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BMS Protocol #: CA163-157

ICE Trial

RANDOMIZED OPEN-LABEL NEO-ADJUVANT PHASE II STUDY COMPARING IXABEPILONE (I) Vs IXABEPILONE PLUS CETUXIMAB (IC) IN TRIPLE NEGATIVE BREAST CANCER PATIENTS (ICE)

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STUDY SUMMARY

Title:	Randomized open-label neo-adjuvant phase II study comparing ixabepilone (I) vs. ixabepilone plus cetuximab (IC) in Triple negative breast cancer patients
Objectives:	Primary Objective: 1. To determine the pathologic complete response rate (pCR) (breast and axilla) of Ixabepilone versus Ixabepilone when combined with cetuximab in patients with invasive breast adenocarcinoma T1N1-N3M0 or T2-4 N0-3M0 disease who are Triple negative and who are candidates for preoperative chemotherapy. Secondary Objective(s): 1. To evaluate overall objective response rate in both treatment groups. 2. To assess toxicity of each regimen. Exploratory Objectives: 1. To correlate the expression of biomarkers such as PTEN, EGFR and EGFR-pathway associated genes (including, but not limited to, TGF-α, CRYAB, NRAS, KRAS, AKT3, PTEN, MEK1 and KRAS amplicon genes) with the objective clinical and pathologic response. 2. To potentially explore other gene/protein expression patterns, mutations and gene/chromosomal copy number alterations related to sensitivity or resistance to ixabepilone, other anti-microtubule agents and cetuximab. These additional exploratory analyses may include, but are not limited to, beta tubulin isoforms, ABC transporters, TACC3, HCAPG, GTSE1, Kallikreins and defined gene models associated with ixabepilone response.

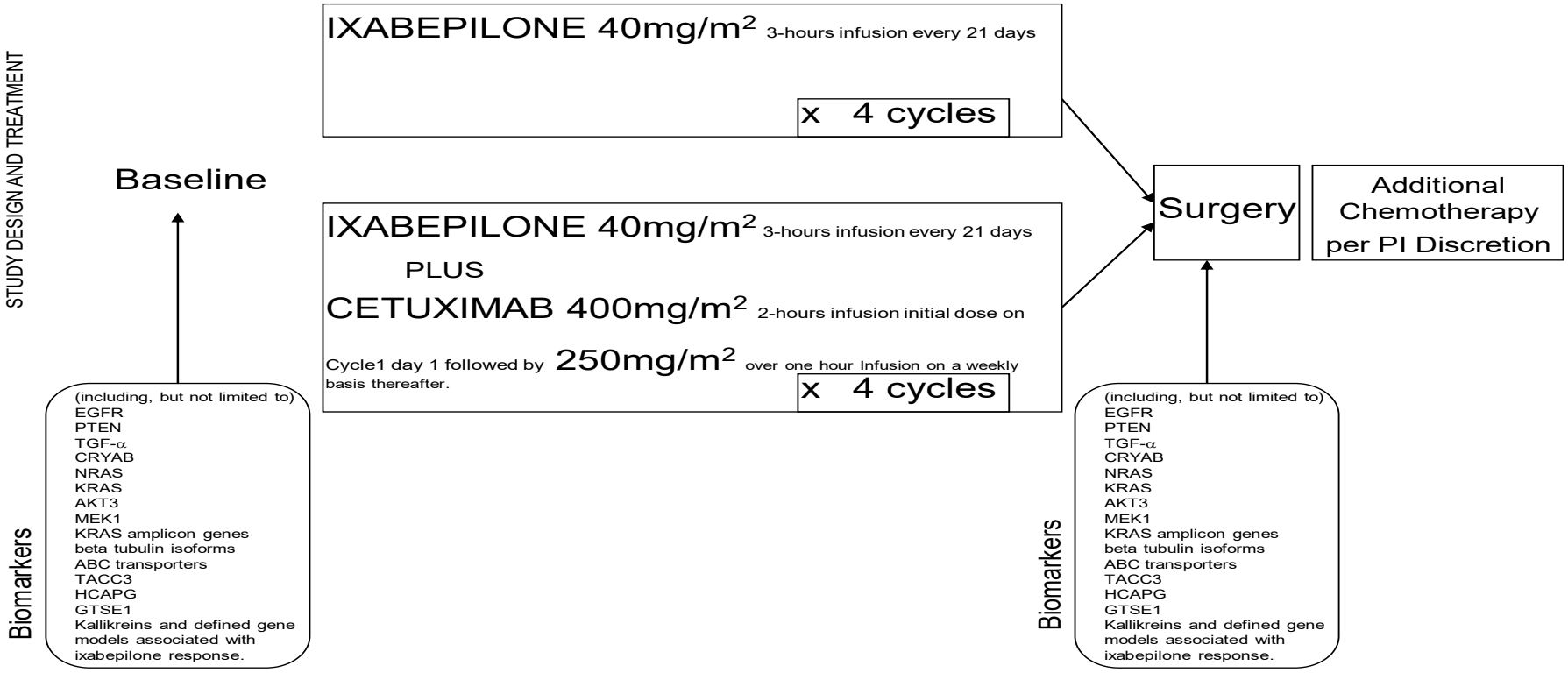
Study population:	-Patients with either T1N1-N3M0 or T2-4 N0-3M0 disease who are Triple negative and who are candidates for preoperative chemotherapy.
Sample Size:	116 patients.
Number of Centers:	Single Institution: The Methodist Hospital Research Institute.
Treatment Plan:	This is a randomized open-label phase II trial; 116 triple negative breast cancer patients will be randomized equally between 1) Ixabepilone or 2) Ixabepilone plus Cetuximab. The study population will include subjects ≥ 18 years of age diagnosed with invasive ductal carcinoma (IDC). Randomization will be stratified by disease stage (T1N1-3M0 or T2-4 N0-3M0). Prior to randomization, all patients will have a physical assessment, imaging, a blood draw, and a diagnostic biopsy prior to the start of treatment. The study doctor will determine if the subject qualified to be enrolled in this study. If subjects are enrolled, the duration of their participation is expected to be approximately 6 months Treatment Arm 1 Patients will receive Ixabepilone 40mg/m^2 given over approximately 3-hours on day 1. Patients will receive a total of four cycles of this drug with each cycle given 21 (+/- 3) days. Subjects will be premedicated approximately 1 hour before the ixabepilone infusion with 1) both an oral H1 blocker (diphenhydramine 50 mg or equivalent) and an oral H2 blocker (e.g., ranitidine 150 300 mg or cimetidine 300–800 mg or nizatidine 150-300 mg or famotidine 20–40 mg);

	or 2) with premedication per institutional standards. Premedication with steroids is indicated in subjects who developed a hypersensitivity reaction to ixabepilone in previous cycles. Patients with severe reaction (see dose modification chart below)should not be retreated. Treatment Arm 2 Patients will receive Ixabepilone 40mg/m^2 given over approximately 3-hours on day 1followed by cetuximab given on day 1 at 400 mg/m^2 as an initial loading dose (first infusion) administered as approximately a 120-minute IV infusion (maximum infusion rate 5 mL/min) followed by 250 mg/m^2 infused over approximately 60 minutes (maximum infusion rate 5 mL/min) as subsequent weekly dose. Patients will receive a total of 4 cycles of this combination with each cycle being 21 (+/-2) days. Subjects will be premedicated approximately 1 hour before the ixabepilone infusion with either 1) both an oral H1 blocker (diphenhydramine 50 mg or equivalent) and an oral H2 blocker (e.g., ranitidine 150-300 mg or cimetidine 300–800 mg or nizatidine 150 - 300 mg or famotidine 20–40 mg); or 2) with premedication per institutional standards. Premedication with steroids is indicated in subjects who developed a hypersensitivity reaction to ixabepilone in previous cycles. Patients with severe reaction (see dose modification chart below)should not be retreated. Subjects who are to receive cetuximab are to be premedicated with an H_1 antagonist (e.g., 50 mg of diphenhydramine IV or equivalent).
Inclusion Criteria:	Inclusion Criteria: • Patients with histologic confirmation of invasive breast carcinoma. • Patients must have intact primary tumor.

	• Patients ≥ 18 years. • Invasive breast carcinoma must be determined to be ER (-), PR (-) and HER2Neu (-) prior to study entry. • Patients should have T1N1-3M0 or T2-4 N0-3M0. • Patients with bilateral breast cancer are eligible. • Patients with second primary breast cancers are eligible. • Patients should have a Karnofsky performance scale of ≥ 70%. • Patients must have clinically measurable disease by physical exam to be treated in the neoadjuvant setting. • Patients should have adequate bone marrow function, as defined by peripheral granulocyte count of ≥1500/mm^3, and platelet count ≥100000mm^3. • Patients must have adequate liver function with a bilirubin within normal laboratory values. Alkaline phosphatase and transaminases (ALT and AST) may be up to 1.5 x upper limit of normal (ULN) of the institution. • Patients should have adequate renal function with creatinine levels within normal range. • Patients should have a normal left ventricular ejection fraction (LVEF) of ≥ 50%. • Negative serum or urine pregnancy test for a woman of childbearing potential (WOCBP). • WOCBP must use a reliable and appropriate contraceptive method during the study and six months after chemotherapy is completed. WOCBP are women who are not menopausal for 12 months or had no previous surgical sterilization. • Patients must sign an informed consent indicating that they are aware of the investigational nature of the study, in keeping with institutional policy
Exclusion Criteria:	Exclusion Criteria: • Patients with a history of other invasive malignancies diagnosed

	and treated within the previous 5 years, except non-melanoma skin cancer and non-invasive cervical cancer. • Her2Neu, ER and PR positive patients should be excluded. • Patients with an organ allograft or other history of immune compromise. • Prior treatment with any investigational drug within the preceding 4 weeks. • Chronic treatment with systemic steroids or another immunosuppressive agent. • Uncontrolled chronic viral infections or patients requiring parenteral antibiotics will be excluded. Patients taking highly active anti-viral therapies and with controlled disease will be allowed to participate. • Patients with an active, bleeding diathesis or on oral anti-vitamin K medication (except low dose coumarin defined as 1 mg a day) • Other concurrent and/or uncontrolled medical disease which could compromise participation in the study (i.e., uncontrolled diabetes, uncontrolled hypertension, severe infection, severe malnutrition, unstable angina, or congestive heart failure - New York Heart Association Class III or IV, ventricular arrhythmias, active ischemic heart disease, myocardial infarction within six months, chronic liver or renal disease, active upper GI tract ulceration). • Patients with a pre-existing peripheral neuropathy(> grade1).
Statistical Design:	Blocked randomization will be used to randomly assign patients either to arm 1 or arm 2. A total of 116 patients will be enrolled to allow for 6.5% dropout before randomization of 108 patients, given sample size of 54 patients in each arm to be able to detect 25% difference in pathological complete response rates between the arm with Ixabelpilone plus Cetuximab and the arm with Ixabepilone with 81.9% power and a two-sided significance level of 0.05.

RANDOMIZED OPEN-LABEL NEO-ADJUVANT PHASE II STUDY COMPARING
IXABEPILONE (I) Vs IXABEPILONE PLUS **CE**TUXIMAB (IC) IN TRIPLE NEGATIVE BREAST CANCER PATIENTS (ICE).



Randomized open-label neo-adjuvant phase II study comparing ixabepilone (I) vs. ixabepilone plus cetuximab (IC) in Triple negative breast cancer patients.

1.0 Synopsis:

Ixabepilone and capecitabine combination has demonstrated to be an active regimen in patients with metastatic breast cancer (MBC) after failing an anthracycline and a taxane regimen. Cetuximab is active in tumors that express EGFR with demonstrated activity in head and neck and colorectal tumors. A proportion of breast cancers are known to express EGFR. Cetuximab's mechanism of action suggests the possibility of efficacy in breast cancer patients and several studies show that it may be efficacious in Triple Negative Breast Cancer (TNBC). This study seeks to evaluate Ixabepilone alone or in combination with cetuximab as a possible way to increase antitumor activity.

This is a randomized open-label phase II trial, one hundred sixteen triple negative breast cancer patients will be randomized equally between 1) Ixabepilone or 2) Ixabepilone plus Cetuximab. Randomization will be stratified by disease stage (T1N1-3M0 or T2-4 N0-3M0).
At the baseline all patients will receive a physical assessment, imaging, a blood draw, and a biopsy prior to the start of treatment.

1.1 Primary Objective(s):
To determine the pathologic complete response rate (pCR) (breast and axilla) of Ixabepilone versus Ixabepilone when combined with cetuximab in patients with invasive breast adenocarcinoma T1N1-N3M0 or T2-4 N0-3M0 disease who are Triple negative and who are candidates for preoperative chemotherapy.

1.2 Secondary Objective(s):
To evaluate overall objective response rate in both treatment groups.
To assess safety and toxicity of each regimen.

1.3 Exploratory Objective(s):

To correlate the expression of biomarkers such as PTEN, EGFR and EGFR-pathway associated genes (including, but not limited to TGF-α, CRYAB, NRAS, KRAS, AKT3, PTEN, MEK1 and KRAS amplicon genes) with the objective clinical and pathologic response.

For all samples available, the following tests will be conducted:

- Immunohistochemical detection of EGFR, and the PTEN expression in tumor tissue samples. For those EGFR-pathway associated genes (including, but not limited to, TGF-α, CRYAB, NRAS, KRAS, AKT3, PTEN, MEK1 and KRAS amplicon genes) an mRNA-based assay will be used.
- Additional exploratory biomarker analysis may be performed on tumor or blood samples during or following the study. Such exploratory analyses may include assessments of somatic gene mutation, gene/chromosome copy number alteration and gene/protein expression for genes/proteins related to sensitivity or resistance to ixabepilone, other micro-tubule agents and cetuximab. These exploratory biomarkers may include, but are not limited to, beta tubulin isoforms, ABC transporters, TACC3, HCAPG, GTSE1, Kallikreins and gene models associated with ixabepilone response.

These analyses will allow for the correlation of biomarkers expression and response to treatment. Tumor tissue samples will be obtained from the original paraffin blocks or slides containing tumor tissue from the initial diagnostic work-up (see section 7.1).

In addition to the diagnostic core needle biopsy for biomarkers at baseline a second search for the same biomarkers will be done at the time of surgery.

The following tests will be conducted (prioritized in the order below):

- EGFR expression by IHC
- PTEN expression by IHC
- Estrogen receptor expression by IHC (at baseline only)

- Progesterone receptor expression by IHC (at baseline only)
- Her2 neu expression by IHC and/or FISH (at baseline only)
- Ki67 (MIB-1) by IHC (at baseline only)
- TGF-α expression by mRNA-based assay
- CRYAB expression by mRNA-based assay
- NRAS expression by mRNA-based assay
- KRAS expression by mRNA-based assay
- AKT3 expression by mRNA-based assay
- MEK1 expression by mRNA-based assay
- KRAS amplicon genes expression by mRNA-based assay
- Beta tubulin isoforms
- ABC transporters
- TACC3
- HCAPG
- GTSE1
- Kallikreins and defined gene models associated with ixabepilone response.
- Additional exploratory biomarkers related to ixabepilone, anti-microtubule and cetuximab sensitivity or resistance

The goal of this clinical research study is to compare the effectiveness of each study group of chemotherapy in triple negative breast cancer patients, and to evaluate the pathologic response rate for each group. We will be collecting tissue from the primary tumor for biomarker testing; the procedure will be done at baseline, and at the time of surgery.

