 15, 22, 29, 36, 57, 64, 85, and at the termination visit.**

[6] If a patient has a positive anti-drug antibody result at the Termination visit, they may be asked to return to the clinical site for additional monthly follow-up, for up to 12 months after their last dose for a repeat anti-drug antibody test

[7] Both serum and urine samples will be collected at each visit The urine sample should not be from the first morning urine

[8] Labs may be drawn within 1 day of Day 1 Confirmatory labs for eligibility criteria must be drawn and resulted within 7 days of Day 1

[9] For females of child bearing potential only urine (dipstick) pregnancy test to be collected and resulted prior to dosing of ACE-011/placebo

[10] Urinalysis to be performed at screening, prior to administration ACE-011/placebo (Days 1 and 85) and termination visit Assessments (central lab only): pH, specific gravity, protein, glucose, ketones, blood, leukocyte esterase, and nitrite Microscopy required only to follow-up abnormal findings

[11] Screening- any concomitant treatments taken 28 days prior to day 1 including up to 90 days after the last dose of ACE-011/placebo

[12] AEs will be reported from day 1 post dose including up to 90 days after the last dose of ACE-011/placebo Additional AEs noted after 90 days from the last dose of ACE-011/placebo will be reported at the discretion of the investigator SREs and SAEs will be followed through out the study If a patient is followed for survival only, SREs and SAEs will be captured up to 90 days from the last dose of ACE-011/placebo

[13] Females of child bearing potential- collection of menstrual cycle information start and end date pf last menstrual period since the last assessment

[14] A CT/MRI scan acquired within 6 weeks of initiating the current chemotherapy regimen may be used for the screening assessment Head, neck, thoracic, abdominal, and pelvic scans are to be acquired One measurable or non measurable lesion per RECIST is to be identified by the scan Brain (head) CT only to be done to rule out CNS metastases in patients with suspicious clinical symptoms or prior history of CNS metastases

[15] Follow-up CT/MRI scans may be acquired +/- 5 day window for both Day 64 and Day 113 Additional CT/MRI scans acquired during the post treatment follow up period will be performed at the discretion of the investigator

[16] Hip and lumbar spine BMD assessment Refer to Imaging Acquisition Guidelines for guidelines regarding washout period for contrast (if used) for radiological images, prior to collection of DXA

[17] Bone scans will be acquired within 4 weeks of initiating the current chemotherapy regimen for those patients who have bone metastases identified Follow up bone scan may be acquired +/- 5 day window Day 281

[18] All study visit day dates (inclusive of windows) are calculated from Day 1(day of initial ACE-011/placebo administration) Patients who discontinue ACE-011/placebo due to ACE-011/placebo related toxicities (section 9 6 2 1) will enter the treatment follow-up period

[19] Chemotherapy regimens will be administered to the patient as per standard of care Continuation of the same chemotherapy regimen may be done through the follow up period of the study at the discretion of the investigator

[20] Patients who terminate treatment early due to disease progression and/or begin a new chemotherapy regimen will complete the day 281/Termination visit procedures and be followed for survival only (7 months from the last dose of ACE-011/placebo) If a patient withdraws from the study (section 8 3), the Day 281/Termination visit procedures are to be completed and the patient will be followed for survival If withdrawal reason is due to ESA administration or ACE-011 related toxicity, the patient should enter the treatment follow up period

8. SELECTION AND WITHDRAWL OF PATIENTS

8.1. Number of Patients

The study will include three dose levels of ACE-011 at 0.1 mg/kg, 0.3 mg/kg and 0.5 mg/kg and a placebo group. A total of 105 will be treated with ACE-011 or placebo.

8.2. Entry Criteria

8.2.1. Patient Inclusion Criteria

1. Women ≥ 18 years of age.
2. Histologically confirmed diagnosis of breast cancer documented by cytology or biopsy.
3. Evidence of metastatic breast cancer with a minimum of one measurable lesion or non measurable lesion per RECIST criteria (Appendix 1).
4. Receiving a multicycle myelosuppressive chemotherapy regimen including one of the following: anthracycline, taxane, gemcitabine or vinorelbine.
5. Anticipated treatment with the same myelosuppressive chemotherapy regimen for a minimum of 9 weeks after Day 1 of ACE-011/placebo administration.
6. Hemoglobin value between ≥ 6.5 to < 11.0 g/dL (≥ 65 to < 110 g/L).
7. ≥ 30 days must have elapsed (from Day 1) since previous treatment with erythropoiesis-stimulating agent (including concurrent treatment with IV iron) for chemotherapy induced anemia.
8. ≥ 7 days have elapsed (from Day 1) since the last RBC blood transfusion and receipt of ≤ 2 units of blood in the past 30 days.
9. ECOG Performance status of 0 – 2 (Appendix 2).
10. Adequate renal (creatinine ≤ 1.5 x ULN or creatinine clearance ≥ 40 mL/min) and hepatic function (bilirubin ≤ 1.5 x ULN; AST/ALT ≤ 2.5 x ULN).
11. Females of childbearing potential participating in the study are to use adequate birth control measures (i.e. oral contraceptives or two forms of barrier method birth control) during study participation. Females of childbearing potential must have a negative serum pregnancy test within 7 days to ACE-011/placebo administration (Day 1). A female of childbearing potential is a sexually mature woman who has not undergone a hysterectomy or who has not been postmenopausal for at least 24 consecutive months (i.e. who has had menses at some time in the preceding 24 months).
12. Life expectancy of ≥ 6 months.
13. Is willing to adhere to the study visit schedule, understand and comply with all protocol requirements.
14. Understand and sign a written informed consent.

8.2.2. Patient Exclusion Criteria

1. At the time of screening patients who have any grade ≥ 3 toxicity (as per NCI CTCAE v3.0 (Appendix 3)) (except for the following disease related toxicities: hematological events- anemia, thrombocytopenia or neutropenia or non hematological events- nausea, vomiting, fatigue, or muscle or bone/joint pain).
2. Prior radiation therapy to > 20% of the whole skeleton. Use of palliative radiation if the area being treated is <15% of body surface area with no pelvic radiation or no more than 10% of the bone marrow reserve is radiated is permitted during patient participation in the study, at the discretion of the investigator.
3. Have received more than 5 prior chemotherapy treatment regimens for metastatic breast cancer.
4. Clinically significant pulmonary, endocrine, neurologic, gastrointestinal, hepatic or genitourinary disease unrelated to underlying hematologic disorder.
5. Patients with heart failure as classified by the New York Heart Association (NYHA) classification of 3 or higher (Appendix 4).
6. History of thrombosis, deep vein thrombosis (DVT), pulmonary emboli, or embolic stroke.
7. Untreated CNS metastases (exception: CNS metastases treated with whole brain radiotherapy > 6 months prior to randomization).
8. Diagnosis of a myeloid malignancy or known history of myelodysplasia.
9. History of second malignancy within 5 years (except excised and cured basal cell carcinoma, squamous cell carcinoma of the skin or cervical carcinoma in situ).
10. Patients with a recent history (within 14 days of Day 1) of administration of systemic (IV or oral) antibiotics. Patients should be free of infection at least 14 days after the last dose of systemic antibiotic treatment (from Day 1).
11. Uncontrolled hypertension. If hypertension is considered clinically stable, systolic blood pressure (BP) must be < 150 mmHg and diastolic BP must be < 100 mmHg.
12. Known history of hepatitis C antibody (HCV), hepatitis B surface antigen (HBsAg and HB core Ab), or human immunodeficiency virus (HIV) antibody.
13. Deficiency in iron (serum ferritin < 100 ng/mL (< 224.7 pmol/L)), vitamin B_{12}, or folate.
14. History of anemia as a result of inherited hemoglobinopathy such as sickle cell anemia or thalassemia.
15. History of autoimmune or hereditary hemolysis or active gastrointestinal bleeding (within the previous 12 months as compared to Day 1).
16. Patient received treatment with another investigational drug or device within 1 month prior to Day 1, or if the half life of the previous product is known, within 5 times the half life prior to dosing, whichever may be longer.
17. Pregnant or lactating females.
18. History of severe allergic or anaphylactic reactions or hypersensitivity to recombinant proteins or excipients in the investigational product.

19. Major surgery within 30 days prior to Day 1 (patients must have completely recovered from any previous surgery prior to Day 1).

8.3. Patient Withdrawal Criteria

Patients will be informed that they have the right to withdraw from the study at any time for any reason without prejudice to their medical care.

Patients must be withdrawn from the study for any of the following reasons:

- Patient request
- Patient is unwilling or unable to comply with the protocol
- ESA administration during the treatment period
- Medical reason, such as cancer or treatment related toxicity, or at the discretion of the Investigator and/or the Medical Monitor(s)

The reasons for withdrawal must be recorded in the patient's case report form (CRF). The Investigator must notify the Medical Monitor and the contract research organization (CRO) immediately when a patient has been discontinued/withdrawn due to an AE. All patients who are withdrawn from the study should complete the tests and evaluations scheduled for Day 281/Termination at the time of withdrawal. Discontinued/withdrawn patients should be followed for survival only for 7 months after the last dose of ACE-011/placebo. Patients who withdraw due to ESA administration during the treatment period or due to ACE-011 related toxicity should enter the treatment follow-up period.

9. TREATMENT OF PATIENTS

9.1. Concomitant Medications

9.1.1. General Concomitant Medication Usage

During screening, and during the study, patients may take only stable doses of medications for chronic conditions that are not specifically excluded by the protocol (see Sections 8.2.1 Patient Inclusion Criteria and 8.2.2 Patient Exclusion Criteria). The Medical Monitor should be consulted by the Investigator if there are any concerns about what constitutes a stable dose or what is a chronic condition. Concomitant medications will be recorded in the CRF through 90 days from the last dose of ACE-011/placebo.

9.1.2. Concomitant treatment for anemia

Concurrent therapy with a new prescription medication related to treatment of anemia during the course of the study must first be discussed with the Investigator and Medical Monitor prior to administration, unless appropriate medical care necessitates that medication should begin before the Medical Monitor can be consulted.

9.1.2.1. Iron supplementation

Iron supplementation within 7 days of Day 1 (besides an oral general multivitamin with iron) will not be allowed. If a patient becomes iron replete during study treatment (ferritin < 100 ng/mL (< 224.7 pmol/L) or transferrin < 20%), treatment with iron supplementation is at the discretion of the investigator.

9.1.2.2. Erythropoiesis-stimulating agents (ESAs)

If concurrent treatment with erythropoiesis-stimulating agent is required in the opinion of the investigator, the erythropoiesis-stimulating agent label instructions are to be followed. The patient is to discontinue further treatment with ACE-011/placebo and enter the treatment follow-up period of the study. See Table 7 for Schedule of Events.

9.1.2.3. RBC Transfusions

Concurrent treatment for chemotherapy induced anemia with blood transfusions is recommended when hemoglobin value is < 8 g/dL or at investigator discretion if the hemoglobin value is above 8 g/dL and associated with anemia symptoms such as hemodynamic or pulmonary compromise that require treatment. If a transfusion is given to a patient during the treatment period, ACE-011/placebo should be administered no sooner than 7 days from the date of the transfusion. After the transfusion, the hemoglobin value should be assessed no later than 7 days from the date of the transfusion. On the day of ACE-011/placebo administration, the hemoglobin value and blood pressure will be assessed. See Figure 1 for ACE-011/placebo dose modification rules.

9.2. Chemotherapy

Chemotherapy treatment for metastatic breast cancer is to be given as per standard of care at the site. ACE-011/placebo administration should be given prior to chemotherapy administration on ACE-011/placebo administration days. Continuation of treatment with the same chemotherapy regimen through the treatment or post-treatment follow-up period of the study is at the discretion of the investigator.

9.3. Treatment Compliance

Each dose of ACE-011/placebo will be administered as a SC injection at the clinical site by the study staff and will be documented in the study record. Monitoring for patient compliance with the treatment regimen is therefore unnecessary.

9.4. Rescreening

If a patient receives a screening number through Interactive Web/Voice Response System (IVRS/IWRS) and does not continue to meet eligibility criteria (prior to randomization), the patient may be re-screened once during the enrollment period of the study. Evaluation and decision to re-screen a patient will be done by the site, Medical Monitor, and Acceleron (as needed). All screening procedures are to be re-performed to verify patient eligibility. The screening DXA scan is to be re-done if more than 45 days has elapsed since the initial screening DXA scan. Informed consent must be signed by the patient if more than 14 days has elapsed since initially signing consent.

Patients who have been randomized but have not received the first dose of ACE-011/placebo and no longer meet eligibility criteria will require further evaluation by the site, Medical Monitor, and Acceleron (as needed) for continued participation in the study.

9.5. Randomization

Patients will be randomized to limit scientific bias within the study. Randomization assignments will be generated through a computerized system, provided by an Interactive Web/Voice Response System (IWRS/IVRS). Patients will be stratified according to frequency of the planned chemotherapy regimen (weekly chemotherapy vs. less frequent chemotherapy administration).

9.5.1. Blinding

The Investigators, patients, and sponsor or representative will remain blinded to the treatment arm assignment of each patient. The unblinded statistician will remain uninvolved in the study conduct until database lock and unblinding of the data has occurred.

Safety parameters will be reviewed on an ongoing basis throughout the study including adverse events, serious adverse events, laboratory listings, and vital signs. If substantial toxicity trends are observed in one treatment group versus another, the DMC may request unblinding the treatment code assignment schema.

9.5.2. Unblinding

In the event of a medical emergency for an individual patient in which knowledge of the study medication is critical to the patient's medical management, the investigator may break the blind for that patient. However, prior to breaking the blind, every effort must be made by the investigator to first discuss the need to break the blind with the sponsor or sponsor representative. Further, it must be determined by the investigator that breaking the treatment blind is necessary information for the medical management of that patient. If the blind is broken, the patient must withdraw from the study by completing the Day 281/Termination visit procedures and be followed for survival only

9.6. Treatments Administered

9.6.1. Selection of Doses in the Study

Single dose administration of ACE-011 up to 3.0 mg/kg was previously demonstrated to be safe and to have durable effects on markers of bone formation and resorption as well as red blood cells following a single IV administration. A dose-limiting pharmacodynamic effect of ACE-011 with increases in hemoglobin, hematocrit and RBCs was established in healthy post-menopausal women at the 1.0 mg/kg dose level following multiple SC administration. This study will confirm the safety in following repeated SC administration of ACE-011 and further evaluate the potential efficacy in patients with chemotherapy induced anemia. The safety and preliminary efficacy of ACE-011 following multiple doses up to 0.5 mg/kg will be assessed using a parallel randomization design.

9.6.2. Selection and Timing of Dosing for Each Patient

Patients will be enrolled and receive their assigned dose of ACE-011/placebo every 28 days (i.e. on Days 1, 29, 57, and 85). Patients will be receiving chemotherapy, concurrently for metastatic breast cancer. After completion of ACE-011/placebo treatment, patients will return to the site monthly for 3 follow-up assessments (i.e., Days 99, 113 and 141). Subsequently, the patient will return to the site for 4 post treatment follow-up assessments (i.e. Days 169, 197, 225, and 253). The patient will return to the site for a termination visit approximately 1 month after the post treatment follow-up period (Day 281). Patients will be discontinued from the ACE-011/placebo treatment for reasons listed in section 8.3 of the protocol, for unacceptable toxicity, if the chemotherapy regimen is discontinued or for disease progression that requires the initiation of another chemotherapy treatment.

9.6.2.1. ACE-011 Treatment Related Toxicity

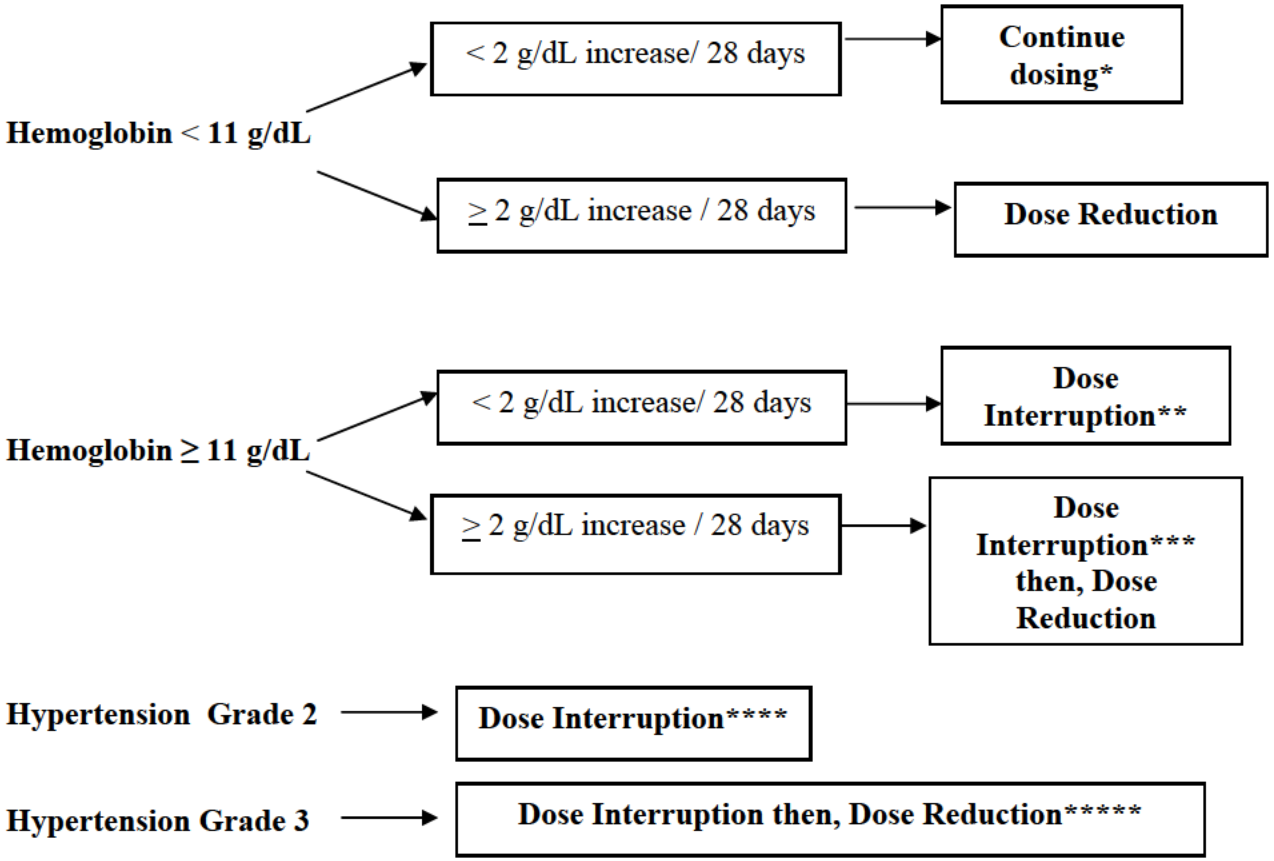
Patients with grade ≥3 toxicity related to ACE-011 will be discontinued from ACE-011/placebo treatment except for the following. Patients with grade 3 hypertension that decreases to grade ≤ 1 within 7 days after treatment with anti-hypertensive therapy may continue on ACE-011/placebo treatment. Dose reduction rules will apply at the subsequent scheduled dosing day(s). Patients with a hemoglobin level above the upper limit of normal at any time during the

study after Day 1 will be discontinued. Patients who discontinue treatment with ACE-011/placebo will enter the treatment follow-up period.

9.6.2.2. ACE-011 Dose Modification Rules

Throughout the study blood pressure and hemoglobin values will be evaluated for patient safety. The local hemoglobin values and blood pressure values will be evaluated at each study visit and on each ACE-011/placebo dosing day. The hemoglobin values from the previous 28 days between each ACE-011/placebo dosing day and blood pressure will be reviewed and evaluated for dose modifications. The parameters below outline rules for dosing modifications (Figure 1). Table 8 outlines the dose reduction levels of ACE-011.

Figure 1: ACE-011/Placebo Dose Modification Rules (after initial dose of ACE-011/placebo)



* Patients who have received a blood transfusion in the past 28 days should continue with dosing at the same ACE-011/placebo dosing level if the transfusion was given no sooner than 7 days from the dosing day and the hemoglobin level is < 11 g/dL and hypertension ≤ Grade 1 on the day of dosing

** ACE-011/placebo should be held until the following scheduled treatment visit

*** ACE-011/placebo should be held and at the following scheduled treatment visit, a dose reduction of ACE-011/placebo will be administered based on the evaluation of the hemoglobin and blood pressure at that time

**** ACE-011/placebo should be held until resolution to ≤ Grade 1 (NCI CTCAE v3 0) and may then be resumed at the following scheduled treatment visit.

***** ACE-011/placebo should be held until resolution to ≤Grade 1 (NCI CTCAE v3 0) within 7 days after treatment with anti- hypertensive therapy and may continue on a reduced dose of ACE-011/placebo treatment for the subsequent scheduled dosing day

Table 8: ACE-011/Placebo Dose Reduction Levels

When required per the dose modification rules above (Figure 1), ACE-011/placebo dose(s) should be reduced as follows for dose 2 (Day 29), dose 3 (Day 57) and/or dose 4 (Day 85):

Dose	Dose reduction #1	Dose Reduction #2	Dose Reduction #3
Placebo	0.1 mg/kg	0.05 mg/kg	0.03 mg/kg
0.1 mg/kg	0.05 mg/kg	0.03 mg/kg	0.01 mg/kg
0.3 mg/kg	0.1 mg/kg	0.05 mg/kg	0.03 mg/kg
0.5 mg/kg	0.3 mg/kg	0.1 mg/kg	0.05 mg/kg

Blood pressure and hemoglobin values must be evaluated at each dosing day for consideration of administration of ACE-011/placebo.

The dose level for the initial dose reduction will be maintained unless subsequent dose reductions need to be administered, upon evaluation of the dose modifications listed above. A patient may have up to 3 dose reductions in the study.

Dose reduction steps and the administration of the reduced dose(s) will be conducted in a manner that will preserve the blinded status of the original treatment group of each patient. Patients in the placebo group who are designated by the investigator to undergo a dose reduction will continue to receive placebo.

10. STUDY PROCEDURES AND SCHEDULE

10.1.1. Written Informed consent

Patients will be required to sign an Institutional Review Board/Institutional Ethics Committee (IRB/IEC) approved informed consent form (ICF) prior to any study related procedures, including screening evaluations.

Screen failure information will be maintained, including but not limited to, reason for failure.

10.1.2. Screening (Within 14 days of dosing)

The following will be collected within 14 days prior to the initial dosing Day 1. Confirmatory labs for eligibility criteria must be drawn and resulted within 7 days of Day 1:

- Informed consent
- Assessment of inclusion/exclusion criteria
- Complete Medical History and Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), temperature (°C) and height.
- FACT-Fatigue survey
- 12 lead Electrocardiogram (ECG) performed at two time intervals (approximately one hour apart) in triplicate at 3 minute intervals (±1 min)
- Serum iron, total iron binding capacity (TIBC), transferrin and serum ferritin
- Vitamin B12 and serum folate levels
- Coagulation
- Serum chemistry
- Hematology
- LH, FSH, testosterone, progesterone, and estradiol
- Serum Pregnancy Test for females of childbearing potential- to be assessed within 7 days of Day 1.
- Urinalysis
- Evaluation of transfusion frequency (including history of transfusion up to 8 weeks prior to Day 1)
- Documentation of concomitant medications (any treatments taken 28 days prior to Day 1)

- Females of childbearing potential- collection of menstrual cycle information of the start and end date of the last menstruation.
- CT/MRI scan: A CT/MRI scan acquired within 6 weeks of initiating the current chemotherapy regimen may be used for the screening assessment. One measurable or non-measurable lesion per RECIST v1.1 is to be identified by the scan.
    - Scans of the anatomical areas of head (if applicable), neck, thoracic, abdominal, and pelvic are to be available for evaluation. If one of the anatomical areas listed has not be acquired, prior to screening, then a CT/MRI must be done, during screening for this study, to ensure a scan of the head (if applicable), neck, thoracic, abdominal, and pelvic is available.
        - Brain (head) CT only to be done to rule out CNS metastases in patients with suspicious clinical symptoms or prior history of CNS metastases.
- Dual-energy X-ray absorptiometry (DXA) of lumbar spine and hip
    - Refer to Imaging Acquisition Guidelines for guidelines regarding washout period for contrast (if used) for radiological images, prior to acquiring DXA scan.
- Bone scans (only for patients with bone metastases) acquired within 4 weeks of initiating the current chemotherapy regimen may be used for the screening assessment.

10.1.3. Initial Dosing: Day 1

Patients will be dosed on Day 1. Results from screening evaluations must be reviewed prior to randomization to confirm patient eligibility. A patient will be considered randomized once a multi-digit patient identification number has been assigned to the patient through the IVRS/IWRS. After assignment of the patient identification number and the inclusion/exclusion criteria have been met, ACE-011/placebo administration may begin.

Chemotherapy regimen will be administered per standard of care at the site. The following tests must be performed prior to ACE-011/placebo administration:

- Confirm eligibility of patient by inclusion/exclusion criteria (local lab hemoglobin value and central lab values). The local hemoglobin value should be drawn prior to ACE-011/placebo administration (up to 1 day prior to Day 1).
- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- FACT-Fatigue survey
- 12 lead Electrocardiogram (ECG) performed at two time intervals (approximately one hour apart) in triplicate at 3 minute interval (±1 min)

- Serum iron, total iron binding capacity, transferrin and serum ferritin
- Coagulation
- Chemistry
- Hematology
- Serum erythropoietin marker
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test
- Bone biomarkers- osteocalcin (OC), procollagen type 1 N-terminal propeptide (P1NP), serum bone specific alkaline phosphatase (BSAP), C-terminal type 1 collagen telopeptide (CTX), tartrate-resistant acid phosphatase isoform-5b (TRACP-5b), and urine N-telopeptide (uNTX).
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions the on collection of serum and urine for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Females of childbearing potential- Urine (dipstick) pregnancy test to be collected and resulted prior to ACE-011/placebo dosing
- Urinalysis
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events (after ACE-011/placebo administration)
- Females of childbearing potential-collection of menstrual cycle information of the start and end date of the last menstruation.
- Patients will be administered ACE-011/placebo before the chemotherapy regimen begins during the study visits.

