HER2 Gene Amplification Testing by Fluorescence in situ Hybridization (FISH): Comparison of the American Society of Clinical Oncology (ASCO)-College of American Pathologists (CAP) Guidelines with FISH Scores used for enrollment in Breast Cancer International Research Group (BCIRG) Clinical Trials

Press, et al

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BCIRG
Breast Cancer International Research Group

BCIRG 006

MULTICENTER PHASE III RANDOMIZED TRIAL COMPARING DOXORUBICIN AND CYCLOPHOSPHAMIDE FOLLOWED BY DOCETAXEL (AC→T) WITH DOXORUBICIN AND CYCLOPHOSPHAMIDE FOLLOWED BY DOCETAXEL AND TRASTUZUMAB (HERCEPTIN®) (AC→TH) AND WITH DOCETAXEL, CARBOPLATIN AND TRASTUZUMAB (TCH) IN THE ADJUVANT TREATMENT OF NODE POSITIVE AND HIGH RISK NODE NEGATIVE PATIENTS WITH OPERABLE BREAST CANCER CONTAINING THE HER2 ALTERATION.

Drug Name/Project Number: BCIRG 006
Sponsor Number: TAX GMA 302

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*Please note that the breast cancer recurrence substudy is contained in a document separate from the original BCIRG 006 protocol. References throughout the substudy protocol refer to specific sections outlined in the BCIRG 006 main protocol.

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TABLE OF CONTENTS

I STUDY SUMMARY - BCIRG 006

TITLE

A MULTICENTER PHASE III RANDOMIZED TRIAL COMPARING DOXORUBICIN AND CYCLOPHOSPHAMIDE FOLLOWED BY DOCETAXEL (AC→T) WITH DOXORUBICIN AND CYCLOPHOSPHAMIDE FOLLOWED BY DOCETAXEL AND TRASTUZUMAB (AC→TH) AND WITH DOCETAXEL, CARBOPLATIN AND TRASTUZUMAB (TCH) IN THE ADJUVANT TREATMENT OF NODE POSITIVE AND HIGH RISK NODE NEGATIVE PATIENTS WITH OPERABLE BREAST CANCER CONTAINING THE HER2 ALTERATION.

OBJECTIVES

Primary Objective

To compare disease-free survival after treatment with doxorubicin and cyclophosphamide followed by docetaxel (Taxotere®) (AC→T) with doxorubicin and cyclophosphamide followed by docetaxel and trastuzumab (Herceptin®) (AC→TH) and with docetaxel in combination with carboplatin and Herceptin® (TCH) in the adjuvant treatment of node positive and high risk node negative patients with operable breast cancer containing the HER2 alteration.

Secondary Objectives

- *For all patients*

To compare overall survival between the 3 above mentioned arms.

To compare cardiac toxicity between the 3 above mentioned arms.

To compare toxicity and quality of life between the 3 above mentioned arms.

To evaluate pathologic and molecular markers for predicting efficacy in these patient groups.

In addition, an independent socio-economic study will be conducted in parallel with the clinical study.

- *For patients having consented to the optional substudies*

To correlate baseline peripheral levels of the shed HER2 extracellular domain (ECD) with baseline FISH results and to determine whether peripheral levels of HER2 ECD measured at different timepoints constitute a prognostic and/or predictive factor of disease free survival and survival.

To evaluate genetic and biochemical markers for predicting risk of developing cardiac dysfunction and later cardiac events in these patient groups.

STUDY DESIGN AND DOSAGE REGIMEN

Prospective, non-blinded randomized phase III trial. Three thousand, one hundred and fifty patients (3,150 patients) will be post-surgically stratified at inclusion according to institution, nodal status (node negative, node positive 1-3 nodes, node positive 4 or more nodes), of hormonal receptor status (estrogen and/or progesterone receptor positive versus negative) and will be randomized to receive adjuvant therapy with either:

AC→T: Doxorubicin 60 mg/m² IV in combination with cyclophosphamide 600 mg/m² IV on an every 3 week basis for 4 cycles followed by docetaxel 100 mg/m² as 1 hour IV infusion on an every 3 week basis for 4 cycles.

AC→TH: Doxorubicin 60 mg/m² IV in combination with cyclophosphamide 600 mg/m² IV on an every 3 week basis for 4 cycles. Three weeks after the last cycle of AC, Herceptin® 4 mg/kg loading dose by IV infusion over 90 minutes on Day 1 of Cycle 5 will be administered, followed by Herceptin® 2 mg/kg by IV infusion over 30 minutes weekly starting Day 8; and docetaxel 100 mg/m² administered by IV infusion over 1 hour on Day 2 of Cycle 5, then on day 1 on an every 3 week basis for all subsequent cycles (total 4 cycles of docetaxel). Beginning three weeks after the last cycle of chemotherapy, Herceptin® 6 mg/kg by IV infusion over 30 minutes will be given every 3 weeks. Herceptin® will continue for 1 year from date of first administration i.e. Herceptin® administration will stop at 1 year from its initiation, regardless of the number of doses received or missed. For those cycles where docetaxel and Herceptin® are due to be administered on the same day, docetaxel to be administered first, except for the first cycle, when Herceptin® loading dose is given on day 1 and docetaxel on day 2.

TCH: Docetaxel / Carboplatin / Herceptin: Herceptin® 4 mg/kg loading dose by IV infusion over 90 minutes on Day 1 of Cycle 1 only, followed by Herceptin® 2 mg/kg by IV infusion over 30 minutes weekly starting on Day 8 until three weeks after the last cycle of chemotherapy. Docetaxel 75 mg/m² will be administered on Day 2 of Cycle 1, then on Day 1 of all subsequent cycles by IV infusion over 1 hour followed by carboplatin at target AUC = 6 mg/mL/min administered by IV infusion over 30-60 minutes repeated every 3 weeks. A total of six cycles of docetaxel and carboplatin will be administered every 3 weeks. Herceptin® will continue weekly during treatment with chemotherapy and then every 3 weeks during follow-up period for 1 year from date of first administration i.e. Herceptin® administration will stop at 1 year from its initiation, regardless of the number of doses received or missed. During the follow-up period, Herceptin® will be administered at 6 mg/kg by IV infusion over 30 minutes every 3 weeks. For those cycles where chemotherapy and Herceptin® are due to be administered on the same day, docetaxel will be administered first, followed by carboplatin followed by Herceptin® except for the first cycle, when Herceptin® loading dose is given on day 1 and docetaxel/carboplatin on day 2.

Dose reduction and/or treatment delay and treatment discontinuation are planned for the 3 arms in case of severe hematological and/or non-hematological toxicities.

Indication for Hormonal therapy

Tamoxifen (20 mg p.o. daily) for 5 years will be administered starting 3 to 4 weeks after the last course of chemotherapy for patients with positive estrogen and/or progesterone receptors.

Post menopausal patients are allowed to switch to Anastrozole in case of Tamoxifen related severe toxicities (e.g. hot flushes, vaginal bleeding, vaginal discharge, thromboembolic events). Anastrazole will be given at the dose of 1 mg daily. Of note, the total duration of the hormonal therapy, i.e. Tamoxifen followed by Anastrozole, should not exceed 5 years. Only post-menopausal patients are eligible to receive Anastrozole.

Post menopausal women without contraindications to the use of Tamoxifen and who have already started Tamoxifen, can receive a sequential therapy consisting of Tamoxifen for 2 to 3 years followed by Anastrozole or Exemestane for a maximum of 5 years of hormonal therapy.