2.0 Study Flow chart or Schedule of Events

Procedure	Screening/Baseline Visit	Prior to Each Cycle	End-of- Treatment Visit [a]	Follow-up
Eligibility Assessments				
Obtain Informed Consent	X			
Confirm Inclusion/Exclusion Criteria Met [b]	X			
Obtain Medical History	X			
Safety Assessments				
Karnofsky performance scale [a b]	X	X	X	
Record Weight and Height	X	X	X	
Document Vital Signs	X	X	X	
Assess Signs and Symptoms and Adverse Events [c, o]	X	X	X	
Perform Pregnancy Test in WOCBP [d]	X			
Perform mammogram [e]	X		X	
Perform ultrasound of breast and nodal region [e]	X		X	
(MRI)*				
Perform Laboratory Tests [fghi]	X	X	X	X
Prepare Paraffin Block/Slides [j]	X		X	
Obtain Blood Sample for biomarkers [k] **	X		X	
Perform Electrocardiogram [l]	X		X	
Perform Echocardiogram [l]	X			
Efficacy Assessments				
Evaluate Tumor Response RECIST Criteria [m]	X		X	
Clinical Drug Supplies				
Premed Before Infusion [n]		X		

a. Evaluate subjects who discontinue the protocol for reasons other than disease progression every 4 weeks until progression or until they receive additional anti-cancer therapy.

b. Karnofsky Performance Status of =/> 70% is required for study entry.

c. Document signs and symptoms and adverse events at least every 4 weeks until all study drug-related toxicities resolve, stabilize, return to baseline, or are deemed irreversible.

d. Pregnancy tests (serum or urine) should be performed within 72 hours of study start, or whenever pregnancy is suspected.

e. Mammogram and ultrasound should have been performed any time within 90 days prior to beginning of the study treatment and within 3 to 4 weeks of the neoadjuvant therapy completion before surgery. *MRI (magnetic resonance imaging) should be performed only if requested by the physician.

f. Laboratory tests comprise albumin, alkaline phosphatase, ALT, AST, total bilirubin, blood urea nitrogen, calcium, chloride, creatinine, glucose, lactic dehydrogenase, magnesium (at baseline only for the ixabepilone alone arm), potassium, phosphorus, and sodium, as well as complete blood count (Hemoglobin, Hematocrit, RBC, WBC, Platelets) with differential (Neutrophils, Lymphocytes, Monocytes, Eosinophils, Basophils). CrCL "CrCl = [(140 - age) x IBW] / (Scr x 72) (x 0.85 for females)" before every cycle for patients with an abnormal creatinine.

g. Perform laboratory tests within 72 hours of beginning each cycle. Perform baseline laboratory tests within four (4) weeks before administration of study drug

h. Perform complete blood count (Hemoglobin, Hematocrit, RBC, WBC, Platelets) with differential (Neutrophils, Lymphocytes, Monocytes, Eosinophils, Basophils) at week 1 and week 2 for the first 4 cycles and thereafter at the expected time of nadir (lowest count of any type of blood cell).

i. Laboratory tests comprise of calcium, magnesium, and potassium before every dose of cetuximab therapy and for at least 8 weeks following the completion of cetuximab. Replete electrolytes as necessary.

j. Send tissue slides to the central laboratory (see lab manual). Collected at baseline, and at the time of surgery [UPTEN, EGFR and EGFR-pathway associated genes (TGF-a, CRYAB, NRAS, KRAS, AKT3, PTEN, MEK1 and KRAS amplicon genes). Additional exploratory biomarkers may include (but are not limited to) beta tubulin isoforms, ABC transporters, TACC3, HCAPG, GTSE1, Kallikreins and defined gene models associated with ixabepilone response. Tumor biomarkers will be assessed on a formalin-fixed, paraffin-embedded (TFFPET) tumor sample from a previous biopsy (baseline) and from the surgical specimen. A minimum of 15 slides will be collected at each time.

k. Serum sample for biomarkers expression at baseline, and at the time of surgery. ** Optional (Serum sample for biomarkers expression will be done at later time).

l. A 12-lead ECG with QT measurement should be performed at screening, at the end of study/early termination, and whenever medically indicated. An Echocardiogram should be performed at screening only and whenever medically indicated.

m. Evaluate tumor response using RECIST criteria before chemotherapy and before surgery (from randomization to disease progression or until surgery).

n. Subjects who are to receive ixabepilone are to be premedicated approximately 1 hour before the infusion with both an oral H1 blocker (diphenhydramine 50 mg or equivalent) and an oral H2 blocker (eg, ranitidine 150-300 mg or cimetidine 300–800 mg or nizatidine 150-300 mg or famotidine 20–40 mg); premedication with steroids is indicated in subjects who developed a hypersensitivity reaction to ixabepilone in previous cycles. Subjects who are to receive cetuximab are to be premedicated with an H1 antagonist (e.g., 50 mg of diphenhydramine IV)

o. Monitor the subject for hypersensitivity reactions during and after all infusions.

2.1 Study Procedures by Visit and Treatment Cycle

Laboratory analyses and clinical examinations are to be repeated for a total treatment duration time of 4 to 6 months. Radiologic examinations are to be performed before chemotherapy and before surgery (from randomization to disease progression or until surgery).

The maximum number of treatment cycles allowed is 4.

The sequence of events for up to 4 cycles is described below.

2.2 Screening/Baseline visit

Informed consent must be obtained before performing any study-related procedures. Screening procedures may be done up to 4 weeks prior the start of neo-adjuvant therapy. Screening evaluation will include blood work, pregnancy testing, medical history, physical exam (including tumor measurements of the breast and lymph node, height, weight, blood pressure, heart rate and temperature) electrocardiogram, and echocardiogram. Tumor measurements will be recorded as the largest dimension in centimeters. Medical history should include relevant underlying conditions and concomitant medications. Baseline signs and symptoms are to be recorded and followed throughout the trial. A mammography and ultrasound will be performed to assess disease status.

Prior to initiation of neo-adjuvant therapy, a core tumor breast biopsy and blood sample will be obtained for baseline biomarkers expression studies.

If a patient entered the study (i.e. signed informed consent) prior to histological confirmation of breast cancer, this biopsy may serve the dual purpose of confirming the diagnosis and obtaining tissue for biomarkers research.

Following confirmation of eligibility, a blocked randomization will be used to randomize patients to either arm 1 or arm 2. A master sequence of randomization numbers for each arm will be generated.

For those study patients who voluntarily agree to have an optional serum samples taken for biomarkers expression; blood samples will be stored and the biomarkers expression will be done at later time (see lab manual). Patients will be informed that they will not be excluded from the clinical study if they do not wish to participate in the (blood samples) biomarkers study.

•The biomarkers blood sample must be collected after a patient number has been obtained. (see CA163157 Procedure Manual).

• A minimum of 15 tissue slides from the tumor breast biopsy or lymph nodes (when available) will be collected at baseline and send to the central laboratory. Note: please collect all tissue slides (15 at baseline and 15 after surgery) at the site before send it out to the central lab (see lab manual)

2.2.1 Radiological Evaluation Visit

• Perform mammogram and ultrasound of the breast and nodal region to determine baseline tumor status using RECIST criteria. This radiological evaluation should be completed within 90 days before study drug administration. If more than 90 days have elapsed since the baseline radiological evaluation, the mammogram and ultrasound must be repeated before the initial infusion of study drug is administered. If, at any time, is suspected progression diseases a complete radiological evaluation must be performed.

MRI (magnetic resonance imaging), only if requested by physician).

2.3 Cycle 1 Clinic Visit

• Following confirmation of eligibility, a blocked randomization will be used to randomize patients to either arm 1 or arm 2. A master sequence of randomization numbers for each arm will be generated (see CA163157 Protocol Manual). Once the subject is randomized, administration of study drug must begin within 72 hours. NOTE: If more than 72 hours have elapsed between the blood sampling for laboratory analysis and administration of ixabepilone or cetuximab, then the analyses must be repeated. (Except for cycle 1 for which baseline labs obtained within 2 weeks are accepted.)

• laboratory tests within 72 hours of beginning each cycle comprise albumin, alkaline phosphatase, ALT, AST, total bilirubin, blood urea nitrogen, calcium, chloride, creatinine, glucose, lactic dehydrogenase, magnesium (at baseline only for the ixabepilone alone arm), potassium, phosphorus, and sodium, as well as complete blood count (Hemoglobin, Hematocrit, RBC, WBC, Platelets) with differential (Neutrophils, Lymphocytes, Monocytes, Eosinophils, Basophils).

CrCL "CrCl = [(140 - age) x IBW] / (Scr x 72) (x 0.85 for females)" before every cycle for patients with an abnormal creatinine.

• Laboratory tests comprise of calcium, magnesium, and potassium before every dose of cetuximab therapy (before every cycle for the ixabepilone plus cetuximab arm).

• Perform a physical examination; record weight and height, karnofsky performance scale, document vital signs. Assess signs and symptoms and document baseline adverse events.

• Subjects who are to receive ixabepilone are to be premedicated approximately 1 hour before the infusion with either 1)both an oral H1 blocker (diphenhydramine 50 mg or equivalent) and an oral H2 blocker (eg, ranitidine 150-300 mg or cimetidine 300–800 mg or nizatidine 150-300 mg or famotidine 20–40 mg); or 2) with premedication per institutional standards. Premedication with steroids is indicated in subjects who developed a hypersensitivity reaction to ixabepilone in previous cycles. Subjects who are to receive cetuximab are to be premedicated with an H_1 antagonist (e.g., 50 mg of diphenhydramine IV or equivalent)

• Infuse ixabepilone or ixabepilone plus cetuximab accordingly with the randomization arm.

2.3.1 Cycle 1 Post-Infusion Laboratory Visits

• Perform platelet count and complete blood count with differential at week 1 (+/- 3 days) and week 2(+/- 3 days) for the first 4 cycles and thereafter at the expected time of nadir (lowest count of any type of blood cell).

2.3.2 Cycle 2 Pre-Infusion Laboratory Visit

• Within 72 hours before beginning the Cycle 2 infusion, obtain blood sample for analysis of albumin, alkaline phosphatase, ALT, AST, total bilirubin, blood urea nitrogen, calcium, chloride, creatinine, glucose, lactic dehydrogenase, potassium, phosphorus, and sodium, as well as complete blood count (Hemoglobin, Hematocrit, RBC, WBC, Platelets) with differential (Neutrophils, Lymphocytes, Monocytes, Eosinophils, Basophils).

CrCL "CrCl = [(140 - age) x IBW] / (Scr x 72) (x 0.85 for females)" before every cycle for patients with an abnormal creatinine.

• Laboratory tests comprise of calcium, magnesium, and potassium before every dose of cetuximab therapy (before every cycle for the ixabepilone plus cetuximab arm).

2.3.2.1 Cycle 2 Clinic Visit

• Perform a physical examination to include tumor measurements of the breast and lymph node; record weight and height, karnofsky performance scale, document vital signs. Assess signs and symptoms and document baseline adverse events.

• Subjects who are to receive ixabepilone are to be premedicated approximately 1 hour before the infusion with either 1) both an oral H1 blocker (diphenhydramine 50 mg or equivalent) and an oral H2 blocker (eg, ranitidine 150-300 mg or cimetidine 300–800 mg or nizatidine 150-300 mg or famotidine 20–40 mg); or 2) with premedication per institutional standards. Premedication with steroids is indicated in subjects who developed a hypersensitivity reaction to ixabepilone in previous cycles.

•Subjects who are to receive cetuximab are to be premedicated with an H_1 antagonist (e.g., 50 mg of diphenhydramine IV or equivalent).

• Infuse ixabepilone or ixabepilone plus cetuximab accordingly with the study arm.

2.3.2.2 Cycle 2 Post-Infusion Laboratory Visits

• Perform platelet count and complete blood count with differential at week 1(+/- 3 days) and week 2 (+/- 3 days) for the first 4 cycles and thereafter at the expected time of nadir (lowest count of any type of blood cell).

2.3.3 Cycle 3 Pre-Infusion Laboratory Visit

• Within 72 hours before beginning the Cycle 3 infusion, obtain blood sample for analysis of albumin, alkaline phosphatase, ALT, AST, total bilirubin, blood urea nitrogen, calcium, chloride, creatinine, glucose, lactic dehydrogenase, potassium, phosphorus, and sodium, as well as complete blood count (Hemoglobin,

Hematocrit, RBC, WBC, Platelets) with differential (Neutrophils, Lymphocytes, Monocytes, Eosinophils, Basophils).

CrCL "CrCl = [(140 - age) x IBW] / (Scr x 72) (x 0.85 for females)" before every cycle for patients with an abnormal creatinine.

• Laboratory tests comprise of calcium, magnesium, and potassium before every dose of cetuximab therapy (before every cycle for the ixabepilone plus cetuximab arm).

2.3.3.1 Cycle 3 Clinic Visit

• Perform a physical examination to include tumor measurements of the breast and lymph node; record weight and height, karnofsky performance scale, document vital signs. Assess signs and symptoms and document baseline adverse events.

• Subjects who are to receive ixabepilone are to be premedicated approximately 1 hour before the infusion with either 1) both an oral H1 blocker (diphenhydramine 50 mg or equivalent) and an oral H2 blocker (eg, ranitidine 150-300 mg or cimetidine 300–800 mg or nizatidine 150-300 mg or famotidine 20–40 mg); or 2) with premedication per institutional standards. Premedication with steroids is indicated in subjects who developed a hypersensitivity reaction to ixabepilone in previous cycles.

•Subjects who are to receive cetuximab are to be premedicated with an H_1 antagonist (e.g., 50 mg of diphenhydramine IV or equivalent).

• Infuse ixabepilone or ixabepilone plus cetuximab accordingly with the study arm.

2.3.3.2 Cycle 3 Post-Infusion Laboratory Visits

• Perform platelet count and complete blood count with differential at week 1(+/- 3 days) and week 2 (+/- 3 days) for the first 4 cycles and thereafter at the expected time of nadir (lowest count of any type of blood cell).

2.3.4 Cycle 4 Pre-Infusion Laboratory Visit

• Within 72 hours before beginning the Cycle 4 infusion, obtain blood sample for analysis of albumin, alkaline phosphatase, ALT, AST, total bilirubin, blood urea nitrogen, calcium, chloride, creatinine, glucose, lactic dehydrogenase, potassium,

phosphorus, and sodium, as well as complete blood count (Hemoglobin, Hematocrit, RBC, WBC, Platelets) with differential (Neutrophils, Lymphocytes, Monocytes, Eosinophils, Basophils).

CrCL "CrCl = [(140 - age) x IBW] / (Scr x 72) (x 0.85 for females)" before every cycle for patients with an abnormal creatinine.

• Laboratory tests comprise of calcium, magnesium, and potassium before every dose of cetuximab therapy (before every cycle for the ixabepilone plus cetuximab arm).

2.3.4.1 Cycle 4 Clinic Visit

• Perform a physical examination to include tumor measurements of the breast and lymph node; record weight and height, karnofsky performance scale, document vital signs. Assess signs and symptoms and document baseline adverse events.

• Subjects who are to receive ixabepilone are to be premedicated approximately 1 hour before the infusion with either 1)both an oral H1 blocker (diphenhydramine 50 mg or equivalent) and an oral H2 blocker (eg, ranitidine 150-300 mg or cimetidine 300–800 mg or nizatidine 150-300 mg or famotidine 20–40 mg); or 2) with premedication per institutional standards. Premedication with steroids is indicated in subjects who developed a hypersensitivity reaction to ixabepilone in previous cycles.

•Subjects who are to receive cetuximab are to be premedicated with an H_1 antagonist (e.g., 50 mg of diphenhydramine IV or equivalent).

• Infuse ixabepilone or ixabepilone plus cetuximab accordingly with the study arm.

2.3.4.2 Cycle 4 Post-Infusion Laboratory Visits

• Perform platelet count and complete blood count with differential at week 1(+/- 3 days) and week 2 (+/- 3 days) for the first 4 cycles and thereafter at the expected time of nadir (lowest count of any type of blood cell).

2.4 End-of-Treatment/Early Termination Visit

Within 3 - 4 weeks of completing cycle 4 or one (1) week after the patient has withdrawn from the study for any reason other than completed treatment.

• Obtain blood sample for analysis of albumin, alkaline phosphatase, ALT, AST, total bilirubin, blood urea nitrogen, calcium, chloride, creatinine, glucose, lactic dehydrogenase, potassium, phosphorus, and sodium, as well as complete blood count (Hemoglobin, Hematocrit, RBC, WBC, Platelets) with differential (Neutrophils, Lymphocytes, Monocytes, Eosinophils, Basophils) .
CrCL "CrCl = [(140 - age) x IBW] / (Scr x 72) (x 0.85 for females)" before every cycle for patients with an abnormal creatinine.
• Laboratory tests comprise of calcium, magnesium, and potassium for at least 8 weeks following the completion of cetuximab therapy. Replete electrolytes as necessary.
• Perform a physical examination to include tumor measurements of the breast and lymph node; record weight and height, karnofsky performance scale, document vital signs. Assess signs and symptoms and document baseline adverse events.
• For those study patients who voluntarily agree to have an optional serum samples taken for biomarkers expression; blood samples will be stored and the biomarkers expression will be done at later time (see lab manual). Patients will be informed that they will not be excluded from the clinical study if they do not wish to participate in the (blood samples) biomarkers study.
• Perform electrocardiogram at the end of the study/early termination visit.
• Perform mammogram and ultrasound of breast and nodal region to determine tumor status using RECIST criteria.

2.5 Post-neoadjuvant Surgical Intervention

Within 3 - 4 weeks of completing neoadjuvant therapy, patients will undergo surgery of the breast and axilla according to local guidelines. Patients will remain in the study throughout the neoadjuvant therapy, regardless of type, until the completion of surgical intervention and the collection of the post-neoadjuvant therapy biopsy. Portions of tumor tissue from the primary site, or lymph nodes (when available), will be obtained for post treatment biomarkers expression studies.

Note: please collect all tissue slides (15 at baseline and 15 after surgery) at the site before send it out to the central lab (see lab manual)

2.6 End of Study Assessments/ Follow-up

Vital signs including blood pressure, heart rate and temperature will be performed during this visit. Toxicity assessment will be continuous throughout the study. CTC Version 3.0 (dated June 10, 2003) will be used to grade toxicities. For the patients on the ixabepilone plus cetuximab arm the labs will be done for at least 8 weeks and will include calcium, magnesium, and potassium following the completion of cetuximab therapy. Replete electrolytes as necessary.