After ACE-011/placebo and chemotherapy are administered, the patient may leave the clinic, based on the clinical judgment of the staff.

All study visit day dates (inclusive of windows) are calculated from Day 1(day of initial ACE-011/placebo administration).

10.1.4. ACE-011/placebo administration: Days 29, 57, and 85 (± 3 days)

Chemotherapy regimen will be administered per standard of care at the site. ACE-011/placebo will be administered approximately every 28 days. The following procedures must be performed prior to ACE-011/placebo administration:

- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- FACT-Fatigue survey
- Serum iron, total iron binding capacity, transferrin and serum ferritin
- Coagulation
- Serum chemistry
- Hematology
- Serum erythropoietin marker
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test
- Bone biomarkers- OC, BSAP, P1NP, CTX, TRACP-5b, and uNTX
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions the collection of serum and urine for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Females of childbearing potential -Urine (dipstick) pregnancy test to be collected and resulted prior to ACE-011/placebo dosing
- Urinalysis (Day 85 only)
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events
- Females of childbearing potential- collection of menstrual cycle information of the start and end date of the last menstruation.

Hemoglobin (HgB) (by review of local lab values) and blood pressure (BP) measurement must be assessed prior to ACE-011/placebo dose administration. Local Hgb must be done within 1 day prior to the ACE-011/placebo dosing day.

10.1.5. Additional visits during Treatment Period: Days 8, 15, 22, 36 (± 1 day), 43, 64 and 71 (± 2 days)

Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- 12 lead ECG (Days 8, 36, and 64 only- to be done in triplicate at 3 minute intervals (±1 min)
- Serum folate (Day 36 only)
- Coagulation
- Serum chemistry
- Hematology
- Back-up blood sample to be drawn for future testing
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events
- CT/MRI scan (Day 64 only, +/- 5 day window). Same modality and anatomical areas as acquired prior to enrollment should be used for follow up scans. Tumor response assessments to be evaluated by the investigator.

If patient terminates the treatment period early, prior to the Day 85 of ACE-011/placebo, the patient will enter the treatment period follow up. However, if patient terminates ACE-011/placebo early and begins a new chemotherapy regimen, the patient should complete the Day 281/Termination visit procedures and be followed for survival only.

10.1.6. Treatment Period Follow-up visits : Days 99, 113 (± 3 days) and 141 (± 7 days)

Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Physical Examination (Days 113 and 141 only)
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- 12 lead ECG-including (Day 113 only- to be done in triplicate at 3 minute intervals (±1 min)
- Serum folate (Day 113 only)
- Coagulation
- Serum chemistry
- Hematology
- Serum erythropoietin marker (Day 141 only)

- LH, FSH, testosterone, progesterone, and estradiol (Days 113 and 141 only)
- Anti-drug antibody test (Days 113 and 141 only)
- Bone biomarkers- OC, BSAP, P1NP, CTX, TRACP-5b, and uNTX (Days 113 and 141 only)
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions on the collection of serum and urine for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events
- Females of childbearing potential- collection of menstrual cycle information of the start and end date of the last menstruation. (Days 113 and 141 only)
- CT/MRI scan (Day 113 only, +/- 5 day window). Same modality and anatomical areas as acquired prior to enrollment should be used for follow up scans. Tumor response assessments to be evaluated by the investigator.
- DXA of lumbar spine and hip (Day 113 only)
    - Refer to Imaging Acquisition Guidelines for guidelines regarding washout period for contrast (if used) for radiological images, prior to acquiring DXA scan.

10.1.7. Post Treatment Period Follow-up visits : Days 169, 197, 225, and 253 (± 7 days)

If a CT/MRI and bone scan (if applicable) is performed as per standard of care during the post treatment period follow up, results may be collected within the CRF to capture the status of the disease. Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Hematology
- Evaluation of transfusion frequency
- Documentation of concomitant medications (up to 90 days from the last dose of ACE-011/placebo)
- Evaluation of adverse events (up to 90 days from the last dose of ACE-011/placebo)

10.1.8. Termination Visit: Day 281 (± 7 days)

Day 281 is the final visit. If a patient terminates the study early, the Day 281 procedures should be followed. Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- FACT-Fatigue survey
- 12 lead ECG (in triplicate at 3 minute intervals (±1 min)
- Serum iron, total iron binding capacity, transferrin and serum ferritin
- Serum folate
- Coagulation
- Serum chemistry
- Hematology
- Serum erythropoietin marker
- Anti-drug antibody test
- Bone biomarkers- OC, BSAP, P1NP, CTX, TRACP-5b, and uNTX
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions on the collection of serum and urine for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Females of childbearing potential -Urine (dipstick) pregnancy test
- Urinalysis
- Evaluation of transfusion frequency
- Documentation of concomitant medications (up to 90 days from the last dose of ACE-011/placebo)
- Evaluation of adverse events (up to 90 days from the last dose of ACE-011/placebo)
- DXA of lumbar spine and hip
    - Refer to Imaging Acquisition Guidelines for guidelines regarding washout period for contrast (if used) for radiological images, prior to acquiring DXA scan.

- Bone scan (Day 281, +/- 5 day window). Follow-up scan to be acquired if performed at baseline and the scan was abnormal.

If a patient has a positive anti-drug antibody result at Day 281/Termination visit, the patient may be asked to return to the clinical site for additional monthly anti-drug antibody testing, up to 12 months after their last dose.

10.2. Discontinuation of study

The Sponsor may terminate this study, after consultation with the Investigator, at any time for safety or administrative reasons. The Sponsor may terminate the study if the occurrence of serious adverse events (SAEs) or other findings suggest unacceptable risk to the health of the patients.

11. INVESTIGATIONAL PRODUCT MATERIALS AND MANAGEMENT

11.1. Reference Product

The placebo (control agent) to be used in this study will be sterile normal saline (0.9% Sodium Chloride for Injection) administered as a SC injection. Sterile, normal saline will be supplied by the investigational site's pharmacy, as applicable. The manufacturer's directions for placebo storage and handling are to be followed, as are standard clinical practices for ensuring sterility of the placebo.

11.2. Investigational Product Packaging and Labeling

ACE-011 will be supplied in 2 mL clear glass vials with gray stoppers and red flip-top seals that contain 1 mL of ACE-011. The investigational drug product consists of ACE-011 in PBS at a nominal concentration of approximately 50 mg/mL.

11.3. Investigational Product Storage

ACE-011 is recommended to be stored at ≤ -65℃.

11.4. Investigational Product Preparation

See ACE-011 Handling and Administration Guidelines.

11.5. Administration

ACE-011/placebo will be administered by subcutaneous injection (SC). Subcutaneous injections will be given in the upper arm and/or thigh. Please refer to ACE-011 Handling and Administration Guidelines document for further information.

Dose modifications will be determined by the investigator after reference to the dose modification rules listed in Figure 1. In the case of a dose interruption or dose reduction of ACE-011, the investigator or designee will notify the pharmacy staff, who is unblinded, of the treatment decision for appropriate preparation of the investigational drug to maintain the blind of the study.

11.6. Drug Accountability

Accountability for ACE-011/placebo is the responsibility of the Investigator. Investigational clinical supplies must be received by a designated person at the clinical site and kept in a secured location. The investigational site must maintain accurate records demonstrating dates and amounts of ACE-011/placebo received, to whom it was dispensed (patient-by-patient accounting), and accounts of any ACE-011/placebo accidentally or deliberately destroyed or returned. Unless otherwise notified, all vials of ACE-011/placebo, both used and unused, must be saved for drug accountability. The used vials may be discarded, per the institution's standard

practice, after drug accountability has been completed by the monitor. The Investigator must return all unused vials of ACE-011 to the Sponsor at the end of the study, or the ACE-011 may be destroyed at the clinical site with the permission of the Sponsor. For either scenario, the outcome must be documented on the drug accountability log. The Sponsor will provide direction for the outcome of all unused vials.

11.7. Investigational Product Handling and Disposal

Please refer to the ACE-011 Handling and Administration Guidelines, provided under separate cover, for detailed drug handling, administration, and storage instructions.

12. ASSESSMENT OF EFFICACY

Efficacy measurements will include assessments for hematopoietic response which is defined as an increase in hemoglobin of ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs, at each dose level. The hematopoietic endpoints will be determined by the monitoring of hematologic laboratory values and RBC transfusion and/or ESA administration. Secondary endpoints associated with assessment of disease progression will be evaluated by CT/MRI scan and bone mineral density by DXA scans, respectively. Bone biomarkers and incidence of skeletal related events (SREs) will also be evaluated for all patients. SREs are defined as pathologic fracture, radiation therapy to bone, surgery to bone, or spinal cord compression. Quality of life assessment (FACT-Fatigue) will be evaluated for each patient.

13. ASSESSMENT OF SAFETY

13.1. Safety Parameters

13.1.1. Adverse Events

All patients will be assessed for safety by monitoring adverse events, clinical laboratory tests, vital signs and physical examinations. AE information will be collected throughout the study. See Section 14 Adverse Events for a full description of AE definitions, AE and SAE reporting procedures, and emergency procedures.

13.1.2. Clinical Laboratory Tests

The following laboratory safety assessments will be drawn at the investigational site's local laboratory according to the laboratory collection recommendations. The samples will then be sent to the central lab for value reporting. Please refer to Lab Reference Manual for further information. Hemoglobin and hematocrit lab tests will be drawn and evaluated by the local laboratory at screening and at each visit day throughout the study. The local hemoglobin laboratory value will be used in the evaluation of ACE-011/placebo dose modifications. The sponsor may request additional safety tests be performed based on ongoing data review during the study.

- Iron studies: Serum iron, total iron binding capacity (TIBC), transferrin and serum ferritin on specified days.
- Nutritional tests: Serum folate and vitamin B12
- Coagulation (central lab only): prothrombin time (PT), partial thromboplastin time (PTT), and fibrinogen
- Serum chemistry (central lab only): AST, ALT, lactate dehydrogenase (LDH), total bilirubin, indirect bilirubin, alkaline phosphatase, total protein, albumin, sodium, potassium, chloride, carbon dioxide, calcium, phosphorus, blood urea nitrogen (BUN), creatinine, uric acid, glucose, amylase, and lipase.
- Hematology: complete blood count (CBC) with differential and platelet. CBC includes red blood cells (RBC), white blood cells (WBC), hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC). Reticulocyte percent, erythropoietin levels and peripheral blood smear will be collected on specified days.
- Bone biomarkers: (serum) osteocalcin (OC), procollagen type 1 N-terminal propeptide (P1NP), serum bone specific alkaline phosphatase (BSAP), C-terminal type 1 collagen telopeptide (CTX), tartrate-resistant acid phosphatase isoform-5b (TRACP-5b), and urine N-telopeptide (uNTX).
- Pregnancy test for females of child bearing potential

- Urinalysis (central lab only): including determination of pH, specific gravity, protein, glucose, ketones, blood, leukocyte esterase and nitrite. Microscopy required only to follow-up abnormal findings.

13.1.3. Other Safety Assessments

- Physical examinations
- Vital signs: weight (kg), heart rate (beats/min), seated blood pressure (mmHg) and temperature (°C). Height will be collected at screening only.
- FACT-Fatigue survey
- 12-lead ECG (done in triplicate)
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test
- CT/MRI scan
- DXA
- Bone scans (for those patients with baseline bone metastases)

13.2. Pharmacokinetics

See Appendix 5 for further details regarding PK sampling.

14. ADVERSE EVENT

Adverse Event

An adverse event (AE) is any untoward medical occurrence in a patient administered a pharmaceutical product, which does not necessarily have a causal relationship with the treatment. An AE can be any unfavorable and unintended sign (e.g., including an abnormal laboratory finding), symptom, or disease temporally associated with the use of the investigational drug, whether or not it is considered to be investigational drug related. This includes any newly occurring event or previous condition that has increased in severity or frequency since the administration of investigational drug (treatment-emergent).

Serious Adverse Event

A serious adverse event (SAE) is any AE, occurring at any dose level and regardless of causality that:

- Results in death.
- Is life-threatening: Life-threatening means that the patient was at immediate risk of death from the reaction as it occurred, (i.e., it does not include a reaction which hypothetically might have caused death had it occurred in a more severe form).
- Requires inpatient hospitalization or prolongation of existing hospitalization.
- Results in persistent or significant disability/incapacity.
- Is a congenital anomaly/birth defect.
- Is an important medical event: an important medical event is an event that may not result in death, be life-threatening, or require hospitalization but may be considered an SAE when, based upon appropriate medical judgment, it may jeopardize the patient and may require medical or surgical intervention to prevent one of the outcomes listed in the definitions for SAEs. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.
- A hospitalization for an elective procedure will not be considered a SAE.

SAEs will be captured throughout the patient participation in the study through Day 281/Termination visit. SAEs will be captured up to 90 days from the last dose of ACE-011/placebo for those patients who are followed for survival only.

Unexpected Adverse Event

An unexpected AE is an AE that is not reported in the Investigator Brochure or in the case of an AE already described, the severity of which is not consistent with that described in the Investigator Brochure.

14.1. Adverse Event Classification

The following classifications should be used when evaluating the relationship of AEs and SAEs to the investigational drug.

None:	No relationship between the experience and the administration of investigational drug; related to other etiologies such as concomitant medications or patient's clinical state.
Unlikely:	The current state of knowledge indicates that a relationship is unlikely.
Possibly:	A reaction that follows a plausible temporal sequence from administration of the investigational drug and follows a known response pattern to the suspected investigational drug. The reaction might have been produced by the patient's clinical state or other modes of therapy administered to the patient.
Probably:	A reaction that follows a plausible temporal sequence from administration of the investigational drug and follows a known response pattern to the suspected investigational drug. The reaction cannot be reasonably explained by the known characteristics of the patient's clinical state or other modes of therapy administered to the patient.
Definitely:	A reaction that follows a plausible temporal sequence from administration of the investigational drug and follows a known response pattern to the suspected investigational drug and can be confirmed with a positive re-challenge test or supporting laboratory data.

Severity of AEs will be graded by the investigator using the National Cancer Institute (NCI; US) Common Terminology Criteria for Adverse Events (CTCAE) version 3.0 guidelines (Appendix 3).

14.2. Recording Adverse Events

Patients will be evaluated and questioned generally for AEs during the course of the study, starting from baseline (Day 1). The Medical History CRF should be comprehensive of patient's medical history up to Day 1. All AEs occurring after ACE-011/placebo administration up to 90 days from the last dose of ACE-011/placebo are to be documented on AE CRF. The specific disease or syndrome rather than individual associated signs and symptoms should be identified by the investigator and recorded on the CRF.

Specifically, skeletal related event information is to be captured and reported for the duration of the patient participation in the study through Day 281/Terminiation visit. SREs will be captured up to 90 days from the last dose of ACE-011/placebo for those patients who are followed for survival only.

Serious Adverse Events (SAEs)

For SAEs, a SAE form must be completed with as much information as possible and submitted in the time frame described in Section 14.3.

When new significant information is obtained as well as when the outcome of an event is known, the investigator should record the information on a new SAE form. If the patient was hospitalized, a copy of the discharge summary must be included as part of the patient medical file. In all instances, the investigator should follow up with patients until the outcome of the SAE is known.

14.3. Reporting Adverse Events

All AEs spontaneously reported by the patient and/or in response to an open question from study personnel or revealed by observation, physical examination or other diagnostic procedures will be recorded on the AE CRF. Any clinically relevant change in laboratory assessments or other clinical findings is considered an AE and must be recorded on the AE CRF. Clinically significant laboratory abnormalities are those that are identified as such by the investigator and/or those that require intervention. All AEs are to be followed until the event resolves or the clinical course is stabilized.

Serious Adverse Events

For SAEs, a SAE form must be completed with as much information as possible and submitted in the time frame described below.

All SAEs that occur during the course of the study (from the signing of the ICF and throughout patient participation in the study procedures) must be reported by the Investigator to PPD and Sponsor by faxing the SAE form within 24 hours from the point in time when the Investigator becomes aware of the SAE. All SAEs and deaths must be reported regardless of relationship to investigational drug.

SAE Reporting:

PPD Medical Monitor
PPD Inc.


If there are serious, unexpected AEs that are also associated with the use of the investigational drug, the Sponsor will notify the appropriate regulatory agency(ies) and all appropriate parties on an expedited basis. It is the responsibility of the Investigator to promptly notify the Institutional Review Board (IRB) of all unexpected SAEs involving risk to human patients.

14.4. Emergency Sponsor Contact

In the event of a medical emergency (i.e., an event that requires immediate attention regarding the treatment of a patient, operation of the clinical study, and/or the use of investigational drug), investigational site personnel must immediately contact the Medical Monitor.

15. STATISTICS

Statistics Analysis Overview

The study will be considered complete with regard to the primary endpoint once all eligible subjects have completed up to 2 months after the last dose of ACE-011. Patients will be randomized to one of three ACE-011 treatment groups (0.1, 0.3 and 0.5 mg/kg) or placebo with a ratio of 2:2:2:1. Patients will be stratified according to administration of either single agent weekly chemotherapy or less frequent chemotherapy regimens.

15.1. Determination of Sample Size

With an anticipated dropout rate of 20%, a sample size of 30 patients per dose level is estimated to achieve at least 80% power with 2 sided significance level of 0.05 to differentiate targeted response rate of p1=0.45 from null hypothesis p0=0.20. An additional 15 patients will be enrolled to the placebo group for safety comparison. A total of 105 patients will be randomized to three ACE-011 dose groups (0.1, 0.3, and 0.5 mg/kg) and placebo with a ratio of 2:2:2:1.

15.2. Analysis Populations

For this study, the following populations will be defined and used in the analysis and presentation of the data.

Modified Intent-to-Treat (MITT) population: The MITT population is defined as all patients randomized who received at least one dose of ACE-011/placebo.

Per-Protocol (PP) population: The PP population is defined as all patients randomized who have confirmed chemotherapy induced anemia, received at least one dose of ACE-011 and on study (treatment) for at least 2 months (57 days).

Safety population: The safety population is defined as all patients who received at least one dose of ACE-011 or placebo.

15.3. Background and Demographic Characteristics

Summary statistics (mean, standard deviation, median, minimum and maximum) will be provided for those variables measured on a continuous scale. The frequency distribution (number and percentage) will be provided for those variables measured on a nominal scale.

15.3.1. Patient Disposition

Patient disposition (analysis population allocation, entered, discontinued, along with primary reason for discontinuation) will be summarized using frequency and percent for each dose group. A summary of patients enrolled by site will be provided.

15.3.2. ACE-011

ACE-011 treatment exposure (total dosage received) will be summarized for each dose group as well as for each cycle for each dose level. Listings for dose adjustment will be provided.

15.3.3. Concomitant Therapy

By-patient listings will be provided for all concomitant medications and therapies taken during the study. All concomitant medications will be coded and categorized by the WHO drug coding system. Usage frequency of each coded concomitant medications will be summarized by each dose group.

15.4. Efficacy Evaluation

15.4.1. Primary Efficacy Analysis

The primary efficacy endpoint is the hematopoietic response rate at each dose level. It is defined as the proportion of patients who have a hematopoietic response of an increase in hemoglobin of ≥ 1.0 g/dL for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs. The ACE-011 treatment period is defined as from the time of first ACE-011 dose to up to 2 months after the last dose of ACE-011 treatment. To be considered as a responder, all hemoglobin measurements during a 28 consecutive day period or longer must have an increase of ≥ 1.0 g/dL from baseline. If RBC transfusions and/or ESAs are required, no hemoglobin measurements within 28 days will be used to determine the hemoglobin response. Primary efficacy analysis will be based on PP population. Primary efficacy analysis will also be conducted on MITT population as supportive to the PP based analysis. Patients who do not receive ACE-011 will be excluded from primary efficacy analysis. Patients who discontinue the ACE-011 treatment prematurely, with no sufficient hemoglobin measurement, will be counted as non-responder in the calculation of response rate.

A 95% confidence interval (CI) for the response rate for each dose level will be provided to assess whether 95% CI of response rate contains null hypothesis response rate, p0=0.20. The response rate and 95% CI will also be provided for patients with chemotherapy on weekly basis versus those with chemotherapy on a less frequent basis for each dose level as well as overall (all three doses levels analyzed together).

An exploratory analysis with the Kaplan Meier approach will be considered to account for those patients who discontinue the ACE-011 treatment prematurely if the drop out rate is greater than 20%. Exploratory analysis of comparing hematopoietic response rates between active and placebo will be performed using Fisher's exact test. Due to the nature of the study (phase 2 dose determination study), no multiplicity adjustment will made for the multiple comparisons.

15.4.2. Secondary Efficacy Analyses

The secondary efficacy endpoints include:

- Proportion of patients achieving an increase in hemoglobin of ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for 28 consecutive days during the treatment period and up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs.
- Duration of hematopoietic response (days) defined as the first time hemoglobin increases at least ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL from baseline to the last time there is hemoglobin ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL increase from baseline. Duration of response is only calculated for a responder and will be at least 28 days. The duration of response will be at least 28 days is only calculated for a patient who meets the primary efficacy endpoint.
- Time to achieve hematopoietic response (days) defined as time from first dose of ACE-011 to the first time a hemoglobin result at least ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL increase from baseline and maintained for a period of at least 28 consecutive days.
- Proportion of patients receiving red blood cell (RBC) transfusions and/or ESAs in each dose group as well as within each cycle of each ACE-011 dose level.
- Objective tumor response rate for each dose level using RECIST criteria.
- Progression-Free Survival is defined as the time from the start date of current chemotherapy regimen to the first observation of documented disease progression or death due to any cause. If the subject has not progressed or died at the time of final analysis, PFS will be censored at the time of last tumor assessment.

The statistical analysis for binary endpoints will be similar to the primary efficacy analysis. For time to event endpoints, the Kaplan-Meier method will be used to estimate the survival curve and median time to event. These analyses will be performed for each dose group and all ACE-011 groups combined.

15.5. Safety Evaluation

Safety variables will be tabulated and presented for all patients who receive ACE-011. Adverse events will be coded using the Medical Dictionary for Regulatory Activities (MedDRA). Incidence of treatment-emergent adverse events will be presented by system organ class (SOC) and preferred term. Adverse events will also be presented by severity, and relationship to investigational drug. Change from baseline in clinical laboratory parameters and vital signs will be summarized across time. Shift tables will be presented for selected laboratory parameters. Physical examination results will be presented in listings.

Data from all patients who receive one or more doses of ACE-011/placebo will be included in the safety analyses. To assess clinical safety, adverse events, vital sign measurements and clinical laboratory information will be summarized by visit. Adverse events and laboratory results will also be summarized by severity grade (NCI CTCAE v3.0). Descriptive statistics will be generated and shift tables provided as appropriate.

15.6. Pharmacokinetic Evaluation

Listing of individual patient serum ACE-011 concentrations, actual blood sampling times and graphs of concentration vs. time will be prepared for each dose group. Summaries of PK parameters will be summarized by dose group. The trough concentrations will be summarized for all patients who provide pharmacokinetic samples by dose group. Exploratory analyses will be performed to correlate trough concentrations with efficacy, bone biomarker data, and safety data, however these analyses will be data-driven and only conducted if warranted by the data.

15.6.1. Anti-drug Antibody Data

The results of anti-drug and neutralizing antibodies will be presented over time. Exploratory analysis will be performed on the potential effect of anti-drug antibody on ACE-011 PK and drug exposure if anti-drug antibody tests are deemed positive.

15.7. Interim Analysis

There is no planned interim analysis for this study.

15.8. Deviation from Original Analysis Plan

Deviations from the statistical analyses outlined in this protocol will be indicated in the statistical analysis plan; any further modifications will be noted in the final clinical study report.

16. DIRECT ACCESS TO SOURCE DATA/DOCUMENTS

16.1. Study Monitoring

The Clinical Monitor will arrange to visit the Investigator sites at regular intervals during the study. The monitoring visits must be conducted according to the applicable ICH and GCP guidelines to ensure protocol adherence, quality of data, drug accountability, compliance with regulatory requirements, and continued adequacy of the investigational site and its facilities. During these visits, CRFs and other data related to the study will be reviewed and any discrepancies or omissions will be resolved. The Clinical Monitor will be given access to study relevant source documents (including medical records) for purposes of source data verification.

During and/or after completion of the study, quality assurance officers named by the Sponsor or the regulatory authorities may wish to perform on-site audits. The Investigator is expected to cooperate with any audit and provide assistance and documentation (including source data) as requested.

16.2. Audits and Inspections

The Investigators and clinical sites will permit trial related monitoring, audits, IRB review, and regulatory inspections as requested by FDA, the Sponsor or designee, including direct access to source data/documents (i.e., original medical records, laboratory reports, hospital documents, progress reports, signed ICFs, etc.) in addition to CRFs.

17. QUALITY CONTROL AND QUALITY ASSURANCE

17.1. Data Quality Control and Quality Assurance

17.1.1. Investigator responsibility

The investigator is responsible for ensuring the study is conducted according to the protocol, Code of Federal Regulations (CFR), GCP, and any other applicable regulatory requirements. The investigator responsibilities are outlined in these documents along with ensuring that documentation of a signed informed consent, is obtained prior to patient participation in the study.

17.1.2. Protocol modifications

The investigator may not modify the protocol without agreement from the Sponsor and prior review or approval by the IRB. Any deviations from the protocol should be documented by the investigator or designee.

17.2. Financing and Insurance

Financing and insurance are addressed in a separate document.