Post menopausal patients who have not yet started hormonal therapy can receive 5 years of Anastrozole or sequential therapy consisting of Tamoxifen for 2 to 3 years followed by Anastrozole or Exemestane for a maximum of 5 years of hormonal therapy.

Post menopausal patients who have completed 5 years Tamoxifen are allowed to continue the hormonal treatment with letrozole for a maximum of 3 years.

Note: - the use of AIs should be accurately documented and reported on page FU6 of the CRF,

- the same approach is followed for all patients treated in the BCIRG 006/TAX GMA 302 study in order to avoid treatment imbalance between the 3 arms.
- Each center will be requested to provide BCIRG data center with their institution guideline related to the use of aromatase inhibitor by completing a questionnaire.

Radiation – Either Arm

Patients treated with lumpectomy will undergo postoperative radiation therapy after completion of chemotherapy and resolution of any side effect. Postmastectomy radiation therapy, and ipsilateral nodal radiation therapy, may be used at the discretion of the treating radiation oncologist. This will be done according to the guidelines at each institution. Radiation guidelines will be requested from each institution prior to study start at that institution.

Surgery, Axillary Lymph Node Assessment – Either Arm

Definitive surgical treatment must be either mastectomy with axillary lymph node involvement assessment or breast conserving surgery with axillary lymph node involvement assessment, for operable breast cancer (T1-3, clinical N0-1, M0). Margins of resected specimen from definitive surgery must be histologically free of invasive carcinoma and/or ductal carcinoma in situ. The finding of lobular carcinoma in-situ will not be scored as a positive margin. For positive lymph node involvement assessment, axillary lymph node dissection must be performed with at least one axillary lymph node (pN1) among a minimum of 6 resected lymph nodes. For negative lymph node involvement assessment, 0 (pN0) among a minimum of 6 resected nodes OR a negative sentinel node biopsy (pN0) may be used for the axillary lymph node assessment. Only centers with established experience in sentinel lymph node biopsy will be allowed to use such a procedure for the purpose of this study.

High Risk Node Negative Patients

High risk node negative patient will be defined as a patient with negative (pN0) lymph node involvement, and at least 1 of the following factors:
Tumor size > 2 cm, estrogen receptor and progesterone receptor status is negative, histologic and/or nuclear grade 2-3, or age < 35 years.

Estrogen and/or Progesterone Receptor Status

Hormonal receptor status on the primary tumor sample must be known prior to randomization. ER-positive tumors can be defined as positive by the dextran-coated charcoal or sucrose-density gradient method, or positive (using individual laboratory criteria) by the enzyme immunoassay method (EIA), or by immunocytochemical assay. Those not definitively negative i.e “borderline”, etc, will also be considered positive.

Patients whose tumor is estrogen receptor negative with progesterone receptor status unknown or undetermined, MUST have the PR assayed in order to determine hormonal receptor status.

Patients whose tumor is progesterone receptor negative with estrogen receptor status unknown or undetermined, MUST have the ER assayed in order to determine hormonal receptor status.

Shed HER2 extracellular domain (ECD) (optional)

Serum samples are to be collected at baseline, at end of chemotherapy for AC→TH and AC→T arms or 6 weeks after the end of chemotherapy visit for TCH arm (FUp1a), and then every 6 months during the first five years of follow-up or until disease relapse, withdrawn consent or death whichever comes first and, at the time of relapse.

Cardiac genetic and biochemical markers (optional) - *(not applicable for France)*

Blood sample is to be collected at baseline for cardiac genetic markers.
Plasma samples are to be collected at baseline, cycle 4, end of chemotherapy for AC→TH and AC→T arms or 6 weeks after the end of chemotherapy visit for TCH arm (FUp1a) and then every 6 months during the first three years of follow-up, at year 5 of follow-up (FU #14) and at any time of clinical evidence of cardiac failure. If the 60 months FUP visit has already happened for a patient when amendment 5 becomes available, plasma sample should be scheduled as soon as the patient has signed her informed consent, at time of LVEF8.

PROPHYLACTIC PREMEDICATION REGIMEN

Docetaxel Prophylaxis

Patients receiving docetaxel in either of the arms AC→T or AC→TH or TCH will receive the following prophylactic premedication:

Dexamethasone 8 mg (or equivalent) orally for 6 doses

1. Night before chemotherapy
2. Morning of chemotherapy
3. 1 hour before docetaxel infusion (may be given orally or intravenous)
4. Night of chemotherapy
5. Morning the day after chemotherapy
6. Evening the day after chemotherapy

Antiemetic Prophylaxis

Antiemetic prophylaxis is mandatory for all patients. Selection of antiemetics is at the discretion of the investigator.

SELECTION OF PATIENTS

A INCLUSION CRITERIA

1 Written informed consent prior to beginning specific protocol procedures, including expected cooperation of the patients for the treatment and follow-up, must be obtained and documented according to local regulatory requirements.

2. Histologically proven breast cancer with an interval between definitive surgery that includes axillary lymph node involvement assessment and registration of less than or equal to 60 days. If the definite surgery and the axillary node dissection are performed in two different days, both days should be within the 60 days window. A central pathology review may be performed post randomization for confirmation of diagnosis and molecular studies. The same block used for HER2 determination prior to randomization may be used for the central pathology review. See Appendix 3* for details on this process.

3 Definitive surgical treatment must be either mastectomy **with** axillary lymph node involvement assessment, or breast conserving surgery **with** axillary lymph node involvement assessment for operable breast cancer . Margins of resected specimen from definitive surgery must be histologically free of invasive adenocarcinoma and/or ductal carcinoma in situ (DCIS). The finding of lobular carcinoma in-situ will not be scored as a positive margin.

4 Patients must be either lymph node positive or high risk node negative. Lymph node positive patients will be defined as patients having invasive adenocarcinoma with at least one axillary lymph node (pN1) showing evidence of tumor among a minimum of six resected lymph nodes.
High risk lymph node negative patients will be defined as patients having invasive adenocarcinoma with either 0 (pN0) among a minimum of 6 resected lymph nodes or negative sentinel node biopsy (pN0) AND at least one of the following factors: tumor size > 2 cm, ER and PR status is negative, histologic and/or nuclear grade 2-3, or age < 35 years.

5 Tumor must show the presence of the HER2 gene amplification by Fluorescence In-Situ Hybridization (FISH analysis) by a designated central laboratory (see Appendix 3 for complete details).

6. Estrogen and/or progesterone receptor analysis performed on the primary tumor prior to randomization. Results must be known at the time of randomization.

(Note: Patients whose tumor is estrogen receptor negative with progesterone receptor status unknown or undetermined, MUST have the PR assayed in order to determine hormonal receptor status. Patients whose tumor is progesterone receptor negative with estrogen receptor status unknown or undetermined, MUST have the ER assayed in order to determine hormonal receptor status.)

7 Age ≥ 18 years and age ≤ 70 years. The upper age limit is not meant to be exclusionary but rather is based on the lack of safety data for the TCH regimen in women >70 years of age.

8 Karnofsky Performance status index ≥ 80%.

9 Normal cardiac function must be confirmed by LVEF (echocardiography or MUGA scan) and ECG within 3 months prior to registration. The result of the echocardiography or MUGA must be equal to or above the lower limit of normal for the institution.