End of study visit will occur approximately 30 days after surgery. The patients on the ixabepilone arm with continuing diagnosis of toxicity related to neoadjuvant chemotherapy will be seen by the Investigator at least every 4 weeks until all study drug related toxicities resolve, stabilize, return to baseline or are deemed irreversible as judged by the investigator. The patients on the ixabepilone plus cetuximab arm with continuing diagnosis of toxicity related to neoadjuvant chemotherapy will be seen by the Investigator at least every week (eight (8) weeks total) until all study drug related toxicities resolve, stabilize, return to baseline or are deemed irreversible as judged by the investigator.

2.7 Details of Procedure

2.7.1 Safety Assessments

Medical history will include relevant underlying conditions. Baseline signs and symptoms are to be recorded and followed throughout the trial. Toxicity assessments will be continuous during treatment and in post-treatment follow-up and occur every 4 weeks until all study drug related toxicities resolve, stabilize, return to baseline or are deemed irreversible as judged by the investigator. CTC Version 3.0 (June 10, 2003) will be used to grade toxicities. Vital signs including blood pressure, heart rate and temperature should be performed at each physical exam. Perform baseline laboratory tests within two (2) weeks before administration of study drug, baseline serum chemistries (lactate dehydrogenase (LDH), glucose, electrolytes, albumin, blood urea nitrogen (BUN), creatinine,

alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase, total bilirubin, magnesium, calcium and phosphorus) and hematology (Complete Blood Count (CBC) + differential + platelets) will be done at screening. Additionally, CBC + differential and platelets and serum chemistry panel must be performed within 72 hours prior to each subsequent chemotherapy cycle.

Laboratory tests may be done more frequently if medically indicated.

If CTC Grade 4 hematologic toxicity is seen, repeat CBC + differential + platelets should be repeated every 3 - 4 days until recovery.

2.7.2 Efficacy Assessments

A clinical assessment as well as mammography and ultrasound will be performed to assess disease status at baseline.

Clinical tumor response in the primary site and in the axilla will be assessed after each cycle of treatment and prior to the next treatment by physical exam. Three to four weeks after the last treatment with the study drugs patients will be assessed for disease status via mammography and ultrasound as well as clinical assessment. Disease progression suspected from clinical tumor response assessments during any cycle should be confirmed by mammography and ultrasound.

3.0 Biomarkers Research Assessment

3.0.1 Biomarkers Exploratory Research Tumor Tissue Collection

Two samples of tumor tissue will be collected during the course of the study. The first sample will be collected prior to treatment with the study drug and the second at the time of the surgery. These samples will be used to obtain mRNA for analysis of tumor gene expression. Tumor biomarkers will be assessed on a formalin-fixed, paraffin-embedded (FFPE) tumor sample from a previous biopsy (baseline) and from the surgical specimen. A minimum of 15 slides will be collected at each time. Details for processing, storing and shipping these samples will be provided by BMS in the CA163157 Procedure Manual.

3.0.2 Biomarkers Exploratory Research Blood Collection

This procedure is optional (Serum sample for biomarkers expression will be done at later time).

One venous blood sample of 2 mLs will be collected. The specimen will be obtained prior to treatment with the study drugs. This sample will be used to obtain mRNA for analysis of gene expression. Details for collection, processing, storing and shipping these samples will be provided in the CA163157 Procedure Manual.

3.0.3 Biomarkers Exploratory Research Tumor Tissue Collection

Any patient, who switches to another neoadjuvant therapy prior to surgery, will be asked to undergo an additional breast biopsy. This breast biopsy is optional and will require an additional consent. The biopsy must be obtained within 21 days after last dose of study drug and prior to next therapeutic regimen.

Details for processing storing and shipping these samples will be provided by BMS in the CA163157 Procedure Manual.

4.0 Introduction, Background and Rationale

Breast cancer is the most common cancer in women. It kills 376,000 women a year worldwide, and there are about 900,000 new cases annually. Estimates for 2007 indicate that breast cancer will cause 180,510 cases of new cancers in the United States, accounting for 26% of new cancer cases in women and 40,460 cancer deaths. Approximately 50% of women are diagnosed with localized disease. Although many of these women will have an initial good response to treatment, 50% to 75% of those diagnosed with early disease will eventually relapse. Metastatic breast cancer is largely incurable using conventional therapy, with a median survival of 2 to 3 years from manifestation of metastases. [1]

4.0.1 Current Chemotherapy

Many chemotherapeutic agents have been demonstrated to be active in MBC, including alkylating agents, anti-metabolites, anthracyclines, vinca alkaloids,

podophyllotoxins, and taxanes. Anthracyclines and taxanes are generally considered the most active cytotoxic drug classes in breast cancer. As a result, both classes of agents are commonly used in the metastatic as well as the adjuvant setting. Docetaxel and paclitaxel, alone or in combination with other chemotherapeutic agents, have shown significant response rates in first- or second-line treatment of MBC. As second-line monotherapy, taxanes have had response rates in the range of 28% to 55%. Despite high activity, however, relapse is common and long-term disease control is rare. Additionally, many subjects fail to respond to taxanes and anthracyclines. Options are limited for subjects who have failed prior treatment with both an anthracycline and a taxane. Capecitabine alone , and in combination with ixabepilone are currently the only chemotherapy agents specifically approved in regions around the world for the treatment of metastatic breast cancer in subjects who have failed or are resistant to both an anthracycline and a taxane.[2,3,4]

4.0.2 Epothilones

The epothilone are a new class of non-taxane tubulin polymerization agents obtained by fermentation of the myxobacterium *Sorangium cellulosum*.[5] The chief components of the fermentation process are epothilones A and B. In 1994, the National Cancer Institute discovered that the epothilones possess potent cytotoxic activity. The cytotoxic activities of the epothilones, like those of the taxanes, have been linked to stabilization of microtubules, which results in mitotic arrest at the G2/M transition.[6,7] Importantly, the epothilones are active against various taxane-resistant cell lines, including those with overexpression of the multidrug resistance gene and BIII tubulin isoforms, or have tubulin mutations. Ixabepilone is a semi-synthetic derivative of the natural product epothilone B specifically designed to overcome the metabolic instability of the natural product as demonstrated in preclinical models. Similar to paclitaxel, ixabepilone blocks cells in the mitotic phase of the cell division cycle and is a highly potent cytotoxic agent capable of killing cancer cells at low nanomolar concentrations.

An important feature of ixabepilone is its antitumor activity in a broad range of taxane sensitive and taxane-resistant preclinical human tumor models. Phase 1

studies of ixabepilone have demonstrated antitumor activity in cancers of the ovary, cervix, colon, stomach, and breast, as well as in melanoma, non-small-cell lung carcinoma, and non-Hodgkin's lymphoma. Phase 2 studies of monotherapy ixabepilone have demonstrated notable activity in subjects with both taxane-sensitive and taxane-resistant breast cancer as well as in other tumor types. A Phase 1/2 study of ixabepilone plus capecitabine has established that the combination is active in subjects who have failed prior anthracycline and taxane and that the two agents can be safely administered together, each at efficacious doses. Subset analysis from the recently completed CA163046 study showed that subjects with HER2 (ErbB2) positive metastatic breast cancer previously treated with a taxane and an anthracycline benefit from ixabepilone plus capecitabine over capecitabine alone.

Preclinical Metabolism and Potential for Drug Interactions

In vitro studies suggest that CYP3A4 is the CYP isozyme primarily responsible for the oxidative metabolism of ixabepilone in humans. Therefore, coadministration of inhibitors of CYP3A4 may affect the pharmacokinetics of ixabepilone. In vitro studies also suggest that ixabepilone is a weak inhibitor of human CYP3A4. Given that ixabepilone is administered on an intermittent schedule (q21 days), is a weak inhibitor of CYP3A4 in vitro (IC50 of approximately 3700 ng/mL), and typically has peak plasma concentrations of less than 400 ng/mL when administered as a 3-hour infusion, there is a low likelihood of a clinically important effect on the pharmacokinetics of coadministered medications that are dependent on CYP3A4 metabolism.

Phase 1 Ixabepilone Monotherapy Studies

To date, approximately 2700 subjects have been treated with ixabepilone in the Bristol-Myers Squibb (BMS)-sponsored Phase 1, Phase 2, and Phase 3 studies.[8] Ixabepilone is also being independently evaluated in studies conducted by the National Cancer Institute Cancer Therapy Evaluation Program.

BMS has sponsored two Phase 1 studies of ixabepilone monotherapy, which were designed to establish a recommended dose for Phase 2 studies. CA163-001 evaluated ixabepilone administered intravenously (IV) on Day 1 of a 21-day

cycle (q3 weeks). CA163-002 evaluated ixabepilone administered IV on a weekly schedule. CA163-001 is summarized below; CA163-002 is summarized in the Investigator Brochure.[8] (Of note, the National Cancer Institute has also sponsored Phase 1 monotherapy studies, which evaluated a variety of schedules, including a daily x 5 days q21-day schedule). CA163-001 was an open-label dose-escalation study of ixabepilone administered once every 3 weeks as a 1-hour infusion in subjects with advanced malignancies. Sixty-one (61) subjects were treated at dose levels of 7.4, 15, 30, 50, 57, and 65 mg/m^2. At doses below 50 mg/m^2, no dose-limiting toxicities (DLT) were observed. First-course DLT was experienced by 2 out of 3 subjects at 65 mg/m^2 (Grade 3 neuropathy and prolonged Grade 4 neutropenia; prolonged Grade 4 neutropenia) and by all 3 subjects at 57 mg/m^2 (Grade 3 arthralgia and myalgia in 2 subjects; Grade 4 neutropenia with pneumococcal sepsis and death in 1 subject). After the maximum tolerated dose was established as 50 mg/m^2, a cohort of 10 additional subjects were treated at this dose. Severe toxicity requiring a dose reduction was observed in only 1 of these subjects (febrile neutropenia). Other toxicities included fatigue, weakness, constipation, diarrhea, nausea, vomiting, rash, alopecia, and low-grade neuropathy. Antitumor activity seen among 22 subjects treated at 50 mg/m^2 included a complete response in ovarian cancer and partial responses in non-small-cell lung cancer and melanoma. Due to cumulative neuropathy observed with the 1-hour schedule, this study was expanded at doses of 40 and 50 mg/m^2 administered as a 3-hour infusion. As of July 2002, data were available for 12 subjects treated at 40 mg/m^2 over 3 hours for a total of 35 cycles. The most notable toxicity was neutropenia. Grade 4 neutropenia was observed in 2 subjects (17%) but no episodes of febrile neutropenia were reported. Sensory or painful peripheral neuropathy of any grade was reported in 4 (33%) subjects, all Grade 1. There were no treatment discontinuations due to drug toxicity in this dose cohort. H1 and H2 histamine blockers were introduced as routine premedication after hypersensitivity reactions (HSRs) were reported early in the Phase 1 program. No subsequent HSRs were reported in the study. Other common safety events included

myalgias (8%) and stomatitis/pharyngitis (8%). No deaths occurred on-study that were deemed related to study drug. One partial response was reported in this dose cohort in a patient with ovarian cancer.

CA163-031: A Combined Phase 1/Phase 2 Study

CA163-031 was an open-label, multicenter, dose-escalation study of the combination of ixabepilone and capecitabine for the treatment of taxane- and anthracycline-pretreated MBC. The primary objective of the study was to determine the recommended Phase 2 and Phase 3 doses of ixabepilone and capecitabine. However, once safety and preliminary efficacy data identified ixabepilone 40 mg/m^{2}plus capecitabine 2000 mg/m^2 as the recommended dose, the protocol was amended to enroll additional subjects at this dose to estimate overall response rate. [9]

CA163-031 Efficacy Summary

No objective responses were observed among the 6 subjects in the ixabepilone 8 mg/m^2 + capecitabine 1650 mg/m^{2}dose cohort. One partial response was observed among the 6 subjects in the ixabepilone 10 mg/m^2 + capecitabine 1650 mg/m^2 dose cohort. In the second phase of the study, 62 subjects were treated with the doses selected in the first phase: 40 mg/m^2 ixabepilone, given on Day 1 of each cycle, and 1000 mg/m^2 capecitabine, administered twice daily (total daily dose: 2000 mg/m^2) on Days 1–14 of each cycle. Of these 62 subjects, 50 were evaluable for response. The objective response rate was 30% (95% CI: 17.9%, 44.6%), including 1 complete response (2%) and 14 partial responses (28%). Stable disease was observed in 16 (32%) subjects and progressive disease in 14 (28%). Median time to response was 6.0 weeks; duration of response was 6.9 months; and progression-free survival was 3.8 months. The overall response rate among subjects with ER-/PR-/HER2-negative tumors (n=22) was 23%. [9]

CA163-031 Safety Summary (Ixabepilone 40 mg/m^2 + Capecitabine 2000 mg/m^2 Dose Cohort)

A total of 62 subjects received the recommended Phase 2/Phase 3 dose of ixabepilone 40 mg/m^2 plus capecitabine 2000 mg/m^2. Of the 30 subjects who received this dose in the Phase 1 segment, 2 developed dose-limiting toxicity of

Grade 3 hand-foot syndrome. Two of the 62 subjects (3%) in this dose cohort died on study or within 30 days of the last dose of study drug. One death was due to disease progression; the other was due to septic shock that was judged to be probably related to study drug. Fifteen subjects (24%) in this dose cohort had at least 1 serious adverse event (SAE). SAEs that occurred in more than 1 subject were nausea (4 subjects), vomiting (4 subjects), neutropenia (2 subjects), stomatitis (2 subjects), dehydration (2 subjects), dyspnea (2 subjects), and convulsions (2 subjects). Most subjects (95%) in this dose cohort had at least 1 treatment-related adverse event (TRAE). The most frequently reported TRAEs of any grade were fatigue (79%), nausea (74%), hand-foot syndrome (63%), alopecia (63%), myalgia (60%), paresthesia (55%), and diarrhea (52%). Treatment-related neutropenia was reported in 4 (7%) subjects. [9]

CA163-031 Safety Summary (Other Dose Cohorts)

Of the 6 subjects in the ixabepilone 8 mg/m^2+ capecitabine 1650 mg/m^2 dose cohort, none died and 1 subject (17%) developed an SAE of pyrexia. TRAEs that occurred in ≥ 16.7% of subjects were these: paresthesia (67%), alopecia (50%), erythrodysesthesia syndrome (50%), diarrhea (50%), nausea (50%), vomiting (50%), constipation (33%), myalgia (33%), headache (33%), neuropathic pain (33%), and rash (33%). The following TRAEs occurred in 16.7% of subjects: stomatitis, abdominal discomfort, abdominal pain, upper abdominal pain, sensitivity of teeth, fatigue, injection site irritation, pyrexia, hepatic pain, pharyngitis, skin infection, anorexia, arthralgia, back pain, muscle spasm, pain in extremity, peripheral neuropathy, peripheral sensory neuropathy, cough, respiratory tract congestion, palmar-plantar erythrodysesthesia syndrome, increased lacrimation, diarrhea, nail disorder, skin discoloration, hot flush, and orthostatic hypotension. Of the 6 subjects in the ixabepilone 10 mg/m^2 + capecitabine 1650 mg/m^2 dose cohort, none died and 1 subject each (17%) developed an SAE of abdominal pain, nausea, or deep-vein thrombosis. TRAEs that occurred in ≥ 16.7% of subjects were these: alopecia (83%), fatigue (83%), myalgia (83%), diarrhea (83%), nausea (83%), paresthesia (67%), palmar-plantar erythrodysesthesia syndrome (67%), headache (50%), constipation

(50%), dyspepsia (50%), stomatitis (50%), increased lacrimation (50%), abdominal pain (33%), upper abdominal pain (33%), and vomiting (33%). The following TRAEs occurred in 16.7% of subjects: pyrexia, catheter site infection, nasopharyngitis, upper respiratory tract infection, anorexia, decreased appetite, vulvovaginal discomfort, blister, nail disorder, rash, and skin exfoliation. Of the 30 subjects in the ixabepilone 40 mg/m^2 + capecitabine 1650 mg/m^2 dose cohort, 5 died: 3 of disease progression, 1 of pulmonary embolism, and 1 of pneumonia and terminal cardiovascular dysfunction. The SAEs of neutropenia and dehydration each occurred in 7% of subjects.

The following SAEs occurred in 3.3% of subjects: febrile neutropenia, stomatitis, vomiting, fatigue, noncardiac chest pain, pyrexia, urinary tract infection, increased AST, and decreased platelet count.