17.3. Confidentiality

To maintain patient privacy, all CRFs, drug accountability records, study reports and communications will identify the patient by the assigned patient identification number. The Investigator will grant monitor(s) and auditor(s) from the Sponsor or designee and regulatory authority(ies) access to the patient's original medical records for verification of data gathered on the CRFs and to audit the data collection process. The patient's confidentiality will be maintained and will not be made publicly available to the extent permitted by the applicable laws and regulations.

All information regarding the investigational product supplied by the Sponsor to the Investigator is privileged and confidential information. The Investigator agrees to use this information to accomplish the study and will not use it for other purposes without consent from the Sponsor. It is understood that there is an obligation to provide the Sponsor with complete data obtained during the study. The information obtained from the clinical study will be used towards the development of the investigational product and may be disclosed to regulatory authority(ies), other Investigators, corporate partners, or consultants as required.

17.4. Publication Policy

All information concerning ACE-011 is considered confidential and shall remain the sole property of the Sponsor. The Investigator agrees to use this information only in conducting the study and shall not use it for any other purposes without the Sponsor's written approval. The Investigator agrees not to disclose the Sponsor's confidential information to anyone except to

people involved in the study that need such information to assist in conducting the study, and then only on like terms of confidentiality and nonuse.

It is understood by the Investigator that the information developed from this clinical study will be used by the Sponsor in connection with the development of ACE-011, and therefore may be disclosed as required to regulatory agencies. To allow for the use of the information derived from clinical studies, it is understood that there is an obligation to provide the Sponsor with complete test results and all data developed in the study.

No publication or disclosure of study results will be permitted except as specified in a separate, written, agreement between the Sponsor and the Investigator.

17.5. Protocol Amendments

Protocol amendments that impact patient safety change the scope of the investigation, or affect the scientific quality of the study must be approved by the IRB and submitted to the appropriate regulatory authorities before implementation of such modifications to the study.

In the event that the protocol needs to be modified immediately to eliminate an apparent hazard to a patient, the Sponsor will amend and implement the protocol change and subsequently notify the regulatory authorities and/or the IRB, as appropriate.

18. ETHICS

18.1. Institutional Review Board or Independent Ethics Committee

The Investigator must obtain written IRB approval of the protocol, approval for relevant supporting information and all types of patient recruitment and advertisement and ICF prior to starting the study. The IRB will meet all Food and Drug Administration (FDA) requirements governing IRBs (Code of Federal Regulations, Title 21, Part 56).

The Sponsor or designee must approve the ICF submitted to the investigational site's IRB. All patient recruitment and advertisements must be submitted to the Sponsor or designee prior to submission to the IRB, for review.

18.2. Ethical Conduct of the Study

The Sponsor and the Investigator must comply with all instructions, regulations, and agreements in this protocol and in the applicable International Conference on Harmonisation (ICH) and GCP guidelines, and must also conduct the study in accordance with local regulations.

18.3. Written Informed Consent

18.3.1. Informed Consent

Informed written consent is required from each patient prior to any testing under this protocol, including screening tests and evaluations. The consent form, as specified by the clinical site's IRB, must follow the Protection of Human Patients regulations listed in the Code of Federal Regulations, Title 21, Part 50.

The background of the proposed study and the benefits and risks of the procedures and study must be explained to the patients. It is the responsibility of the Investigator to obtain consent and to provide the patient with a copy of the signed and dated ICF. Confirmation of a patient's informed consent must also be documented in the patient's medical record prior to any testing under this protocol, including screening tests and evaluations.

All ICFs used in this study must be approved by the appropriate IRB and by the Sponsor or designee. The ICF must not be altered without the prior agreement of the relevant IRB and the Sponsor.

18.3.2. Patient Data Protection

Prior to any testing under this protocol, including screening tests and evaluations, patients must authorize the release and use of protected health information (PHI), as required by local law.

The patient will not be identified by name in the CRF or in any study reports. These reports will be used for research purposes only. The Sponsor, its designee, and various government health agencies may inspect the records of this study. Every effort will be made to keep the patient's personal medical data confidential.

19. DATA HANDLING AND RECORDKEEPING

19.1. Case Report Form Completion

CRFs will be completed for each randomized patient. It is the Investigator's responsibility to ensure the accuracy, completeness, and timeliness of the data reported in the patient's CRF. Source documentation supporting the CRF data should indicate the patient's participation in the study and should document the dates and details of study procedures, AEs, and patient status.

Investigators will maintain copies of the CRFs at the clinical site. For patients who discontinue or terminate from the study, the CRFs will be completed as much as possible, and the reason for the discontinuation or termination clearly and concisely specified on the appropriate CRF.

19.2. Retention of Records

The Investigator will maintain all study records according to ICH-GCP and applicable regulatory requirements. Records will be retained for at least 2 years after the last marketing application approval or 2 years after formal discontinuation of the clinical development of the investigational product or according to applicable regulatory requirements. If the Investigator withdraws from the responsibility of keeping the study records, custody must be transferred to a person willing to accept the responsibility. The Sponsor must be notified in writing if a custodial change occurs.

20. REFERENCES

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21. APPENDICES

Appendix 1: Response Evaluation Criteria in Solid Tumors

Response Evaluation Criteria in Solid Tumors (RECIST) v1.1	
Terms	Definition
Measurable disease	The presence of at least one measurable lesion by CT or MRI. If the measurable disease is restricted to a solitary lesion, its neoplastic nature should be confirmed by cytology/histology. All measurements should be taken and recorded in metric notation, using a ruler or calipers. All baseline evaluations should be performed as closely as possible to the beginning of treatment and never more than 4 weeks before the beginning of the treatment.
Measurable lesions	Lesions that can be accurately measured in least one dimension by CT scan with longest diameter $\geq$ 10 mm (CT scan thickness no greater than 5 mm). The same method of assessment and the same technique should be used to characterize each identified and reported lesion at baseline and during follow-up.
Non-measurable lesion	All other lesions, including small lesions (longest diameter <10mm or pathological lymph nodes with P10 to <15mm short axis) as well as truly non-measurable lesions. Lesions considered truly non-measurable include: leptomeningeal disease, ascites, pleural or pericardial effusion, inflammatory breast disease, lymphangitic involvement of skin or lung, abdominal masses/abdominal organomegaly identified by physical exam that is not measurable by reproducible imaging techniques.

RECIST Criteria-Response Evaluation

Evaluation of target lesions	
Complete Response (CR)	Disappearance of all target lesions. Any pathological lymph nodes (whether target or non-target) must have reduction in short axis to <10 mm.
Partial Response (PR):	At least a 30% decrease in the sum of diameters of target lesions, taking as reference the baseline sum diameters.
Stable Disease (SD):	Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum diameters while on study.
Progressive Disease (PD):	At least a 20% increase in the sum of diameters of target lesions, taking as reference the smallest sum on study (this includes the baseline sum if that is the smallest on study). In addition to the relative increase of 20%, the sum must also demonstrate an absolute increase of at least 5 mm. (Note: the appearance of one or more new lesions is also considered progression).

Evaluation of non-target lesions	
Complete Response (CR):	Disappearance of all non-target lesions and normalisation of tumour marker level. All lymph nodes must be non-pathological in size (<10mm short axis).
Non-CR/Non-PD	Persistence of one or more non-target lesion(s) and/or maintenance of tumour marker level above the normal limits.
Progressive Disease (PD):	Unequivocal progression (see comments below) of existing non-target lesions. (Note: the appearance of one or more new lesions is also considered progression).

Evaluation of best overall response	The best overall response is the best response recorded from the start of the treatment until disease progression/recurrence (taking as reference for PD the smallest measurements recorded since the treatment started). In general, the patient's best response assignment will depend on the achievement of both measurement and confirmation criteria. Patients with a global deterioration of health status requiring discontinuation of treatment without objective evidence of disease progression at that time should be classified as having "symptomatic deterioration". Every effort should be made to document the objective progression even after discontinuation of treatment. In some circumstances it may be difficult to distinguish residual disease from normal tissue. When the evaluation of complete response depends on this determination, it is recommended that the residual lesion be investigated (fine needle aspirate/biopsy) to confirm the complete response status.

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Appendix 2: ECOG Performance Status

The ECOG scale is used to assess a patient's quality of life in an evaluation by a health professional of the daily activities and how the activities are affected by the disease of the patient.

Grade	ECOG
0	Fully active, able to carry on all pre-disease performance without restriction
1	Restricted in physically strenuous activity but ambulatory and able to carry out work of a light or sedentary nature, e.g., light house work, office work
2	Ambulatory and capable of all selfcare but unable to carry out any work activities. Up and about more than 50% of waking hours
3	Capable of only limited selfcare, confined to bed or chair more than 50% of waking hours
4	Completely disabled. Cannot carry on any selfcare. Totally confined to bed or chair
5	Dead

Oken, M.M., Creech, R.H., Tormey, D.C., Horton, J., Davis, T.E., McFadden, E.T., Carbone, P.P. (1982) Toxicity And Response Criteria Of The Eastern Cooperative Oncology Group. Am J Clin Onco 5:649-655.

Appendix 3: National Cancer Institute (NCI) Common Terminology Criteria for Adverse Events (CTCAE) Version 3.0

See http://ctep.cancer.gov/reporting/ctc.html

Appendix 4: New York Heart Association- Classification of heart failure

Class 1 - Class 1 heart failure - no limitation of activities. No symptoms from ordinary activities.
Class 2 - Class 2 heart failure- mild limitation of activity. Comfortable with rest or mild exertion.
Class 3 - Class 3 heart failure- marked limitation of activity and be comfortable only at rest.
Class 4 –Class 4 heart failure- complete rest, confined to bed or chair; any physical activity brings on discomfort and symptoms occur at rest.

National Heart, Lung, and Blood Institute, National Institutes of Health. New York Heart Association Classification. 2008.

Appendix 5- Pharmacokinetic Sampling

Pharmacokinetics

Blood samples will be collected from approximately one-third of patients at selected study sites for the evaluation of serum concentrations of ACE-011. Pre-dose PK samples will be collected on Day 1 and Day 85.

Blood samples for PK should be collected in a fasted state (defined by no food or drink except water for at least 4 hours prior to the study procedure) with the exception of the Day 1 post 4 hour dose and the Day 85 post 4 hour dose. Pre-dose samples should be collected prior to ACE-011/placebo dosing (Day 1, 29, 57, and 85). The post 4 hour collection (± 10 minute window) on Day 1 and Day 85 is calculated to be 4 hours post the administration of ACE-011/placebo. Chemotherapy regimens scheduled will be administered to the patient as per standard of care. Collection, handling, and shipping procedures for blood samples are provided in the Study Reference Guide.

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Schedule of Events cont.

	Screen	Treatment Period														Post Treatment Follow up Period				Term Visit[20]
ACE-011/Placebo Dose period		#1				#2			#3			#4	Follow-up							
Day	-14-0	1	8 (± 1d)	15 (± 1d)	22 (± 1d)	29 (± 3d)	36 (± 1d)	43 (± 2d)	57 (± 3d)	64 (± 2d)	71 (± 2d)	85 (± 3d)	99 (± 3d)	113 (± 3d)	141 (±7d)	169 (± 7d)	197 (± 7d)	225 (± 7d)	253 (± 7d)	281 (± 7d)
PK Sampling		X X- Post 4 hours	X	X	X	X	X		X	X		X X-Post 4 hours		X						
ACE-011/Placebo administration		X				X			X			X								
Chemo regimen		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

Protocol Amendment – Summary of Changes

CLINICAL STUDY PROTOCOL

A Phase 2, Double-Blind, Randomized, Placebo-Controlled Study of ACE-011 for the Treatment of Chemotherapy Induced Anemia in Patients with Metastatic Breast Cancer

STUDY DRUG:	**ACE-011**
PROTOCOL NUMBER:	**A011-08**
SPONSOR:	**Celgene Corporation** **86 Morris Avenue** **Summit, NJ 07901**
MEDICAL MONITOR:	**PPD Inc.**

PROTOCOL DATE:	**13 March 2009**
AMENDMENT #1:	**22 April 2009**
AMENDMENT #2:	**21 May 2009**
AMENDMENT #3:	**07 October 2009**
AMENDMENT #4:	**04 February 2010**

Summary:
Amendment 04 includes the following changes:

- Administrative changes
- Addition of Celgene Corporation at the Sponsor and deletion of Acceleron Pharma as the Sponsor
- Amend inclusion criteria #4 to include capecitabine
- Amend inclusion criteria #10 to add AST/ALT ≤ 5x ULN when there is evidence of liver metastases.
- Amend exclusion criteria # 2 with the deletion of "Use of palliative radiation if the area being treated is <15% of body surface area with no pelvic radiation or no more than 10% of the bone marrow reserve is radiated is permitted during patient participation in the study, at the discretion of the investigator."
- Addition of language to exclusion #3 "not including the current chemotherapy regimen"
- Amend language of exclusion #6 to "Recent history of thrombosis, deep vein thrombosis (DVT), pulmonary emboli, or embolic stroke, occurring within the last 6 months; if on anticoagulation therapy must be clinically stable as determined by the Investigator."
- Amend language of exclusion #10 "Administration of IV antibiotics or febrile (temperature elevation > 38 ° C) within 14 days of Day 1."
- Addition of language to exclusion #12 "or active hepatitis C."
- Amend language of exclusion criteria #13 "Clinically significant iron (transferrin saturation < 20%), vitamin B_{12}, or folate deficiency."
- Deletion of the following visits days from the schedule of events: Day 22, 36, 64, 99, 197, and 253.
- Decrease in frequency of the collection of ECGs, coagulation, chemistry and anti-drug antibody blood samples.
- Deletion of urine NTX biomarker, serum ferritin and transferrin blood sample collection.
- Addition of blood pressure measurements during the follow up period of the study.
- Addition of text regarding blood pressure measurements for those patients with a history of CNS metastases.
- Amend language within Section 14-Adverse Events and Section 15- Statistics

Protocol Location	Change
Cover page, Signature page and Table 1	Change in sponsor • Deletion of Acceleron Pharma • Addition of Celgene Corporation • Administrative changes

Section 2. Protocol Synopsis	*Name of Sponsor* o Deletion of Acceleron Pharma o Addition of Celgene Corporation *Study center* o Amend the number of sites to 35-45 centers *Duration of study* o Language was amended to state that patients may be asked to return to the clinic every 3 months if a positive anti-drug antibody is noted, until a negative result is noted. *Safety* o Deletion of "assessment of quality of life". o Amended the frequency of the ECG collection timepoints.
Section 2. Protocol Synopsis and Section 6.3 Exploratory objectives	*Objectives* o Deletion of urine NTX
Section 2. Protocol Synopsis-Inclusion/Exclusion criteria and Section 8-Selection	*Inclusion criteria (Section 8.2.1 Patient inclusion criteria)* o Amend inclusion criteria #4 to include capecitabine o Amend inclusion criteria #10 to add AST/ALT ≤ 5x ULN when there is evidence of liver metastases. *Exclusion criteria (Section 8.2.2 Patient exclusion criteria)* o Addition of language to exclusion #3 "not including the current chemotherapy regimen" o Amend language of exclusion #6 to "Recent history of thrombosis, deep vein thrombosis (DVT), pulmonary emboli, or embolic stroke, occurring within the last 6 months; if on anticoagulation therapy must be clinically stable as determined by the Investigator." o Amend language of exclusion #10 to be "Administration of IV antibiotics or febrile (temperature elevation > 38 ° C) within 14 days of Day 1." o Addition of language to exclusion #12 "or active hepatitis C." o Amend language of exclusion criteria #13 "Clinically significant iron (transferrin saturation < 20%), vitamin B_{12}, or folate deficiency." o Amend timeframe for exclusion criteria #15 from 12 months to 6 months for active gastrointestinal bleeding.

Schedule of Events (Table 7)	o Formatting o Deletion of the following visits days from the schedule of events: Day 22, 36, 64, 99, 197, and 253. o Decrease in frequency of the collection of ECGs, coagulation, chemistry and anti-drug antibody blood samples. o Deletion of urine NTX biomarker, serum ferritin and transferrin blood sample collection. o Addition of blood pressure measurements during the follow up period of the study. o Deletion of assessment of collection of menstrual cycle information o Addition of the following language to footnote 1: For patients with previous history of CNS metastases, if BP value increases by one or more NCI CTCAE v3.0 grades, the patient must return within 7 days to have BP assessed again. Patients with a recent change in anti-hypertensive medication or a change in the past month should be seen weekly for BP measurements. o Clarification provided regarding ECG collection o Deleted language regarding withdraw criteria in footnote 19 and 20
Section 5.4.3 A011-04: A Phase 2a Study in Patients with Osteolytic Lesions of Multiple Myeloma	o Replaced language regarding data review with “Please refer to the Investigator Brochure for further information.”

Section 7. Investigational Plan	○ Administrative changes ○ Language was amended to state that patients may be asked to return to the clinic every 3 months if a positive anti-drug antibody is noted, until a negative result is noted.
Section 8. Selection	*Section 8.3 Patient Withdrawal Criteria* ○ Clarification of reason for withdrawal and the visit(s) to be complete upon patient withdrawal.
Section 9. Treatment of Patients	Updated language through to reflect the changes in protocol visits *Section 9.1.1 General Concomitant Medication Usage* ○ Addition of language- "Patients with a recent change in anti-hypertensive medication or a change in the past month should be seen weekly for BP measurements." *Section 9.1.2.1 Iron supplementation* ○ Replace serum ferritin level with transferrin saturation < 20 % *Section 9.6 Treatments Administered* ○ Amended language regarding withdrawal criteria to reference section 8.3 only ○ Deletion of language regarding chemotherapy treatment
Section 10. Study Procedures and Schedule	Administrative changes *Section 10.1.2 Screening (within 14 days of Day 1)* • Amended language regarding the ECGs: At two different time intervals (approximately 1 hour apart) triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min) with the use of a digital ECG machine ○ If a local ECG machine is used, a single (12 lead) ECG is to be performed. • Replace transferrin and serum ferritin with transferrin saturation % • Deletion of: Females of childbearing potential- collection of menstrual cycle information of the start and end date of the last menstruation. • Screening bone scan collection timeframe amended from 4 weeks prior to start of chemotherapy regimen to 6 weeks prior. *Section 10.1.3 Initial Dosing: Day 1* ○ Deletion of the following language: A patient will be considered randomized once a multi-digit patient identification number has been assigned to the patient through the IVRS/IWRS. ○ Added the following language : "and the patient has been randomized (through IVRS/IWRS)" ○ Amended language regarding the ECGs: Triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min) with the use of a digital ECG

	machine o If a local ECG machine is used, a single (12 lead) ECG is to be performed. o Replace transferrin and serum ferritin with transferrin saturation % o Deletion of urine NTX o Deletion of: Females of childbearing potential- collection of menstrual cycle information of the start and end date of the last menstruation. *Section 10.1.4 ACE-011/placebo administration: Days 29, 57, and 85 (± 3 days)* o Replace transferrin and serum ferritin with transferrin saturation % on Day 57 only o Collection of coagulation and anti-drug antibody blood samples on Day 57 only o Amended language regarding ECGs: Triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min) with the use of a digital ECG machine (Day 57 only) o If a local ECG machine is used, a single (12 lead) ECG is to be performed. o Deletion of urine NTX o Deletion of: Females of childbearing potential- collection of menstrual cycle information of the start and end date of the last menstruation. *Section 10.1.5 Additional visits during the Treatment Period: Days 8, 15, (± 1 day), 43 and 71 (± 2 days)* o Patients with previous history of CNS metastases, if blood pressure (BP) value increases by one or more grade(s) per the NCI CTCAE v3.0, the patient must return within 7 days to have BP assessed again o Amended language regarding ECGs : Triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min) with the use of a digital ECG machine (Day 8 only) o If a local ECG machine is used, a single (12 lead) ECG is to be performed. o Deletion of collection of serum chemistry, folate and coagulation o *Section 10.1.6 Treatment Period Follow-up visits : Days 113 and 141(± 7 days)* Amended language regarding ECGs : Triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min) with the use of a digital ECG machine (Day 113 only) o If a local ECG machine is used, a single (12 lead) ECG is to be performed. o Deletion of urine NTX

	○ Extended window for CT/MRI scan from +/- 5 days to +/- 7 days *Section 10.1.7 Post treatment follow up period visits: Day 169 and 225 (± 7 days)* ○ Addition of blood pressure value collection *Section 10.1.8 Termination Visit: Day 281 (± 7 days)* ○ Replace transferrin and serum ferritin with transferrin saturation % ○ Amended language regarding ECGs: Triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min) with the use of a digital ECG machine ○ If a local ECG machine is used, a single (12 lead) ECG is to be performed. ○ Deletion of urine NTX and serum folate ○ Extended window for bone scan from +/- 5 days to +/- 7 days ○ Additional language regarding positive anti-drug antibody test follow up
Section 13 Assessment of Safety	*Section 13.1.2 Clinical laboratory tests* ○ Deletion of serum ferritin, transferrin, and uNTX
Section 14. Adverse Event	○ Formatting. ○ Additional text added regarding AEs, SAEs, and Pregnancy
Section 16. Direct Access to Source Data/Documents Section 19. Data Handling and Record Keeping	○ Additional language added for clarification purposes.

CLINICAL STUDY PROTOCOL

A Phase 2, Double-Blind, Randomized, Placebo-Controlled Study of ACE-011 for the Treatment of Chemotherapy Induced Anemia in Patients with Metastatic Breast Cancer

STUDY DRUG:	**ACE-011**
PROTOCOL NUMBER:	**A011-08**
IND NUMBER:	**103,362**
SPONSOR:	**Celgene Corporation** **86 Morris Avenue** **Summit, NJ 07901**
MEDICAL MONITOR:	**PPD Inc.** PPD [redacted]
PROTOCOL DATE:	**13 March 2009**
AMENDMENT #1:	**22 April 2009**
AMENDMENT #2:	**21 May 2009**
AMENDMENT #3:	**07 October 2009**
AMENDMENT #4:	**04 February 2010**

Signature Page

Celgene Corporation Approval

Signature: PPD _____ **Date:** 24 Feb 2010

Name (print): _____

Investigator Agreement: I have read the protocol and agree to conduct the study as outlined in the protocol. The study will be conducted in accordance to current United States Food and Drug Administration (FDA) regulations, International Conference of Harmonization (ICH) Guidelines, Good Clinical Practices (GCP), and local ethical and legal requirements.

Signature: _____ **Date:** _____

Name (print): _____

Institution Name and Address:

_____

_____

_____

PROCEDURES IN CASE OF EMERGENCY

Table 1: Emergency Contact Information

Role in Study	Name	Address and Telephone number
PPD Medical Monitor	PPD	PPD Inc. 1800 Perimeter Park Drive, Suite 275 Morrisville, NC 27560-7200 USA PPD

2. PROTOCOL SYNOPSIS

Name of Sponsor/Company: Celgene Corporation 86 Morris Avenue Summit, NJ 07901
Name of Investigational Product: ACE-011
Name of Active Ingredient: ACE-011 is a fully human fusion protein consisting of the extracellular domain (ECD) of the activin receptor IIA (ActRIIA) linked to the human IgG1 Fc domain. Mechanism of Action: ACE-011 is a disulfide-linked, glycosylated, dimeric protein. ACE-011 competes with the activin receptor IIA that binds a number of TGF-β superfamily ligands including activin, myostatin (growth differentiation factor [GDF]-8), and GDF-11.
Title of Study: A Phase 2, Double-Blind, Randomized, Placebo-Controlled Study of ACE-011 for the Treatment of Chemotherapy Induced Anemia in Patients with Metastatic Breast Cancer.
Study center(s): approximately 35-45 centers
Phase of development: 2
Objectives **Primary:** 1. To evaluate the proportion of patients in each ACE-011 dose regimen who achieve a hematopoietic response during the treatment period including up to 2 months after the last dose of ACE-011 treatment of chemotherapy induced anemia in patients with metastatic breast cancer. Hematopoietic response is defined as an increase in hemoglobin of ≥ 1.0 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs. **Secondary:** 1. To evaluate the safety and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer. 2. To evaluate the percentage of patients who achieve an increase in hemoglobin of ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs. 3. To estimate the duration of the hematopoietic response associated with each ACE-011 dose regimen. 4. To estimate the time to the hematopoietic response associated with each ACE-011 dose regimen. 5. To estimate the proportion of patients receiving red blood cell (RBC) transfusions

and/or ESAs within each cycle of ACE-011 with each dose regimen.

6. To estimate the effect of each ACE-011 dose regimen on progression free survival (PFS) and objective response rate (ORR).

Exploratory objectives:

1. To estimate the effect of each ACE-011 dose regimen on serum bone biomarkers (OC, BSAP, P1NP, CTX, and TRACP-5b), bone mineral density (dual-energy X-ray absorptiometry [DXA] scans of lumbar spine and hip) and incidence of skeletal related events (SREs).
2. To evaluate the quality of life in relation to the treatment of chemotherapy induced anemia in patients with metastatic breast cancer during the study.

Methodology: This is a Phase 2, double-blind, randomized, placebo-controlled study to evaluate the efficacy, safety, and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer. Patients will be randomized to one of three treatment arms or placebo:

1. ACE-011 0.1 mg/kg subcutaneously (SC) every 28 days for up to 4 doses.
2. ACE-011 0.3 mg/kg SC every 28 days for up to 4 doses.
3. ACE-011 0.5 mg/kg SC every 28 days for up to 4 doses.
4. Placebo SC every 28 days for up to 4 doses.