10 Laboratory requirements: (within 14 days prior to registration)

a) Hematology:
   i) Neutrophils ≥ 2.0 10^9/L
   ii) Platelets ≥ 100 10^9/L
   iii) Hemoglobin ≥ 10 g/dL

b) Hepatic function:
   i) Total bilirubin ≤ 1 UNL
   ii) ASAT (SGOT) and ALAT (SGPT) ≤ 2.5 UNL
   iii) Alkaline phosphatase ≤ 5 UNL
   iv) Patients with ASAT and/or ALAT > 1.5 x UNL **associated** with alkaline phosphatase > 2.5 x UNL are not eligible for the study.

c) Renal function:
   i) Creatinine ≤ 175 µmol/L (2 mg/dL)
   ii) If creatinine between 140 and 175 µmol/L, the calculated creatinine clearance should be ≥ 60 mL/min.

11 Complete staging work-up within 3 months prior to registration. All patients will have contralateral mammography and/or ultrasound (mammogram is preferred), chest X-ray (PA and lateral) and/or CT and/or MRI, abdominal ultrasound and/or CT scan and/or MRI, and bone scan. In cases of positive bone scans, bone X-ray or bone MRI evaluation is mandatory to rule out the possibility of metastatic bone scan positivity. Other tests may be performed as clinically indicated (see appendix 5).

12 Patients must be accessible for treatment and follow-up. Patients registered on this trial must be treated and followed at participating centers which will include Principal or Co-investigator's sites.

13 Negative pregnancy test (urine or serum) within 7 days prior to registration for all women of childbearing potential.

B EXCLUSION CRITERIA

1 Prior systemic anticancer therapy for breast cancer (immunotherapy, hormonotherapy, chemotherapy).

2 Prior anthracycline therapy, taxoids (paclitaxel, docetaxel) or platinum salts for any malignancy.

3 Prior radiation therapy for breast cancer.

4 Bilateral invasive breast cancer.

5 Pregnant, or lactating patients. Patients of childbearing potential must implement adequate non-hormonal contraceptive measures during study treatment (chemotherapy, Herceptin® and tamoxifen therapy) and must have negative urine or serum pregnancy test within 7 days prior to registration.

6 Any T4 or N2 or known N3 or M1 breast cancer.

7 Pre-existing motor or sensory neurotoxicity of a severity ≥ grade 2 by NCI criteria.

8 Cardiac disease that would preclude the use of doxorubicin, docetaxel and Herceptin®.
- a) any documented myocardial infarction
- b) angina pectoris that requires the use of antianginal medication
- c) any history of documented congestive heart failure
- d) Grade 3 or Grade 4 cardiac arrhythmia (NCI CTC, version 2.0)
- e) clinically significant valvular heart disease
- f) patients with cardiomegaly on chest x-ray or ventricular hypertrophy on ECG, unless they demonstrate by echocardiography or MUGA scan within the past 3 months that the LVEF is ≥ the lower limit of normal for the radiology facility;
- g) patients with poorly controlled hypertension i.e. diastolic greater than 100 mm/Hg. (Patients who are well controlled on medication are eligible for entry
- h) patients who currently receive medications (digitalis, beta-blockers, calcium channel-blockers, etc) that alter cardiac conduction, if these medications are administered for cardiac arrhythmia, angina or congestive heart failure. If these medications are administered for other reasons (ie hypertension), the patient will be eligible.

9 Other serious illness or medical condition:
- a) history of significant neurologic or psychiatric disorders including psychotic disorders, dementia or seizures that would prohibit the understanding and giving of informed consent
- b) active uncontrolled infection
- c) active peptic ulcer, unstable diabetes mellitus
- d) patients with symptomatic, intrinsic lung disease resulting in dyspnea

10 Past or current history of neoplasm other than breast carcinoma, except for:
- a) curatively treated non-melanoma skin cancer
- b) in situ carcinoma of the cervix
- c) other cancer curatively treated and with no evidence of disease for at least 10 years
- d) ipsilateral ductal carcinoma in-situ (DCIS) of the breast
- e) lobular carcinoma in-situ (LCIS) of the breast
- f) DCIS involving the contralateral breast removed by mastectomy

11 Current therapy with any hormonal agent such as raloxifene, tamoxifen, or other selective estrogen receptor modulators (SERMs), either for osteoporosis or prevention of breast cancer. Patients must have discontinued these agents prior to randomization.

12 Chronic treatment with corticosteroids **unless** initiated > 6 months prior to study entry **and** at low dose (≤ 20 mg methylprednisolone or equivalent).

13 Concurrent treatment with ovarian hormonal replacement therapy. Prior treatment must be stopped prior to randomization.

14 Definite contraindications for the use of corticosteroids.

15 Concurrent treatment with other experimental drugs. Participation in another clinical trial with any investigational not marketed drug within 30 days prior to study entry.

16 Concurrent treatment with any other anti-cancer therapy.

17 Male patients, as no clinical efficacy or safety data are available from phase I-II studies.

NUMBER OF PATIENTS / ENROLLMENT & FOLLOW-UP PERIOD

Sample Size:	Total of 3,150 patients will be randomized (1, 050 patients per treatment arm)
Enrollment start:	March 2001
Enrollment stop:	March 2004
Clinical Follow-up:	10 years
1st interim analysis:	300 events
2nd interim analysis:	450 events
3rd interim analysis:	650 events
Main analysis:	900 events
First follow-up analysis:	3 years after main analysis
Second follow-up analysis:	5 years after main analysis

DURATION OF TREATMENT

All included patients in each arm will receive a fixed number of cycles of treatment.

AC → T: AC x 4 → T x 4
AC → TH: AC x 4 → T x 4 with Herceptin® x 1 year
TCH: TC x 6 cycles with Herceptin® x 1 year

EFFICACY EVALUATION

An intent-to-treat (ITT) analysis will be conducted for all randomized patients. In addition, an analysis will be conducted among the eligible patients.

Disease-Free Survival (DFS) is defined as the interval from the date of randomization to the date of local, regional or metastatic relapse or the date of second primary cancer (with the exception of curatively treated non-melanoma skin cancer or in situ carcinoma of the cervix – see Exclusion Criteria 10a, 10b) or death from any cause whichever occurs first (see section 8.3.1).

Survival will be measured from the date of randomization to the date of death of any cause.

STATISTICAL CONSIDERATIONS

The sample size determination was done based on the following assumptions:
The primary objective of this trial is to show that the treatments under investigation differ in terms of disease-free survival (DFS).
The following assumptions were initially made:

- the proportions of patients who have no axillary lymph nodes involved (N0), 1 to 3 axillary lymph nodes involved (N1-3) and 4 or more axillary lymph nodes involved trial (N4+) will be, respectively, 20%, 50% and 30%

- the DFS at 5 years of patients receiving AC→T in these strata are, respectively, equal to about 67%, 57% and 42%
- the overall DFS of all patients receiving AC→T will be equal to about 55% (20% x 67% + 50% x 57% + 30% x 42%)
- it is of clinical interest to detect a 7% improvement in 5-year DFS (i.e. an increase from 55% to 62%)
- the overall error rate for a false positive outcome (α) is set to 5%, using two-sided significance tests. Since the three pairwise treatment comparisons will be of interest in the final analysis, the error rate for each comparison is set at a conservative level of 0.017
- the error rate for a false negative outcome (β) is set to 20% i.e the power of the trial is set to 80% for the difference of clinical interest

A total of 3,150 patients are necessary to have sufficient power to compare AC→T with AC→TH with TCH for all randomized patients, assuming an anticipated ineligible rate of 3%. 1, 050 patients will be necessary in each treatment group. This sample size calculation takes into account the fact that 1 interim analysis will be performed when 50% of the events (654 events) have been observed and the final analysis will take place when a total of 1,308 events have taken place among all patients.