TRAEs that occurred in ≥ 7% of subjects were these: alopecia (70%), nausea (70%), paresthesia (67%), palmar-plantar erythrodysesthesia syndrome (67%), vomiting (53%), diarrhea (57%), myalgia (53%), constipation (37%), stomatitis (37%), peripheral neuropathy (37%), arthralgia (23%), rash (23%), increased lacrimation (23%), dyspnea (20%), dizziness (20%), nail disorder (20%), fatigue (17%), peripheral sensory neuropathy (17%), dehydration (13%), decreased appetite (13%), dysgeusia (13%), neuropathic pain (13%), hypoesthesia (13%), anorexia (10%), and cough (10%). The following TRAEs occurred in 7% of subjects: neutropenia, oral hypoesthesia, retching, pyrexia, pain, decreased weight, joint swelling, shoulder pain, headache, ageusia, pruritus, skin discoloration, and hot flush. [9]

CA163-009: Phase 2 Study in Taxane-Resistant MBC

CA163-009 was an international multi-institution study conducted in subjects with MBC who were resistant to paclitaxel or docetaxel. Ixabepilone was administered on Day 1 of an every-21-day cycle. To be eligible, subjects must have had tumor progression within 4 months of their last dose of a taxane-containing regimen if administered in the metastatic setting, or within 6 months of taxane therapy if administered in the adjuvant setting. The taxane regimen must have been the last regimen received by the subject. Subjects were required to have

dimensionally measurable disease and Eastern Cooperative Oncology Group (ECOG) performance status of 0-1. At the onset of the study, ixabepilone was administered as a 50 mg/m^2 infusion over 1 hour, repeated every 21 days. Due to the severity of peripheral neuropathy observed in the first subjects enrolled in this study, as well as in the CA163-010 study, the infusion duration was extended to 3 hours. The dose of ixabepilone was then reduced to 40 mg/m^{2}due to excessive myelosuppression and mucositis observed in another study in subjects treated with the 50 mg/m^2 over 3-hour regimen.

A total of 49 subjects were treated. Median age was 54 years. Results in evaluable subjects were Partial Response: 6 (12%); Stable Disease: 20 (41%); and Progressive Disease: 21 (43%).[10]

CA163-010: Phase 2 Study in Taxane-Sensitive MBC

CA163-010 was a multinational, multicenter, nonrandomized, open-label study of ixabepilone in subjects with taxane-sensitive MBC. Ixabepilone was administered on Day 1 of an every-21-day cycle. Subjects must have received an anthracycline in the adjuvant setting but no prior therapy for metastatic disease. Prior taxane therapy in the adjuvant setting was permitted provided treatment was greater than 1 year before enrollment. Subjects had to have at least one bi-dimensionally measurable lesion and an ECOG performance status of 0-1. At the onset of the study, ixabepilone was administered as a 50 mg/m^2 infusion over 1 hour, repeated every 21 days. Due to the severity of peripheral neuropathy observed in the first subjects enrolled in this study, as well as in CA163-009, the infusion duration was extended to 3 hours. The dose of ixabepilone was then reduced to 40 mg/m^{2}due to excessive myelosuppression and mucositis observed in another study in subjects treated with the 50 mg/m^2 over 3-hour regimen. A total of 65 subjects were treated. Median age was 52 years. Results in evaluable subjects were Partial Response: 27 (42%); Stable Disease: 23 (35%); and Progressive Disease: 13 (20%).[11]

CA163-080: Phase 2 Study in Treatment-Naive MBC

CA163-080 was a single-arm, open-label, multicenter, multinational study of ixabepilone as neoadjuvant therapy in treatment-naive MBC. Ixabepilone was

given in a dose of 40 mg/m^2 as a 3-hour IV infusion every 3 weeks. A total of 161 subjects were treated. Median age was 55 years. Results in evaluable subjects were Primary Best Overall Response Rate (Complete Response [CR] or Partial Response [PR]): 98/161 (61%); Pathologic Complete Response in Breast [pCR_B]: 29/161 (18%); and Pathologic Complete Response in Breast and Lymph Nodes [pCR_{BL}]: 17/161 (11%). Independent assessment of subject responses yielded pCR_B and pCR_{BL} rates of 14% and 9%, respectively. In subjects with ER-negative, ER-/PR-negative, and triple-negative tumors, as assessed by locally read immunohistochemistry (IHC) and fluorescence in situ hybridization (FISH), investigator-assessed pCR_B rates were 29%, 33%, and 26%, respectively. The pCR_{BL} rates were 19%, 23%, and 19%, respectively.

No deaths were reported. Twelve (8%) subjects reported SAEs. Among these subjects, 10 (6%) reported at least 1 SAE assessed as possibly, probably, or certainly related to ixabepilone therapy. These SAEs included peripheral or sensory neuropathy, febrile neutropenia, neutropenia, vomiting, and hypersensitivity, each reported in 2 subjects; and mucositis, constipation, diarrhea, stomatitis, lymphopenia, leukopenia, thrombocytopenia, increased ALT, and increased AST, each reported in 1 subject.

Treatment-related adverse events were reported in 97% of subjects. The most common (≥ 15% of subjects) were alopecia (86%), myalgia (50%), arthralgia (30%), asthenia (27%), neutropenia (21%), peripheral sensory neuropathy (20%), nausea (19%), fatigue (19%), and diarrhea (15%). Most were Grade 1 to 2. Grade 3/4 treatment-related adverse events were reported in 25% of subjects. The most common Grade 3/4 event was neutropenia (14%).[12]

CA163-081: Phase 2 Study in Refractory MBC

CA163-081 was a single-arm, open-label, multicenter, multinational, two-stage design study of ixabepilone in MBC that was resistant to an anthracycline, a taxane, and capecitabine. A total of 126 subjects were treated. Data from this study have been published. Of the 126 treated subjects, 113 were evaluable for response. Subjects were heavily pretreated: 88% had received ≥ 2 lines of prior chemotherapy in the metastatic setting. Independent Radiology Facility (IRF)-

assessed objective response rate (ORR) was 11.5% (95% CI: 6.3%–18.9%) for response-evaluable subjects. Investigator-assessed ORR for all treated subjects was 18.3% (95% CI: 11.9%–26.1%). Half (50%) of subjects achieved stable disease (SD), with 14.3% achieving SD ≥ 6 months. Median duration of response and progression-free survival were 5.7 months and 3.1 months, respectively. Median overall survival was 8.6 months. Ixabepilone demonstrated activity in 5/42 IRF response-evaluable subjects (ORR: 12%) with triple-negative tumors (tumors that did not express ER, PR, or HER2). Subjects received a median of 4 treatment cycles (range 1-16), and 25% of subjects received ≥ 8 cycles. Grade 3-4 treatment-related events included peripheral sensory neuropathy (14%), fatigue/asthenia (13%), myalgia (8%), and stomatitis/mucositis (6%). Resolution of Grade 3-4 peripheral sensory neuropathy occurred after a median period of 5.4 weeks. Thus, ixabepilone demonstrated clear activity and a manageable safety profile in subjects with MBC resistant to anthracycline, taxane, and capecitabine. Responses were durable and notable in subjects who had not previously responded to multiple prior therapies. [13]

Phase 3 Metastatic Breast Cancer Studies: CA163-046 [14]and CA163-048[15]

Two Phase 3 open-label, randomized, multicenter, multinational studies in MBC have completed enrollment. CA163-046 has enrolled subjects with taxane-refractory MBC who have received prior treatment with or are refractory to an anthracycline. CA163-048 has enrolled subjects with MBC who have received prior treatment with an anthracycline and a taxane. Both studies have two treatment arms: ixabepilone plus capecitabine or capecitabine alone.

These results supported the use of ixabepilone plus capecitabine in two Phase 3 studies in patients with breast cancer. Studies CA163046 and CA163048 were randomized, multicenter, multinational studies comparing ixabepilone plus capecitabine versus capecitabine alone in patients with metastatic breast cancer. The primary endpoint of CA163046 was progression-free survival in patients previously treated with or resistant to an anthracycline and who are taxane resistant. The primary endpoint for CA163048 is overall survival in patients previously treated with an anthracycline and a taxane. [15]

Study 046, an international Phase 3 study, was designed to compare ixabepilone plus capecitabine with capecitabine alone in anthracycline-pretreated or -resistant and taxane-resistant locally advanced MBC. Participants were required to have measurable disease with tumors that progressed rapidly in the adjuvant or metastatic setting after receiving both anthracyclines and taxanes. Eight percent of patients were receiving first-line treatment, having relapsed within one year of prior anthracycline/taxane therapy in the adjuvant setting; treatment for the remaining 92% of patients was as either second- or third-line therapy. Of the 85% of patients with progression in the metastatic setting following prior taxane, 42% progressed on taxane therapy. A total of 737 patients received ixabepilone (40 mg/m^2 as a 3-hour intravenous (IV) infusion on Day 1 of a 21-day cycle) plus capecitabine (2000 mg/m^2 orally administered in two divided doses each on Days 1 through 14 of a 21-day cycle) (n=369) or capecitabine alone (2500 mg/m^2 on the same schedule as the same schedule) (n=368). The primary endpoint was progression free survival (PFS) evaluated by blinded Independent Radiology Review Committee (IRRC). The addition of ixabepilone to capecitabine resulted in a clinically meaningful and statistically robust improvement in PFS in patients with MBC resistant to anthracyclines and taxanes. The improvement in PFS by the addition of ixabepilone to capecitabine was consistent across subpopulations, including patients with visceral metastases, >2 sites of metastatic disease, ≥65 years of age, or with HER2+ breast cancer. [14]

CA163048 compared ixabepilone in combination with capecitabine to capecitabine alone in patients with metastatic or locally advanced breast cancer previously treated with anthracyclines and taxanes. A total of 1198 patients were treated: 595 patients received ixabepilone (40 mg/m^2 as a 3-hour intravenous (IV) infusion on Day 1 of a 21-day cycle) plus capecitabine (2000 mg/m^2 orally administered in two divided doses each on Days 1 through 14 of a 21-day cycle) and 603 patients received capecitabine alone (2500 mg/m^2 on the same schedule). In this study overall survival (OS) was the primary endpoint. Results demonstrated statistically significant and clinically meaningful superiority in PFS and improved RR over capecitabine alone that translated into a modest

improvement in OS favoring the combination which did not meet statistical significance. [15]

Both Phase 3 studies showed a statistically significant and clinically meaningful improvement in PFS in favor of the combination of ixabepilone and capecitabine over capecitabine alone. Results of these studies showed an improvement in median PFS of 1.6 to 1.8 months and a 21 to 25% reduction in the risk of progression. The benefit in PFS was supported by significant improvements in objective response rate (ORR) in favor of the combination, including a more than doubling of the ORR in CA163046 (35% vs 14%) as assessed by the IRRC. The analysis of OS in both studies showed a 0.9 hazard ratio (HR) favoring the combination that did not reach statistical significance but was consistent with the absence of a detrimental effect. No other Phase 3 studies conducted in patients with breast cancer previously treated with anthracyclines and a taxane have demonstrated a survival benefit. [14]

Peripheral Neuropathy

Some subjects in these studies have experienced ixabepilone-related sensory neuropathy that is most often cumulative and reversible. Less frequently, severe motor neuropathy, usually presenting with lower extremity weakness, has been observed. In addition, there have been rare reports of autonomic neuropathy. Neuropathy may be dose- and time dependent. In the event of severe peripheral neuropathy, ixabepilone treatment was to be discontinued and not resumed until the neuropathy returned to baseline or to ≤ Grade 1, as defined by the National Cancer Institute (NCI) Common Terminology Criteria for Adverse Events (CTCAE). If the neuropathy was greater than Grade 1, dose reductions were warranted, provided the subject qualified for additional treatment. Peripheral neuropathy was a dose-limiting toxicity in Phase 1 monotherapy studies of ixabepilone at doses above 50 mg/m^{2}every 3 weeks, and cumulative neuropathy has commonly been reported in the Phase 2 studies. The neuropathy is dose dependent and may also be schedule related. The neuropathy is cumulative in nature and gradually lessens following the discontinuation of therapy. Subjects are typically described as presenting with sensory manifestations (dysesthesias

and paresthesias, involving hands and feet). Neuropathic pain has also been reported, although less frequently
(Grade 3/4-related in 3% of 439 subjects treated across all BMS Phase 2 studies). Motor neuropathy has been reported only as a related Grade 3/4 adverse event in 1% of all Phase 2 subjects, and it has only rarely been described in the absence of sensory or painful neuropathy. To date, autonomic neuropathy has been reported in only two subjects, both enrolled in Phase 2 breast cancer studies. One subject developed orthostatic hypotension and gastroparesis after 5 cycles of therapy and the other developed gastroparesis after 14 cycles of therapy. When ixabepilone dose reductions were necessary due to toxicity, the first reduction was to be to a dose of 32 mg/m^2. The use of the 32 mg/m^2 dose has been established in at least two other ixabepilone studies: the CA163-011 study of subjects with non-small cell lung cancer and the CA163-116 study of subjects with metastatic pancreatic cancer. A total
dose per cycle of 30 mg/m^2 has also been administered in subjects with breast cancer (6 mg/m2 daily times 5 every 3 weeks) in studies led by the National Cancer Institute.

Hypersensitivity Reactions

The diluent used with ixabepilone is a BMS-purified polyoxyethylated castor oil (Cremophor®EL) and has the potential for inducing a hypersensitivity reaction. In clinical studies of ixabepilone, anaphylaxis and severe hypersensitivity reactions (dyspnea requiring bronchodilators, hypotension requiring treatment, syncope, bradycardia) that were reported as serious adverse events have occurred in less than 1% of subjects. Routine premedication, including H1 and H2 antihistamines (not corticosteroids) was instituted after 2 subjects developed severe hypersensitivity adverse events (dyspnea and bronchospasm) in the weekly Phase 1 monotherapy study. Since premedication was instituted, the incidence of the NCI CTCAE term “hypersensitivity” of any grade is < 1% in 439 subjects treated in all BMS-sponsored Phase 2 studies. In addition, 17% of subjects had a CTCAE term of “allergy/immunology, other” (of any grade) reported; the majority of incidents consisted of skin rash.

4.0.3 **Cetuximab**

Cetuximab is a genetically engineered version of a mouse antibody that contains both human and mouse components. It can be produced in large quantities in the laboratory. This new monoclonal antibody is believed to work by targeting a natural protein called "epidermal growth factor receptor" (EGFR) on the surface of cancer cells, interfering with their growth. Cetuximab is a chimeric monoclonal antibody, an epidermal growth factor receptor (EGFR) inhibitor with demonstrated activity in colorectal cancer and head and neck cancer.
Cetuximab is believed to operate by binding to the extra cellular domain of the EGFR of cells that express EGFR, which include “cancer cells”, preventing ligand binding and activation of the receptor. This blocks the downstream signaling of EGFR resulting in impaired cell growth and proliferation. Cetuximab has also been shown to mediate antibody dependent cellular cytotoxicity (ADCC). For patients with tumors that express EGFR and who no longer responded to treatment with irinotecan alone or in combination with other chemotherapy drugs, the combination treatment of cetuximab and irinotecan shrank tumors in 22.9% of patients and delayed tumor growth by approximately 4.1 months. For patients who received cetuximab alone, the tumor response rate was 10.8% and tumor growth was delayed by 1.5 months. The efficacy and safety of cetuximab alone or in combination with irinotecan were studied in a randomized, controlled trial with 329 patients and also in combination with irinotecan in 138 patients in which all patients received both drugs. Cetuximab was further evaluated as a single agent in a third clinical trial with 57 patients. Safety data from an additional 111 patients treated only with cetuximab was also evaluated. All of the trials included patients with EGFR-expressing metastatic colorectal cancer, whose disease had progressed after receiving irinotecan. (Cetuximab) is indicated for the treatment of EGFR-expressing, metastatic colorectal carcinoma; In combination with irinotecan for patients who are refractory to irinotecan-based chemotherapy, and as a single agent for patients who are intolerant to irinotecan-based therapy.
The effectiveness of cetuximab in EGFR-expressing mCRC is based on objective response rates. Currently, no data are available that demonstrate an

improvement in disease-related symptoms or increased survival with cetuximab in combination with irinotecan for the treatment of EGFR-expressing, metastatic colorectal carcinoma. [16, 17]

Squamous Cell Carcinoma of the Head and Neck (SCCHN)

Cetuximab is indicated in combination with radiation therapy for the initial treatment of locally or regionally advanced squamous cell carcinoma of the head and neck, and as a single agent, is indicated for the treatment of patients with recurrent or metastatic squamous cell carcinoma of the head and neck for whom prior platinumbased therapy has failed.