Number of patients (planned): 105 patients

Diagnosis and main criteria for inclusion

Inclusion Criteria:

1. Women ≥ 18 years of age.
2. Histologically or cytologically confirmed diagnosis of breast cancer.
3. Evidence of metastatic breast cancer with a minimum of one measurable lesion or non measurable lesion per RECIST v1.1 criteria (Appendix 1).
4. Receiving a multicycle myelosuppressive chemotherapy regimen including one of the following: anthracycline, taxane, gemcitabine, vinorelbine or capecitabine.
5. Anticipated treatment with the same myelosuppressive chemotherapy regimen for a minimum of 9 weeks after Day 1 of ACE-011/placebo administration.
6. Hemoglobin value between ≥ 6.5 to < 11.0 g/dL (≥ 65 to < 110 g/L).
7. ≥ 30 days must have elapsed (from Day 1) since previous treatment with erythropoiesis-stimulating agent (including concurrent treatment with IV iron) for chemotherapy induced anemia.
8. ≥ 7 days have elapsed (from Day 1) since the last RBC blood transfusion and receipt of ≤ 2 units of blood in the past 30 days.
9. ECOG Performance status of 0 – 2 (Appendix 2).
10. Adequate renal (creatinine ≤ 1.5 x ULN or creatinine clearance ≥ 40 mL/min) and hepatic function [bilirubin ≤ 1.5 x ULN; AST/ALT ≤ 2.5 x ULN (AST/ALT ≤ 5 x ULN when there is evidence of liver metastases)].
11. Females of childbearing potential participating in the study are to use adequate birth control measures (i.e. oral contraceptives or two forms of barrier method birth control) during study participation. Females of childbearing potential must have a negative serum pregnancy test within 7 days to ACE-011/placebo administration (Day 1). A female of childbearing potential is a sexually mature woman who has not undergone a hysterectomy or who has not been postmenopausal for at least 24 consecutive months (i.e. who has had menses at some time in the preceding 24 months).
12. Life expectancy of ≥ 6 months.
13. Is willing to adhere to the study visit schedule, understand and comply with all protocol requirements.
14. Understand and sign a written informed consent.

Exclusion Criteria:

1. At the time of screening patients who have any grade ≥ 3 toxicity (as per NCI CTCAE v3.0 (Appendix 3)) (except for the following disease related toxicities: hematological events- anemia, thrombocytopenia or neutropenia or non hematological events- nausea, vomiting, fatigue, or muscle or bone/joint pain).
2. Prior radiation therapy to > 20% of the whole skeleton.
3. Have received more than 5 prior chemotherapy treatment regimens for metastatic breast cancer (not including the current chemotherapy regimen).
4. Clinically significant pulmonary, endocrine, neurologic, gastrointestinal, hepatic or genitourinary disease unrelated to underlying hematologic disorder.
5. Patients with heart failure as classified by the New York Heart Association (NYHA) classification of 3 or higher (Appendix 4).
6. Recent history of thrombosis, deep vein thrombosis (DVT), pulmonary emboli, or embolic stroke, occurring within the last 6 months; if on anticoagulation therapy must be clinically stable as determined by the Investigator.
7. Untreated CNS metastases or CNS metastases treated with whole brain radiotherapy < 6 months prior to Day 1.
8. Diagnosis of a myeloid malignancy or known history of myelodysplasia.
9. History of second malignancy within 5 years (except excised and cured basal cell carcinoma, squamous cell carcinoma of the skin or cervical carcinoma in situ).
10. Administration of IV antibiotics or febrile (temperature elevation > 38 ° C) within 14 days of Day 1.
11. Uncontrolled hypertension. If hypertension is considered clinically stable, systolic blood pressure (BP) must be < 150 mmHg and diastolic BP must be < 100 mmHg.
12. Known history of hepatitis B surface antigen (HBsAg and HB core Ab), human immunodeficiency virus (HIV) antibody or active hepatitis C.
13. Clinically significant iron (transferrin saturation < 20%), vitamin B_{12}, or folate deficiency.
14. History of anemia as a result of inherited hemoglobinopathy such as sickle cell anemia or thalassemia.
15. History of autoimmune or hereditary hemolysis; active gastrointestinal bleeding (within the last 6 months as compared to Day 1).
16. Patient received treatment with another investigational drug or device within 1 month prior to Day 1, or if the half life of the previous product is known, within 5 times the half life prior to dosing, whichever may be longer.
17. Pregnant or lactating females.
18. History of severe allergic or anaphylactic reactions or hypersensitivity to recombinant proteins or excipients in the investigational product.
19. Major surgery within 30 days prior to Day 1 (patients must have completely recovered from any previous surgery prior to Day 1).

Investigational product, dosage and mode of administration:

ACE-011/placebo will be administered as a SC injection, approximately every 28 days, on days 1, 29, 57, and 85 of the treatment period. Each patient will be randomized to one of three treatment arms or placebo:

Dose	Number of Patients
0.1 mg/kg	30
0.3 mg/kg	30
0.5 mg/kg	30
Placebo	15
Total patients enrolled (planned): 105	

Patients with grade ≥ 3 toxicity related to ACE-011 will be discontinued from ACE-011/ placebo treatment except for the following: Patients with grade 3 hypertension that decreases to grade ≤ 1 within 7 days after treatment with anti-hypertensive therapy may continue on ACE-011/placebo treatment. Dose reduction rules will apply at the subsequent scheduled dosing day(s). Patients with a hemoglobin level above the upper limit of normal at any time during the study after Day 1 will be discontinued.

ACE-011/placebo dose modification rules are based on hemoglobin values and blood pressure measurements during each treatment cycle. ACE-011/placebo dose interruption and dose reduction rules will be implemented for those patients with hemoglobin of ≥ 11 g/dL, an increase of ≥ 2 g/dL/ 28 days, or hypertension ≥ Grade 2 within each cycle (see Figure 1).

Duration of study: Patients will be followed for a minimum of 7 months following their last dose of ACE-011/placebo. Study participation for each patient is approximately 10.5 months (14 day screening period, 3 month treatment period with a 2 month treatment follow up period, and a 5 month post treatment follow-up period). The start date of the chemotherapy regimen used in this protocol to treat metastatic breast cancer will be recorded. If a patient begins a new chemotherapy regimen during the study, the patient will discontinue further ACE-011/placebo treatment, complete the Day 281/Termination visit procedures and be followed for survival only. If a patient has a positive anti-drug antibody result at the Day 281/Termination visit, they may be asked to return to the clinical site every 3 months until a negative result is obtained. The study is complete when the last patient enrolled has completed the last protocol required follow up visit.

Criteria for evaluation

Efficacy: The primary endpoint is to establish the proportion of patients who have a hematopoietic response of an increase in hemoglobin of ≥ 1.0 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs.

The proportion of patients with an increase in hemoglobin of ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for 28 consecutive days during the treatment period up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs will be

evaluated as a secondary efficacy endpoint.

The hematopoietic endpoints will be determined by the monitoring of hematologic laboratory values, RBC transfusion and/or ESAs administration. The evaluation of response will be from the first dose of ACE-011 through 2 months after the last dose (for each patient). Additional endpoints will be evaluated with the assessment of disease progression by CT/MRI and bone scan and bone mineral density by DXA scans. Bone biomarkers will be evaluated in all patients and incidence of SREs will be recorded, as applicable. SREs are defined as pathologic fracture, radiation therapy to bone, surgery to bone, or spinal cord compression.

Safety: Throughout the study, safety will be evaluated by the sponsor, medical monitor and principal investigator. Additionally, the DMC will review safety parameters for patients enrolled in the study. All patients will be assessed for safety by monitoring adverse events, clinical laboratory tests, vital signs and physical examinations on an ongoing basis. ECGs will also be performed during screening, prior to dosing on the first cycle, during the first and third cycle, Day 113 (28 days after the last dose of ACE-011) and at Day 281/Termination visit to monitor safety. Blood pressure and lab values will be assessed at each study visit during the treatment and treatment follow up period.

Pharmacokinetics: The blood samples will be collected at selected study sites from approximately one-third of patients for the evaluation of serum concentrations of ACE-011. Pre-dose PK samples will be collected on Day 1. See Appendix 5 for PK sampling schedule.

Statistical methods

Sample size determination: With an anticipated dropout rate of 20%, a sample size of 30 patients per dose level is planned to achieve at least 80% power with 2 sided significance level of 0.05 to differentiate targeted response rate of p1=0.45 from null hypothesis p0=0.20. An additional 15 patients will be enrolled into the placebo group for safety comparison. A total of 105 patients will be randomized to three ACE-011 dose groups or placebo with a ratio of 2:2:2:1.

Efficacy analysis: The primary endpoint is the proportion of patients who have a hematopoietic response of an increase in hemoglobin of ≥ 1.0 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs. The proportion of responders will be determined together with 95% confidence interval.

For binary endpoints, the proportion of responders and 95% confidence interval will be calculated. For time to event endpoints, the Kaplan-Meier method will be used to estimate the survival curve and median time to event. Due to the nature of the study, no multiplicity adjustment will be made for efficacy analysis.

Safety analysis: Data from all patients who receive at least one dose of ACE-011/placebo will be included in the safety analyses. To assess clinical safety, adverse events, vital sign measurements and clinical laboratory information will be summarized. Adverse events and laboratory results will also be summarized by severity grade (NCI CTCAE v3.0). Changes from baseline clinical laboratory values and vital signs will be summarized over time. Descriptive statistics and shift tables will be generated as appropriate.

Schedule of Events

	Screen	Treatment Period										Post Treatment Follow up Period		Term Visit[20]
ACE-011/placebo Dose period		#1			#2		#3		#4	Follow-up				
Day	-14-0	1	8 (± 1d)	15 (± 1d)	29 (± 3d)	43 (± 2d)	57 (± 3d)	71 (± 2d)	85 (± 3d)	113 (± 7d)	141 (± 7d)	169 (± 7d)	225 (± 7d)	281 (± 7d)
Informed consent	X													
Inclusion/exclusion criteria	X	X[2]												
Medical history	X													
Physical examination	X	X[2]			X[2]		X[2]		X[2]	X	X			X
Vital signs[1]	X	X[2]	X	X	X[2]	X	X[2]	X	X[2]	X	X	X	X	X
FACT-Fatigue	X	X[2]			X[2]		X[2]		X[2]					X
12 lead ECG[3]	X	X[2]	X				X[2]			X				X
Serum iron, transferrin saturation % and TIBC	X[9]	X[2]					X[2]							X
Vitamin B12	X[9]													
Serum folate	X[9]													
Coagulation[4]	X[9]	X[2]					X[2]							X
Serum chemistry[5]	X[9]	X[2]			X[2]		X[2]		X[2]	X	X			X
Hematology[6]	X[9]	X[2]	X	X	X[2]	X	X[2]	X	X[2]	X	X	X	X	X
Serum Erythropoietin marker		X[2]			X[2]		X[2]		X[2]		X			X
LH, FSH, testosterone, progesterone, estradiol	X[9]	X[2]			X[2]		X[2]		X[2]	X	X			
Anti-drug antibody testing [7]		X[2]					X[2]			X	X			X
Bone biomarker[8]		X[2]			X[2]		X[2]		X[2]	X	X			X
Back up blood sample for future testing		X[2]	X	X	X[2]	X	X[2]	X	X[2]	X	X			X
Pregnancy Test	X[9]	X[10]			X[10]		X[10]		X[10]					X
Urinalysis[11]	X	X[2]							X[2]					X[10]
Evaluate transfusion frequency	X	X[2]	X	X	X[2]	X	X[2]	X	X[2]	X	X	X	X	X
Documentation of conmeds	X[12]	X	X	X	X	X	X	X	X	X	X	X		

Schedule of Events cont.

	Screen	Treatment Period										Post Treatment Follow up Period		Term Visit[20]
ACE-011/Placebo Dose period		#1			#2		#3		#4	Follow-up				
Day	-14-0	1	8 (± 1d)	15 (± 1d)	29 (± 3d)	43 (± 2d)	57 (± 3d)	71 (± 2d)	85 (± 3d)	113 (± 7d)	141 (± 7d)	169 (± 7d)	225 (± 7d)	281 (± 7d)
Evaluation of AEs and SAEs[13]	X-SAE only	X	X	X	X	X	X	X	X	X	X	X	X-SRE and SAE only	X
CT/MRI scan	X[14]									X[15]				
Dual-energy X-ray absorptiometry (DXA[16])	X									X				X
Bone scans[17]	X													X
ACE-011/placebo administration[18]		X			X		X		X					
Chemo regimen[19]		X	X	X	X	X	X	X	X	X	X	X	X	X

[1] Vital signs include weight, heart rate, seated systolic and diastolic blood pressure (BP), and temperature will be assessed at each visit Height will be collected at screening only For patients with previous history of CNS metastases, if BP value increases by one or more NCI CTCAE v3 0 grades, the patient must return within 7 days to have BP assessed again Patients with a recent change in anti-hypertensive medication or a change in the past month should be seen weekly for BP measurements

[2] On dosing days (1, 29, 57 and 85), study procedure to be done prior to dosing of ACE-011/placebo

[3] Use of Digital ECG machine-Screening- At two different time intervals (approximately 1 hour apart) triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min), On all other specified days, one time interval of triplicate (12 lead) ECGs is to be performed at 3 minute intervals (± 1min), prior to dosing as applicable If a local ECG machine is used, a single (12 lead) ECG is to be performed

[4] Coagulation (central lab only) prothrombin time (PT), partial thromboplastin time (PTT), fibrinogen

[5] Chemistry (central lab only): AST, ALT, lactate dehydrogenase (LDH), total bilirubin, indirect bilirubin, alkaline phosphatase, total protein, albumin, sodium, potassium, chloride, carbon dioxide, calcium, phosphorus, blood urea nitrogen (BUN), creatinine, uric acid, glucose, amylase, and lipase

[6] Hematology (local lab) hemoglobin and hematocrit On dosing days (1, 29, 57 and 85), local hemoglobin and hematocrit values are to be drawn and resulted (up to 1 day) prior to dosing to apply dose modification rules for ACE-011/placebo Hematology (central lab only) complete blood count (CBC) with differential and platelet, CBC includes red blood cells (RBC), white blood cells (WBC), hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), Peripheral blood smear will also be done on Days 1, 57 and at the termination visit **Reticulocyte should be done on days 1, 8, 15, 29, 57, 85, and at the termination visit.**

[7] If a patient has a positive anti-drug antibody result at the Termination visit, they may be asked to return to the clinical site every 3 months until a negative result is obtained

[8] Serum samples will be collected at each visit

[9] Labs may be drawn within 1 day prior to Day 1 Confirmatory labs for eligibility criteria must be drawn and resulted within 7 days of Day 1

[10] For females of child bearing potential only urine (dipstick) pregnancy test to be collected and resulted prior to dosing of ACE-011/placebo on dosing days

[11] Urinalysis to be performed at screening, prior to administration ACE-011/placebo (Days 1 and 85) and termination visit Assessments (central lab only): pH, specific gravity, protein, glucose, ketones, blood, leukocyte esterase, and nitrite Microscopy required only to follow-up abnormal findings

[12] Screening- any concomitant treatments taken 28 days prior to day 1 including up to 90 days after the last dose of ACE-011/placebo

[13] AEs will be reported from day 1 post dose including up to 90 days after the last dose of ACE-011/placebo Additional AEs noted after 90 days from the last dose of ACE-011/placebo will be reported at the discretion of the investigator SREs and SAEs will be followed through out the study If a patient is followed for survival only, SREs and SAEs will be captured up to 90 days from the last dose of ACE-011/placebo

[14] A CT/MRI scan acquired within 6 weeks of initiating the current chemotherapy regimen may be used for the screening assessment Head, neck, thoracic, abdominal, and pelvic scans are to be acquired One measurable or non measurable lesion per RECIST v1 1 is to be identified by the scan Brain (head) CT only to be done to rule out CNS metastases in patients with suspicious clinical symptoms or prior history of CNS metastases

[15] Follow-up CT/MRI scans may be acquired +/- 7 day window for Day 113 Additional CT/MRI scans acquired during the post treatment follow up period will be performed at the discretion of the investigator

[16] Hip and lumbar spine BMD assessment Refer to Imaging Acquisition Guidelines for guidelines regarding washout period for contrast (if used) for radiological images, prior to collection of DXA

[17] Bone scans will be acquired within 6 weeks of initiating the current chemotherapy regimen for those patients who have bone metastases identified Follow up bone scan, when the baseline bone scan was abnormal, may be acquired +/- 7 day window for Day 281

[18] All study visit day dates (inclusive of windows) are calculated from Day 1(day of initial ACE-011/placebo administration)

[19] Chemotherapy regimens will be administered to the patient as per standard of care Continuation of the same chemotherapy regimen may be done through the follow up period of the study at the discretion of the investigator

[20] If a patient withdraws from the study (section 8 3), the Day 281/Termination visit procedures are to be completed and the patient will be followed for survival If withdrawal reason is due to ESA administration or ACE-011 related toxicity, the patient should enter the treatment follow up period

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4. LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

The following abbreviations and specialist terms are used in this study protocol.

Table 2: Abbreviations and Specialist Terms

Term	Definition
ACTH	Adrenocorticotropic hormone
ActRIIA	Activin receptor IIA
AE	Adverse event
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
AUC	Area under the plasma concentration - time curve
$AUC_{(0\text{-last})}$	AUC from time 0 to time of last quantifiable sample
$AUC_{(0\text{-}\infty)}$	AUC extrapolated to infinity
BMD	Bone mineral density
BP	Blood pressure
BSAP	Bone specific alkaline phosphatase
BUN	Blood urea nitrogen
CBC	Complete Blood Count
CFR	Code of Federal Regulations (US)
CH	Constant domain
CI	Confidence interval
CIA	Chemotherapy induced anemia
CL	Clearance
CL/F	Total clearance
C_{max}	Maximum observed concentration
CNS	Central nervous system
CO_2	Carbon dioxide
CRF	Case report form
CRO	Clinical research organization
CT	Computed tomography
CTCAE	Common Terminology Criteria for Adverse Events
CTX	Serum C-terminal type I collagen telopeptide
DLT	Dose-limiting toxicity
DXA	Dual-energy x-ray absorptiometry
DMC	Data Monitoring Committee
DVT	Deep vein thrombosis
ECD	Extracellular domain
ECG	Electrocardiogram
ECOG	Eastern Cooperative Oncology Group
EDF	Erythroid differentiation factor
ESA	Erythropoiesis-stimulating agents
F	Bioavailability (absolute)
FACT	Functional Assessment of Chronic Illness
FDA	Food and Drug Administration
FSH	Follicle-stimulating hormone
GCP	Good Clinical Practices
GH	Growth hormone

Term	Definition
HBV	Hepatitis B virus
Hct	Hematocrit
HCV	Hepatitis C virus
Hgb	Hemoglobin
HIV	Human immunodeficiency virus
ICF	Informed consent form
ICH	International Conference on Harmonization
IEC	Independent Ethics Committee
IND	Investigational New Drug
IP	Intraperitoneal
IRB	Institutional Review Board
IV	Intravenous
IVRS	Interactive Voice Response System
Kd	Binding coefficient
λz	Elimination rate constant
LDH	Lactate dehydrogenase
LH	Luteinizing hormone
MCH	Mean corpuscular hemoglobin
MCHC	Mean corpuscular hemoglobin concentration
MCV	Mean corpuscular volume
MedDRA	Medical Dictionary for Regulatory Activities
MITT	Modified Intent to Treat (population)
MPT	Melphalan, Prednisolone,Thalidomide
MRI	Magnetic resonance imaging
NOAEL	No adverse events level
NYHA	New York Heart Association
OC	Osteocalcin
ORR	Objective response rate
OVX	Ovariectomized
PD	Progressive disease
PFS	Progression free survival
PHI	Protected health information
P1NP	Serum intact procollagen type I N terminal propeptide
PK	Pharmacokinetic
PP	Per-protocol (population)
PT	Prothrombin time
PTH	Parathyroid hormone
PTT	Partial thromboplastin time
RAP-011	Murine version of ACE-011, ActRIIA mFc
RBC	Red blood cell (count)
RECIST	Response Evaluation Criteria in Solid Tumors
SAE	Serious adverse event
SAP	Statistical analysis plan
SC	Subcutaneous
SERM	Selective estrogen receptor modulator
SHAM	Sham-operated
SPEP	Serum protein electrophoresis
SRE	Skeletal-related event
T1/2	Elimination half-life
TIBC	Total iron binding capacity

Term	Definition
Tmax	Time to Cmax
TRACP-5b	Serum tartrate-resistant acid phosphatase isoform-5b
ULN	Upper limit of normal
Vz/F	Volume of distribution
WBC	White blood cell (count)

5. INTRODUCTION

5.1. Indication and Rationale

This study is designed to evaluate the efficacy, safety, and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer.

Treatment of patients with metastatic breast cancer with myelosuppressive chemotherapy is frequently associated with anemia. Chemotherapy induced anemia (CIA) is a significant problem for patients with cancer, causing fatigue and reduced quality of life (1). Patients with CIA are currently treated with blood transfusion and/or erythropoiesis-stimulating agents (2). However, with these treatment options CIA is still an area of unmet medical need. Erythropoiesis-stimulating agents (ESAs) can successfully mitigate transfusion need in a proportion of these patients, but concerns have emerged regarding apparent negative effects on survival and/or tumor progression in certain patient populations. The murine surrogate to ACE-011, RAP-011, has been tested as a single agent in breast cancer cell lines MDA-MB-231 and MCF-7 and no effect on enhanced proliferation of these cell lines has been observed in vitro. Therefore, treatment with ACE-011 may provide a distinct benefit/risk profile to patients with chemotherapy induced anemia.

In both a Phase 1 single dose and multiple dose study of ACE-011 in postmenopausal women, increases in hemoglobin and hematocrit were observed following ACE-011 treatment and remained elevated over the course of study. The observed hemoglobin and hematocrit effects of ACE-011 were dose and time dependent. Please refer to the Investigator Brochure for further information.

Based on the effect of ACE-011 on hematopoiesis and consistent biological phenomena observed in both non clinical and clinical studies, it is hypothesized that the blockade of ActRIIA receptor signaling impedes the terminal differentiation step in erythropoiesis to allow additional rounds of cell replication before cells enter the final differentiation phase. The result is a substantial increase in mature erythrocytes released into the circulation. Since this proposed mechanism is different to that of known ESAs, ACE-011 may provide a different clinical profile in the treatment of CIA.

This study will provide information on the pharmacodynamics (PD) properties regarding the ability of ACE-011 to increase hemoglobin in patients with CIA.

5.2. Description of ACE-011

ACE-011 (ActRIIA-IgG1) is a fully human fusion protein consisting of the extracellular domain (ECD) of activin receptor IIA (ActRIIA) linked to the human IgG1 Fc domain, which includes the heavy chain hinge and constant domains, CH2 and CH3. ACE-011 avidly binds to activin A with a binding coefficient (Kd) approximately 8.9 pM and prevents its binding to endogenous receptors, thereby inhibiting biological effects of activin.

5.3. Activin Biology

Activin A was initially identified as a gonadal differentiation factor involved in modulating follicle stimulating hormone (FSH) secretion from the pituitary (3). Subsequently, the pleiotropic nature of activin A has become more apparent (4). There is a growing body of data suggesting a role for activin A in bone remodeling, specifically as a negative regulator of bone growth (5). Before the two molecules were shown to be identical (6), activin A was also described as erythroid differentiation factor (EDF), affecting red blood cells in the later stages of maturation (7). The mechanism(s) by which activin A influences erythropoiesis remain under investigation and, in fact, there are data from studies in vitro and in animals that support erythropoiesis-stimulatory (8,9) and -inhibitory effects (10).

At the cellular level, activin A binds initially to the high-affinity Type II receptor (ActRIIA). The ligand-bound ActRIIA then recruits the low-affinity Type I receptor (ActRIA or ALK-4). The receptor heterocomplex, through its cytoplasmic protein kinase activity, then activates the Smad signal cascade to eventually influence nuclear transcriptional factors (11, 12). The competitive binding of activin A in the blood by the ACE-011 soluble fusion protein can result in inhibition of ActRIIA receptor signaling pathway by impeding biological processes attributed to activin A.

Activin has also been attributed to erythroid differentiation and has been reported to have proerythrocytic effects and to induce terminal differentiation of RBCs. Inhibition of activin may lead to increases in proliferation of the erythroid lineage. Based on its pharmacologic effects, ACE-011 (ActRIIA-IgG1) is being developed for the treatment of bone loss associated with various disease states (e.g., osteoporosis and treatment of osteolytic lesions in patients with multiple myeloma), and treatment of anemia associated with a variety of disorders, such as chemotherapy induced anemia.

The extracellular domain sequence of ActRIIA is completely conserved among numerous species including mouse, rat, cynomolgus monkey and humans, thus ACE-011 is active in these animals. However, in order to reduce the potential immunogenicity of the fully human molecule, ACE-011, in mice and to maximize the opportunity to maintain exposures in chronic models, a murine surrogate molecule was constructed by exchanging the human immunoglobulin Fc sequence portion of ACE-011 with its murine IgG2a homolog. The resultant construct is referred to as RAP-011 (ActRIIA-mIgG2aFc).

5.3.1. Pharmacology Studies

RAP-011 was evaluated in a broad range of animal pharmacology studies to assess the effects of inhibition of activin A on the biological processes attributed to that regulatory protein. RAP-011 has been shown to have significant effects on the red cell compartment. RAP-011 treatment of mice at 10, 30 and 50 mg/kg by intraperitoneal injection (IP) twice per week for 3 months resulted in a 16-26% increase in red blood cell (RBC) counts compared to control animals. Rats treated with RAP-011 at 0.3, 3 and 30 mg/kg once per week for 3 months showed RBC count increases of 6-15% over control animals. Finally, in cynomolgus monkeys treated with 10, 30 or

50 mg/kg of ACE-011 twice per month for 3 months there was a 21- 24% increase in RBC counts compared to control animals. In this study, the RBC count increase was apparent as early as 2 weeks following the initial dose of ACE-011.

While the mechanism(s) underlying the stimulation effect of ACE-011 on erythropoiesis are not yet fully understood, it is hypothesized that a blockade of ActRIIA receptor signaling impedes the terminal differentiation step in erythropoiesis to allow additional rounds of erythroid cell replication before cells enter the terminal differentiation phase. The result is a substantial increase in mature erythrocytes released into circulation.