The randomization will be centralized and stratified at time of inclusion for institution, for node status: node negative, node positive 1-3 nodes and node positive ≥ 4 or more, and for hormonal receptor status (estrogen and/or progesterone receptor positive versus negative)

The group sequential design, according to Peto's method, will be used to a significance level of 0.001 for the interim analysis. This allows the use of an unadjusted level of 0.05 for the final analysis.

With the sample size as stated in the primary endpoint, this trial has 80% power (at a significance level of 0.05) to detect an absolute difference in overall survival of approximately 5%. If overall survival at 5 years in the control arm is between 70% and 80%, the detectable difference in one of the experimental arms is between 5.5% and 4.7%, respectively. This translates into a reduction in relative risk between 21% and 26%.

Updated data on DFS from BCIRG 001 study (TAC arm) have been used to modify the number of events required for interim and final analyses, by assuming 7% absolute advantage in 5 years DFS in favor of one of the Herceptin containing regimen with a power of 80% and $\alpha = 0.05$ as originally planned.

Among the node positive and Her 2 neu positive by FISH patients treated with TAC in the BCIRG 001 trial, 73% of patients were disease free at a median follow-up of 55 months which translates into an estimated 5 year Disease Free Survival of 70% among the patients randomized to the arm without Herceptin, i.e., AC followed by T. As a consequence, the revised calculations are based on a presumed 5 year DFS of 70% in AC-T treated patients.

Additional interim analyses have also been added and a "step-down" testing procedure has been proposed in order to compare the control arm (AC-T) to each Herceptin-containing arm (AC-TH and TCH) at a level equal to $\alpha / 2$ to account for multiple testing, if both of these comparisons reach statistical significance, then compare the two Herceptin-containing arms at level α, otherwise stop.

The revised schedule for conducting the analyses is the following: three interim analyses will be conducted when respectively 300, 450 and 650 events have been observed; and the main analysis will take place when 900 events are observed. In order to avoid any confusion, the previously called "final" analysis is now called the "main" analysis, to reflect the fact that two follow-up analyses will be performed after this analysis.

With the revised assumptions as well as the final number of randomized patients of 3,222, the trial is powered to detect a 7% difference between the control arm and the Herceptin-containing arm, assuming the 5-year DFS in the control arm (AC-T) is 70% [i.e. a 23.7% reduction in relative risk].

With the sample size as stated in the primary endpoint, this trial has 80% power (at a significance level of 0.05) to detect an absolute difference in the 5 years overall survival of approximately 5%. If overall survival at 5 years in the control arm is between 70% and 80%, the detectable difference in one of the experimental arms is between 5.5% and 4.7%, respectively. This translates into a reduction in the relative risk between 21% and 26%.
If the overall survival at 5 years in the control arm is between 80 and 90%, the detectable difference in one of the experimental arm is between 4.7% and 3.4%, respectively. This translates into a reduction in the relative risk between 26% and 35%.

Cardiac Safety Analysis

A secondary objective of the trial is to compare the cardiac toxicity between the 3 arms, in order to determine the safety of Herceptin® with these chemotherapy regimens.

Evaluable Patients for the Cardiac Safety Analysis: All patients randomized to the study must have a normal baseline exhocardiography or MUGA to be eligible. All patients randomized to the study, with the required normal baseline echocardiography or MUGA will be considered evaluable for the cardiac safety evaluation.

Timing of Analyses: Analyses of cardiac toxicity will take place when 100 randomized patients per arm, 300 randomized patients per arm, and 500 randomized patients per arm, as well as all patients randomized in the study, respectively and on an intent-to-treat basis, have been followed up and including the timing of LVEF5. LVEF5 corresponds to the follow-up #1 (this corresponds to 3 months after the end of chemotherapy visit for the AC-T and AC-TH arms, and 4 ½ months after the end of chemotherapy visit for the TCH arm).

At each of these analyses, cardiac deaths, congestive heart failure events, grade 3 or 4 arrhythmias, grade 3 or 4 cardiac ischemia / infarction events, and asymptomatic left ventricular decreases will be reviewed and assessed as outlined in the statistical section 8.4.

At each analysis, the two-tailed significance level of each analysis will be set at 0.05. This level of significance is not adjusted to take repeated analyses into account, and hence it will be merely indicative of a potential increase in incidence that needs to be scrutinized by the IDMC. Assuming a baseline incidence of cardiac deaths and symptomatic cardiac events of 1% in the control arm, the analyses will have approximately the following power to detect a difference of at least 4% in either treatment arm: 40% with 300 patients, 80% with 900 patients, 95% with 1,500 patients and >99.9% with the 3,222 patients randomized in the study. The statistical power to detect a 4% difference would be slightly higher than these figures should the baseline incidence be lower than 1%, and slightly lower than these figures should the baseline incidence be higher than 1%.

Scheduled LVEFs have been planned for all randomized patients in order to evaluate asymptomatic changes in left ventricular ejection fraction from baseline (see section VI). The data on asymptomatic decreases in left ventricular ejection fraction will be collected and reviewed at each of the assessment timelines defined above. Clinically significant asymptomatic cardiac abnormality is an absolute decline of LVEF of > 15 % points from baseline and a value below LLN.

In addition, there will be an on-going safety evaluation through the Serious Adverse Event reporting system. If an unexpected high incidence of cardiotoxicity is observed prior to the analyses mentioned above, an IDMC meeting will be called to evaluate.

The IDMC will recommend the Steering Committee to discontinue one treatment arm according to the cardiac toxicity incidence and/or the data collected from the asymptomatic decrease in LVEF. In this case, the trial will continue until accrual reaches the target sample size of 1,050 patients in each of the remaining two other treatment arms. Since in this case there will be no need to carry out pairwise comparisons, the final analysis will use an unadjusted level of significance of 5% and will have a power of 90% (instead of 80%) to detect the clinically relevant difference of 7% between the two treatment arms being compared.

II INTRODUCTION AND BACKGROUND

2.1 Introduction

Breast cancer is a leading cancer site in women around the world. More than 796,000 new cases (21% of all cancer sites) were diagnosed in 1999 with 314,000 reported deaths in women (14.1%). In the United States, 182 800 new cases (30.4% of all cancers in US women) and 40,800 deaths (15.2% of all cancer deaths) are estimated to occur in the year 2000 [1]. In Canada, an estimated 31, 000 new cases of breast cancer will be diagnosed (30.7% of all cancer) with an estimated 8200 deaths from breast cancer (18.8% of all cancer) for the year 2000 [2]. In the European community, an estimated 135 000 new cases per year (24% of all cancer cases) and 58 000 recorded deaths per year (18% of all cancer deaths) are reported [3].

Surgery is the main modality of treatment in patients with breast cancer. Surgery and/or radiotherapy can control local-regional disease in the majority of patients. However, more than 60% will ultimately die due to widespread disease [4].

In the past 10 years, adjuvant hormonal or cytostatic treatment has been increasingly used [5]. Ongoing studies show that adjuvant treatment can prolong time to recurrence and probably survival in some subsets of patients [6,7].

2.2 Role of Systemic Therapy in Adjuvant Treatment of Breast Cancer

Adjuvant systemic therapy is defined as the administration of chemotherapy or hormonal therapy after primary surgery for breast cancer in order to control clinically occult micro-metastases.