Study 1 was a randomized, multicenter, controlled trial of 424 patients with locally or regionally advanced SCCHN. Patients with Stage III/IV SCCHN of the oropharynx, hypopharynx, or larynx with no prior therapy were randomized (1:1) to receive either cetuximab plus radiation therapy or radiation therapy alone. Stratification factors were Karnofsky Performance Status (60–80 versus 90–100), nodal stage (N0 versus N+), tumor stage (T1–3 versus T4 using American Joint Committee on Cancer 1998 staging criteria), and radiation therapy fractionation (concomitant boost versus once-daily versus twice-daily). Radiation therapy was administered for 6–7 weeks as once daily, twice daily, or concomitant boost. Cetuximab was administered as a 400 mg/m2 initial dose beginning one week prior to initiation of radiation therapy, followed by 250 mg/m2 weekly administered 1 hour prior to radiation therapy for the duration of radiation therapy (6–7 weeks).

Of the 424 randomized patients, the median age was 57 years, 80% were male, 83% were Caucasian, and 90% had baseline Karnofsky Performance Status ≥80. There were 258 patients enrolled in US sites (61%). Sixty percent of patients had oropharyngeal, 25% laryngeal, and 15% hypopharyngeal primary tumors; 28% had AJCC T4 tumor stage. Fifty-six percent of the patients received radiation therapy with concomitant boost, 26% received once-daily regimen, and 18% twice-daily regimen. The main outcome measure of this trial was duration of locoregional control. Overall survival was also assessed. Results are presented in the following Table.

Study 1: Clinical Efficacy in Locoregionally Advanced SCCHN

	Cetuximab + Radiation (n=211)	Radiation Alone (n=213)	Hazard Ratio (95% CI[a])	Stratified Log-rank p-value
Locoregional control				
Median duration (months)	24.4	14.9	0.68 (0.52–0.89)	0.005
Overall survival				
Median duration (months)	49.0	29.3	0.74 (0.57–0.97)	0.03

[a] CI = confidence interval

Study 2 was a single-arm, multicenter clinical trial in 103 patients with recurrent or metastatic SCCHN. All patients had documented disease progression within 30 days of a platinum-based chemotherapy regimen. Patients received a 20-mg test dose of cetuximab on Day 1, followed by a 400-mg/m2 initial dose, and 250 mg/m2 weekly until disease progression or unacceptable toxicity. The median age was 57 years, 82% were male, 100% Caucasian, and 62% had a Karnofsky Performance Status of ≥80.

The objective response rate was 13% (95% confidence interval 7%–21%). Median duration of response was 5.8 months (range 1.2–5.8 months).[16]

The immunohistochemical expression of epidermal growth factor receptor (EGFR) in 241 patients with recurrent breast cancer, out of the 241 patients, EGFR expression was positive in 87 (36%) patients and negative in 154 (64%)

patients with recurrent breast cancer. EGFR expression was positive in 71 of 170 (42%) cases of primary tumor samples and in 28 of 103 (27%) biopsy samples of recurrent breast cancer. An inverse relationship between EGFR and estrogen receptor (ER) status was noted. EGFR expression was positive in 68 of 109 (62%) cases with negative ER status while EGFR expression was positive in 17 of 130 (13%) cases with positive ER status ($p<0.0001$).

Patients with positive EGFR expression had a signi□cantly worse post relapse survival ($p<0.0001$) compared to those with EGFR negative expression. Patients with a negative ER status had a signi□cantly worse post relapse survival ($p<0.0001$) than those with a positive ER status. Additionally, patients with positive EGFR expression had a signi□cantly ($p = 0.0021$) worse post relapse survival compared to those with negative EGFR expression in patients who were ER positive. No difference in post relapse survival was observed in patients who were ER negative.

There was a signi□cant difference in treatment response ($p = 0.0014$) and objective response rate ($p = 0.0075$) between patients with positive and negative EGFR expression in patients receiving chemotherapies; whereas the objective response in patients receiving hormonal therapy was not statistically signi□cant. However, there was a statistical difference in the treatment response ($p = 0.0209$) in hormonal therapies. Additionally, there was a signi□cant difference in the treatment response ($p = 0.0196$) and the objective response rate ($p = 0.0061$) between cases with positive and negative EGFR expression. EGFR negative/ER positive tumors had a better treatment response compared to the EGFR positive/ER negative tumors which had a worse treatment response. Patients with a positive ER status had a better response to hormonal therapies than those with ER negative tumors.

Response to first line treatment and EGFR expression are both independently signi□cant ($p = 0.0016$) factors for post relapse survival. Additionally, in a multivariate analysis, ER status was determined to be an independently signi□cant ($p = 0.0104$) factor for post relapse survival; however, EGFR expression was not a signi□cant factor for post relapse survival. Patients who

responded to first line treatment and who showed positive EGFR expression had a signi□cantly (p = 0.0008) shorter time to progression and signi□cantly (p < 0.0001) worse post relapse survival than those with negative EGFR expression. [18]

Cetuximab Pre-Clinical Data

Howard Hughes Medical Institute researchers have found that four aberrant genes work together to promote the growth of primary breast tumors. Cooperation among the four genes also enables cancerous cells to escape into the bloodstream and penetrate through blood vessels into lung tissues. Although shutting off these genes individually can slow cancer growth and metastasis, the researchers found that turning off all four together had a far more dramatic effect on halting cancer growth and metastasis. Metastasis occurs when cells from a primary tumor break off and invade another organ. It is the deadliest transformation that a cancer can undergo, and therefore researchers have been looking for specific genes that propel metastasis. Massagué et al at the Memorial Sloan-Kettering Cancer Center, also found that they could reduce the growth and spread of human breast tumors in mice by simultaneously targeting two of the proteins produced by these genes, using drugs already on the market. The researchers are exploring clinical testing of combination therapy with the drugs – cetuximab (Erbitux®) and celecoxib (Celebrex®) – to treat breast cancer metastasis. In an earlier study, Massagué and colleagues had identified 18 genes whose abnormal activity is associated with breast cancer's ability to spread to the lungs. In the new study published in Nature, they focused on four of these genes. These genes, which code for proteins called epiregulin, COX2, and matrix metalloproteinases 1 and 2, were already known to help regulate growth and remodeling of blood vessels. The researchers silenced various combinations of the four genes in human breast cancer cells that had metastasised to the lung, and then tested these cells in mice. To silence the four genes, the scientists used a technique called RNA interference, in which RNA molecules are tailored to suppress expression of target genes. The aggressive

metastatic tumor cells of these genes decreased both their ability to grow large aggressive tumors in the mouse mammary gland and also the ability to release cells from these tumors into the circulation. “The remarkable thing was that while silencing these genes individually was effective, silencing the quartet nearly completely eliminated tumor growth and spread." Microscopic analysis of blood vessel structure in the tumors revealed that knocking down all four genes greatly reduced growth of the tangle of blood vessels typically seen in tumors. Further experiments revealed that the tumor blood vessels that did form allowed fewer cancer cells to escape into circulation. The researchers next explored how loss of the four abnormal genes affected the metastatic capability of the cells in the lung. They injected cells deficient in the four genes directly into the circulatory system of mice. "When these cells reached the lung capillaries, they just got stuck there," Cetuximab is an antibody that blocks the action of epiregulin and is used to treat advanced colorectal cancer. Celecoxib is an inhibitor of COX2 that is used as an anti-inflammatory, and is being tested in clinical trials against many types of cancer. The researchers also tested whether cetuximab and celecoxib would work effectively in concert to reduce metastasis in mice. The combination of these two inhibitory drugs was effective, even though the drugs individually were not very effective.
The establishment of distant metastases depends on the capacity of small numbers of cancer cells to regenerate a tumor after entering a target tissue. The mechanisms that confer this capacity remain to be defined. Here we identify a role for the transcriptional inhibitors of differentiation Id1 and Id3 as selective mediators of lung metastatic colonization in the triple negative [TN, i.e., lacking expression of estrogen receptor and progesterone receptor, and lacking Her2 (human epidermal growth factor receptor 2) amplification] subgroup of human breast cancer. Although broad expression of Id1 has recently been documented in tumors of the rare metaplastic subtype, here we report that rare Id1-expressing cells are also present in the more common TN subset of human breast tumors but not in other subtypes. We also provide evidence that Id1 expression is enriched in clinically obtained hormone receptor negative lung metastases. Functional studies demonstrate that Id1 and its closely related family member Id3 are required for tumor initiating functions, both in

the context of primary tumor formation and during metastatic colonization of the lung microenvironment. In vivo characterization of lung metastatic progression reveals that Id1 and Id3 facilitate sustained proliferation during the early stages of metastatic colonization, subsequent to extravasation into the lung parenchyma. These results shed light on the proliferative mechanisms that initiate metastatic colonization, and they implicate Id1 and Id3 as mediators of this malignant function in the TN subgroup of breast cancers.[19]

Case Report

Baltic[20] reported a case of a 39 year old woman who underwent a left breast mastectomy in 1998 due to in□ltrative ductal carcinoma (T2 N0 M0). No other therapy was applied at that time. In 2005, multiple liver metastases in the liver were con□rmed by MRI. Hormonal status of the tumor showed ER and PR expression along with EGFR expression de□ned by immunohistochemistry. The CMF regimen along with Cetuximab as initiated. Four months after therapy began, MRI finding revealed signi□cant reduction on the number and size of metastases in liver segments 6 and 8, mainly in the subcapsular region of the liver.

TBCRC 001: EGFR inhibition with cetuximab added to carboplatin in metastatic triple-negative (basal-like) breast cancer. Basal-like breast cancer (BBC) has low expression of ER, PR, and HER2, and is thus often called triple negative (TN). Preclinical studies suggest that BBC depend on EGFR/HER1 for proliferation. TBCRC 001 is a multicenter randomized phase II study of cetuximab (C) alone and combined with carboplatin (P) in TN metastatic BrCa (MBC). Methods: Eligibility required measurable disease, ≤ 3 prior chemotherapy, no prior platinum or EGFR inhibitor. Patients (pts) in Arm 1 received C alone (400 mg/m^2, then 250 mg/m^2 weekly) with P (AUC 2, 3 of 4 weeks) added upon progression (PD). Pts in Arm 2 received C+P throughout. Primary endpoint was objective response (RR, CR+PR). Results: 102 TN patients were enrolled; Arm 1 = 31 pts (closed in 3/07), Arm 2 = 71 (closed 10/07). Mean age 51, 68% white, 26% African American. 87% received prior adjuvant chemo, 54% prior chemo for MBC (24% > 1 prior). Arm 1: PR 6% (2 pts, > 40 weeks), stable disease (SD) 4%, and clinical benefit (CB=PR or SD > 6mos) 10%. Arm 2: RR 18%

(13 pts), SD 9% (6), and CB 27% (19). PR did not appear to differ by line of therapy: 14% 1st-line, 31% 2nd-line, 17% 3rd-line. Most pts progressed rapidly; median PFS was 2.0 months. The regimen was tolerable, grade 3 toxicity occurring in > 5% pts were: Arm 1 - rash only (10% pts); Arm 2 - rash (6%), fatigue (6%), and N/V (6%). Two grade 4 infusion reactions occurred in Arm 2. Biopsies from 16 mets confirmed subtype: basal-like (13), claudin-low (2), HER2+/ER- (1). Preliminary pharmacodynamics on paired biopsies (before/after 1-2 weeks therapy) in 9 pts reveal that 7 had gene expression evidence of activity across the EGFR pathway: 2 decreased with treatment (1PR, 1SD); 5 had no change or increased activity with treatment (1SD, 4 PD). Final response rates and further correlative endpoints will be presented. Conclusions: C alone was well tolerated but has low activity in TN MBC. C+P has 18% RR and 27% CB. Many pts progressed rapidly, consistent with the aggressive behavior of this tumor subtype. Serial biopsies offer insight into EGFR pathway modulation by therapy. Translational studies confirming molecular subtype, identifying biologic effects of EGFR inhibition and platinum, evaluating circulating tumor cells, and studying the BRCA1 pathway are ongoing[21].

A phase II trial of irinotecan plus cetuximab in patients with metastatic breast cancer and prior anthracycline and/or taxane-containing therapy (N0436). Background: Irinotecan has a 23% response rate (RR) in patients (pts) with previously treated metastatic breast cancer (MBC) (Perez, JCO 2004). Epidermal growth factor receptor (EGFR) is overexpressed in MBC, especially in those with ER-/PR- and HER2-negative tumors (triple-negative, [TNeg]). Cetuximab is a monoclonal antibody against EGFR with additive activity to irinotecan in colorectal cancer with acceptable toxicity. Methods: This one-stage phase II study enrolled pts with MBC, measurable disease, and prior anthracycline and/or taxane therapy. Pts received cetuximab 400 mg/m^2 day 1 cycle 1 then 250 mg/m^2 weekly and irinotecan 80 mg/m^2 days 1, 8 of each 21-day cycle. Primary endpoint was confirmed RR by RECIST criteria. Planned sample size was 36. Genotyping for UGT1A1 polymorphisms was performed. Results: 19 eligible pts enrolled from 2/06 to 9/06 with a median age of 49 yrs (range: 28-76), 74% had visceral disease, 32% were ER and/or PR positive, 11%

were HER2 positive, 58% were TNeg. 79% had prior chemotherapy for MBC. Pts received a median of 2 cycles (range: 1-11) of treatment. Confirmed RR was 11% (95% CI: 1-33%), with 1 PR and 1 CR. One pt had stable disease for 11 cycles. RR and clinical benefit rate (CBR: RR + stable > 6 months) for Tneg vs non- Tneg was 18% vs 0% (p=0.49) and 27% vs 0% (p=0.23). 12 pts progressed on therapy within 2 cycles. With low response rate and rapid progression, the study closed early. NCI CTC version 3 grade 3-4 adverse events occurred in 10 pts; only dermatologic toxicity (derm tox) had a > 5% incidence (26%). Patients with grade $\geq$ 2 derm tox had a CBR of 21% (3/14) vs 0% in patients with grade 0-1. 18 pts have progressed with a median time to progression of 1.4 mos (95% CI: 1.1-2.2 mos). 15 pts have died with a median overall survival of 9.4 mos (95% CI: 3.3-16.1 mos). UGT1A1 results will be presented. Conclusions: Irinotecan and cetuximab has minimal activity in this population. Tolerability is acceptable. There was a numerically higher CBR in Tneg pts and patients with grade $\geq$ 2 derm tox, but small pt numbers limit conclusions. Better predictive factors for response to EGFR inhibition are needed. NCI CA-25224, CA-63848, CA-37404, CA- 60276, Bristol-Myers Squibb, Imclone systems[22]**.**

Preliminary results of a randomized phase II study of weekly Irinotecan /carboplatin with or without cetuximab in patients with metastatic breast cancer. O'Shaughnessy and colleagues also presented data on the use of cetuximab in a randomized phase 2 study of weekly irinotecan (90 mg/m^2) and carboplatin (AUC 2), with or without cetuximab (250 mg/m^2). In the overall intention-to-treat study population, response rate improved marginally from 28% to 33% with the addition of cetuximab. Approximately half of the study population (78 patients) had triple-negative tumors. In that subgroup, cetuximab was associated with a response rate of 49% compared with 30% when chemotherapy was used alone. No significant differences in PFS or overall survival were observed in any subgroup, and significant toxicity led to dose reductions in starting doses of the chemotherapy agents. Based on the available evidence, there is little reason to believe that either single-agent cetuximab or a small molecule tyrosine kinase inhibitor of EGFR will have substantial activity in the

triple-negative setting. It remains unknown whether these agents will prove useful in this context when combined with chemotherapy[23]

4.0. 4. Biomarkers Rationale Assessments:

EGFR

Epidermal growth factor receptors (EGFRs) are the prototypic members of a family of growth factors derived from membrane-anchored precursors. All members of this family are characterized by the presence of at least one EGFR structural unit in their extracellular domain. Increased expression and activation of receptor tyrosine kinases occurs frequently in human breast carcinomas.

Roughly 35% breast cancers express EGFR as ascertained by IHC and its expressed with a 2-fold higher frequency in hormone receptor negative breast cancers compared to hormone receptor positive breast cancers .[24,25,26]

A wide variety of *in vitro* and *in vivo* biological effects have been ascribed to EGF and other members of the EGF family. EGF promotes proliferation and differentiation of mesenchymal and epithelial cells. EGF family ligand-receptor signaling is implicated in a wide variety of human carcinomas. Several therapies targeting these receptors are currently in clinical trials and therapeutic strategies including blockage of individual receptors with monoclonal antibodies and inhibition of tyrosine kinase function. [27, 28, 29] Insufficient data exists to suggest a link between EGFR protein expression and response to EGFR inhibitors in breast cancer. [30]

PTEN

PTEN is a tumor suppressor that negatively regulates the PI3 kinase pathway, which is downstream of EGFR. Loss of PTEN protein expression, attributed to loss of heterozygosity and/or post-transcriptional modification, is exhibited in 16 – 37% of breast cancers[31, 32]. PTEN-loss is significantly associated with HR-

negative breast cancers. [33, 34, 35] Studies has been shown to confer resistance to EGFR inhibitors *in vitro* in breast cancer cells and to predict response to Erbitux in metastatic colorectal cancers [36, 37, 38].