RAP-011 administered at 10 mg/kg to mice three days prior to administration of paclitaxel at 30 mg/kg was sufficient to prevent decreases in red blood cell parameters seen three days later. Mice receiving paclitaxel alone had decreased hematocrit from 43% to 38% three days later. RAP-011 administered 3 days prior to paclitaxel injection was sufficient to keep the hematocrit above 42% at three days and up to two weeks after paclitaxel administration. Therefore prophylactic treatment with RAP-011 was able to prevent paclitaxel induced anemia in mice.

RAP-011 has also been shown to significantly increase bone mineral density and strength in normal animals and in a variety of animal models of bone loss (13, 14, 15). In a murine ovariectomy (OVX)-induced model of osteopenia, RAP-011 (10 mg/kg IV, twice per week x 12 weeks) increased trabecular BMD > 25% in 6-12 weeks compared to the phosphate buffered saline (PBS)-treated controls ($p<0.002$). RAP-011 treatment was able to reverse the accelerated bone loss associated with ovariectomy. As expected sham-operated (SHAM) mice (i.e., intact ovaries) started the study with a greater trabecular BMD than OVX mice. PBS-treated SHAM mice showed a 10% decrease in trabecular BMD after 12 weeks of treatment compared to baseline, while SHAM-RAP-011 mice had a 32% increase in trabecular BMD over the same period. These data demonstrate that RAP-011 is able to increase trabecular BMD in both the normal state and in a model of established bone loss. The femurs of both the RAP-011 treated OVX and SHAM mice showed statistically significant increases in all measured parameters of bone strength (i.e., maximum load, stiffness, energy, ultimate strength, and elastic modulus). In summary, treatment with RAP-011 produced bone of superior quality consistent with an anabolic agent.

RAP-011 was evaluated in a mouse model of skeletal metastasis using luciferase-tagged, human MDA-MB-231 breast cancer cells (estrogen receptor negative). Mice were pretreated for 2 weeks with RAP-011 (10 mg/kg, biweekly, SC) prior to intracardiac injection of tumor cells. Treatment with RAP-011 was continued for an additional 5 weeks following tumor implantation. A parallel study was conducted to examine the effect of RAP-011 treatment, as described above, on survival of MDA-MB-231 bearing mice.

The data demonstrate that RAP-011 treatment acts to inhibit MDA-MB-231 metastasis to bone and may also play a role in inhibiting tumor growth in soft tissue. Furthermore, in the presence of tumor cells, RAP-011 is able to prevent lytic disease through an anabolic mechanism of bone formation. Lastly, inhibition of tumor growth and metastasis with RAP-011 leads to a greater than 25% survival benefit in this model.

The efficacy of RAP-011 was examined in an orthotopic model of breast cancer using luciferase-tagged human MCF-7 breast cancer cells (estrogen receptor positive). Mice were pretreated for 2 weeks with RAP-011 (10 mg/kg, biweekly, SC) prior to the implantation of tumor cells into the mammary fat pad of female nude mice. Treatment with RAP-011 was continued for an additional 7 weeks following tumor implantation at which time mice were sacrificed and assessed for tumor burden. RAP-011 effectively lowered the tumor burden in mice as detected by bio-luminescence. In addition, a 60% reduction in tumor weight was observed in the RAP-011 treated mice.

The ability of RAP-011 to repair osteolytic lesions caused by metastatic disease was investigated in mice. In this model athymic nude mice received an intratibial injection of MDA-MB-231-Luc cells to mimic bone metastasis. Mice were treated with a chemotherapy regimen of paclitaxel (20 mg/kg, IP, every three days) starting seven days after tumor injection and continuing for the duration of the study. On study day 42 mice with a detectable tumor burden by bioluminescent imaging were divided into two groups and began treatment with RAP-011 (10 mg/kg, SC, biweekly) or vehicle. Prior to dosing, μCT scans of the tumor bearing tibia were conducted to identify osteolytic lesions. RAP-011 treatment continued for four weeks (to study day 70) and a final μCT scan of the tibia was performed. On study day 70 osteolytic lesions remained the same or had progressed further when compared to study day 42 in the vehicle treated mice. While some mice showed progression of osteolytic lesions (most likely related to tumor burden) on study day 70, a majority of mice treated with RAP-011 demonstrated repair of the osteolytic lesions seen on study day 42. Therefore treatment with RAP-011 has the ability to repair osteolytic lesions caused by tumors and after cytoxic chemotherapy with paclitaxel.

5.3.2. Toxicology

ACE-011 has been evaluated for toxicological effects in two species, Sprague-Dawley rats and cynomolgus monkeys. The dose levels ranged from 0.3 to 30 mg/kg in rats and from 1 to 50 mg/kg in monkeys. These dose ranges were designed to support Phase 1a single-dose levels of 0.01 to 3.0 mg/kg IV and Phase 1b multiple doses of 0.1 to 2 mg/kg (monthly, SC). Weekly dosing in animals was designed to provide continuous, but fluctuating serum concentrations of ACE-011, which would be mimicked by a one-month dosing interval in humans.

Initial studies utilized IV dosing to identify target organs while longer-term studies (3 months in rats; 3 and 6 months in monkeys) utilized SC dosing to mimic the intended dosing regimen for patients. The expected pharmacologic effect, increased red blood cells (RBCs), hemoglobin (Hgb), hematocrit (HCT) and reticulocytes, was observed in all studies, presumably based on the ability of ACE-011 to inhibit activin. A second expected pharmacologic effect of ACE-011, based on inhibition of activin, was the reversible reduction in sperm production and testicular tubular damage in rats. Since male cynomolgus monkeys were sexually immature in these studies, it was not possible to monitor this effect in this species.

Effects on the adrenal gland were only seen in rats and were more pronounced in female rats. This toxicological observation may not be relevant for predicting effects in humans since adrenal toxicity was not seen in cynomolgus monkeys. Dose-limiting toxicity and no observable adverse effect levels (NOAELs) were primarily based on renal toxicity in both rats and monkeys. There

was some indication that kidney toxicity was an indirect effect, based on the formation of antibody/antigen complexes, but since these studies were not designed to investigate toxicological mechanisms, a definitive cause of renal impairment and renal damage was not determined. Development of antibodies to ACE-011 was noted in all of these studies, which is an expected immune reaction in rats and monkeys dosed with a human protein.

The no observable adverse effect level (NOAELs) from the 3 month SC studies was 3 and 30 mg/kg in rats and monkeys, respectively. Because the kidney findings were observed at all dose levels, the NOAEL from the 6 month monkey study is < 10 mg/kg.

5.4. Summary of Clinical Experience

5.4.1. A011-01: A Phase 1a Study in Healthy Postmenopausal Women

ACE-011 was first studied in a randomized, Phase 1a, single dose, dose escalation study in healthy, post-menopausal females (16). ACE-011 was diluted in normal saline administered as an IV infusion (over approximately 1 hour) or as a SC injection on Day 1. Dose levels were 0.01, 0.03, 0.1, 0.3, 1.0, and 3.0 mg/kg IV and 0.03 and 0.1 mg/kg SC. A total of 48 subjects were enrolled; 5 active and 1 placebo at each of the IV and SC dose levels. All subjects were followed for 4 months following a single dose administration.

The pharmacokinetics of ACE-011 was linear for all IV and SC doses. Across IV doses ranging from 0.1 through 3.0 mg/kg, the mean clearance (CL) ranged from 0.092 to 0.128 mL/h/kg, the volume of distribution ranged from 73.7 to 110 mL/kg, and the mean T1/2 ranged from 23.7 to 31.8 days, with no apparent dependence on dose. After SC administration of 0.03 and 0.1 mg/kg, ACE-011 was completely absorbed, and the mean T1/2 was approximately 30 days, with no apparent dependence on dose.

The most commonly occurring treatment-emergent adverse events (AEs; i.e., those occurring in more than 1 subject in any treatment group) were headache, infusion site reaction, injection site hemorrhage, and toothache. The majority of the injection site reactions and injection site hemorrhages were in the first IV cohort and were related to infiltration of the IV site. The majority of treatment-emergent AEs were mild in severity and were judged to be unrelated to ACE-011. No deaths, serious AEs (SAEs), or AEs leading to discontinuation were reported. Changes in red blood cells (RBCs), hemoglobin, reticulocytes, liver function tests, glucose, uric acid, amylase and lipase occurred in some subjects; however, none of these laboratory results were reported as adverse events. Mild, transient elevations in pancreas enzymes, liver enzymes, or glucose were reported as AEs in five patients. There were no clinically significant changes from baseline in vital signs, physical examination, endocrine function, or electrocardiogram (ECG) data. Preservation of adrenal cortical function was monitored through evaluation of serum electrolytes (sodium and potassium levels) and through cortisol response to adrenocorticotropic hormone (ACTH) stimulation. No clinically significant findings were observed at doses up to 3.0 mg/kg IV, and ACE-011 was well tolerated in healthy, post-menopausal female volunteers in single dose levels up to 0.1 mg/kg SC and 3.0 mg/kg IV, the highest dose levels tested in this study.

5.4.2. A011-02: A Phase 1b Study in Healthy Postmenopausal Women

ACE-011 was studied in a Phase 1b, single-center, randomized, double-blind, placebo-controlled, multi-dose, dose-escalating study to evaluate the safety, tolerability and pharmacodynamics of ACE-011 in healthy postmenopausal women. Four cohorts of 10 subjects each were planned at the following dose levels: 0.1, 0.3, 1.0, and 2.0 mg/kg administered subcutaneously. Within each cohort, subjects were to be randomized to either active or placebo treatment in an 8:2 ratio. Subjects were to receive one SC injection of ACE-011 or placebo every 28 days for a total of 4 doses. All subjects were to be followed for 12 weeks after the last dose.

The treatment phase of the study was terminated early after a dose-limiting pharmacodynamic effect was observed. While on study, one subject in the 1.0 mg/kg cohort experienced a Serious Adverse Event (SAE) of progressive and persistent hypertension that was attributed to a rapid and significant rise in hemoglobin and hematocrit levels. Due to symptoms of headaches, nausea, eye pain, dizziness and vomiting approximately 1 week following the second dose, the subject was hospitalized for monitoring and evaluation of her hypertension. The head MRI, fundoscopy, and ECG performed were reported as normal and headache symptoms resolved following therapeutic phlebotomy. The SAE resolved and the subject was discharged from the hospital the following day. The hypertension was monitored closely and managed initially with antihypertensive medication. The hypertension resolved by the end of the study without need of any antihypertensive medication. The subject received aspirin prophylactically until the end of the study and continued taking ibuprofen as needed for headaches. Further details can be found in the Investigator Brochure.

Based on the magnitude of the hematopoietic response, the Sponsor suspended dose escalation to the 2.0 mg/kg dose level and further dosing in all cohorts as a result of this dose-limiting pharmacodynamic effect at the 1.0 mg/kg dose level. In total, 31 subjects were enrolled and treated. Dose levels of ACE-011 administered included 0.1, 0.3, and 1.0 mg/kg (Cohorts 1, 2, and 3 respectively). All subjects randomized to active treatment in Cohort 1 received all 4 planned doses of ACE-011. Due to early discontinuation of study drug, subjects randomized to active treatment in Cohort 2 received 3 doses of ACE-011, and subjects randomized to active treatment in Cohort 3 received 2 doses of ACE-011. Subjects randomized to placebo treatment received between 1 and 4 doses of study treatment.

In the analysis of the unblinded data, after the administration of the first dose, a dose and time dependent increase in hemoglobin values was observed (see Table 3 below):

Table 3: A011-02: A Phase 1b Study in Healthy Postmenopausal Women: Hemoglobin Evaluation

Evaluation Time Point	Mean Change from Baseline (g/dL)			
	Placebo N=7*	ACE-011 0.1 mg/kg N=8	ACE-011 0.3 mg/kg N=8	ACE-011 1.0 mg/kg N=8
Baseline Mean, pre-Dose 1	13.20	13.11	13.30	12.71
Day 8	0.17	0.68	0.85	1.21
Day 15	-0.27	0.43	0.44	1.75
Day 29	0.27	0.61	1.21	2.68**
Day 36	0.56	0.64	1.89	2.96
Day 43	-0.02	0.89	1.21	2.85
Day 57	0.27	1.28	1.64**	2.09
Day 64	0.53	1.11	2.49	2.21
Day 71	-0.10	1.34	2.09	1.66
Day 85	0.38	1.18**	2.55	1.86
Day 92	0.00***	1.04	1.60***	3.80***
Day 99	-0.20	1.21	3.20***	1.28***
Day 113	-0.02	1.30	1.29	1.04
Day 141	0.30	0.95	0.34	2.30
Day 169	0.20***	0.06		2.00***

*The number of placebo subjects with data decreases over time as a result of the early discontinuation of the study (i.e., there were placebo subjects in each dosing cohort). There were seven placebo subjects with data at baseline, Days 8, 15, and 36; six subjects with data at Days 29, 43, 57, and 85; five subjects with data on Days 71 and 113; four subjects with data on Day 64; three subjects with data on Day 141; two subjects with data on Day 99; and one subject with data on Days 92 and 169.

** Number of doses administered per treatment group: 0.1 mg/kg 4 doses; 0.3 mg/kg 3 doses; 1.0 mg/kg 2 doses. Data beyond this study day are considered follow-up results.

***n=1

No severe or life-threatening events were reported. The incidence of the most common treatment-emergent AEs (i.e., occurring in more than one subject in any treatment group) is presented below (see Table 4). The most notable AEs were those related to increases in hematologic laboratory measures in the 1.0 mg/kg dose group: hematocrit, hemoglobin, and RBC count. AEs of increased hemoglobin and/or hematocrit were reported for seven of the eight subjects in this dose group. These events were reported as mild or moderate elevations; all were considered probably related to study drug treatment. Three of the subjects in the 1.0 mg/kg group with elevated hemoglobin levels underwent phlebotomies and all hemoglobin elevations were resolved by the end of the follow-up period. No erythroid lineage AEs were reported in the 0.1 or 0.3 mg/kg treatment groups.

Table 4: A011-02: Incidence of Most Common Adverse Events

System Organ Class and Preferred Term	Dose Group			
	Placebo N=7 n (%)	0.1 mg/kg N=8 n (%)	0.3 mg/kg N=8 n (%)	1.0 mg/kg N=8 n (%)
GASTROINTESTINAL DISORDERS				
Diarrhea	0 (0.0%)	2 (25.0%)	0 (0.0%)	0 (0.0%)
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS				
Asthenia	0 (0.0%)	1 (12.5%)	2 (25.0%)	0 (0.0%)
Fatigue	2 (28.6%)	0 (0.0%)	1 (12.5%)	2 (25.0%)
INFECTIONS AND INFESTATIONS				
Viral upper respiratory tract infection	0 (0.0%)	4 (50.0%)	0 (0.0%)	0 (0.0%)
INJURY, POISONING AND PROCEDURAL COMPLICATIONS				
Limb injury	0 (0.0%)	1 (12.5%)	0 (0.0%)	2 (25.0%)
INVESTIGATIONS				
Hematocrit increased	0 (0.0%)	0 (0.0%)	0 (0.0%)	6 (75.0%)
Hemoglobin increased	0 (0.0%)	0 (0.0%)	0 (0.0%)	7 (87.5%)
Red blood cell count increased	0 (0.0%)	0 (0.0%)	0 (0.0%)	3 (37.5%)
METABOLISM AND NUTRITION DISORDERS				
Decreased Appetite	1 (14.3%)	0 (0.0%)	2 (25.0%)	0 (0.0%)
MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS				
Arthralgia	2 (28.6%)	0 (0.0%)	2 (25.0%)	0 (0.0%)
Muscle spasms	0 (0.0%)	1 (12.5%)	0 (0.0%)	2 (25.0%)
NERVOUS SYSTEM DISORDERS				
Dizziness	0 (0.0%)	0 (0.0%)	2 (25.0%)	2 (25.0%)
Headache	2 (28.6%)	4 (50.0%)	3 (37.5%)	2 (25.0%)
Paresthesia	1 (14.3%)	0 (0.0%)	0 (0.0%)	3 (37.5%)
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS				
Oropharyngeal pain	2 (28.6%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
VASCULAR DISORDERS				
Hot flush	0 (0.0%)	2 (25.0%)	0 (0.0%)	1 (12.5%)

Note: Table presents AEs reported in more than one subject in any treatment group.

Headache was reported frequently in all treatment groups with no apparent dose response trend, though the reports in the higher dose groups were more often considered possibly related to treatment. Most headaches were reported as mild (one headache was reported as moderate).

Paresthesia and dizziness were reported more frequently in the ACE-011 groups, though the events were generally not considered drug related. Other frequently reported events (e.g. fatigue, upper respiratory infection) did not appear to increase with dose and were generally mild.

Hemoglobin levels for all subjects with elevations had returned to within normal limits by the end of the study.

Adrenal cortical function was monitored through evaluation of serum electrolytes (sodium and potassium levels) and through cortisol response to adrenocorticotropic hormone (ACTH) stimulation. All ACTH simulation test results were normal.

PK results confirmed that the PK of ACE-011 is linear following the first SC doses of all three dose levels tested, 0.1, 0.3, and 1.0 mg/kg. The terminal half life ($T_{1/2}$) of ACE-011 following the last dose in all three dose groups was identical, with mean $T_{1/2}$ being approximately 23 days. The mean CL/F ranged from 3.05 to 3.90 mL/d/kg, the mean Vz/F ranged from 97.47 to 103.03 mL/kg, and the mean $T_{1/2}$ ranged from 20.92 to 23.34 days in all 3 dose levels, with no apparent dependence on dose.

Bone mineral density was assessed by dual-energy X-ray absorptiometry (DXA). Mean change and mean percent change in BMD results from baseline to study end are summarized in the table below. A dose-dependent increase in the BMD of the total hip from baseline to end of study was observed, with a significant and rapid increase of 2.4% in the 1.0 mg/kg dose group, compared to a 0.7% decrease in the placebo group. BMD results for lumbar spine showed slight increases of 0.4% to 1.0% from baseline to study end in all active treatment groups, compared with a 0.5% decrease in the placebo group (see Table 5 below):

Table 5: A011-02: A Phase 1b Study in Healthy Postmenopausal Women: BMD

	Treatment Group							
	Placebo N=7		**ACE-011 0.1 mg/kg N=8**		**ACE-011 0.3 mg/kg N=8**		**ACE-011 1.0 mg/kg N=8**	
	Mean Change	Mean Percent Change	Mean Change	Mean Percent Change	Mean Change	Mean Percent Change	Mean Change	Mean Percent Change
Lumbar Spine (g/cm^2)	-0.0020	-0.5%	0.0099	0.7%	0.0082	1.0%	0.0051	0.4%
Total Hip (g/cm^2)	-0.0062	-0.7%	0.0050	0.6%	0.0075	0.9%	0.0220	2.4%

Number of doses administered and day of last dose per treatment group: 0.1 mg/kg 4 doses (Day 85); 0.3 mg/kg 3 doses (Day 57); 1.0 mg/kg 2 doses (Day 29). Data beyond this study day are considered follow-up results.

5.4.3. A011-04: A Phase 2a Study in Patients with Osteolytic Lesions of Multiple Myeloma

Study A011-04 is an ongoing, non-IND, Phase 2a, multi-center, randomized, multiple-dose study to evaluate the safety, tolerability and efficacy of ACE-011 in patients with osteolytic lesions of multiple myeloma. Safety evaluations include adverse events (AEs), clinical laboratory tests, standard 12-lead electrocardiogram (ECG), vital signs, Eastern Cooperative Oncology Group

(ECOG) performance status and physical examinations. Additionally, the study includes the assessment of biochemical markers of bone formation and resorption, skeletal related events (SREs), bone mineral density (BMD) by duel-energy x-ray absorptiometry (DXA) and bone pain by visual analog scale (VAS).

In this study, patients are randomized in a 4:1 ratio to one of three dose levels of ACE-011 (0.1, 0.3 and 0.5 mg/kg) or placebo, to be administered to patients every 28 days by subcutaneous injection, for up to four doses over a 3-month period. The test article is being evaluated in combination with the anti-myeloma therapy of melphalan (4 mg/m^2 on days 1-7), prednisolone (40 mg/m^2 on days 1-7) and thalidomide (100 mg per day) (MPT). The sites, Sponsor and Sponsor representatives are blinded to treatment assignment.

As of 10 March 2009, 30 patients have been randomized, enrollment is complete and treatment is ongoing. Fifteen patients have received multiple doses of their assigned treatment; 7 patients have received 4 doses, 3 patients have received 3 doses and 5 patients have received 2 doses. Of the remaining 15 patients, 12 patients have received their first dose of ACE-011 or placebo and 3 patients have not yet received their first dose. Per the monitored preliminary data on 18 patients presented to the Independent Data Monitoring Committee (DMC) on 9 February 2009, there were 8 Grade 3/severe AEs reported in 4 patients, including neutropenia, platelet count decreased, pain in extremity, back pain and anemia. All Grade 3 AEs were determined to be not related to ACE-011 and the Grade 3 hematologic adverse events were considered either probably or definitely related to the MPT treatment.

Following preliminary analysis of the blinded central laboratory data, increases in hemoglobin values were observed within 28 days after administration of the first dose of ACE-011/placebo. Per data available, 11 out of 30 patients had $\geq$ 1 g/dL increase in hemoglobin within the first 28 days on the study, 4 patients achieved a $\geq$ 2 g/dL increase within their first 28 days on study, while 8 patients achieved a $\geq$ 2 g/dL increase in hemoglobin through Day 85.

Including all visits as of 10 March 2009 through Day 85, 9 out of 30 patients had a dose interruption due to a hemoglobin value of > 13 g/dL according to the dose modification rules defined in the protocol and 1 of the 9 patients, who had a history of hypertension, also had a dose interruption due to Grade 2 hypertension. Three of the 9 patients that required a dose interruption due to a hemoglobin value of > 13 g/dL, as per protocol, had hemoglobin levels above 13 g/dL prior to their first dose. Taken together, these data, suggest a beneficial pharmacodynamic effect of ACE-011 on erythropoiesis in a patient population with cancer chemotherapy induced anemia. No patients have had a dose reduction in ACE-011/placebo treatment or discontinued treatment.

One (1) subject died on PPD due to sudden death (including progression of disease). PPD The subject had received the first dose of ACE-011/placebo and started cycle 1 MPT on PPD. The last study dose was administered on PPD On PPD the patient developed grade 2 blood pressure increase, which resolved with the patient's blood pressure medication. The following day (PPD the patient's blood pressure in the morning was back to baseline. The patient then died on PPD, 18 days after last

study dose, due to the described sudden death. No autopsy was performed. The Investigator assessed event causality as possibly related to ACE-011/placebo and probably related to MPT.

One SAE of atrial fibrillation with syncope occurred resulting in study discontinuation. This was in a patient with prior history of Chronic Atrial Fibrillation. On further review this patient's inclusion into the study was a protocol violation. The Investigator assessed event causality as unrelated to ACE-011/placebo and possibly related to MPT.

One SAE of prolonged hospitalization due to pneumonia, that was considered not related to ACE-011, was reported in a PPD enrolled in the study. On PPD four days after the first dose of ACE-011/placebo and the initial doses of melphalan, prednisolone and thalidomide (MPT), the patient presented with elevated temperatures up to 39° C, cough and fatigue. Chest x-ray performed on PPD revealed a pneumonic infiltration in the lower lobe of the left lung. On PPD hospitalization of the patient was prolonged due to the event. The patient was treated with moxifloxacin, doripenem, meropenem and fluconazole.

On PPD, the pneumonic infiltration resolved, as confirmed by chest x-ray. This SAE resolved upon discharge of the patient from the hospital on PPD. Pneumonia is considered a labeled event per the current package insert for thalidomide. The Investigator considered the event of pneumonia unrelated to ACE-011 and possibly related to MPT.

An SAE of pain was reported in a PPD and was found to have a pathological fracture of the right upper femoral bone approximately 2.5 months after last dose of ACE-011/placebo, approximately 5 weeks after last dose of Melphalan-Prednisolone and one day after the last dose of Thalidomide. Radiologic evaluations demonstrated progressive bone tissue destruction at presentation. The subject was casted, and then underwent coxal bandaging. The event resolved with sequelae. The investigator considered the events of pain in the right leg and pathological fracture of the upper third of the right femur unrelated to ACE-011/placebo and unrelated to MPT.

Please refer to the Investigator Brochure for further information.

5.5. Potential Risks for Human Use

The 1-month safety study in rats suggested that ACE-011 may affect sperm count and motility in a dose-dependent manner, although these findings appeared to be reversible upon drug discontinuation. These reproductive effects are likely a result of the expected biological activity of activin inhibition.

The 1-month safety study in rats also suggested that ACE-011 induces adrenocortical necrosis in rats. At present, the mechanism by which ACE-011 has induced this lesion in rats and the basis for its apparent species specificity are unknown; the finding has not been observed in mice or cynomolgus monkeys at similar dose levels. Adrenal cortical function was monitored in human subjects receiving ACE-011 by the evaluation of serum electrolytes including sodium and potassium levels and by the evaluation of cortisol response to ACTH stimulation in both the

Phase 1a single dose and Phase 1b multiple dose healthy volunteer clinical studies. These parameters were evaluated up to doses of 3.0 mg/kg IV and 1.0 mg/kg SC, and no clinically significant perturbations in adrenal function were observed.

Laboratory findings from the Phase 1a single dose study showed elevations in hematology results, pancreas and liver enzymes, and uric acid. The most commonly seen treatment-emergent adverse events in this study were headache, infusion site reaction, injection site hemorrhage, and toothache.

Based on data from the Phase 1b study, the most notable AEs were increases in hematologic laboratory measures in the 1.0 mg/kg dose group: hematocrit, hemoglobin, and RBC count. AEs of increased hemoglobin and/or hematocrit were reported for seven of the eight subjects in this dose group. These events were reported as mild or moderate elevations; all were considered probably related to study drug treatment. No erythroid lineage AEs were reported in the 0.1 or 0.3 mg/kg treatment groups. Hematologic parameters will be monitored carefully in this study.