2.2.1 Adjuvant Chemotherapy

A number of chemotherapy protocols have shown effectiveness in the adjuvant setting of breast cancer. The most optimal regimen has not yet been identified. Several regimens represent acceptable alternatives [8]. They range from various CMF
(cyclophosphamide, methotrexate, 5-FU) chemotherapy regimens to anthracycline containing regimens such as AC (doxorubicin, cyclophosphamide), CAF (cyclophosphamide, doxorubicin, 5-FU), FAC (5-FU, doxorubicin, cyclophosphamide), AVCF (doxorubicin, vincristine, cyclophosphamide, 5-FU), or FEC (5-FU, epirubicin, cyclophosphamide)[9-15]. Numerous randomized trials have compared CMF regimens or variations to anthracycline containing polychemotherapies including FAC [11], initially developed by the CALGB (Cancer Leukemia Group B).

The impact of anthracycline-containing polychemotherapy appears real, but modest when compared to other regimens in the adjuvant setting [11-15]. Overall, in both node-negative and node-positive patients, as confirmed by the update of the meta-analysis performed by the Early Breast Cancer Trialists' Collaborative Group, adjuvant chemotherapy significantly improves disease-free and overall survival in young patients and to a lesser extent in older patients [7].

Among the novel chemotherapeutic drugs introduced in the 1990's (taxanes, vinorelbine, gemcitabine, 5-fluorouracil prodrugs…), the taxanes have emerged as very effective compounds and the available results suggest that they will be remembered in the future as the breast cancer chemotherapy of the 1990's. However, the impact of the taxanes on the natural history of breast cancer is yet to be defined, despite the trend of results suggesting that these agents could have the potential for significant advancement in the management of breast cancer.

Two different strategies have been pursued in assessing the potential role of taxanes in adjuvant setting and have led to several large phase III trials. The first strategy is related to the concept of sequential chemotherapy for which both paclitaxel and docetaxel are being investigated. This has led to testing protocols such as the CALGB ATC program (A, doxorubicin followed by paclitaxel followed by C, cyclophosphamide) or AC followed by paclitaxel or docetaxel (CALGB; NSABP, National Surgical Adjuvant Breast and Bowel Project) or AT (docetaxel) followed by CMF (Breast Adjuvant Study Team and IBCSG/ International Breast Cancer Study Group) or FEC (5-fluorouracil, epidoxorubicin, cyclophosphamide) followed by docetaxel (French Cooperative Group). The second strategy follows the classical polychemotherapy concept

for which quasi-exclusively docetaxel-based combinations are being studied due to the enhancement of doxorubicin-related cardiotoxicity observed when paclitaxel is administered with doxorubicin [16,17]. Protocols such as TAC (docetaxel, doxorubicin, cyclophosphamide) at doses 75/50/500 mg/m² have been compared to FAC (Breast Cancer International Research Group, BCIRG trial 001) in patients with node positive breast cancer or AT (docetaxel) at doses 60/60 mg/m² to AC (Eastern Cancer Oncology Group, ECOG) in patients with high risk node negative or 1-3 positive nodes.

Published results to date comparing a taxane-containing regimen to standard chemotherapy as adjuvant therapy in primary breast cancer come from the CALGB trial 9344 presented in 1998 [18]. The results suggest that the sequential addition of paclitaxel to AC may offer improved overall survival and disease-free survival in patients with node-positive primary breast cancer. These results were based on a median follow-up of 18 months in 3170 randomized patients. At this first pre-planned interim analysis (450 events), the sequential addition of paclitaxel reduced the recurrence rate by 22% (p=0.0077) and the death rate by 26% (p=0.0390). However, this trial has been criticized for its short follow-up and the fact that it compared 4 cycles of chemotherapy (AC) versus 8 cycles (AC followed paclitaxel). An updated analysis at 52 months follow-up was recently presented at the NIH Consensus Development Conference. Results confirmed the statistically significant advantage of AC followed by Taxol® in terms of disease-free survival, but overall survival was no longer significantly different than with AC. Of note, the larger advantage in favor of AC followed by Taxol® was observed in ER negative patients. [6].

Recently, results from the NSABP-B28 study comparing 4 cycles of AC (60/600) to 4 cycles of AC followed by 4 cycles of paclitaxel (225) were presented at the NIH Consensus Conference in Washington [6]. Based on a median follow-up of 34 months, there was no statistical difference between the two arms either in terms of survival (133 deaths on the control arm, 136 on the treatment arm, relative risk 1.00, 95 percent CI=[0.78 to 1.27], p=0.98) or in DFS (282 events on the control arm, 269 on the treatment arm, relative risk =0.93 percent CI=[0.78 to 1.10], p=0.38). However, a strong trend in favor of AC followed by Taxol® was observed in patients not receiving Tamoxifen who are ER or PgR negative [6].

Preliminary results from the MD Anderson Study comparing 8 cycles of FAC to 4 cycles of paclitaxel followed by 4 cycles of FAC in 524 adjuvant patients with operable breast cancer has shown no difference in DFS or OS with a median follow-up of 36 months to date [19]. The small sample size in this study may have attributed to no statistical difference being observed.

The negative results from the NSABP B28 together with the latest results from the CALGB 9344 and MDACC study, keep open the question as to which is superior, the combination or sequence in the adjuvant setting. The sequential AC followed by a taxane regimen can be considered as a solid standard option for current Herceptin® studies.

In 2000, the first generation of adjuvant trials comparing taxane-anthracycline containing combination to classical anthracycline-containing polychemotherapy are either completed or nearing completion. The trend is now to open the second generation of adjuvant trials with taxanes, in which both arms contain taxanes. The American Intergroup is using the sequential approach and is asking a taxane question, comparing AC followed by either paclitaxel or docetaxel (weekly or q3weekly). The sequential strategy is being directly compared to the polychemotherapy strategy by NSABP with the B30 trial: AC (60,600 mg/m²) x 4 followed by docetaxel (100 mg/m²) x 4 vs AT [60/60 mg/m²] x 4 vs TAC [60/60/600 mg/m²] x 4). In this program, a sequence with 8 courses is being compared to 4 courses of docetaxel-doxorubicin based polychemotherapy using the doublet-based docetaxel / doxorubicin at 60/60 mg/m² (favoring the increase dose of doxorubicin with 60 mg/m² instead of 50 mg/m² and decreasing the dose of docetaxel from 75 mg/m² to 60 mg/m²). In parallel to this present trial, BCIRG is conducting a randomized trial (BCIRG 005) comparing the same sequence used in the NSABP B30 trial (AC (60,600 mg/m²) x 4 followed by docetaxel (100 mg/m²) x 4) to TAC [75/50/500 mg/m²] given 6 times in the adjuvant treatment of patients with positive axillary nodes and no amplification of the HER2 gene.

Considering the importance of the HER2 as a prognostic and predictive factor in breast cancer and the recent development of trastuzumab (Herceptin®), the humanized monoclonal antibody that selectively targets the extracellular domain of the human epidermal growth factor receptor 2 protein (HER2), new adjuvant studies are planned for patients who overexpress or amplify HER2. The NSABP-B31 study is comparing 4 cycles of AC (60, 600 mg/m²) followed by 4 cycles of paclitaxel 175 mg/m² to AC (60,600 mg/m²) followed by 4 cycles of paclitaxel 175 mg/m² with weekly Herceptin® for 1 year. The second American study, from the Intergroup, is comparing four cycles of AC followed by four cycles of paclitaxel (doses as above) to four cycles of AC followed by weekly paclitaxel with Herceptin® for 1 year to four cycles of AC followed by four cycles of paclitaxel every 3 weeks with Herceptin®. The proposed BCIRG 006 study is comparing four cycles of AC followed by four cycles of docetaxel (100 mg/m²) to AC followed by docetaxel (100 mg/m²) with Herceptin® for 1 year versus the combination of 6 cycles of docetaxel (75 mg/m²) and carboplatin (AUC 6 mg/mL/min) with Herceptin® for 1 year (see study summary).