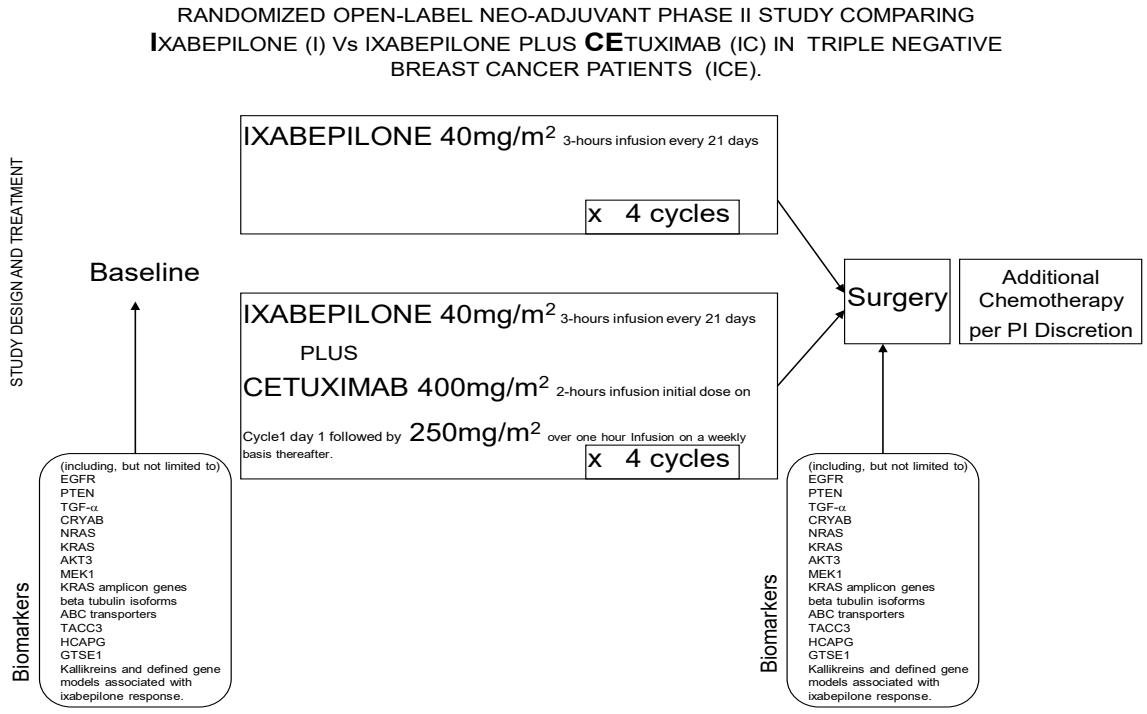
EGFR-associated signature genes

Hoadley et al showed that the expression of a number of EGFR-pathway related genes are associated with the basal-like subtype, including: TGF-α, CRYAB, NRAS, KRAS, AKT3, PTEN, MEK1 and KRAS amplicon genes. In addition, this group identified three gene signatures that correlated with EGFR activity in vitro that could be identified in two breast cancer patient cohorts. While the genes in these signatures do not appear to be directly related to the EGFR pathway, they may still have predictive value. [39]

Other exploratory biomarkers

Additional exploratory biomarkers related to ixabepilone, anti-microtubule and cetuximab sensitivity and resistance may be assessed. Markers shown to have an association with paclitaxel resistance, include beta tubulin isoforms, specifically beta III tubulin [40] and ABC transporter expression, specifically MDR1 and MRP7[41]. The capacity of these biomarkers to predict response to ixabepilone has not been established, as yet. Preliminary analyses have suggested that several gene models, including TACC3, HCAPG, GTSE1, Kallikreins and pre-defined gene sets, may be able to differentiate ixabepilone activity from paclitaxel activity. These models were derived from a comparison of gene expression patterns of pathological CR patients in microarray data available from two neoadjuvant breast cancer studies, CA163-080 (ixabepilone monotherapy) and MDA-133 (paclitaxel-FAC combination therapy) [42].

5.0 Study Design



6.0 Study Population

-Patients with either T1N1-N3M0 or T2-4 N0-3M0 disease who are triple negative and who are candidates for preoperative chemotherapy.

Patients will receive a physical assessment, imaging, a blood draw, and a biopsy prior to the start of treatment.

6.1 Inclusion Criteria:

-Patients with histologic confirmation of invasive breast carcinoma.

-Patients must have intact primary tumor.

-Age ≥ 18 years.

-Invasive breast carcinoma must be determined to be ER (-), PR (-) and HER2Neu (-) prior to study entry.
-Patients should have T1N1-3M0 or T2-4 N0-3M0.
-Patients with bilateral breast cancer are eligible.
-Patients should have a Karnofsky performance scale of ≥ 70%.
-Patients must have clinically measurable disease by physical exam to be treated in the neoadjuvant setting.
-Patients should have adequate bone marrow function, as defined by peripheral granulocyte count of ≥ 1500/mm^3, and platelet count ≥ 100000mm^3.
-Patients must have adequate liver function with a bilirubin within normal laboratory values. Alkaline phosphatase and transaminases (ALT and AST) may be up to 1.5 x upper limit of normal (ULN) of the institution.
-Patients should have adequate renal function with creatinine levels within normal range.
-Patients should have a normal left ventricular ejection fraction (LVEF) of ≥50%.
-Negative serum or urine pregnancy test for a woman of childbearing potential (WOCBP).
-WOCBP must use a reliable and appropriate contraceptive method during the study and six months after chemotherapy is completed. WOCBP are women who are not menopausal for 12 months or had no previous surgical sterilization.
-Patients must sign an informed consent indicating that they are aware of the investigational nature of the study, in keeping with institutional policy.

6.2 Exclusion Criteria:
-Patients with a history of other invasive malignancies diagnosed and treated within the previous 5 years, except non-melanoma skin cancer and non-invasive cervical cancer.
-Her2Neu, ER and PR positive patients should be excluded.
-Patients with an organ allograft or other history of immune compromise.
-Prior treatment with any investigational drug within the preceding 4 weeks.
-Chronic treatment with systemic steroids or another immunosuppressive agent.

- Uncontrolled chronic viral infections or patients requiring parenteral antibiotics will be excluded. Patients taking highly active anti-viral therapies and with controlled disease will be allowed to participate.
-Patients with an active, bleeding diathesis or on oral anti-vitamin K medication (except low dose coumadin defined as 1 mg a day)
-Other concurrent and/or uncontrolled medical disease which could compromise participation in the study (i.e., uncontrolled diabetes, uncontrolled hypertension, severe infection, severe malnutrition, unstable angina, or congestive heart failure
- New York Heart Association Class III or IV, ventricular arrhythmias, active ischemic heart disease, myocardial infarction within six months, chronic liver or renal disease, active upper GI tract ulceration).
-Patients with a pre-existing peripheral neuropathy > grade 1.

6.3 Randomization

Blocked randomization will be used to randomize patients to either arm 1 or arm 2. A master sequence of randomization numbers for each arm will be generated.

7.0 Treatment Plan/Methods

This is a randomized open-label phase II trial; 116 triple negative breast cancer patients will be randomized equally between 1) Ixabepilone or 2) Ixabepilone plus Cetuximab. The study population will include patients≥ 18 years of age diagnosed with invasive mammary carcinoma. Patients will be assigned to treatment arms using a stratified blocked randomization, with strata based on disease stage (T1N1-3M0 or T2-4 N0-3M0). All patients will receive a physical assessment, imaging, a blood draw, and a core needle biopsy (CNB) a biopsy prior to the start of treatment.

7.1 Treatment Arm 1

Patients will receive Ixabepilone 40mg/m^2 given over approximately 3-hours on day 1 Patients will receive a total of four cycles of this combination with each cycle given 21(+/- 3) days.

Subjects will be premedicated approximately 1 hour before the ixabepilone infusion with either 1) both an oral H1 blocker (diphenhydramine 50 mg or equivalent) and an oral H2 blocker (e.g., ranitidine 150 300 mg or cimetidine 300–800 mg or nizatidine 150-300 mg or famotidine 20–40 mg); or 2) with premedication per institutional standards. Premedication with steroids is indicated in subjects who developed a hypersensitivity reaction to ixabepilone in previous cycles. Patients with severe reaction should not be retreated (see dose modification chart below).

7.2 Treatment Arm 2

Patients will receive Ixabepilone 40mg/m^2 given over approximately 3-hours on day 1 followed by Cetuximab given on day 1 at 400 mg/m^2 as an initial loading dose (first infusion) administered as a 120-minute IV infusion (maximum infusion rate 5 mL/min) followed by 250 mg/m^2 infused over 60 minutes (maximum infusion rate 5 mL/min) as a subsequent weekly dose. Patients will receive a total of 4 cycles of this combination with each cycle being 21 (+/-3) days.

Subjects will be premedicated approximately 1 hour before the ixabepilone infusion with either 1)both an oral H1 blocker (diphenhydramine 50 mg or equivalent) and an oral H2 blocker (e.g., ranitidine 150-300 mg or cimetidine 300–800 mg or nizatidine 150 300 mg or famotidine 20–40 mg); or 2) with premedication per institutional standards. Premedication with steroids is indicated in subjects who developed a hypersensitivity reaction to ixabepilone in previous cycles. Patients with severe reaction should not be retreated. Subjects who are to receive cetuximab are to be premedicated with an H_1 antagonist (e.g., 50 mg of diphenhydramine IV or equivalent)

7.3 Dose modifications:

7.3.1 Ixabepilone dose modifications:

The dose of Ixabepilone may be decreased depending on how well the patient tolerates the study treatment. Subjects should be evaluated during treatment by periodic clinical observation and laboratory tests including complete blood cell counts. If toxicities are present, treatment should be delayed to allow recovery.

If toxicities recur, an additional 20% dose reduction should be made.

The dose levels of Ixabepilone will be reduced as described in the table below:

Dose Level	Ixabepilone
1	(Starting Dose) 40 mg/m^2
-1	32 mg/m^2
-2	25 mg/m^2

Patients requiring dose reductions below dose level -2 must discontinue Ixabepilone, with the following exception: Patients with responding disease may continue treatment on study after consultation with the sponsor, TMHRI.

No dose re-escalation will be allowed after dose reduction.

Dose Modification - Study Drug Related Non-Hematologic Toxicities

Toxicity	Ixabepilone
Grade 2	
Grade 2 except Gr 2 Neuropathy lasting ≥ 7 days [a]	No change
Grade 2 Neuropathy lasting ≥ 7 days	Decrease 1 dose level [b,c]

Dose Modification - Study Drug Related Non-Hematologic Toxicities

Toxicity	Ixabepilone
Grade 3	
Grade 3 Neuropathy [c]	Decrease 1 dose level [a, b]
Grade 3 Neuropathy lasting < 7 days	Decrease 1 dose Level [a, b]

Grade 3 Neuropathy lasting ≥ 7 days	Discontinue Ixabepilone [d]
Grade 4	
Grade 4 toxicity	Discontinue Ixabepilone [d]

a. Delay until toxicity resolved to baseline or ≤ Grade 1.

b. Patients requiring more than two dose reductions will discontinue Ixabepilone except those who appear to be benefiting from treatment in which case treatment can be continued after consultation with and approval by the Sponsor "The Methodist Hospital Research Institute" (TMHRI).

c. Excludes Grade 3 fatigue/asthenia and transient arthralgia/myalgia for which no dose reduction is required.

d. Responding patients who have sufficiently recovered from toxicity during previous cycle may be considered for retreatment after dose reduction only after discussion with and agreement by Sponsor (TMHRI).

Dose Modification - Hematologic Toxicities

Toxicity	Ixabepilone
Grade 3:	
Grade 3 Platelets with: significant bleeding or requiring platelet	Decrease 1 dose Level [a,b]

transfusion	
Grade 4	
Grade 4 Platelets	Decrease 1 dose Level [a,b]
Grade 4 Neutrophils lasting ≥ 7 days	Decrease 1 dose Level [a,c]
Febrile Neutropenia any grade	Decrease 1 dose Level [a,c]

a. Patients requiring more than 2 dose reductions will discontinue Ixabepilone except those who appear to be benefiting from treatment in which case treatment can be continued after consultation with and approval by the Sponsor (TMHRI)

b. Delay until platelet count ≥ 100,000/mm^3

c. Delay until neutrophil count ≥ 1,500/mm [43, 44]

7.3.2. Cetuximab Dose Modifications:

Infusion Reactions

Reduce the infusion rate by 50% for NCI CTC Grade 1 or 2 and non-serious NCI CTC Grade 3–4 infusion reactions.

Immediately and permanently discontinue cetuximab for serious infusion reactions, requiring medical intervention and/or hospitalization.[45]

Dermatologic Toxicity: Recommended dose modifications for severe (NCI-CTC Grade 3 or 4) acneform rash are:

Cetuximab Dose Modification Guidelines for Rash

Severe Acneform Rash Modification	cetuximab	Outcome	cetuximab Dose
1st occurrence	Delay infusion 1 to 2 weeks	Improvement	Continue at 250 mg/m^2
		No Improvement	Discontinue cetuximab

2nd occurrence	Delay infusion 1 to 2 weeks	Improvement	Reduce dose to 200 mg/m^2
		No Improvement	Discontinue cetuximab
3rd occurrence	Delay infusion 1 to 2 weeks	Improvement	Reduce dose to 150 mg/m^2
		No Improvement	Discontinue cetuximab
4th occurrence	Discontinue cetuximab		

Patients who experience unacceptable cetuximab toxicity or who are determined to have progressive disease as defined by RECIST during any cycle will be discontinued from the ixabepilone plus cetuximab arm. However if the toxicity is related with cetuximab only; then ixabepilone will be given on a regular basis until the neoadjuvant cycles are completed.
Patients who are deemed not appropriate for further chemotherapy will proceed to local therapy (surgical intervention or radiation therapy).

7.4 Discontinuation of Subjects from Treatment:
Subjects MUST discontinue study treatment (investigational or noninvestigational treatment) for any of the following reasons:
• Withdrawal of informed consent (subject's decision to withdraw for any reason).
• Any clinical adverse event (AE), laboratory abnormality, or intercurrent illness which, in the opinion of the investigator, indicates that continued participation in the study is not in the best interest of the subject.
• Pregnancy.
• Termination of the study by the sponsor, by the local IRB or by the supporting/funding organization Bristol-Myers Squibb (BMS)
• Loss of ability to freely provide consent through imprisonment or involuntary incarceration for treatment of either a psychiatric or physical (e.g., infectious disease) illness.
All subjects who discontinue study treatment should comply with protocol-specified follow-up procedures. The only exception to this requirement is when a

subject withdraws consent for all study procedures or loses the ability to consent freely (i.e., is imprisoned or involuntarily incarcerated for the treatment of either a psychiatric or physical illness).

If a subject was withdrawn before completing the study, the reason for withdrawal must be entered on the appropriate case report form (CRF) page.

7.5 Surgical Intervention

Approximately 3-4 weeks following completion of neoadjuvant chemotherapy a clinical assessment and repeat mammography and ultrasound will be performed. The patient will then proceed to breast and axillary surgery as judged by the treating physician. **Surgery should be performed approximately 3 - 4 weeks after the last dose of neoadjuvant ixabepilone or Ixabepilone plus cetuximab regimen** or at resolution of possible hematological or infective complication. Reasons for delaying surgery in individual patients must be recorded in source documentation.

8.0 CRITERIA FOR RESPONSE

8.0.1 PATHOLOGICAL RESPONSE: The histologic response after treatment will be evaluated in the primary tumor and in the axillary lymph nodes removed by axillary dissection. In the primary tumor, cellular modifications will be evaluated in both the infiltrating tumoral component and in the possible ductal component, if applicable:

− Analysis of the viable residual infiltrating component (% of total tumoral mass)
− Analysis of the residual ductal component (% of total tumoral mass)
− Determination of the mitotic index.

In the axillary lymph nodes: analysis of the viable residual component.

The criteria of pathological response will be defined according to the Sataloff Classification [46].

The response in the primary tumor site will be graded from A to D.

Primary site response:

T-A Total or near total therapeutic effect.

T-B Subjectively greater than 50% therapeutic effect but less than total or near total.

T-C Less than 50% therapeutic effect, but effect evident

T-D No therapeutic effect.

Axillary lymph node response:

N-A Evidence of therapeutic effect, no metastatic disease.

N-B No nodal metastasis or therapeutic effect.

N-C Evidence of therapeutic effect but nodal metastasis still Present.

N-D Viable metastatic disease, no therapeutic effect

pCR corresponds to pathological grades T-A; N-A, N-B.

8.0.2 RECIST Criteria

The main endpoint in this study is to determine the pathologic complete response rate (pCR) (breast and axilla) of Ixabepilone versus Ixabepilone when combined with cetuximab in patients with invasive breast adenocarcinoma T1N1-N3M0 or T2-4 N0-3M0 disease who are Triple negative and who are candidates for preoperative chemotherapy.