Headache was reported frequently in all treatment groups with no apparent dose response trend, though the reports in the higher dose groups were more often considered possibly related to treatment. Most headaches were reported as mild (one headache was reported as moderate). Paresthesia and dizziness were reported more frequently in the ACE-011 groups, though the events were generally not considered drug related. Other frequently reported events (e.g. fatigue, upper respiratory infection) did not appear to increase with dose and were generally mild.

In the Phase 1b multiple-dose healthy volunteer study, some subjects treated with ACE-011 showed elevations in blood pressure. One subject who had received two doses of ACE-011 at the 1.0 mg/kg dose level had a serious adverse event. The patient was hospitalized for further evaluation of increases in blood pressure and symptoms that may have been caused by the increase in blood pressure. The symptoms that the subject experienced included headache, dizziness, nausea, vomiting, and elevated hematology values (increases in the number of red blood cells). This serious adverse event was judged to be probably related to the study drug and resolved following discharge from hospital the following day. Throughout the course of the follow up period hematologic parameters were monitored frequently until the hemoglobin and hematocrit levels returned to within normal limits. Further details are outlined in the Investigator Brochure.

As with all biologics there is the potential for anti-drug antibodies that can be associated with increased drug clearance and hypersensitivity reactions. An immune response to ACE-011 has been seen in monkeys that received the drug for 1 month and longer. Some of those animals developed kidney inflammation possibly related to that immune response. Anti-drug antibody formation will be assessed in this study to examine the immunogenicity of ACE-011, and to monitor reversibility of any AEs over time.

Please refer to the Investigator Brochure for more information.

6. TRIAL OBJECTIVES AND PURPOSE

6.1. Primary objective

1. To evaluate the proportion of patients in each ACE-011 dose regimen who achieve a hematopoietic response during the treatment period including up to 2 months after the last dose of ACE-011 treatment of chemotherapy induced anemia in patients with metastatic breast cancer. Hematopoietic response is defined as an increase in hemoglobin of ≥ 1.0 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs.

6.2. Secondary objectives

1. To evaluate the safety and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer.
2. To evaluate the percentage of patients who achieve an increase in hemoglobin of ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for a consecutive 28 day period during the treatment period including up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs.
3. To estimate the duration of the hematopoietic response associated with each ACE-011 dose regimen.
4. To estimate the time to the hematopoietic response associated with each ACE-011 dose regimen.
5. To estimate the proportion of patients receiving red blood cell (RBC) transfusions and/or ESAs within each cycle of ACE-011 with each dose regimen.
6. To estimate the effect of each ACE-011 dose regimen on progression free survival (PFS) and objective response rate (ORR).

6.3. Exploratory objectives

1. To estimate the effect of each ACE-011 dose regimen on serum bone biomarkers (OC, BSAP, P1NP, CTX, and TRACP-5b), bone mineral density (DXA scans of lumbar spine and hip) and incidence of skeletal related events (SREs).
2. To evaluate the quality of life in relation to the treatment of chemotherapy induced anemia in patients with metastatic breast cancer during the study.

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7. INVESTIGATIONAL PLAN

7.1. Overall Study Design and Plan

This is a Phase 2, double-blind, randomized, placebo-controlled study to evaluate the efficacy, safety, and tolerability of ACE-011 for the treatment of chemotherapy induced anemia in patients with metastatic breast cancer. A total of approximately 105 patients will be enrolled.

The Data Monitoring Committee (DMC) is responsible for reviewing safety. The DMC will be comprised of a minimum of 3 members. The DMC will meet regularly during the study to review serious adverse events (SAEs), adverse events (AEs), laboratory results, and vital signs. Safety data will be reviewed throughout the study by the DMC, Medical Monitor and the Investigator. DMC responsibilities, membership, meeting frequencies and procedures will be outlined in the DMC charter.

Patients will be evaluated for study inclusion/exclusion criteria by the Investigator. Patients who meet the study entry criteria will be enrolled within 14 days of the screening visit. Local lab values for hemoglobin and central lab values will be utilized for the evaluation of patient eligibility. Central labs will also be used in the evaluation of patient safety throughout the study. Local lab values for hemoglobin and hematocrit will be collected and reported from screening (for eligibility) through the Day 281/Termination visit. Three cohorts of 30 patients each are planned at the following doses of ACE-011: 0.1 mg/kg, 0.3 mg/kg and 0.5 mg/kg. Fifteen patients will be enrolled into the placebo cohort (as shown in Table 6).

Table 6: Study Design

Dose	Number of Patients
0.1 mg/kg	30
0.3 mg/kg	30
0.5 mg/kg	30
Placebo	15
Total patients enrolled (planned): 105	

Each eligible patient will be randomized to one of the four cohorts to receive a dose administered as a subcutaneous injection every 28 days for up to 4 doses. Dosing will be administered on days 1, 29, 57, and 85. Concurrent treatment with a myelosuppressive chemotherapy regimen for metastatic breast cancer will also be administered per standard of care at the site. Patients will be stratified according to administration of either single agent weekly chemotherapy or less frequent chemotherapy regimens.

Blood samples will be collected from approximately one-third of patients participating in the study at selected study sites for the evaluation of serum concentrations of ACE-011. Pre-dose PK samples will be collected as specified in Appendix 5 (PK sampling schedule.)

Patients will be assessed for safety by monitoring adverse events, clinical laboratory tests, vital signs and physical examinations on an ongoing basis. ECGs will be performed during screening, prior to dosing on the first cycle, during the first and third cycle, Day 113 (28 days after the last dose of ACE-011) and at Day 281/ Termination visit to monitor safety. Blood pressure and lab values will be assessed at each study visit during the treatment and treatment follow up period.

Local laboratory hemoglobin values and blood pressure values will be evaluated prior to dosing per the dose modification criteria, as applicable.

Patients will be followed for a minimum of 7 months following their last dose of ACE-011/placebo. Study participation for each patient is approximately 10.5 months (14 day screening period, 3 month treatment period with a 2 month treatment follow up period, and a 5 month post treatment follow-up period). If a patient discontinues the study due to progression of disease (metastatic breast cancer) and/or begins another chemotherapy regimen, the patient will discontinue ACE-011/placebo treatment, complete the Day 281/Termination visit procedures, and be followed for survival only (a minimum 7 months from the last dose of ACE-011/placebo).

Anti-drug antibody testing will be performed at Day 281/Termination visit and if positive, the patient may be asked to return every 3 months until a negative result is obtained

Back up (serum) blood samples will be collected for future testing for the evaluation of novel tumor markers, such as endoglin and ALK-1 (activin receptor-like kinase-1). The samples will the stored at the central lab until this evaluation is performed.

Table 7: Schedule of Events

Schedule of Events														
	Screen	Treatment Period										Post Treatment Follow up Period		Term Visit[20]
ACE-011/placebo Dose period		#1			#2		#3		#4	Follow-up				
Day	-14-0	1	8 (± 1d)	15 (± 1d)	29 (± 3d)	43 (± 2d)	57 (± 3d)	71 (± 2d)	85 (± 3d)	113 (± 7d)	141 (± 7d)	169 (± 7d)	225 (± 7d)	281 (± 7d)
Informed consent	X													
Inclusion/exclusion criteria	X	X[2]												
Medical history	X													
Physical examination	X	X[2]			X[2]		X[2]		X[2]	X	X			X
Vital signs[1]	X	X[2]	X	X	X[2]	X	X[2]	X	X[2]	X	X	X	X	X
FACT-Fatigue	X	X[2]			X[2]		X[2]		X[2]					X
12 lead ECG[3]	X	X[2]	X				X[2]			X				X
Serum iron, transferrin saturation % and TIBC	X[9]	X[2]					X[2]							X
Vitamin B12	X[9]													
Serum folate	X[9]													
Coagulation[4]	X[9]	X[2]					X[2]							X
Serum chemistry[5]	X[9]	X[2]			X[2]		X[2]		X[2]	X	X			X
Hematology[6]	X[9]	X[2]	X	X	X[2]	X	X[2]	X	X[2]	X	X	X	X	X
Serum Erythropoietin marker		X[2]			X[2]		X[2]		X[2]		X			X
LH, FSH, testosterone, progesterone, estradiol	X[9]	X[2]			X[2]		X[2]		X[2]	X	X			
Anti-drug antibody testing [7]		X[2]					X[2]			X	X			X
Bone biomarker[8]		X[2]			X[2]		X[2]		X[2]	X	X			X
Back up blood sample for future testing		X[2]	X	X	X[2]	X	X[2]	X	X[2]	X	X			X
Pregnancy Test	X[9]	X[10]			X[10]		X[10]		X[10]					X
Urinalysis[11]	X	X[2]							X[2]					X[10]
Evaluate transfusion frequency	X	X[2]	X	X	X[2]	X	X[2]	X	X[2]	X	X	X	X	X
Documentation of conmeds	X[12]	X	X	X	X	X	X	X	X	X	X	X		

Schedule of Events cont.

	Screen	Treatment Period										Post Treatment Follow up Period		Term Visit[20]
ACE-011/Placebo Dose period		#1			#2		#3		#4	Follow-up				
Day	-14-0	1	8 (± 1d)	15 (± 1d)	29 (± 3d)	43 (± 2d)	57 (± 3d)	71 (± 2d)	85 (± 3d)	113 (± 7d)	141 (± 7d)	169 (± 7d)	225 (± 7d)	281 (± 7d)
Evaluation of AEs and SAEs[13]	X-SAE only	X	X	X	X	X	X	X	X	X	X	X	X-SRE and SAE only	X
CT/MRI scan	X[14]									X[15]				
Dual-energy X-ray absorptiometry (DXA[16])	X									X				X
Bone scans[17]	X													X
ACE-011/placebo administration[18]		X			X		X		X					
Chemo regimen[19]		X	X	X	X	X	X	X	X	X	X	X	X	X

[1] Vital signs include weight, heart rate, seated systolic and diastolic blood pressure (BP), and temperature will be assessed at each visit Height will be collected at screening only For patients with previous history of CNS metastases, if BP value increases by one or more NCI CTCAE v3 0 grades, the patient must return within 7 days to have BP assessed again Patients with a recent change in anti-hypertensive medication or a change in the past month should be seen weekly for BP measurements

[2] On dosing days (1, 29, 57 and 85), study procedure to be done prior to dosing of ACE-011/placebo

[3] Use of Digital ECG machine-Screening- At two different time intervals (approximately 1 hour apart) triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min), On all other specified days, one time interval of triplicate (12 lead) ECGs is to be performed at 3 minute intervals (± 1min), prior to dosing as applicable If a local ECG machine is used, a single (12 lead) ECG is to be performed

[4] Coagulation (central lab only) prothrombin time (PT), partial thromboplastin time (PTT), fibrinogen

[5] Chemistry (central lab only): AST, ALT, lactate dehydrogenase (LDH), total bilirubin, indirect bilirubin, alkaline phosphatase, total protein, albumin, sodium, potassium, chloride, carbon dioxide, calcium, phosphorus, blood urea nitrogen (BUN), creatinine, uric acid, glucose, amylase, and lipase

[6] Hematology (local lab) hemoglobin and hematocrit On dosing days (1, 29, 57 and 85), local hemoglobin and hematocrit values are to be drawn and resulted (up to 1 day) prior to dosing to apply dose modification rules for ACE-011/placebo Hematology (central lab only) complete blood count (CBC) with differential and platelet, CBC includes red blood cells (RBC), white blood cells (WBC), hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), Peripheral blood smear will also be done on Days 1, 57 and at the termination visit **Reticulocyte should be done on days 1, 8, 15, 29, 57, 85, and at the termination visit.**

[7] If a patient has a positive anti-drug antibody result at the Termination visit, they may be asked to return to the clinical site every 3 months until a negative result is obtained

[8] Serum samples will be collected at each visit

[9] Labs may be drawn within 1 day prior to Day 1 Confirmatory labs for eligibility criteria must be drawn and resulted within 7 days of Day 1

[10] For females of child bearing potential only urine (dipstick) pregnancy test to be collected and resulted prior to dosing of ACE-011/placebo on dosing days

[11] Urinalysis to be performed at screening, prior to administration ACE-011/placebo (Days 1 and 85) and termination visit Assessments (central lab only): pH, specific gravity, protein, glucose, ketones, blood, leukocyte esterase, and nitrite Microscopy required only to follow-up abnormal findings

[12] Screening- any concomitant treatments taken 28 days prior to day 1 including up to 90 days after the last dose of ACE-011/placebo

[13] AEs will be reported from day 1 post dose including up to 90 days after the last dose of ACE-011/placebo Additional AEs noted after 90 days from the last dose of ACE-011/placebo will be reported at the discretion of the investigator SREs and SAEs will be followed through out the study If a patient is followed for survival only, SREs and SAEs will be captured up to 90 days from the last dose of ACE-011/placebo

[14] A CT/MRI scan acquired within 6 weeks of initiating the current chemotherapy regimen may be used for the screening assessment Head, neck, thoracic, abdominal, and pelvic scans are to be acquired One measurable or non measurable lesion per RECIST v1 1 is to be identified by the scan Brain (head) CT only to be done to rule out CNS metastases in patients with suspicious clinical symptoms or prior history of CNS metastases

[15] Follow-up CT/MRI scans may be acquired +/- 7 day window for Day 113 Additional CT/MRI scans acquired during the post treatment follow up period will be performed at the discretion of the investigator

[16] Hip and lumbar spine BMD assessment Refer to Imaging Acquisition Guidelines for guidelines regarding washout period for contrast (if used) for radiological images, prior to collection of DXA

[17] Bone scans will be acquired within 6 weeks of initiating the current chemotherapy regimen for those patients who have bone metastases identified Follow up bone scan, when the baseline bone scan was abnormal, may be acquired +/- 7 day window for Day 281

[18] All study visit day dates (inclusive of windows) are calculated from Day 1(day of initial ACE-011/placebo administration)

[19] Chemotherapy regimens will be administered to the patient as per standard of care Continuation of the same chemotherapy regimen may be done through the follow up period of the study at the discretion of the investigator

[20] If a patient withdraws from the study (section 8 3), the Day 281/Termination visit procedures are to be completed and the patient will be followed for survival If withdrawal reason is due to ESA administration or ACE-011 related toxicity, the patient should enter the treatment follow up period

8. SELECTION AND WITHDRAWL OF PATIENTS

8.1. Number of Patients

The study will include three dose levels of ACE-011 at 0.1 mg/kg, 0.3 mg/kg and 0.5 mg/kg and a placebo group. A total of approximately 105 patients will be treated with ACE-011 or placebo.

8.2. Entry Criteria

8.2.1. Patient Inclusion Criteria

1. Women ≥ 18 years of age.
2. Histologically or cytologically confirmed diagnosis of breast cancer.
3. Evidence of metastatic breast cancer with a minimum of one measurable lesion or non measurable lesion per RECIST v1.1 criteria (Appendix 1).
4. Receiving a multicycle myelosuppressive chemotherapy regimen including one of the following: anthracycline, taxane, gemcitabine, vinorelbine or capecitabine.
5. Anticipated treatment with the same myelosuppressive chemotherapy regimen for a minimum of 9 weeks after Day 1 of ACE-011/placebo administration.
6. Hemoglobin value between ≥ 6.5 to < 11.0 g/dL (≥ 65 to < 110 g/L).
7. ≥ 30 days must have elapsed (from Day 1) since previous treatment with erythropoiesis-stimulating agent (including concurrent treatment with IV iron) for chemotherapy induced anemia.
8. ≥ 7 days have elapsed (from Day 1) since the last RBC blood transfusion and receipt of ≤ 2 units of blood in the past 30 days.
9. ECOG Performance status of 0 – 2 (Appendix 2).
10. Adequate renal (creatinine ≤ 1.5 x ULN or creatinine clearance ≥ 40 mL/min) and hepatic function [bilirubin ≤ 1.5 x ULN; AST/ALT ≤ 2.5 x ULN (AST/ALT ≤ 5 x ULN when there is evidence of liver metastases)].
11. Females of childbearing potential participating in the study are to use adequate birth control measures (i.e. oral contraceptives or two forms of barrier method birth control) during study participation. Females of childbearing potential must have a negative serum pregnancy test within 7 days to ACE-011/placebo administration (Day 1). A female of childbearing potential is a sexually mature woman who has not undergone a hysterectomy or who has not been postmenopausal for at least 24 consecutive months (i.e. who has had menses at some time in the preceding 24 months).
12. Life expectancy of ≥ 6 months.
13. Is willing to adhere to the study visit schedule, understand and comply with all protocol requirements.

14. Understand and sign a written informed consent.

8.2.2. Patient Exclusion Criteria

1. At the time of screening patients who have any grade ≥ 3 toxicity (as per NCI CTCAE v3.0 (Appendix 3)) (except for the following disease related toxicities: hematological events- anemia, thrombocytopenia or neutropenia or non hematological events- nausea, vomiting, fatigue, or muscle or bone/joint pain).
2. Prior radiation therapy to > 20% of the whole skeleton.
3. Have received more than 5 prior chemotherapy treatment regimens for metastatic breast cancer (not including the current chemotherapy regimen).
4. Clinically significant pulmonary, endocrine, neurologic, gastrointestinal, hepatic or genitourinary disease unrelated to underlying hematologic disorder.
5. Patients with heart failure as classified by the New York Heart Association (NYHA) classification of 3 or higher (Appendix 4).
6. Recent history of thrombosis, deep vein thrombosis (DVT), pulmonary emboli, or embolic stroke occurring within the last 6 months; if on anticoagulation therapy must be clinically stable as determined by the Investigator.
7. Untreated CNS metastases or CNS metastases treated with whole brain radiotherapy < 6 months prior to Day 1.
8. Diagnosis of a myeloid malignancy or known history of myelodysplasia.
9. History of second malignancy within 5 years (except excised and cured basal cell carcinoma, squamous cell carcinoma of the skin or cervical carcinoma in situ).
10. Administration of IV antibiotics or febrile (temperature elevation > 38 ° C) within 14 days of Day 1.
11. Uncontrolled hypertension. If hypertension is considered clinically stable, systolic blood pressure (BP) must be < 150 mmHg and diastolic BP must be < 100 mmHg.
12. Known history hepatitis B surface antigen (HBsAg and HB core Ab), human immunodeficiency virus (HIV) antibody or active hepatitis C.
13. Clinically significant iron (transferrin saturation < 20%), vitamin B_{12}, or folate deficiency.
14. History of anemia as a result of inherited hemoglobinopathy such as sickle cell anemia or thalassemia.
15. History of autoimmune or hereditary hemolysis; active gastrointestinal bleeding (within the last 6 months as compared to Day 1).
16. Patient received treatment with another investigational drug or device within 1 month prior to Day 1, or if the half life of the previous product is known, within 5 times the half life prior to dosing, whichever may be longer.
17. Pregnant or lactating females.
18. History of severe allergic or anaphylactic reactions or hypersensitivity to recombinant proteins or excipients in the investigational product.

19. Major surgery within 30 days prior to Day 1 (patients must have completely recovered from any previous surgery prior to Day 1).

8.3. Patient Withdrawal Criteria

Patients will be informed that they have the right to withdraw from the study at any time for any reason without prejudice to their medical care.

Patients must be withdrawn from the study for any of the following reasons:

- Patient request
- Patient is unwilling or unable to comply with the protocol
- ESA administration during the treatment period
- Medical reason, such as beginning a new chemotherapy regimen, cancer or treatment related toxicity, or at the discretion of the Investigator and/or the Medical Monitor(s)
- Patients with a hemoglobin level above the upper limit of normal at any time during the study after Day 1
- Grade ≥ 3 toxicity related to ACE-011 [except grade 3 hypertension that decreases to grade ≤ 1 within 7 days after treatment with anti-hypertensive therapy (section 9.6.2.1)]
- Pregnancy

The reasons for withdrawal must be recorded in the patient's case report form (CRF). The Investigator must notify the Medical Monitor and the contract research organization (CRO) immediately when a patient has been discontinued/withdrawn due to an AE. All patients who are withdrawn from the study should complete the tests and evaluations scheduled for Day 281/Termination at the time of withdrawal. Discontinued/withdrawn patients should be followed for survival only for 7 months after the last dose of ACE-011/placebo. If a patient begins a new chemotherapy regimen during the study, the patient will discontinue further ACE-011/placebo treatment, complete the Day 281/Termination visit procedures and be followed for survival only. Patients who withdraw due to ESA administration during the treatment period or due to ACE-011 related toxicity should enter the treatment follow-up period.

9. TREATMENT OF PATIENTS

9.1. Concomitant Medications

9.1.1. General Concomitant Medication Usage

During screening, and during the study, patients may take only stable doses of medications for chronic conditions that are not specifically excluded by the protocol (see Sections 8.2.1 Patient Inclusion Criteria and 8.2.2 Patient Exclusion Criteria). The Medical Monitor should be consulted by the Investigator if there are any concerns about what constitutes a stable dose or what is a chronic condition. Patients with a recent change in anti-hypertensive medication or a change in the past month should be seen weekly for BP measurements. Concomitant medications will be recorded in the CRF through 90 days from the last dose of ACE-011/placebo.

9.1.2. Concomitant treatment for anemia

Concurrent therapy with a new prescription medication related to treatment of anemia during the course of the study must first be discussed with the Investigator and Medical Monitor prior to administration, unless appropriate medical care necessitates that medication should begin before the Medical Monitor can be consulted.

9.1.2.1. Iron supplementation

Iron supplementation within 7 days of Day 1 (besides an oral general multivitamin with iron) will not be allowed. If a patient becomes iron replete during study treatment (transferrin saturation < 20%), treatment with iron supplementation is at the discretion of the investigator.

9.1.2.2. Erythropoiesis-stimulating agents (ESAs)

If concurrent treatment with erythropoiesis-stimulating agent is required in the opinion of the investigator, the erythropoiesis-stimulating agent label instructions are to be followed. The patient is to discontinue further treatment with ACE-011/placebo and enter the treatment follow-up period of the study. See Table 7 for Schedule of Events.

9.1.2.3. RBC Transfusions

Concurrent treatment for chemotherapy induced anemia with blood transfusions is recommended when hemoglobin value is < 8 g/dL or at investigator discretion if the hemoglobin value is above 8 g/dL and associated with anemia symptoms such as hemodynamic or pulmonary compromise that require treatment. If a transfusion is given to a patient during the treatment period, ACE-011/placebo should be administered no sooner than 7 days from the date of the transfusion. After the transfusion, the hemoglobin value should be assessed no later than 7 days from the date of the transfusion. On the day of ACE-011/placebo administration, the hemoglobin value and blood pressure will be assessed. See Figure 1 for ACE-011/placebo dose modification rules.

9.2. Chemotherapy

Chemotherapy treatment for metastatic breast cancer is to be given as per standard of care at the site. ACE-011/placebo administration should be given prior to chemotherapy administration on ACE-011/placebo administration days. Continuation of treatment with the same chemotherapy regimen through the follow-up period of the study is at the discretion of the investigator.

9.3. Treatment Compliance

Each dose of ACE-011/placebo will be administered as a SC injection at the clinical site by the study staff and will be documented in the study record. Monitoring for patient compliance with the treatment regimen is therefore unnecessary.

9.4. Rescreening

If a patient receives a screening number through Interactive Voice/Web Response System (IVRS/IWRS) and does not continue to meet eligibility criteria (prior to randomization), the patient may be re-screened once during the enrollment period of the study. Evaluation and decision to re-screen a patient will be done by the site, Medical Monitor, and Acceleron (as needed). All screening procedures are to be re-performed to verify patient eligibility. The screening DXA scan is to be re-done if more than 45 days has elapsed since the initial screening DXA scan. Informed consent must be signed by the patient if more than 14 days has elapsed since initially signing consent.

Patients who have been randomized but have not received the first dose of ACE-011/placebo and no longer meet eligibility criteria will require further evaluation by the site, Medical Monitor, and Acceleron (as needed) for continued participation in the study.

9.5. Randomization

Patients will be randomized to limit scientific bias within the study. Randomization assignments will be generated through a computerized system, provided by an Interactive Voice/Web Response System (IVRS/IWRS). Patients will be stratified according to frequency of the planned chemotherapy regimen (weekly chemotherapy vs. less frequent chemotherapy administration).

9.5.1. Blinding

The Investigators, patients, and sponsor or representative will remain blinded to the treatment arm assignment of each patient. The unblinded statistician will remain uninvolved in the study conduct until database lock and unblinding of the data has occurred.

Safety parameters will be reviewed on an ongoing basis throughout the study including adverse events, serious adverse events, laboratory listings, and vital signs. If substantial toxicity trends are observed in one treatment group versus another, the DMC will have access to the unblinded treatment code assignment schema.

9.5.2. Unblinding

In the event of a medical emergency for an individual patient in which knowledge of the study medication is critical to the patient's medical management, the investigator may request to break the blind for that patient. However, prior to breaking the blind, every effort must be made by the investigator to first discuss the need to break the blind with the sponsor or sponsor representative. Further, it must be determined by the investigator that breaking the treatment blind is necessary information for the medical management of that patient. If the blind is broken, the patient must withdraw from the study by completing the Day 281/Termination visit procedures and be followed for survival only

9.6. Treatments Administered

9.6.1. Selection of Doses in the Study

Single dose administration of ACE-011 up to 3.0 mg/kg was previously demonstrated to be safe and to have durable effects on markers of bone formation and resorption as well as red blood cells following a single IV administration. A dose-limiting pharmacodynamic effect of ACE-011 with increases in hemoglobin, hematocrit and RBCs was established in healthy post-menopausal women at the 1.0 mg/kg dose level following multiple SC administration. This study will confirm the safety in following repeated SC administration of ACE-011 and further evaluate the potential efficacy in patients with chemotherapy induced anemia. The safety and preliminary efficacy of ACE-011 following multiple doses up to 0.5 mg/kg will be assessed using a parallel randomization design.