2.2.2 Adjuvant Hormonotherapy

The role of adjuvant hormonotherapy has also been addressed by the NIH Consensus Development Conference Statement [6] and the Early Breast Cancer Trialists Cooperative Group [7]. Adjuvant hormonal therapy should be recommended to women whose breast tumors contain hormone receptor protein, regardless of age, menopausal status, involvement of axillary lymph nodes, or tumor size. While the likelihood of benefit correlates with the amount of hormone receptor protein in tumor cells, patients with any extent of hormone receptor in their tumor cells may still benefit from hormonal therapy. Such treatment has led to substantial reductions in the likelihood of tumor recurrence, second primary breast cancer, and death persisting for at least 15 years of follow-up.

2.3 Rationale for the use of Herceptin®

2.3.1 Human Epidermal Growth Factor Receptor 2 (HER2)

Growth factors and their receptors are known to play critical roles in development, cell growth, and differentiation. In almost all human tissues a number of these receptors possess intrinsic tyrosine kinase activity that is activated upon interaction of the receptor with its cognate ligand. Abnormal expression of human epidermal growth factor receptor 2 (HER2) is frequently observed in a number of primary human tumors, suggesting that the over expression of this growth factor receptor may contribute to transformation and tumorigenesis. In most of these cases, pathologic HER2 protein overexpression is thought to result from gene amplification and has been correlated with poor clinical outcome in patients with both breast and ovarian cancers. Approximately 25% to 30% of patients with breast and 8-11% of ovarian cancers over express HER2. [20-22] Similar correlations may exist for gastric cancers, non small cell lung cancers, and adenocarcinoma of the salivary gland and endometrium [23].

2.3.1.1 Antibodies Against HER2

Murine monoclonal antibodies (muMAbs) against HER2 were produced to study the basic biology of this protein. Those antibodies directed against the extra cellular domain of the HER2 receptor were shown to inhibit the proliferation of human tumor cells over expressing pI85HER2 [24]. The most encouraging results were obtained with muMAb *4D5,* which produced significant anti proliferative effects *in vitro* against mammalian cancer cells including human breast cell lines that over express the HER2 receptor [23]. muMAb 4D5 has no effect on cell lines that do not over express the receptor [25-29].

Pre clinical *in vivo* studies with muMAb 4D5 were conducted using both human breast and ovarian cancer hetero transplants
from surgically excised human tumor specimens [29,30].The tumors were characterized to determine which had amplification and over expression of the HER2 gene. Results of these studies again established a clear antiproliferative effect against those human tumors characterized by over expression of the HER2 receptor. No effect was seen on tumor xenografts that did not over express the receptor.

Herceptin®, the humanized version of muMAb 4D5, was engineered by inserting the complementarity determining regions (CDRs) of muMAb 4D5 into the framework of a consensus human IgG [26]. Herceptin® binds the extra cellular domain of the HER2 receptor with 3 times greater affinity than does muMAb 4D5. Herceptin® is comparable to muMAb 4D5 in blocking cell proliferation; however, unlike muMAb 4D5, it induces antibody dependent cellular cytotoxicity (ADCC) against tumor cell lines in the presence of human peripheral blood mononuclear cells (PBMCs).

Toxicology studies were conducted in mice and rhesus monkeys (monkeys were chosen as the primary species for toxicology trials because Herceptin® binds to the rhesus monkey HER2 receptor). No drug-related effects were observed.

2.3.1.2 Methods to Detect HER2 Overexpression

Pathologic HER2 overexpression in human breast cancers is almost invariably due to amplification of the HER2 gene [31, 32]. Less than 5% of breast cancers show strong overexpression without gene amplification. Currently, the most frequently performed assay for assessment of HER2 status is immunohistochemistry. However, HER2 immunohistochemistry, especially as performed with FDA-approved assays, is known to have a high rate of both false-positive and false-negative results which are at least in part dependent on the subjective interpretation of the assay in different laboratories. More over, recent data have shown that patients amplifying the oncogene HER2 (FISH positive) were the only ones likely to benefit from Herceptin® based therapy [33]. The relationship between HER2 overexpression and c-erbB2 amplification as measured by FISH was analysed using 623 clinical specimens with a forced 1:1 ratio of positive (2+/3+) to negative (0/1+) results by the Clinical Trials Assay (CTA), 317 CTA+ and 306 CTA- [33]. These specimens were then analyzed by FISH test. The amplification rates by CTA score were 3+, 89.3%; 2+, 23.9%; 1+, 6.7%; 0, 4.2%. The overall 2x2 concordance between the CTA and FISH was 81.3%. The relationship between c-erbB2 amplification status and Herceptin® clinical benefit was then evaluated in 3 pivotal trials. In study H0648g (see also paragraph 2.3.2), the addition of Herceptin® to chemotherapy (AC or paclitaxel) resulted in a response rate of 54% versus 31% with chemotherapy alone and a 30% increase in median survival (26 months versus 20 months) for the FISH positive subgroup. The FISH negative subgroup showed no improvement in response rate (38% versus 38%) and no improvement in survival. In H0649g, response rate in the FISH positive subgroup was 19%. No responses were seen in the FISH negative subgroup including 17 patients demonstrating a 3+ CTA score. In H0650g, the FISH positive subgroup showed a 34% response rate while the FISH negative subgroup demonstrated a 7% response rate (2 patients) [34, 35]. Based on these data, we require that a tumor sample be forwarded to one of the designated central laboratories for FISH determination prior to randomization.

Serum assays for detecting levels of shed extracellular domain (ECD) of HER2 in peripheral blood for comparison with the FISH analysis as a predictive factor in determining a patient's outcome with Herceptin® containing regimens are currently being investigated [36, 37] although results are preliminary at this time. This may also prove to be an efficient means of testing for HER2 and thereby targeting those patients who will benefit most from Herceptin®. We have thus decided to propose an optional substudy to collect serum samples at baseline and during follow-up in order to test the levels of shed ECD of HER2 and to correlate them with the clinical outcome.

2.3.2 Clinical Studies of Herceptin®

Three phase I and 2 phase II studies of Herceptin® have been completed. In the first phase I trial (H0407g), 16 patients with HER2-overexpressing tumors received a single intravenous (IV) dose, ranging from 10 to 500 mg, of Herceptin®. Two patients developed chills during the infusion, and fever developed in 4 patients (1 in each of the dose groups) [38].

In the second phase I trial (H0452g), 17 patients received 8 weekly IV doses, ranging from 10 to 500 mg, of Herceptin®. Adverse events reported were not unexpected given the study population, and no clinically significant severe adverse events were attributed to Herceptin® [38].

In the third phase I trial (H0453g), 15 breast cancer patients received 9 weekly IV doses, ranging from 10 to 500 mg, of Herceptin®, and 3 doses of cisplatin, 100 mg/m², every 4 weeks. Two patients discontinued the study because of adverse events, 1 because of grade 3 renal toxicity and grade 4 thrombocytopenia, and the other because of grade 3 renal toxicity.