The clinical tumor response will be assessed using RECIST criteria at baseline and before surgery.

RECIST criteria offer a simplified, conservative, extraction of imaging data for wide application in clinical trials. They presume that linear measures are an adequate substitute for 2-D methods and register four response categories[47]:

- CR (complete response) = disappearance of all target lesions
- PR (partial response) = 30% decrease in the sum of the longest diameter of target lesions
- PD (progressive disease) = 20% increase in the sum of the longest diameter of target lesions
- SD (stable disease) = small changes that do not meet above criteria

9.0 Study Materials:

Chemistry

The epothilones are a class of tubulin polymerization agents obtained from the fermentation of *Sorangium cellulosum*. Ixabepilone is a semisynthetic derivative of epothilone B that has improved in vitro metabolic stability when compared with its natural precursor.

Pharmaceutical Properties and Formulation

Investigational Product Identification

Ixabepilone for Injection

Ixabepilone is a 16-membered polyketide macrolide, with a chemically modified lactam substitution for the naturally existing lactone. The chemical name for ixabepilone is [1*S*-[1*R**,3*R**(E),7*R**, 10*S**,11*R**,12*R**,16*S**]]-7,11-dihydroxy-8,8,10,12,16-pentamethyl-3-[1-methyl-2-(2-methyl-4-thiazolyl)ethenyl]-17-oxa-4-azabicyclo[14.1.0]heptadecane-5,9-dione. It has a molecular formula of $C_{27}H_{42}N_2O_5S$, and its molecular weight is 506.71 grams/mole.

Ixabepilone for Injection is supplied as a lyophilized (freeze dried), white to off-white, whole or fragmented cake in a vial. The drug product is available in two potencies:

- Ixabepilone, for injection 15 mg supplied with diluent for ixabepilone, 8 mL.
- Ixabepilone, for injection 45 mg supplied with diluent for ixabepilone, 23.5 mL.

Vehicle for Constitution of Ixabepilone for Injection

Vehicle for Constitution of Ixabepilone for Injection is supplied with the drug product for use in constitution of the lyophile. Vehicle for Constitution is provided in vials containing 8.0 mL/via (15mg) and 23.5mL (45mg) the vehicle is a mixture of dehydrated alcohol and cleaned Cremophor (polyoxyethylated castor oil). One vial of the 8.0 mL/vial vehicle product is provided whenever a 15 mg/vial of Ixabepilone for Injection is supplied.

Packaging

Ixabepilone for Injection and the Vehicle for Constitution are packaged in Type I glass vials, stoppered with butyl rubber closures and sealed with aluminum seals. A sufficient excess of drug and vehicle is provided in the respective vials to allow for withdrawal losses.

Handling and Dispensing of Investigational Product

The investigational product should be stored in a secure area according to local regulations. It is the responsibility of the investigator to ensure that the investigational product is dispensed only to study patients. The investigational product must be dispensed only from official study sites by authorized personnel, according to local regulations.

Storage Requirements/Stability

Ixabepilone for Injection should be stored refrigerated at 2°C to 8°C (36°F to 46°F) and protected from light. Vehicle for Constitution should be stored at 2°C to 25°C (36°F to 77°F). If the vehicle is refrigerated, it must be allowed to warm to room temperature before constitution of the lyophile. After initial constitution with the accompanying vehicle, the solution may be stored in the vial for a maximum of 1 hour at room temperature and room light. The constituted solution should not be stored in the syringe. After final dilution with Lactated Ringer's Injection, USP (LRI) to ixabepilone concentrations between 0.2 and 0.6 mg/mL, the solution is stable when stored at room temperature and room light for a maximum of 6 hours. Administration of the entire infusion volume must be completed within the 6-hour time period as noted above.

Lactated Ringer's Injection, USP is specified because it has a pH range of 6 to 7.5, which is required to maintain ixabepilone stability. Other diluents should not be used with ixabepilone.

Preparation and Administration

An ixabepilone kit contains two vials, a vial of ixabepilone for injection which contains ixabepilone powder and a vial containing diluent for ixabepilone. Only the supplied diluent must be used for constituting ixabepilone for injection. The ixabepilone kit must be stored in a refrigerator at 2° C - 8° C (36° F - 46° F) in the original package to protect from light. Prior to constituting ixabepilone for injection, the kit should be removed from the refrigerator and allowed to stand at room temperature for approximately 30 minutes. When the vials are first removed from the refrigerator, a white precipitate may be observed in the diluent vial. This precipitate will dissolve to form a clear solution once the diluent warms to room temperature. To allow for withdrawal losses, the vial labeled as 15 mg ixabepilone for injection contains 16 mg of ixabepilone and the vial labeled as 45 mg ixabepilone for injection contains 47 mg of ixabepilone. The 15-mg ixabepilone kit is supplied with a vial providing 8 mL of the diluent and the 45-mg ixabepilone kit is supplied with a vial providing 23.5 mL of the diluent. After constituting with the diluent, the concentration of ixabepilone is 2 mg/mL.

A. To constitute:

1) With a suitable syringe, aseptically withdraw the diluent and slowly inject it into the ixabepilone for injection vial. The 15-mg ixabepilone is constituted with 8 mL of diluent and the 45-mg ixabepilone is constituted with 23.5 mL of diluent.
2) Gently swirl and invert the vial until the powder in ixabepilone is completely dissolved.

B. To dilute:

Before administration, the constituted solution must be further diluted only with normal saline (NS) or Lactated Ringer's Injection; USP (LRI) supplied in DEHP [di-(2-ethylhexyl) phthalate] free bags. For most doses, a 250 mL bag of LRI is sufficient. However, it is necessary to check the final infusion concentration of each dose based on the volume of NS or LRI to be used. The final concentration

for infusion must be between 0.2 mg/mL and 0.6 mg/mL. To calculate the final infusion concentration, use the following formulas:

Total Infusion Volume = mL of Constituted Solution + mL of LRI

Final Infusion Concentration = Dose of ixabepilone **(mg)/Total Infusion Volume (mL)**

1) Aseptically, withdraw the appropriate volume of constituted solution containing 2 mg of ixabepilone per mL.
2) Aseptically, transfer to an intravenous (IV) bag containing an appropriate volume of LRI to achieve the final desired concentration of ixabepilone.
3) Thoroughly mix the infusion bag by manual rotation.

The infusion solution must be administered through an appropriate in-line filter with a microporous membrane of 0.2 to 1.2 microns. DEHP-free infusion containers and administration sets must be used. Any remaining solution should be discarded according to institutional procedures for antineoplastics.

Incompatibilities

To minimize patient exposure to the plasticizer di-(2-ethylhexyl) phthalate (DEHP), which may be leached by Cremophor from some brands of polyvinyl chloride (PVC) infusion bags or administration sets, diluted ixabepilone solutions should be stored in bottles (glass, polypropylene) or plastic bags (polyethylene, polypropylene, polyolefin, ethylene-vinyl-acetate) and administered either through polyethylene-lined administration sets or through PVC sets plasticized with TOTM (trioctyl trimellitate). Intravenous infusion sets and components typically used to administer paclitaxel have been found to be compatible with infusions of ixabepilone.

The following infusion components have been qualified for use with Ixabepilone for Injection and Vehicle (Cremophor-containing) for Constitution:

Intravenous infusion sets containing an in-line 0.22 micron filter

- Baxter Vented Paclitaxel Set (Catalog #2C7553)
- Abbott Primary IV Plumset (Catalog #11947)

Intravenous infusion sets *not* containing an in-line 0.22 micron filter

- McGaw AccuPro Pump Nitroglycerin IV Set (Catalog #V8333)
- Clintec IV Fat Emulsion Set (Catalog #2C1105)

Filter Extension Set (to be used with intravenous infusion sets *not* containing an in-line filter)

- Braun Filtered Extension Set with 5 Micron Filter (Catalog #FE-5010Y)

Diluted solutions may also be administered using a syringe pump and polyethylene-lined extension sets.

Safety Precautions

Appropriate protective clothing, eye shield, mask, and gloves should be worn and Class II vertical-laminar-airflow safety cabinets should be used during the preparation and handling of ixabepilone.

Investigational Product Records at Investigational Site(s)

The study drug will be kept in the investigational pharmacy at the sponsor site (TMHRI). It is the responsibility of the Investigator to ensure that a current record of investigational product disposition is maintained at each study site where investigational product is inventoried and disposed. Records or logs must comply with applicable regulations and guidelines, and should include:

- Amount received and placed in storage area.
- Amount currently in storage area.
- Label ID number or batch number.

- Dates and initials of person responsible for each investigational product inventory entry/movement.
- Amount dispensed to and returned by each subject, including unique subject identifiers.
- Amount transferred to another area for dispensing or storage.
- Non-study disposition (e.g., lost, wasted, broken).
- Amount destroyed at study site.

Ixabepilone dispensing record/inventory logs and copies of signed packing lists must be maintained at the investigational site. Batch numbers for ixabepilone must be recorded in the drug accountability records.

Destruction of Investigational Product

Investigational product must be destroyed at the site, it is the Investigator's responsibility to ensure that arrangements have been made for the disposal, written authorization has been granted by BMS, procedures for proper disposal have been established according to applicable regulations and guidelines and institutional procedures, and appropriate records of the disposal have been documented.

Drug Ordering and Accountability

Initial Orders - further information to be provided

Following submission and approval of the required regulatory documents, a supply of ixabepilone may be ordered from BMS. Investigators must complete a Drug Request Form and email it to epothilone.supplies@bms.com. Please fax to 866-227-7229 or 203-677-6489 if you cannot send the form electronically.

Ixabepilone vials (15mL) are shipped in quantities of 18. Allow 5 business days for shipment of drug from BMS receipt of the ixabepilone Clinical Supply Request form. Drug is protocol specific, but not patient specific.

All products will be shipped via Federal Express in a temperature-controlled container. Shipments will be made from BMS on Monday through Thursday for

delivery onsite Tuesday through Friday. There will be no weekend or holiday delivery of drug. It is possible that sites may have more than one ixabepilone clinical study ongoing at the same time. It is imperative that only product designated for this protocol number be utilized for this study. To help segregate product for this study from other investigational or marketed product, stickers bearing the BMS protocol number will be provided and should be affixed to the front of the outer carton just above the company names so as not to obscure any marking.

Re-supply

Reorders should be emailed directly to BMS using epothilone.supplies@bms.com for shipment within 5 business days. When assessing need for resupply, institutions should keep in mind the number of vials used per treatment dose, and that shipments may take 5 business days from BMS receipt of request. Ixabepilone vials (15 and 45 mL) are shipped in quantities of 18. Drug is not patient specific. Be sure to check with your pharmacy regarding existing investigational stock to assure optimal use of drug on hand.[48]

Cetuximab[49]
SUPPLY AND STORAGE: Injection: supplied as 50 mL, single-use vial (preservative free) containing 100 mg of cetuximab at a concentration of 2 mg/mL by ImClone.

SOLUTION PREPARATION AND COMPATIBILITY: PARENTERAL ADMINISTRATION: Intermittent infusion via infusion pump or syringe pump run loading dose over 2 hours, and maintenance dose over 1 hour infusion rate should not exceed 5mL/min use NS to flush line at the end of infusion. DOSAGE GUIDELINES: Numerous dosing schedules exist and depend on disease, response and concomitant therapy. Guidelines for dosing also include consideration of absolute neutrophil count (ANC). Dosage may be reduced,

delayed or discontinued in patients with bone marrow depression due to cytotoxic/radiation therapy or with other toxicities.

10. Adverse Events

10.1 Common side effects of ixabepilone:

Because clinical trials are conducted under widely varying conditions, the adverse reaction rates observed in the clinical trials of a drug cannot be directly compared to rates in other clinical trials and may not reflect the rates observed in clinical practice. Unless otherwise specified, assessment of adverse reactions is based on one randomized study (Study 046) and one single-arm study (Study 081). In Study 046, 369 patients with metastatic breast cancer were treated with Ixabepilone 40 mg/m^2 administered intravenously over 3 hours every 21 days, combined with capecitabine 1000 mg/m^{2}twice daily for 2 weeks followed by a 1-week rest period. Patients treated with capecitabine as monotherapy (n=368) in this study received 1250 mg/m^2 twice daily for 2 weeks every 21 days. In Study 081, 126 patients with metastatic or locally advanced breast cancer were treated with Ixabepilone 40 mg/m^2 administered intravenously over 3 hours every 3 weeks. The most common adverse reactions (≥20%) reported by patients receiving Ixabepilone were peripheral sensory neuropathy, fatigue/asthenia, myalgia/arthralgia, alopecia, nausea, vomiting, stomatitis/mucositis, diarrhea, and musculoskeletal pain. The following additional reactions occurred in ≥20% in combination treatment: palmarplantar erythrodysesthesia (hand-foot) syndrome, anorexia, abdominal pain, nail disorder, and constipation. The most common hematologic abnormalities (>40%) include neutropenia, leukopenia, anemia, and thrombocytopenia. NCI CTC grading for febrile neutropenia ranges from Grade 3 to 5. Three patients (1%) experienced Grade 5 (fatal) febrile neutropenia. Other neutropenia-related deaths (9) occurred in the absence of reported febrile neutropenia. Peripheral sensory neuropathy (graded with the NCI CTC scale) was defined as the occurrence of any of the following: areflexia, burning sensation, dysesthesia, hyperesthesia, hypoesthesia, hyporeflexia, neuralgia, neuritis, neuropathy, neuropathy peripheral, neurotoxicity, painful response to

normal stimuli, paresthesia, pallanesthesia, peripheral sensory neuropathy, polyneuropathy, polyneuropathy toxic and sensorimotor disorder.
Peripheral motor neuropathy was defined as the occurrence of any of the following: multifocal motor neuropathy, neuromuscular toxicity, peripheral motor neuropathy, and peripheral sensorimotor neuropathy. Palmar-plantar erythrodysesthesia (hand-foot syndrome) was graded on a 1-3 severity scale in Study 046. A G-CSF (granulocyte colony stimulating factor) or GM-CSF (granulocyte macrophage stimulating factor) was used in 20% and 17% of patients who received Ixabepilone in Study 046 and Study 081, respectively.
The following serious adverse reactions were also reported in 1323 patients treated with Ixabepilone as monotherapy or in combination with other therapies in Phase 2 and 3 studies.
Infections and Infestations: sepsis, pneumonia, infection, neutropenic infection, urinary tract infection, bacterial infection, enterocolitis, laryngitis, lower respiratory tract infection.
Blood and Lymphatic System Disorders: coagulopathy, lymphopenia.
Metabolism and Nutrition Disorders: hyponatremia, metabolic acidosis, hypokalemia, hypovolemia.
Nervous System Disorders: cognitive disorder, syncope, cerebral hemorrhage, abnormal coordination, lethargy.
Cardiac Disorders: myocardial infarction, supraventricular arrhythmia, left ventricular dysfunction, angina pectoris, atrial flutter, cardiomyopathy, myocardial ischemia.
Vascular Disorders: hypotension, thrombosis, embolism, hemorrhage, hypovolemic shock, vasculitis.
Respiratory, Thoracic, and Mediastinal Disorders: pneumonitis, hypoxia, respiratory failure, acute pulmonary edema, dysphonia, pharyngolaryngeal pain
Gastrointestinal Disorders: ileus, colitis, impaired gastric emptying, esophagitis, dysphagia, gastritis, gastrointestinal hemorrhage
Hepatobiliary Disorders: acute hepatic failure, jaundice
Skin and Subcutaneous Tissue Disorders: erythema multiforme

Musculoskeletal, Connective Tissue Disorders, and Bone Disorders: muscular weakness, muscle spasms, trismus

Renal and Urinary Disorders: nephrolithiasis, renal failure

General Disorders and Administration Site Conditions: chills

Investigations: increased transaminases, increased blood alkaline phosphatase, increased gamma-glutamyltransferase.