9.6.2. Selection and Timing of Dosing for Each Patient

Patients will be enrolled and receive their assigned dose of ACE-011/placebo every 28 days (i.e. on Days 1, 29, 57, and 85). After completion of ACE-011/placebo treatment, patients will return to the site monthly for 2 follow-up assessment (Days 113 and 141). Subsequently, the patient will return to the site for 2 post treatment follow-up assessments (i.e. Days 169 and 225). The patient will return to the site for a termination visit approximately 2 months after the post treatment follow-up period (Day 281). Patients will be discontinued from the ACE-011/placebo treatment for reasons listed in section 8.3 of the protocol.

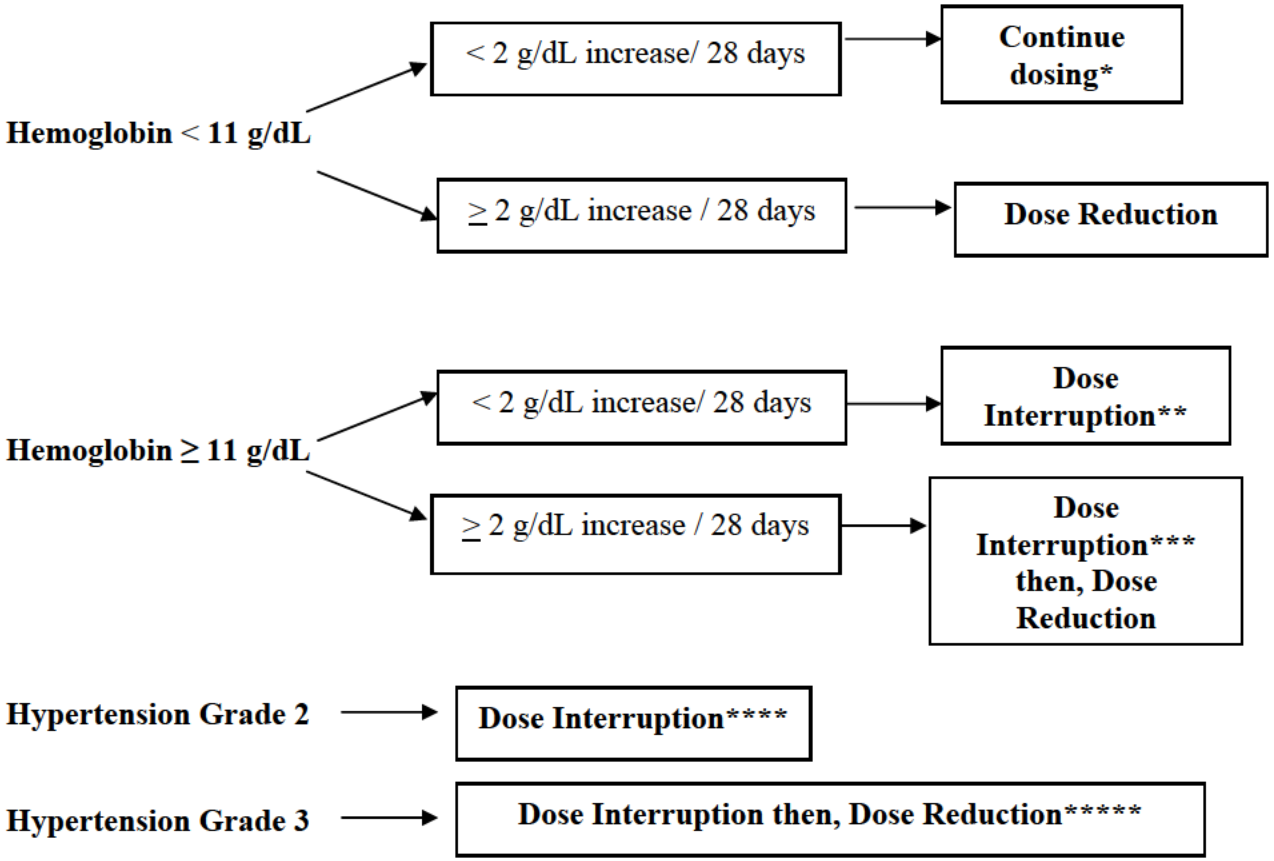
9.6.2.1. ACE-011 Treatment Related Toxicity

Patients with grade ≥3 toxicity related to ACE-011 will be discontinued from ACE-011/placebo treatment except for the following. Patients with grade 3 hypertension that decreases to grade ≤ 1 within 7 days after treatment with anti-hypertensive therapy may continue on ACE-011/placebo treatment. Dose reduction rules will apply at the subsequent scheduled dosing day(s).

9.6.2.2. ACE-011 Dose Modification Rules

Throughout the study blood pressure and hemoglobin values will be evaluated for patient safety. The local hemoglobin values and blood pressure values will be evaluated at each study visit and on each ACE-011/placebo dosing day. The hemoglobin values from the previous 28 days between each ACE-011/placebo dosing day and blood pressure will be reviewed and evaluated for dose modifications. The parameters below outline rules for dosing modifications (Figure 1). Table 8 outlines the dose reduction levels of ACE-011.

Figure 1: ACE-011/Placebo Dose Modification Rules (after initial dose of ACE-011/placebo)



* Patients who have received a blood transfusion in the past 28 days should continue with dosing at the same ACE-011/placebo dosing level if the transfusion was given no sooner than 7 days from the dosing day and the hemoglobin level is < 11 g/dL and hypertension ≤ Grade 1 on the day of dosing

** ACE-011/placebo should be held until the following scheduled treatment visit

*** ACE-011/placebo should be held and at the following scheduled treatment visit, a dose reduction of ACE-011/placebo will be administered based on the evaluation of the hemoglobin and blood pressure at that time

**** ACE-011/placebo should be held until resolution to ≤ Grade 1 (NCI CTCAE v3 0) and may then be resumed at the following scheduled treatment visit.

***** ACE-011/placebo should be held until resolution to ≤Grade 1 (NCI CTCAE v3 0) within 7 days after treatment with anti- hypertensive therapy and may continue on a reduced dose of ACE-011/placebo treatment for the subsequent scheduled dosing day

Table 8: ACE-011/Placebo Dose Reduction Levels

When required per the dose modification rules above (Figure 1), ACE-011/placebo dose(s) should be reduced as follows for dose 2 (Day 29), dose 3 (Day 57) and/or dose 4 (Day 85):

Dose	Dose reduction #1	Dose Reduction #2	Dose Reduction #3
Placebo	0.1 mg/kg	0.05 mg/kg	0.03 mg/kg
0.1 mg/kg	0.05 mg/kg	0.03 mg/kg	0.01 mg/kg
0.3 mg/kg	0.1 mg/kg	0.05 mg/kg	0.03 mg/kg
0.5 mg/kg	0.3 mg/kg	0.1 mg/kg	0.05 mg/kg

Blood pressure and hemoglobin values must be evaluated at each dosing day for consideration of administration of ACE-011/placebo.

The dose level for the initial dose reduction will be maintained unless subsequent dose reductions need to be administered, upon evaluation of the dose modifications listed above. A patient may have up to 3 dose reductions in the study.

Dose reduction steps and the administration of the reduced dose(s) will be conducted in a manner that will preserve the blinded status of the original treatment group of each patient. Patients in the placebo group (dose level 0.3 mg/kg) who are designated by the investigator to undergo a dose reduction will continue to receive placebo.

10. STUDY PROCEDURES AND SCHEDULE

10.1.1. Written Informed consent

Patients will be required to sign an Institutional Review Board/Institutional Ethics Committee (IRB/IEC) approved informed consent form (ICF) prior to any study related procedures, including screening evaluations.

Screen failure information will be maintained, including but not limited to, reason for failure.

10.1.2. Screening (Within 14 days of dosing)

The following will be collected within 14 days prior to the initial dosing Day 1. Labs to confirm eligibility criteria must be drawn and resulted within 7 days of Day 1:

- Informed consent
- Assessment of inclusion/exclusion criteria
- Complete Medical History and Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), temperature (°C) and height.
- FACT-Fatigue survey
- At two different time intervals (approximately 1 hour apart) triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min) with the use of a digital ECG machine
    - If a local ECG machine is used, a single (12 lead) ECG is to be performed.
- Serum iron, transferrin saturation %, and total iron binding capacity (TIBC)
- Vitamin B12 and serum folate levels
- Coagulation
- Serum chemistry
- Hematology
- LH, FSH, testosterone, progesterone, and estradiol
- Serum Pregnancy Test for females of childbearing potential- to be assessed within 7 days of Day 1.
- Urinalysis
- Evaluation of transfusion frequency (including history of transfusion up to 8 weeks prior to Day 1)
- Documentation of concomitant medications (any treatments taken 28 days prior to Day 1)

- CT/MRI scan: A CT/MRI scan acquired within 6 weeks of initiating the current chemotherapy regimen may be used for the screening assessment. One measurable or non-measurable lesion per RECIST v1.1 is to be identified by the scan.
    - Scans of the anatomical areas of head (if applicable), neck, thoracic, abdominal, and pelvic are to be available for evaluation. If one of the anatomical areas listed has not be acquired, prior to screening, then a CT/MRI must be done, during screening for this study, to ensure a scan of the head (if applicable), neck, thoracic, abdominal, and pelvic is available.
        - Brain (head) CT only to be done to rule out CNS metastases in patients with suspicious clinical symptoms or prior history of CNS metastases.
- Dual-energy X-ray absorptiometry (DXA) of lumbar spine and hip
    - Refer to Imaging Acquisition Guidelines for guidelines regarding washout period for contrast (if used) for radiological images, prior to acquiring DXA scan.
- Bone scans (only for patients with bone metastases) acquired within 6 weeks of initiating the current chemotherapy regimen may be used for the screening assessment.

10.1.3. Initial Dosing: Day 1

Patients will be dosed on Day 1. Results from screening evaluations must be reviewed prior to randomization to confirm patient eligibility. After the inclusion/exclusion criteria have been met and the patient has been randomized (through IVRS/IWRS), ACE-011/placebo administration may begin.

Chemotherapy regimen will be administered per standard of care at the site. The following tests must be performed prior to ACE-011/placebo administration:

- Confirm eligibility of patient by inclusion/exclusion criteria (local lab hemoglobin value and central lab values). The local hemoglobin value should be drawn prior to ACE-011/placebo administration (up to 1 day prior to Day 1).
- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- FACT-Fatigue survey
- Triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min) with the use of a digital ECG machine
    - If a local ECG machine is used, a single (12 lead) ECG is to be performed.
- Serum iron, transferrin saturation %, and total iron binding capacity

- Coagulation
- Serum Chemistry
- Hematology
- Serum erythropoietin marker
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test
- Bone biomarkers- osteocalcin (OC), procollagen type 1 N-terminal propeptide (P1NP), serum bone specific alkaline phosphatase (BSAP), C-terminal type 1 collagen telopeptide (CTX), and tartrate-resistant acid phosphatase isoform-5b (TRACP-5b).
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions the on collection of serum for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Females of childbearing potential- Urine (dipstick) pregnancy test to be collected and resulted prior to ACE-011/placebo dosing
- Urinalysis
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events (after ACE-011/placebo administration)
- Patients will be administered ACE-011/placebo before the chemotherapy regimen begins during the study visits.

After ACE-011/placebo and chemotherapy are administered, the patient may leave the clinic, based on the clinical judgment of the staff.

All study visit day dates (inclusive of windows) are calculated from Day 1(day of initial ACE-011/placebo administration).

10.1.4. ACE-011/placebo administration: Days 29, 57, and 85 (± 3 days)

Chemotherapy regimen will be administered per standard of care at the site. ACE-011/placebo will be administered approximately every 28 days. The following procedures must be performed prior to ACE-011/placebo administration:

- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)

- FACT-Fatigue survey
- Serum iron, transferrin saturation %, and total iron binding capacity (Day 57 only)
- Coagulation (Day 57 only)
- Serum chemistry
- Hematology
- Serum erythropoietin marker
- Triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min) with the use of a digital ECG machine (Day 57 only)
    - If a local ECG machine is used, a single (12 lead) ECG is to be performed.
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test (Day 57 only)
- Bone biomarkers- OC, BSAP, P1NP, CTX, and TRACP-5b
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions the collection of serum for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Females of childbearing potential -urine (dipstick) pregnancy test to be collected and resulted prior to ACE-011/placebo dosing
- Urinalysis (Day 85 only)
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events

Hemoglobin (HgB) (by review of local lab values) and blood pressure (BP) measurement must be assessed prior to ACE-011/placebo dose administration. Local Hgb must be done within 1 day prior to the ACE-011/placebo dosing day.

10.1.5. Additional visits during Treatment Period: Days 8, 15 (± 1 day), 43, and 71 (± 2 days)

Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit. Patients with previous history of CNS metastases, if blood pressure (BP) value increases by one or more grade(s) per the NCI CTCAE v3.0, the patient must return within 7 days to have BP assessed again.

- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- Triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min) with the use of a digital ECG machine (Day 8 only)
    - If a local ECG machine is used, a single (12 lead) ECG is to be performed.
- Hematology
- Back-up blood sample to be drawn for future testing
- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events

If patient terminates ACE-011/placebo early and/or begins a new chemotherapy regimen, the patient should complete the Day 281/Termination visit procedures and be followed for survival only.

10.1.6. Treatment Period Follow-up visits : Days 113 and 141 (± 7 days)

Chemotherapy regimen will be administered per standard of care at the site. The following procedures are to be performed during the study visit:

- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- Triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min) with the use of a digital ECG machine (Day 113 only)
    - If a local ECG machine is used, a single (12 lead) ECG is to be performed.
- Serum Chemistry
- Hematology
- Serum erythropoietin marker (Day 141 only)
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test
- Bone biomarkers- OC, BSAP, P1NP, CTX, and TRACP-5b
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions on the collection of serum for the biomarkers.*
- Back-up blood sample to be drawn for future testing

- Evaluation of transfusion frequency
- Documentation of concomitant medications
- Evaluation of adverse events
- CT/MRI scan (Day 113 only, +/-7 day window). Same modality and anatomical areas as acquired prior to enrollment should be used for follow up scans. Tumor response assessments to be evaluated by the investigator.
- DXA of lumbar spine and hip (Day 113 only)
    - Refer to Imaging Acquisition Guidelines for guidelines regarding washout period for contrast (if used) for radiological images, prior to acquiring DXA scan.

10.1.7. Post Treatment Period Follow-up visits : Days 169 and 225 (± 7 days)

If a CT/MRI and bone scan (if applicable) is performed as per standard of care during the post treatment period follow up, results may be collected within the CRF to capture the status of the disease. Chemotherapy regimen will be administered per standard of care at the site, if applicable. The following procedures are to be performed during the study visit:

- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- Hematology
- Evaluation of transfusion frequency
- Documentation of concomitant medications (up to 90 days from the last dose of ACE-011/placebo)
- Evaluation of adverse events (up to 90 days from the last dose of ACE-011/placebo)

10.1.8. Termination Visit: Day 281 (± 7 days)

Day 281 is the final visit. If a patient terminates the study early, the Day 281 procedures should be followed. The following procedures are to be performed during the study visit:

- Physical Examination
- Vital signs including weight (kg), heart rate (beats/min), seated systolic and diastolic blood pressure (mmHg), and temperature (°C)
- FACT-Fatigue survey
- Triplicate (12 lead) ECGs to be performed at 3 minute intervals (± 1min) with the use of a digital ECG machine
    - If a local ECG machine is used, a single (12 lead) ECG is to be performed.
- Serum iron, transferrin saturation %, and total iron binding capacity

- Coagulation
- Serum chemistry
- Hematology
- Serum erythropoietin marker
- Anti-drug antibody test
- Bone biomarkers- OC, BSAP, P1NP, CTX, and TRACP-5b
    - *Please refer to the Study Reference Guide, provided under separate cover, for the Bone Biomarker Collection and Handling Guidelines for instructions on the collection of serum for the biomarkers.*
- Back-up blood sample to be drawn for future testing
- Females of childbearing potential -urine (dipstick) pregnancy test
- Urinalysis
- Evaluation of transfusion frequency
- Documentation of concomitant medications (up to 90 days from the last dose of ACE-011/placebo)
- Evaluation of adverse events (up to 90 days from the last dose of ACE-011/placebo)
- DXA of lumbar spine and hip
    - Refer to Imaging Acquisition Guidelines for guidelines regarding washout period for contrast (if used) for radiological images, prior to acquiring DXA scan.
- Bone scan (+/- 7 day window). Follow-up scan to be acquired if performed at baseline and the scan was abnormal.

If a patient has a positive anti-drug antibody result at the Day 281/Termination visit, they may be asked to return to the clinical site every 3 months until a negative result is obtained,

10.2. Discontinuation of Study

The Sponsor may terminate this study, after consultation with the Investigator, at any time for safety or administrative reasons. The Sponsor may terminate the study if the occurrence of serious adverse events (SAEs) or other findings suggest unacceptable risk to the health of the patients.

11. INVESTIGATIONAL PRODUCT MATERIALS AND MANAGEMENT

11.1. Reference Product

The placebo (control agent) to be used in this study will be sterile normal saline (0.9% Sodium Chloride for Injection) administered as a SC injection. Sterile, normal saline will be supplied by the investigational site's pharmacy, as applicable. The manufacturer's directions for placebo storage and handling are to be followed, as are standard clinical practices for ensuring sterility of the placebo.

11.2. Investigational Product Packaging and Labeling

ACE-011 will be supplied in 2 mL clear glass vials with gray stoppers and red flip-top seals that contain 1 mL of ACE-011. The investigational drug product consists of ACE-011 in PBS at a nominal concentration of approximately 50 mg/mL.

11.3. Investigational Product Storage

ACE-011 is recommended to be stored at ≤ -65℃.

11.4. Investigational Product Preparation

See ACE-011 Handling and Administration Guidelines.

11.5. Administration

ACE-011/placebo will be administered by subcutaneous injection (SC). Subcutaneous injections will be given in the upper arm and/or thigh. Please refer to ACE-011 Handling and Administration Guidelines document for further information.

Dose modifications will be determined by the investigator after reference to the dose modification rules listed in Figure 1. In the case of a dose interruption or dose reduction of ACE-011/placebo, the investigator or designee will notify the pharmacy staff, who is unblinded, of the treatment decision for appropriate preparation of the investigational drug to maintain the blind of the study.

11.6. Drug Accountability

Accountability for ACE-011/placebo is the responsibility of the Investigator. Investigational clinical supplies must be received by a designated person at the clinical site and kept in a secured location. The investigational site must maintain accurate records demonstrating dates and amounts of ACE-011/placebo received, to whom it was dispensed (patient-by-patient accounting), and accounts of any ACE-011/placebo accidentally or deliberately destroyed or returned. Unless otherwise notified, all vials of ACE-011/placebo, both used and unused, must be saved for drug accountability. The used vials may be discarded, per the institution's standard

practice, after drug accountability has been completed by the monitor. The Investigator must return all unused vials of ACE-011 to the Sponsor at the end of the study, or the ACE-011 vials may be destroyed at the clinical site with the permission of the Sponsor. For either scenario, the outcome must be documented on the drug accountability log. The Sponsor will provide direction for the outcome of all unused vials.

11.7. Investigational Product Handling and Disposal

Please refer to the ACE-011 Handling and Administration Guidelines, provided under separate cover, for detailed drug handling, administration, and storage instructions.

12. ASSESSMENT OF EFFICACY

Efficacy measurements will include assessments for hematopoietic response which is defined as an increase in hemoglobin of ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL above the study baseline value for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs, at each dose level. The hematopoietic endpoints will be determined by the monitoring of hematologic laboratory values and RBC transfusion and/or ESA administration. Secondary endpoints associated with assessment of disease progression will be evaluated by CT/MRI scan and bone mineral density by DXA scans, respectively. Bone biomarkers and incidence of skeletal related events (SREs) will also be evaluated for all patients. SREs are defined as pathologic fracture, radiation therapy to bone, surgery to bone, or spinal cord compression. Quality of life assessment (FACT-Fatigue) will be evaluated for each patient.

13. ASSESSMENT OF SAFETY

13.1. Safety Parameters

13.1.1. Adverse Events

All patients will be assessed for safety by monitoring adverse events, clinical laboratory tests, vital signs and physical examinations. AE information will be collected throughout the study. See Section 14 Adverse Events for a full description of AE definitions, AE and SAE reporting procedures, and emergency procedures.

13.1.2. Clinical Laboratory Tests

The following laboratory safety assessments will be drawn at the investigational site's local laboratory according to the laboratory collection recommendations. The samples will then be sent to the central lab for value reporting. Please refer to Lab Reference Manual for further information. Hemoglobin and hematocrit lab tests will be drawn and evaluated by the local laboratory at screening and at each visit day throughout the study. The local hemoglobin laboratory value will be used in the evaluation of ACE-011/placebo dose modifications. The sponsor may request additional safety tests be performed based on ongoing data review during the study.

- Iron studies: Serum iron, transferrin saturation %, total iron binding capacity (TIBC) on specified days.
- Nutritional tests: Serum folate and vitamin B12
- Coagulation: prothrombin time (PT), partial thromboplastin time (PTT), and fibrinogen
- Serum chemistry: AST, ALT, lactate dehydrogenase (LDH), total bilirubin, indirect bilirubin, alkaline phosphatase, total protein, albumin, sodium, potassium, chloride, carbon dioxide, calcium, phosphorus, blood urea nitrogen (BUN), creatinine, uric acid, glucose, amylase, and lipase.
- Hematology: complete blood count (CBC) with differential and platelet. CBC includes red blood cells (RBC), white blood cells (WBC), hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC). Reticulocyte percent, erythropoietin levels and peripheral blood smear will be collected on specified days.
- Bone biomarkers: (serum) osteocalcin (OC), procollagen type 1 N-terminal propeptide (P1NP), serum bone specific alkaline phosphatase (BSAP), C-terminal type 1 collagen telopeptide (CTX), and tartrate-resistant acid phosphatase isoform-5b (TRACP-5b).
- Pregnancy test for females of child bearing potential
- Urinalysis: including determination of pH, specific gravity, protein, glucose, ketones, blood, leukocyte esterase and nitrite. Microscopy required only to follow-up abnormal findings.

13.1.3. Other Safety Assessments

- Physical examinations
- Vital signs: weight (kg), heart rate (beats/min), seated blood pressure (mmHg) and temperature (°C). Height will be collected at screening only.
- 12-lead ECG(s)
- LH, FSH, testosterone, progesterone, and estradiol
- Anti-drug antibody test
- CT/MRI scan
- DXA
- Bone scans

13.2. Pharmacokinetics

See Appendix 5 for further details regarding PK sampling.

14. ADVERSE EVENT

Adverse Event

An adverse event (AE) is any noxious, unintended, untoward medical occurrence that may appear or worsen in a patient during the course of a study, which does not necessarily have a causal relationship with the administration of study medication. An AE can be any unfavorable and unintended sign (e.g. including an abnormal laboratory finding), symptom, or disease temporally associated with the use of the investigational drug, whether or not it is considered to be investigational drug related. This includes any newly occurring event or previous condition that has increased in severity or frequency since the administration of investigational drug (treatment-emergent).

Patients will be evaluated and questioned generally for AEs during the course of the study, starting from baseline (Day 1). All AEs occurring after ACE-011/placebo administration up to 90 days from the last dose of ACE-011/placebo are to be documented on the AE CRF.

Serious Adverse Event

A serious adverse event (SAE) is any AE, occurring at any dose level and regardless of causality that:

- Results in death.
- Is life-threatening: Life-threatening means that the patient was at immediate risk of death from the reaction as it occurred, (i.e., it does not include a reaction which hypothetically might have caused death had it occurred in a more severe form).
- Requires inpatient hospitalization or prolongation of existing hospitalization. Hospitalization is defined as an inpatient admission, regardless of length of stay.
- Results in persistent or significant disability/incapacity.
- Is a congenital anomaly/birth defect.
- Is an important medical event: an important medical event is an event that may not result in death, be life-threatening, or require hospitalization but may be considered an SAE when, based upon appropriate medical judgment, it may jeopardize the patient and may require medical or surgical intervention to prevent one of the outcomes listed in the definitions for SAEs. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.
- A hospitalization for an elective procedure will not be considered a SAE.

SAEs will be captured throughout the patient participation in the study through Day 281/Termination visit (i.e. from the signing of the ICF and throughout patient participation in the study procedures). For patients who discontinue from the study early, SAEs will be captured up to 90 days from the last dose of ACE-011/placebo for those patients who are followed for survival only.

14.1. Adverse Event Classification

The investigator should use the following classifications should be used when evaluating the relationship of AEs and SAEs to the investigational drug (i.e., causality assessment). .

None:	No relationship between the experience and the administration of investigational drug; related to other etiologies such as concomitant medications or patient's clinical state.
Unlikely:	The current state of knowledge indicates that a relationship is unlikely.
Possibly:	A reaction that follows a plausible temporal sequence from administration of the investigational drug and follows a known response pattern to the suspected investigational drug. The reaction might have been produced by the patient's clinical state or other modes of therapy administered to the patient.
Probably:	A reaction that follows a plausible temporal sequence from administration of the investigational drug and follows a known response pattern to the suspected investigational drug. The reaction cannot be reasonably explained by the known characteristics of the patient's clinical state or other modes of therapy administered to the patient.
Definitely:	A reaction that follows a plausible temporal sequence from administration of the investigational drug and follows a known response pattern to the suspected investigational drug and can be confirmed with a positive re-challenge test or supporting laboratory data.

Severity of AEs will be graded by the investigator using the National Cancer Institute (NCI; US) Common Terminology Criteria for Adverse Events (CTCAE) v3.0 guidelines (Appendix 3).