As would be expected in a population receiving cisplatin, episodes of nausea and vomiting occurred frequently. Decreased auditory acuity was reported by 10 patients and required discontinuation of cisplatin in 1 patient. There was no clear relationship between Herceptin® and the incidence of any of these adverse events. Administration of cisplatin did not affect the pharmacokinetics of Herceptin®. Although the small number of patients and the lack of randomization preclude any statement being made about a possible dose effect in tumor responses, 4 of 6 patients in the 250 mg and 500 mg dose groups had objective responses with 3 partial responses (PR, defined as a ≥50% reduction in tumor burden) and one CR which has been durable for greater than 8 years [38].

In a phase II trial of Herceptin® (HO551g), the antibody alone was given to 46 patients with metastatic breast cancer over expressing HER2 received a 250 mg IV loading dose, followed by a 100 mg IV weekly dose for 10 weeks. Twenty-one patients with responses or stable disease at Day 77 entered a maintenance program. Herceptin® was well tolerated. The median time to disease progression was 2.8 months. Of the 43 evaluable patients, *5* (12%) had either a complete response (CR) or PR, 16 (37%) had a minor response or stable disease, and the remaining 22 (51%) had progressive disease. The duration of the 5 responses (1 CR, 4 PRs) ranged from 1-28 months [39].

The results of the initial phase II study have been confirmed in a large multinational phase II trial (H0649g). In this study, 222 women with metastatic breast cancer who had failed 1 or 2 cytotoxic chemotherapy regimens were enrolled. Patients received a loading dose of 4 mg/kg of Herceptin® IV, followed by weekly IV administration of 2 mg/kg. The overall response rate in this study was 15%. The median duration of response was 8 months [40].

In another phase II trial (H0552g) conducted in metastatic breast cancer, 39 patients received a 250 mg IV loading dose of Herceptin®, followed by a 100 mg IV weekly dose for 8 weeks [41]. Cisplatin was given 24 hours after Herceptin® at a dose of 75 mg/m², repeated every 4 weeks. Nineteen patients with responses or stable disease entered a maintenance program. Out of 36 evaluable patients, 9 (25%) had a PR, 9 (25%) had a minor response or stable disease, and the remaining 18 patients (50%) had progressive disease. The median time to disease progression was 3.6 months. The duration of response ranged from 1.6-18 months (median=5.7 months). With one-fourth of the patients having a PR, this study showed an encouraging overall tumor response to Herceptin® plus cisplatin in refractory patients with HER2-overexpressing metastatic breast cancer. Herceptin® was administered safely, and there were few or no changes in vital signs. Adverse events and laboratory abnormalities were not unusual for this patient population. Three patients had an elevated serum creatinine >2.2 mg/dL. Two patients had fever (>38°C) during or after infusion. The observed toxicity did not appear to be greater than that expected with cisplatin therapy, and the observed response rate was higher than that expected with cisplatin treatment alone in this population. However, the lack of a control group precludes any definitive conclusion about the response rate seen in this study.

Antibodies to Herceptin® were not detected in any of the above *5* clinical studies.

A pivotal phase III trial of Herceptin® with chemotherapy (H0648g) has also been reported [42, 43]. In this study, 469 patients with HER2-overexpressing metastatic breast cancer not previously treated with chemotherapy for metastatic disease were entered. The first and larger group consisted of patients with metastatic disease who had received no prior anthracycline therapy in the adjuvant setting. Randomization was to treatment with doxorubicin and cyclophosphamide in standard doses either alone or in combination with Herceptin®. The second group consisted of patients with metastatic disease who had received anthracycline therapy in the adjuvant setting. Randomization in this group was to treatment with Taxol® alone or with Herceptin®. The Taxol® subgroups (n=188) had worse prognostic factors (ie a higher proportion of premenopausal patients, tumors negative for hormonal receptors, positive lymph nodes at time of initial surgery), had more prior therapy (adjuvant chemotherapy, myeloblative chemotherapy, radiotherapy) and a shorter disease-free interval from surgery for primary diagnosis to metastatic disease than did the AC subgroups (n=281). Results are seen in Table 1. A statistically significant improvement in time to disease progression, response rate and overall survival at 2 years was seen with the combination of Herceptin® and chemotherapy in both groups of patients. Patients treated with concurrent administration of Herceptin® and AC, however, had an increased risk of Class III/IV cardiac dysfunction (16%) compared to patients treated with doxorubicin and cyclophosphamide alone (3%). This cardiac risk appears to limit the use of combination of Herceptin® and doxorubicin in first line metastatic and adjuvant strategies for breast cancer. Further

information suggests that cardiac dysfunction is also seen with Herceptin® in association with Taxol® (2%) versus Taxol® alone (1%). Almost all of these patients had received prior anthracyline therefore this still raises concerns about the use of these agents after prior exposure to doxorubicin.

Table 1 Phase III Multicenter Results H0648g

	Combined Results		Paclitaxel Subgroups		AC Subgroups	
	H+Chemo	Chemo Alone	H+P	P alone	H+AC	AC
	(n=235)	(n=234)	(n=92)	(n=96)	(n=143)	(n=138)
Time to Progression						
Median (Months)	7.4	4.6	6.9	2.8	7.8	6.1
p-value (log-rank)	< 0.0001		< 0.0001		<0.002	
Overall Response Rate (%)	50	32	41	17	56	42
p-value (χ^2-test)	< 0.001		< 0.002		0.02	
Survival Time						
Median (Months)	25.1	20.3	22.1	18.4	26.8	21.4
p-value (log rank)	0.046		0.17		0.16	

Despite these observations, several North-American groups cooperative groups are proposing strategies based upon the sequence of doxorubicin-cyclophosphamide (AC) followed by taxane (paclitaxel or docetaxel)-Herceptin®, following the concept of adding Herceptin® to the most recent chemotherapies (taxane-anthracycline based program).

Although Herceptin® has been administered weekly in the pivotal studies, 2 studies are now available looking at the safety, efficacy and pharmacokinetics of Herceptin® when administered every 3 weeks in women with HER2 positive (IHC 2+, 3+) metastatic breast cancer. The first study looked at Herceptin® q3weekly in combination with paclitaxel [44], the second is a monotherapy q3weekly study of Herceptin® (unpublished data).

Initial results from the former study, with Herceptin® and paclitaxel being administered every 3 weeks, were presented at ASCO 2001. Thirty two patients received Herceptin® and paclitaxel administered every 3 weeks. Herceptin® dose consisted of an 8 mg/kg load by IV infusion over 90 minutes followed 6 mg/kg IV infusions over 90 minutes every 3 weeks. Eight cycles of paclitaxel at 175 mg/m² by IV every 3 weeks was planned. Baseline characteristics included median age of 53 years, 63% with prior adjuvant chemotherapy, 63% with prior metatstatic therapy, and 70% with prior anthracycline exposure. Tumor sites at baseline were as follows: 50% of patients had lung involvement, 47% had liver, 66% had bone and 38% had regional involvement, respectively. An average of 6 cycles of paclitaxel and 7 cycles of Herceptin® were administered, respectively. Toxicities seen with the q3weekly regimen were not markedly different from the previous study (H0648g) with Herceptin® having been given weekly. Three patients discontinued treatment due to toxicity. Four patients developed an infusion reaction with each subsequently continuing on treatment. Twelve patients developed myalgia. Two patients had decreases in LVEF of >15%, one of which developed grade 3 cardiac failure.
At the time of this report, 17 patients continued with therapy. Of 32 patients having received the q3weekly combination of Herceptin® and paclitaxel, 9.4% had a complete response, 43.8% had a partial response (overall response rate of 53%) and 25% had stable disease. Duration of response was 6.3 months and time to progression was 10. 9 months.