DRUG INTERACTIONS

Effect of Other Drugs on Ixabepilone *Drugs That May Increase Ixabepilone Plasma Concentrations* CYP3A4 Inhibitors: Co-administration of ixabepilone with ketoconazole, a potent CYP3A4 inhibitor, increased ixabepilone AUC by 79% compared to ixabepilone treatment alone. If alternative treatment cannot be administered, a dose adjustment should be considered. The effect of mild or moderate inhibitors (e.g. erythromycin, fluconazole, or verapamil) on exposure to ixabepilone has not been studied. Therefore, caution should be used when administering mild or moderate CYP3A4 inhibitors during treatment with Ixabepilone, and alternative therapeutic agents that do not inhibit CYP3A4 should be considered. Patients receiving CYP3A4 inhibitors during treatment with Ixabepilone should be monitored closely for acute toxicities (eg, frequent monitoring of peripheral blood counts between cycles of ixabepilone). *Drugs That May Decrease Ixabepilone Plasma Concentrations* CYP3A4 Inducers: ixabepilone is a CYP3A4 substrate. Strong CYP3A4 inducers (eg, dexamethasone, phenytoin, carbamazepine, rifampin, rifampicin, rifabutin, and phenobarbital) may decrease ixabepilone concentrations leading to subtherapeutic levels. Therefore, therapeutic agents with low enzyme induction potential should be considered for coadministration with ixabepilone. St. John's Wort may decrease ixabepilone plasma concentrations unpredictably and should be avoided. Effect of Ixabepilone on Other Drugs Ixabepilone does not inhibit CYP enzymes at relevant clinical concentrations and is not expected to alter the plasma concentrations of other drugs. In patients with cancer who received ixabepilone (40 mg/m^2) in combination with capecitabine (1000 mg/m^2), ixabepilone Cmax decreased by 19%, capecitabine Cmax decreased by 27%,

and 5-fluorouracil AUC increased by 14%, as compared to ixabepilone or capecitabine administered separately. The interaction is not clinically significant given that the combination treatment is supported by efficacy data.

USE IN SPECIFIC POPULATION

Pregnancy Category D: Ixabepilone may cause fetal harm when administered to pregnant women. There are no adequate and well-controlled studies with Ixabepilone in pregnant women. Women should be advised not to become pregnant when taking Ixabepilone. If this drug is used during pregnancy, or if the patient becomes pregnant while taking this drug, the patient should be apprised of the potential hazard to the fetus.

Ixabepilone was studied for effects on embryo-fetal development in pregnant rats and rabbits given IV doses of 0.02, 0.08, and 0.3 mg/kg/day and 0.01, 0.03, 0.11 and 0.3 mg/kg/day, respectively. There were no teratogenic effects. In rats, an increase in resorptions and post-implantation loss and a decrease in the number of live fetuses and fetal weight was observed at the maternally toxic dose of 0.3 mg/kg/day (approximately one-tenth the human clinical exposure based on AUC). Abnormalities included a reduced ossification of caudal vertebrae, sternebrae, and metacarpals. In rabbits, ixabepilone caused maternal toxicity (death) and embryo-fetal toxicity (resorptions) at 0.3 mg/kg/day (approximately one-tenth the human clinical dose based on body surface area). No fetuses were available at this dose for evaluation.

Nursing Mothers: It is not known whether ixabepilone is excreted into human milk. Following intravenous administration of radiolabeled ixabepilone to rats on days 7 to 9 postpartum, concentrations of radioactivity in milk were comparable with those in plasma and declined in parallel with the plasma concentrations. Because many drugs are excreted in human milk and because of the potential for serious adverse reactions in nursing infants from ixabepilone, a decision must be made whether to discontinue nursing or to discontinue ixabepilone taking into account the importance of the drug to the mother.

Pediatric Use: The safety and effectiveness of ixabepilone in pediatric patients have not been established.

Geriatric Use: Clinical studies of ixabepilone did not include sufficient numbers of subjects aged sixty five and over to determine whether they respond differently from younger subjects. Forty-five of 431 patients treated with ixabepilone in combination with capecitabine were ≥65 years of age and 3 patients were ≥75. Overall, the incidence of grade 3/4 adverse reactions were higher in patients ≥65 years of age versus those <65 years of age (82% versus 68%) including grade 3/4 stomatitis (9% versus 1%), diarrhea (9% versus 6%), palmar-plantar erythrodysesthesia syndrome (27% versus 20%), peripheral neuropathy (24% versus 22%), febrile neutropenia (9% versus 3%), fatigue (16% versus 12%), and asthenia (11% versus 6%). Toxicity-related deaths occurred in 2 (4.7%) of 43 patients ≥65 years with normal baseline hepatic function or mild impairment. Thirty-two of 240 breast cancer patients treated with ixabepilone as monotherapy were ≥65 years of age and 6 patients were ≥75. No overall differences in safety were observed in these patients compared to those <65 years of age.

Hepatic Impairment: Ixabepilone was evaluated in 56 patients with mild to severe hepatic impairment defined by bilirubin levels and AST levels. Compared to patients with normal hepatic function (n=17), the area under the curve (AUC0-infinity) of ixabepilone increased by:

• 22% in patients with a) bilirubin >1 – 1.5 x ULN or b) AST >ULN but bilirubin <1.5 x ULN;

• 30% in patients with bilirubin >1.5 – 3 x ULN and any AST level; and

• 81% in patients with bilirubin >3 x ULN and any AST level.

Doses of 10 and 20 mg/m2 as monotherapy were tolerated in 17 patients with severe hepatic impairment (bilirubin >3 x ULN). Ixabepilone in combination with capecitabine must not be given to patients with AST or ALT >2.5 x ULN or bilirubin >1 x ULN.

Dose reduction is recommended when administering Ixabepilone as monotherapy to patients with hepatic impairment Because there is a need for dosage adjustment based upon hepatic function, assessment of hepatic function is recommended before initiation of Ixabepilone and periodically thereafter.

Renal Impairment: Ixabepilone is minimally excreted via the kidney. No controlled pharmacokinetic studies were conducted with Ixabepilone in patients with renal impairment. Ixabepilone in combination with capecitabine has not been evaluated in patients with calculated creatinine clearance of <50 mL/min. ixabepilone as monotherapy has not been evaluated in patients with creatinine >1.5 times ULN. In a population pharmacokinetic analysis of Ixabepilone as monotherapy, there was no meaningful effect of mild and moderate renal insufficiency (CrCL >30 mL/min) on the pharmacokinetics of ixabepilone.

OVERDOSAGE

One case of overdose of Ixabepilone has been reported. The patient mistakenly received 100 mg/m^2 (total dose 185 mg) and was admitted to the hospital for observation. The patient experienced myalgia (grade 1) and fatigue (grade 1) one day after infusion and was treated with a centrally acting analgesic. The patient recovered and was discharged without incident. There is no known antidote for overdosage of Ixabepilone. In case of overdosage, the patient should be closely monitored, and supportive treatment should be administered. Management of overdose should include supportive medical interventions to treat the presenting clinical manifestations. [43, 44]

10.2 Common side effects of cetuximab:

Dermatologic toxicities, including acneform rash, skin drying and fissuring, paronychial inflammation, and infectious sequelae (for example *S. aureus* sepsis, abscess formation, cellulitis, blepharitis, cheilitis) occurred in patients receiving cetuximab therapy. Acneform rash occurred in 76–88% of 1373 patients receiving cetuximab in clinical trials. Severe acneform rash occurred in 1–17 % of patients. Acneform rash usually developed within the first two weeks of therapy and resolved in a majority of the patients after cessation of treatment, although in nearly half, the event continued beyond 28 days. Monitor patients receiving cetuximab for dermatologic toxicities and infectious sequelae. Instruct patients to limit sun exposure during cetuximab therapy. Acne-like rash (88%), nausea, vomiting (55%, 41%) tiredness or weakness (73%), diarrhea (72%), abdominal pain (55%) more serious side-effects are rarer, but include: infusion

reactions (3%), interstitial lung disease (0.5%), sepsis (3%), kidney dysfunction (2%), pulmonary embolism (1%).

Serious infusion reactions, requiring medical intervention and immediate, permanent discontinuation of cetuximab included rapid onset of airway obstruction (bronchospasm, stridor, hoarseness), hypotension, and/or cardiac arrest. Severe (NCI CTC Grade 3 and 4) infusion reactions occurred in 2–5% of 1373 patients in clinical trials, with fatal outcome in 1 patient.

Approximately 90% of severe infusion reactions occurred with the first infusion despite premedication with antihistamines. In patients evaluated during clinical trials, hypomagnesemia occurred in 55% of patients (199/365) receiving cetuximab and was severe (NCI-CTC Grade 3 and 4) in 6–17%. The onset of hypomagnesemia and accompanying electrolyte abnormalities occurred days to months after initiation of cetuximab. Periodically monitor patients for hypomagnesemia, hypocalcemia, and hypokalemia, during and for at least 8 weeks following the completion of cetuximab.

The safety of cetuximab in combination with radiation therapy and cisplatin has not been established. Death and serious cardiotoxicity were observed in a single-arm trial with cetuximab, radiation therapy, and cisplatin (100 mg/m^2) in patients with locally advanced SCCHN. Two of 21 patients died, one as a result of pneumonia and one of an unknown cause. Four patients discontinued treatment due to adverse events. Two of these discontinuations were due to cardiac events.

It is not known whether cetuximab, is secreted in human milk since IgG antibodies, such as cetuximab, can be excreted in human milk. Because many drugs are excreted in human milk and because of the potential for serious adverse reactions in nursing infants from cetuximab, a decision should be made whether to discontinue nursing or to discontinue the drug, taking into account the importance of the drug to the mother. If nursing is interrupted, based on the mean half-life of cetuximab, nursing should not be resumed earlier than 60 days following the last dose of cetuximab. [45, 50, 51]

Adverse Event (AE) related to the study is defined as any untoward medical occurrence in a patient or clinical investigation subject administered a pharmaceutical product and that does not necessarily have to have a causal relationship with this treatment. An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of ixabepilone whether or not considered related to ixabepilone.

During this clinical trial, adverse events can be spontaneously reported or elicited during open-ended questioning, examination, or evaluation of a subject. (In order to prevent reporting bias, subjects should not be questioned regarding the specific occurrence of one or more adverse events.)

A serious AE is any untoward medical occurrence that at any dose:

- results in death,
- is life-threatening (defined as an event in which the patient was at risk of death at the time of the event; it does not refer to an event which hypothetically might have caused death if it were more severe),
- requires inpatient hospitalization or causes prolongation of existing hospitalization,
- results in persistent or significant disability/incapacity,
- is a congenital anomaly/birth defect,
- results in the development of drug dependency or drug abuse,
- is an important medical event (defined as a medical event(s) that may not be immediately life-threatening or result in death or hospitalization but, based upon appropriate medical and scientific judgment, may jeopardize the patient or may require intervention (e.g., medical, surgical) to prevent one of the other serious outcomes listed in the definition above.) Examples of such events include, but are not limited to, intensive treatment in an emergency

room or at home for allergic bronchospasm; blood dyscrasias or convulsions that do not result in hospitalization.)

Also is considered the occurrences of pregnancy or overdose (regardless of adverse outcome), and cancer as events which must be reported as important medical events. [48]

10.3 Reporting of SAEs

Following the subject's written consent to participate in the study, all SAEs should be collected and reported, including those thought to be associated with clinical trial procedures. Following study completion, any SAE thought to be related to study drug or clinical trial procedures should also be reported to the sponsor: The Methodist Hospital Research Institute and the local IRB.

SAE terminology and severity grading will be based on (CTCAEv3).

The following categories and definitions of causal relationship to study drug should be considered for use for this clinical study.

- Certain: There is a known causal relationship between the study drug and the SAE. The event responds to withdrawal of study drug (dechallenge), and recurs with rechallenge when clinically feasible. (>95% certainty)
- Probable: There is reasonable causal relationship between the study drug and the SAE. The event responds to dechallenge. Rechallenge is not required. (65%-95% probability)
- Possible: There is reasonable causal relationship between the study drug and the SAE. Dechallenge information is lacking or unclear. (35%-65% probability of relatedness)
- Not likely: There is temporal relationship to study drug administration, but there is not a reasonable causal relationship between the study drug and the SAE. (5-35% probability of relatedness)
- Not related: There is not a temporal relationship to study drug administration (too early, or late, or study drug not taken), or there is known causal

relationship between the SAE and another drug, concurrent disease, or other circumstance. (<5% chance of relatedness)

- Adverse events classified as "serious" require expeditious handling and reporting to the sponsor (TMHRI) and local IRB to comply with regulatory requirements.
- All serious AEs whether related or unrelated to ixabepilone, must be immediately reported to the sponsor (TMHRI) by the investigator or designee within 24 hours of becoming aware of the event. If only limited information is initially available, follow-up reports are required. The original SAE form must be kept on file at the study site.

All SAEs should be faxed or emailed to TMHRI at:

The Methodist Hospital Research Institute
Fax Number: 713-793-7001
Email: mnsalazar@tmhs.org and afroehlich@tmhs.org

- For studies conducted under an Investigator IND, any event that is both serious and unexpected must be reported to the FDA as soon as possible and, in no event, later than 7 days (death or life-threatening event) or 15 days (all other SAEs) after the investigator's or institution's initial receipt of the information. The Methodist Hospital Research Institute will be provided with a simultaneous copy via facsimile of all adverse events filed with the FDA. SAEs should be reported on the MedWatch Form 3500A, which can be accessed at:

  http://www.accessdata.fda.gov/scripts/MedWatch/

  MedWatch forms should be mailed or faxed to the FDA at:

  MEDWATCH
  5600 Fishers Lane

Rockville, MD 20852-9787
Fax: 1-800-FDA-0178 (1-800-332-0178)

All SAEs are to be simultaneously be faxed to Bristol-Myers Squibb at:

Global Pharmacovigilance and Epidemiology
Bristol-Myers Squibb Company
Fax Number: 609-818-3804
Email:Worldwide.safety@bms.com

- Collection of complete information concerning SAEs is extremely important. Full descriptions of each event will be followed by the sponsor. Thus, follow-up information which becomes available as the SAE evolves, as well as supporting documentation (e.g., hospital discharge summaries and autopsy reports), should be collected subsequently, if not available at the time of the initial report, and immediately sent using the same procedure as the initial SAE report.
- An overdose is defined as the accidental or intentional ingestion of any dose of a product that is considered both excessive and medically important. For reporting purposes, the sponsor considers an overdose, regardless of adverse outcome, as an important medical event.
- AEs should be followed to resolution or stabilization, and reported as SAEs if they become serious. This also applies to subjects experiencing AEs that cause interruption or discontinuation of ixabepilone, or those experiencing AEs that are present at the end of their participation in the study; such subjects should receive post-treatment follow-up as appropriate.

In BMS supported trials, all SAEs must be collected which occur within 30 days of discontinuation of dosing or completion of the patient's participation in the study if the last scheduled visit occurs at a later time. In addition, the Investigator should notify BMS of any SAE that may occur after this time period which they believe to be certainly, probably or possibly related to ixabepilone. [48]

11. Data Management

An outcome database would be created for this specific project.

Using Excel (spread sheet) program

12. Statistical Design and Analyses

12.1 Sample size estimate

The published study CA163-080 has shown that the pathological complete response (pCR) rate is 18% based on a single agent Ixabepilone data, and is the same percentage that we assume for patients that will be treated with ixabepilone (Treatment Arm 1). With total 108 patients (54 in each arm), we will be able to detect 25% difference in pCR rates between the arm with Ixabelpilone plus Cetuximab (Treatment Arm 2) and Arm 1 with 81.9% power and a two-sided significance level of 0.05. A total of 116 patients will be enrolled to allow for 6.5% dropout before randomization of 108 patients. Table 1 provides power estimates to detect hypothesized 25%, 27%, 29%, and 31% difference in pCR rate in the treatment am 1 compared to the treatment arm 2 with given sample size of 54 patients in each arm.

Table 1. Power to detect different differences of pathological complete response rates in the two arms with given sample size of 54 patients in each arm.

Projected pathological complete response rate		Estimated power (%)
Treatment Arm 1	Treatment Arm 2	
18%	0.43%	81.9
	0.45%	87.0
	0.47%	91.0
	0.49%	94.0

12.2 Statistical analysis

All major treatment comparisons between the two randomized groups in this trial will be performed according to the principle of “intention-to-treat”, that is, subjects

will be analyzed according to the treatment arm to which patients were randomized, regardless of compliance to assigned treatment. Summary statistics will be provided for overall response rates and pCR rates by treatment arm. The different rates between treatment arms will be compared by Cochran-Mantel-Haenszel test. . Logistic regression models will be used to assess the association between expression of biomarkers (such as PTEN, EGFR and EGFR-pathway associated genes (including, but not limited to TGF-α, CRYAB, NRAS, KRAS, AKT3, PTEN, MEK1 and KRAS amplicon genes) from the baseline and the overall objective or pathologic response, controlled by treatment arm, age, and disease stage. The unpaired t-test will be applied to compare the difference of expression of biomarkers between the baseline and at surgery in the two arms. Benjamini and Hochberg adjustment will be used to adjust multiple comparisons in expressions of all marks. Safety and toxicity of each regimen will be tabulated by type and grade at the end of chemotherapy. . Radiographic efficacy assessment will be performed on all patients at the end of chemotherapy. Statistical significance is defined as 2-tailed p=0.05 level. Stata Version 10 (College Station, TX) will be used for all statistical analyses.

12.3 Interim data analysis

Safety

Interim data analysis for monitoring safety will be conducted at every cycle for the two arms. Severity of adverse effects will be reported to the Data Monitoring Committee as part of the interim study reports.

Interim data analysis for monitoring efficacy is not applicable for this protocol.

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