14.2. Recording Adverse Events

The Medical History CRF should be comprehensive of patient's medical history up to Day 1. All AEs occurring after ACE-011/placebo administration up to 90 days from the last dose of ACE-011/placebo are to be documented on AE CRF. The specific disease or syndrome rather than individual associated signs and symptoms should be identified by the investigator and recorded on the CRF.

Specifically, skeletal related event information is to be captured and reported for the duration of the patient participation in the study through Day 281/Terminiation visit. SREs will be captured up to 90 days from the last dose of ACE-011/placebo for those patients who are followed for survival only.

Serious Adverse Events (SAEs)

For SAEs, a SAE form must be completed with as much information as possible and submitted in the time frame described in Section 14.3. The Investigator is required to ensure that the data on the SAE form is accurate and consistent. This requirement applies to all SAEs (regardless of relationship to the investigational drug) that occur during the study (from the time the subject signs informed consent through Day 281 or 90 days after last dose of study medication for subjects who are followed for survival only) .

The SAE report should provide a detailed description of the SAE and include copies of hospital records and other relevant documents. If a subject died and an autopsy has been performed, copies of the autopsy report and death certificate are to be sent to the Sponsor or designee as soon as these become available.

When new significant information is obtained as well as when the outcome of an event is known, the investigator should record the information on a new SAE form. If the patient was hospitalized, a copy of the discharge summary must be included as part of the patient medical file. In all instances, the investigator should follow up with patients until the outcome of the SAE is known.

14.3. Reporting Adverse Events

All AEs spontaneously reported by the patient and/or in response to an open question from study personnel or revealed by observation, physical examination or other diagnostic procedures will be recorded on the AE CRF. Any clinically relevant change in laboratory assessments or other clinical findings is considered an AE and must be recorded on the AE CRF. Clinically significant laboratory abnormalities are those that are identified as such by the investigator and/or those that require intervention. All AEs are to be followed until the event resolves or the clinical course is stabilized.

Serious Adverse Events

Any AE that meets any criterion for an SAE requires the completion of an SAE Report Form, in addition to being recorded on the CRF. All SAEs must be reported to the Sponsor or designee by faxing the completed SAE form within 24 hours from the point in time when the Investigator becomes aware of the SAE. This instruction pertains to initial SAE reports as well as any follow-up reports. All SAEs and deaths must be reported regardless of relationship to investigational drug.

SAE Reporting:

PPD Medical Monitor
PPD Inc.
PPD

Please refer to the SAE Completion Guidelines for appropriate contact information.

Where required by local legislation, the Investigator is responsible for informing the IRB/EC of the SAE and providing them with all relevant initial and follow-up information about the event. The Investigator must keep copies of all SAE information on file including correspondence with Celgene and the IRB/EC.

Expedited Reporting of Adverse Events

For the purpose of regulatory reporting, Celgene Drug Safety will determine the expectedness of events suspected of being related to the investigational drug based on the ACE-011 Investigator Brochure.

The Sponsor, or its authorized representative, shall notify the Investigator of the following information:

- Any AE associated with the use of the investigational drug in this study or in other studies that is both serious and unexpected (i.e., SUSAR)
- Any finding from tests in laboratory animals that suggests a significant risk for human subjects including reports of mutagenicity, teratogenicity, or carcinogenicity.

Where required by local legislation, the Investigator shall notify his/her IRB/EC promptly of these new serious and unexpected AE(s) or significant risks to patients.

The Investigator must keep copies of all pertinent safety information on file including correspondence with the Sponsor, or its designee, and the IRB/EC.

14.4. Emergency Sponsor Contact

In the event of a medical emergency (i.e., an event that requires immediate attention regarding the treatment of a patient, operation of the clinical study, and/or the use of investigational drug), investigational site personnel must immediately contact the Medical Monitor.

14.5. Pregnancy

Any pregnancy that occurs during a clinical study with an investigational drug will be reported for tracking purposes only. All pregnancies that are identified during this study need to be followed to conclusion and the outcome reported. Female patients should immediately inform the Investigator of any pregnancies and will immediately be discontinued from study treatment. The patient will remain in the trial and continue to be followed for survival. The Investigator must report all pregnancies to the Sponsor or designee, within 24 hours of awareness, by facsimile, using the Initial Pregnancy Report Form.

The Investigator will follow the female subject until completion of the pregnancy, and must notify the Sponsor about the outcome of the pregnancy as a follow-up to the initial SAE report, using the Follow-up Pregnancy Report Form. If the pregnancy ends in abortion (i.e., spontaneous, therapeutic, etc) this would be considered an AE/SAE if it meets SAE criteria.

If the outcome of the pregnancy was abnormal (i.e., spontaneous or therapeutic abortion [any congenital anomaly detected in an aborted fetus must be documented], stillbirth, neonatal death, or congenital anomaly [including that in an aborted fetus]), the Investigator should follow the procedures for reporting SAEs. All abnormal pregnancy outcomes should be considered medically important (i.e., constitutes an important medical event) even if no other seriousness criteria apply, and must be reported to the Sponsor, or designee, immediately by facsimile, or other appropriate method, within 24 hours of the Investigator's knowledge of the event) using the SAE Report Form, or approved equivalent form.

All neonatal deaths that occur within 28 days of birth should be reported, without regard to causality, as SAEs. In addition, any infant death after 28 days that the Investigator suspects is related to the in utero exposure to the IP should also be reported to the Sponsor, or designee, immediately by facsimile, or other appropriate method, within 24 hours of the Investigator's knowledge of the event using the SAE Report Form, or approved equivalent form.

15. STATISTICS

Statistics Analysis Overview

The study will be considered complete with regard to the primary endpoint once all eligible subjects have completed up to 2 months after the last dose of ACE-011. Patients will be randomized to one of three ACE-011 treatment groups (0.1, 0.3 and 0.5 mg/kg) or placebo with a ratio of 2:2:2:1. Patients will be stratified according to administration of either single agent weekly chemotherapy or less frequent chemotherapy regimens.

15.1. Determination of Sample Size

With an anticipated dropout rate of 20%, a sample size of 30 patients per dose level is estimated to achieve at least 80% power with 2 sided significance level of 0.05 to differentiate targeted response rate of p1=0.45 from null hypothesis p0=0.20. An additional 15 patients will be enrolled to the placebo group for safety comparison. A total of 105 patients are planned to be randomized to three ACE-011 dose groups (0.1, 0.3, and 0.5 mg/kg) and placebo with a ratio of 2:2:2:1.

15.2. Analysis Populations

For this study, the following populations will be defined and used in the analysis and presentation of the data.

Modified Intent-to-Treat (MITT) population: The MITT population is defined as all patients randomized who received at least one dose of ACE-011/placebo.

Per-Protocol (PP) population: The PP population is defined as all patients randomized who have confirmed chemotherapy induced anemia, received at least one dose of ACE-011 and on study (treatment) for at least 2 months (57 days).

Safety population: The safety population is defined as all patients who received at least one dose of ACE-011 or placebo.

15.3. Background and Demographic Characteristics

Summary statistics (mean, standard deviation, median, minimum and maximum) will be provided for those variables measured on a continuous scale. The frequency distribution (number and percentage) will be provided for those variables measured on a nominal scale.

15.3.1. Patient Disposition

Patient disposition (analysis population allocation, entered, discontinued, along with primary reason for discontinuation) will be summarized using frequency and percent for each dose group. A summary of patients enrolled by site will be provided.

15.3.2. ACE-011

ACE-011 treatment exposure (total dosage received) will be summarized for each dose group as well as for each cycle for each dose level. Listings for dose adjustment will be provided.

15.3.3. Concomitant Therapy

By-patient listings will be provided for all concomitant medications and therapies taken during the study. All concomitant medications will be coded and categorized by the WHO drug coding system. Usage frequency of each coded concomitant medications will be summarized by each dose group.

15.4. Efficacy Evaluation

15.4.1. Primary Efficacy Analysis

The primary efficacy endpoint is the hematopoietic response rate at each dose level. It is defined as the proportion of patients who have a hematopoietic response of an increase in hemoglobin of ≥ 1.0 g/dL for 28 consecutive days during the treatment period including up to 2 months after the last dose of ACE-011, in the absence of any RBC transfusions and/or ESAs. The ACE-011 treatment period is defined as from the time of first ACE-011 dose to up to 2 months after the last dose of ACE-011 treatment. To be considered as a responder, all hemoglobin measurements during a 28 consecutive day period or longer must have an increase of ≥ 1.0 g/dL from baseline. If RBC transfusions and/or ESAs are required, no hemoglobin measurements within 28 days will be used to determine the hemoglobin response. Primary efficacy analysis will be based on PP population. Primary efficacy analysis will also be conducted on MITT population as supportive to the PP based analysis. Patients who do not receive ACE-011 will be excluded from primary efficacy analysis. Patients who discontinue the ACE-011 treatment prematurely, with no sufficient hemoglobin measurement, will be counted as non-responder in the calculation of response rate.

A 95% confidence interval (CI) for the response rate for each dose level will be provided to assess whether 95% CI of response rate contains null hypothesis response rate, p0=0.20. The response rate and 95% CI will also be provided for patients with chemotherapy on weekly basis versus those with chemotherapy on a less frequent basis for each dose level as well as overall (all three doses levels analyzed together).

An exploratory analysis with the Kaplan Meier approach will be considered to account for those patients who discontinue the ACE-011 treatment prematurely if the drop out rate is greater than 20%. Exploratory analysis of comparing hematopoietic response rates between active and placebo will be performed using Fisher's exact test. Due to the nature of the study (phase 2 dose determination study), no multiplicity adjustment will made for the multiple comparisons.

15.4.2. Secondary Efficacy Analyses

The secondary efficacy endpoints include:

- Proportion of patients achieving an increase in hemoglobin of ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL for 28 consecutive days during the treatment period and up to 2 months after the last dose of ACE-011 in the absence of any RBC transfusions and/or ESAs.
- Duration of hematopoietic response (days) defined as the first time hemoglobin increases at least ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL from baseline to the last time there is hemoglobin ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL increase from baseline. Duration of response is only calculated for a responder and will be at least 28 days. The duration of response will be at least 28 days is only calculated for a patient who meets the primary efficacy endpoint.
- Time to achieve hematopoietic response (days) defined as time from first dose of ACE-011 to the first time a hemoglobin result at least ≥ 1 g/dL, ≥ 2 g/dL and/or hemoglobin concentration ≥ 11 g/dL increase from baseline and maintained for a period of at least 28 consecutive days.
- Proportion of patients receiving red blood cell (RBC) transfusions and/or ESAs in each dose group as well as within each cycle of each ACE-011 dose level.
- Objective tumor response rate for each dose level using RECIST v1.1 criteria.
- Progression-Free Survival is defined as the time from the start date of current chemotherapy regimen to the first observation of documented disease progression or death due to any cause. If the subject has not progressed or died at the time of final analysis, PFS will be censored at the time of last tumor assessment.

The statistical analysis for binary endpoints will be similar to the primary efficacy analysis. For time to event endpoints, the Kaplan-Meier method will be used to estimate the survival curve and median time to event. These analyses will be performed for each dose group and all ACE-011 groups combined.

15.5. Safety Evaluation

Safety variables will be tabulated and presented for all patients who receive ACE-011. Adverse events will be coded using the Medical Dictionary for Regulatory Activities (MedDRA). Incidence of treatment-emergent adverse events will be presented by system organ class (SOC) and preferred term. Adverse events will also be presented by severity, and relationship to investigational drug. Change from baseline in clinical laboratory parameters and vital signs will be summarized across time. Shift tables will be presented for selected laboratory parameters. Physical examination results will be presented in listings.

Data from all patients who receive one or more doses of ACE-011/placebo will be included in the safety analyses. To assess clinical safety, adverse events, vital sign measurements and clinical laboratory information will be summarized by visit. Adverse events and laboratory results will also be summarized by severity grade (NCI CTCAE v3.0). Descriptive statistics will be generated and shift tables provided as appropriate.

15.6. Pharmacokinetic Evaluation

Listing of individual patient serum ACE-011 concentrations, actual blood sampling times and graphs of concentration vs. time will be prepared for each dose group. Summaries of PK parameters will be summarized by dose group. The trough concentrations will be summarized for all patients who provide pharmacokinetic samples by dose group. Exploratory analyses will be performed to correlate trough concentrations with efficacy, bone biomarker data, and safety data, however these analyses will be data-driven and only conducted if warranted by the data.

15.6.1. Anti-drug Antibody Data

The results of anti-drug and neutralizing antibodies will be presented over time. Exploratory analysis will be performed on the potential effect of anti-drug antibody on ACE-011 PK and drug exposure if anti-drug antibody tests are deemed positive.

15.7. Interim analysis

There is no planned interim analysis for this study.

15.8. Deviation from Original Analysis Plan

Deviations from the statistical analyses outlined in this protocol will be indicated in the statistical analysis plan; any further modifications will be noted in the final clinical study report.

16. DIRECT ACCESS TO SOURCE DATA/DOCUMENTS

16.1. Study Monitoring

The Clinical Monitor will arrange to visit the investigator sites at regular intervals during the study. The monitoring visits must be conducted according to the applicable ICH and GCP guidelines to ensure protocol adherence, quality of data, drug accountability, compliance with regulatory requirements, and continued adequacy of the investigational site and its facilities. During these visits, CRFs and other data related to the study will be reviewed and any discrepancies or omissions will be resolved. The Clinical Monitor will be given access to study relevant source documents (including medical records) for purposes of source data verification.

During and/or after completion of the study, quality assurance officers named by the Sponsor or the regulatory authorities may wish to perform on-site audits. The Investigator is expected to cooperate with any audit and provide assistance and documentation (including source data) as requested.

16.2. Audits and Inspections

The Investigators and clinical sites will permit trial related monitoring, audits, IRB review, and regulatory inspections as requested by FDA, EMEA, or Health Canada the Sponsor or designee, including direct access to source data/documents (i.e., original medical records, laboratory reports, hospital documents, progress reports, signed ICFs, etc.) in addition to CRFs.

If the investigator is contact by any regulatory authority regarding an inspection, he/she should contact Celgene immediately.

17. QUALITY CONTROL AND QUALITY ASSURANCE

17.1. Data Quality Control and Quality Assurance

17.1.1. Investigator responsibility

The investigator is responsible for ensuring the study is conducted according to the protocol, Code of Federal Regulations (CFR), GCP, and any other applicable regulatory requirements. The investigator responsibilities are outlined in these documents along with ensuring that documentation of a signed informed consent, is obtained prior to patient participation in the study.

17.1.2. Protocol modifications

The investigator may not modify the protocol without agreement from the Sponsor and prior review or approval by the IRB. Any deviations from the protocol should be documented by the investigator or designee.

17.2. Financing and Insurance

Financing and insurance are addressed in a separate document.

17.3. Confidentiality

To maintain patient privacy, all CRFs, drug accountability records, study reports and communications will identify the patient by the assigned patient identification number. The Investigator will grant monitor(s) and auditor(s) from the Sponsor or designee and regulatory authority(ies) access to the patient's original medical records for verification of data gathered on the CRFs and to audit the data collection process. The patient's confidentiality will be maintained and will not be made publicly available to the extent permitted by the applicable laws and regulations.

All information regarding the investigational product supplied by the Sponsor to the Investigator is privileged and confidential information. The Investigator agrees to use this information to accomplish the study and will not use it for other purposes without consent from the Sponsor. It is understood that there is an obligation to provide the Sponsor with complete data obtained during the study. The information obtained from the clinical study will be used towards the development of the investigational product and may be disclosed to regulatory authority(ies), other Investigators, corporate partners, or consultants as required.

17.4. Publication Policy

All information concerning ACE-011 is considered confidential and shall remain the sole property of the Sponsor. The Investigator agrees to use this information only in conducting the study and shall not use it for any other purposes without the Sponsor's written approval. The Investigator agrees not to disclose the Sponsor's confidential information to anyone except to

people involved in the study that needs such information to assist in conducting the study, and then only on like terms of confidentiality and nonuse.

It is understood by the Investigator that the information developed from this clinical study will be used by the Sponsor in connection with the development of ACE-011, and therefore may be disclosed as required to regulatory agencies. To allow for the use of the information derived from clinical studies, it is understood that there is an obligation to provide the Sponsor with complete test results and all data developed in the study.

No publication or disclosure of study results will be permitted except as specified in a separate, written, agreement between the Sponsor and the Investigator.

17.5. Protocol Amendments

Protocol amendments that impact patient safety change the scope of the investigation, or affect the scientific quality of the study must be approved by the IRB and submitted to the appropriate regulatory authorities before implementation of such modifications to the study.

In the event that the protocol needs to be modified immediately to eliminate an apparent hazard to a patient, the Sponsor will amend and implement the protocol change and subsequently notify the regulatory authorities and/or the IRB, as appropriate.

18. ETHICS

18.1. Institutional Review Board or Independent Ethics Committee

The Investigator must obtain written IRB approval of the protocol, approval for relevant supporting information and all types of patient recruitment and advertisement and ICF prior to starting the study. The IRB will meet all Food and Drug Administration (FDA) requirements governing IRBs (Code of Federal Regulations, Title 21, Part 56).

The Sponsor or designee must approve the ICF submitted to the investigational site's IRB. All patient recruitment and advertisements must be submitted to the Sponsor or designee prior to submission to the IRB, for review.

18.2. Ethical Conduct of the Study

The Sponsor and the Investigator must comply with all instructions, regulations, and agreements in this protocol and in the applicable International Conference on Harmonisation (ICH) and GCP guidelines, and must also conduct the study in accordance with local regulations.

18.3. Written Informed Consent

18.3.1. Informed Consent

Informed written consent is required from each patient prior to any testing under this protocol, including screening tests and evaluations. The consent form, as specified by the clinical site's IRB, must follow the Protection of Human Patients regulations listed in the Code of Federal Regulations, Title 21, Part 50.

The background of the proposed study and the benefits and risks of the procedures and study must be explained to the patients. It is the responsibility of the Investigator to obtain consent and to provide the patient with a copy of the signed and dated ICF. Confirmation of a patient's informed consent must also be documented in the patient's medical record prior to any testing under this protocol, including screening tests and evaluations.

All ICFs used in this study must be approved by the appropriate IRB and by the Sponsor or designee. The ICF must not be altered without the prior agreement of the relevant IRB and the Sponsor.

18.3.2. Patient Data Protection

Prior to any testing under this protocol, including screening tests and evaluations, patients must authorize the release and use of protected health information (PHI), as required by local law.

The patient will not be identified by name in the CRF or in any study reports. These reports will be used for research purposes only. The Sponsor, its designee, and various government health agencies may inspect the records of this study. Every effort will be made to keep the patient's personal medical data confidential.

19. DATA HANDLING AND RECORDKEEPING

19.1. Case Report Form Completion

CRFs will be completed for each randomized patient. It is the Investigator's responsibility to ensure the accuracy, completeness, and timeliness of the data reported in the patient's CRF. Source documentation supporting the CRF data should indicate the patient's participation in the study and should document the dates and details of study procedures, AEs, and patient status.

Investigators will maintain copies of the CRFs at the clinical site. For patients who discontinue or terminate from the study, the CRFs will be completed as much as possible, and the reason for the discontinuation or termination clearly and concisely specified on the appropriate CRF.

19.2. Retention of Records

The Investigator will maintain all study records according to ICH-GCP and applicable regulatory requirements. Records will be retained for at least 2 years after the last marketing application approval in an ICH region and until there are no pending or contemplated marketing applications in an ICH region or 2 years after formal discontinuation of the clinical development of the investigational product or according to applicable regulatory requirements. If the Investigator withdraws from the responsibility of keeping the study records, custody must be transferred to a person willing to accept the responsibility. The Sponsor must be notified in writing if a custodial change occurs.

20. REFERENCES

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21. APPENDICES

Appendix 1: Response Evaluation Criteria in Solid Tumors

Response Evaluation Criteria in Solid Tumors (RECIST) v1.1	
Terms	Definition
Measurable disease	The presence of at least one measurable lesion by CT or MRI. If the measurable disease is restricted to a solitary lesion, its neoplastic nature should be confirmed by cytology/histology. All measurements should be taken and recorded in metric notation, using a ruler or calipers. All baseline evaluations should be performed as closely as possible to the beginning of treatment and never more than 4 weeks before the beginning of the treatment.
Measurable lesions	Lesions that can be accurately measured in least one dimension by CT scan with longest diameter $\geq$ 10 mm (CT scan thickness no greater than 5 mm). The same method of assessment and the same technique should be used to characterize each identified and reported lesion at baseline and during follow-up.
Non-measurable lesion	All other lesions, including small lesions (longest diameter <10mm or pathological lymph nodes with P10 to <15mm short axis) as well as truly non-measurable lesions. Lesions considered truly non-measurable include: leptomeningeal disease, ascites, pleural or pericardial effusion, inflammatory breast disease, lymphangitic involvement of skin or lung, abdominal masses/abdominal organomegaly identified by physical exam that is not measurable by reproducible imaging techniques.

RECIST Criteria-Response Evaluation

Evaluation of target lesions	
Complete Response (CR)	Disappearance of all target lesions. Any pathological lymph nodes (whether target or non-target) must have reduction in short axis to <10 mm.
Partial Response (PR):	At least a 30% decrease in the sum of diameters of target lesions, taking as reference the baseline sum diameters.
Stable Disease (SD):	Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum diameters while on study.
Progressive Disease (PD):	At least a 20% increase in the sum of diameters of target lesions, taking as reference the smallest sum on study (this includes the baseline sum if that is the smallest on study). In addition to the relative increase of 20%, the sum must also demonstrate an absolute increase of at least 5 mm. (Note: the appearance of one or more new lesions is also considered progression).

Evaluation of non-target lesions	
Complete Response (CR):	Disappearance of all non-target lesions and normalisation of tumour marker level. All lymph nodes must be non-pathological in size (<10mm short axis).
Non-CR/Non-PD	Persistence of one or more non-target lesion(s) and/or maintenance of tumour marker level above the normal limits.
Progressive Disease (PD):	Unequivocal progression (see comments below) of existing non-target lesions. (Note: the appearance of one or more new lesions is also considered progression).

Evaluation of best overall response	The best overall response is the best response recorded from the start of the treatment until disease progression/recurrence (taking as reference for PD the smallest measurements recorded since the treatment started). In general, the patient's best response assignment will depend on the achievement of both measurement and confirmation criteria. Patients with a global deterioration of health status requiring discontinuation of treatment without objective evidence of disease progression at that time should be classified as having "symptomatic deterioration". Every effort should be made to document the objective progression even after discontinuation of treatment. In some circumstances it may be difficult to distinguish residual disease from normal tissue. When the evaluation of complete response depends on this determination, it is recommended that the residual lesion be investigated (fine needle aspirate/biopsy) to confirm the complete response status.

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Appendix 2: ECOG Performance Status

The ECOG scale is used to assess a patient's quality of life in an evaluation by a health professional of the daily activities and how the activities are affected by the disease of the patient.

Grade	ECOG
0	Fully active, able to carry on all pre-disease performance without restriction
1	Restricted in physically strenuous activity but ambulatory and able to carry out work of a light or sedentary nature, e.g., light house work, office work
2	Ambulatory and capable of all selfcare but unable to carry out any work activities. Up and about more than 50% of waking hours
3	Capable of only limited selfcare, confined to bed or chair more than 50% of waking hours
4	Completely disabled. Cannot carry on any selfcare. Totally confined to bed or chair
5	Dead

Oken, M.M., Creech, R.H., Tormey, D.C., Horton, J., Davis, T.E., McFadden, E.T., Carbone, P.P. (1982) Toxicity And Response Criteria Of The Eastern Cooperative Oncology Group. Am J Clin Onco 5:649-655.

Appendix 3: National Cancer Institute (NCI) Common Terminology Criteria for Adverse Events (CTCAE) Version 3.0

See http://ctep.cancer.gov/reporting/ctc.html

Appendix 4: New York Heart Association- Classification of heart failure

Class 1 - Class 1 heart failure - no limitation of activities. No symptoms from ordinary activities.
Class 2 - Class 2 heart failure- mild limitation of activity. Comfortable with rest or mild exertion.
Class 3 - Class 3 heart failure- marked limitation of activity and be comfortable only at rest.
Class 4 –Class 4 heart failure- complete rest, confined to bed or chair; any physical activity brings on discomfort and symptoms occur at rest.

National Heart, Lung, and Blood Institute, National Institutes of Health. New York Heart Association Classification. 2008.

Appendix 5- Pharmacokinetic Sampling

Pharmacokinetics

Blood samples will be collected from approximately one-third of patients at selected study sites for the evaluation of serum concentrations of ACE-011.

Blood samples for PK should be collected in a fasted state (defined by no food or drink except water for at least 4 hours prior to the study procedure) with the exception of the Day 1 post 4 hour dose and the Day 85 post 4 hour dose. Pre-dose samples should be collected within 1 hour of ACE-011/placebo dosing (Day 1, 29, 57, and 85). The post 4 hour collection (± 10 minute window) on Day 1 and Day 85 is calculated to be 4 hours post the administration of ACE-011/placebo. Chemotherapy regimens scheduled will be administered to the patient as per standard of care. Collection, handling, and shipping procedures for blood samples are provided in the Study Reference Guide.

Schedule of Events cont.

	Screen	**Treatment Period**										**Post Treatment Follow up Period**		**Term Visit**
ACE-011/Placebo Dose period		**#1**			**#2**		**#3**		**#4**	**Follow-up**				
Day	**-14-0**	**1**	**8 (± 1d)**	**15 (± 1d)**	**29 (± 3d)**	**43 (± 2d)**	**57 (± 3d)**	**71 (± 2d)**	**85 (± 3d)**	**113 (± 7d)**	**141 (± 7d)**	**169 (± 7d)**	**225 (± 7d)**	**281 (± 7d)**
PK Sampling		X X- Post 4 hours	X	X	X		X		X X- Post 4 hours	X				
ACE-011/Placebo administration		X			X		X		X					
Chemo regimen		X	X	X	X	X	X	X	X	X	X	X	X	X