Preliminary results from the monotherapy program looking at q3weekly administration of Herceptin® became available (unpublished data from Roche). Eighty patients with HER2 overexpressing metastatic breast cancer are expected to be enrolled, with 50 patients having been recruited to date. Patients who were 3+ by immunohistochemistry or positive by FISH analysis were entered. Prior adjuvant therapy is allowed, but patients may not have had prior chemotherapy for metastatic disease. Patients are to receive a loading dose of 8 mg/kg of Herceptin® by IV infusion over 90 minutes, followed by q3weekly IV infusions of 6 mg/kg. At the time of this report, the median number of cycles of treatment received was 3. Number of cycles of treatment reported at this time is as follows:

Cycle Number	1	2	3	4	5	6	7	8	9
Patients treated	50	43	29	18	14	12	4	1	1

Eleven patients have been withdrawn from the study. Reasons include one patient having a cerebrovascular accident resulting in death and the remaining patients withdrawing because of insufficient response. Four serious adverse events have been reported. One patient dying from the cerebrovascular accident was mentioned above. One patient was diagnosed with a benign endometrial polyp for which a dilatation and curettage was required. The third patient was hospitalized for severe pain at the site of her disease (sternum and left supraclavicular fossa). The event resolved and patient continued to receive Herceptin®. The last case was that of a patient who developed an episode of shortness of breath following her loading dose of Herceptin®. This patient had a history of malignant pleural effusion. The event resolved and Herceptin® was discontinued.

Hematological toxicity seen with the q3weekly regimen was minimal and mild with the most severe anemia and neutropenia reported as grade 2, respectively, and grade 1 thrombocytopenia. Of 15 patients who have had 2 measurements of LVEF performed at baseline and at cycle 4, 1 patient had a decrease of LVEF of 24% with a concomitant mild dyspnea. Infusion reactions were mild to moderate and consisted of fatigue, headache, pyrexia, rigors and shivering. Other nonehematologic toxicities reported were mild and moderate. No response data is available from the monotherapy q3weekly study.

2.3.3 Clinical pharmacology of Herceptin®

The pharmacokinetics of Herceptin® was studied in breast cancer patients with metastatic disease.
Preliminary pharmacokinetic data from studies using a loading dose of 4 mg/kg followed by a weekly maintenance dose of 2 mg/kg, have indicated a mean half-life of 5.8 days (range of 1 to 32 days). Additionally, results from a population pharmacokinetic re-analysis of data from single-agent clinical trials of Herceptin® administered weekly have indicated a half life of 28.5 days (95% confidence interval, 25.5-32.8 days) [45]. In an ongoing phase I/II trial investigating a q3 weelkly regimen with Herceptin® (loading Herceptin® dose of 8 mg/kg followed by q3 week dose of 6 mg/kg) in combination with paclitaxel (Study BO15935), the half-life of Herceptin® was estimated to be at least 3-4 weeks (20 days at cycle 4 and 19 days at cycle 10-12). Thus, Herceptin® may be present in the circulation for up to 24 weeks (range 18-24 weeks) after stopping Herceptin® treatment. It is already known that, when used in combination, Herceptin® and anthracyclines are associated with an increased risk of cardiotoxicity. Therefore, the use of anthracyclines after stopping Herceptin® may carry a higher risk of cardiac toxicity. If possible, physicians should avoid anthracycline based therapy for up to 24 weeks after stopping Herceptin® therapy. If anthracyclines are used, the patient's cardiac function should be monitored carefully.

Furthermore, peak serum concentrations and average concentrations (AUC/dosing interval) were higher with the q3weekly regimen than seen with the weekly schedule in the H0648g study. Peak Herceptin® serum concentrations for the q3weekly regimen was 164 μg/mL as compared to 100 μg/mL as seen in the pivotal study. Average concentrations were 81 μg/mL in the q3weekly regimen versus 53 μg/mL in the weekly H0648g study. However, pre-dose concentrations with the q3weekly schedule were lower than seen with the weekly schedule in the H0648g study (39 versus 53.4 μg/mL, respectively).
Despite the difference in AUC and peak concentration, the safety data of the weekly schedule, as previously described (see 2.3.2) does not show substantial difference with the data generated with the q3weekly schedule.

A series of simulations were done based on the pharmacokinetic parameters estimated previously for Herceptin (using a population PK approach on the published data from several studies). The simulations were done for the weekly (4+2 mg/kg) and the 3 weekly (8+6 mg/kg) dosing regimens assuming that no dose was missed and then for a range of time delays in the scheduled doses.
It was assumed that if dosing is re-started after a delay, we want to stay within 20% of the original trough concentrations at steady-state. In the simulations, if re-start of the maintenance dose was associated with a long and gradual come back to the steady-state, a loading dose was added as if we wanted to start the treatment for a new patient.
The results show that if a patient misses a dose by less than a week, continuing with the usual maintenance dose (2mg/kg for the weekly schedule, 6mg/kg for the 3 weekly schedule) would not cause a long lack of exposure and, therefore, there is no need for re-loading the patients. If a patient misses a dose by more than a week, the continuation of the maintenance

dose will not bring the levels back quickly to within the 20% of the original steady-state levels. Without a loading dose it takes a long time (6-8 weeks at least) to get back to within this range. Hence, a delay more than 1 week in dose warrants re-loading the patients.

2.3.4 Synergism between Herceptin® and chemotherapeutic agents

Studies in preclinical models used combination therapy for breast cancer cells that overexpress HER2, and the use of agents that interfere with HER2 function in combination with Taxol® resulted in significant antitumor effects [28,29,46,47,49]. The most compelling preclinical rationale, however, for the combination of carboplatin/Herceptin® is derived from studies designed to determine the effects of combined chemotherapy / Herceptin® *in vitro* and *in vivo* breast cancer models. These studies demonstrate that there are additive and/or synergistic therapeutic effects between a number of chemotherapeutic agents and Herceptin® [29]. The most significant therapeutic interaction is the synergistic effect seen with cisplatin or carboplatin and Herceptin® [28,29]. This synergistic effect results in a two-log increase in cytotoxic killing of HER2 positive cells exposed to the combination. The effect is not seen in cells in which HER2 overexpression does not exist. Antibodies to Herceptin® were detected in a single patient in the above clinical trials, however the patient did not demonstrate an adverse clinical event [42,43,47]. In addition to this synergistic effect with platinum analogues, the preclinical studies demonstrate also synergistic effects with docetaxel [29] while additive effects have been noted with other drugs including Taxol® [48].

Hancock et al demonstrated that a combination of cisplatin and the TAb 250 monoclonal antibody, which is specific to an extracellular epitope of the c-erbB-2 protein (gp 185), significantly enhanced the cytotoxic effect of cisplatin in human breast tumor cell lines and in mice with human tumor xenografts that overexpress c-erbB-2. This synergistic cytotoxicity was apparent over a wide range of antibody concentrations, including concentrations that showed no inhibitory effect alone [47]. Pietras et al also demonstrated this phenomenon in a drug-resistant ovarian cancer cell line with overexpression of p185HER2 [28]. The increase in cytotoxicity may be a result of inhibition of repair of cisplatin induced DNA damage by the antibody [28,46,49].

2.4 Rationale for the use of Herceptin® with docetaxel and platinum based chemotherapies

2.4.1 Docetaxel monochemotherapy

The great majority of phase II studies were performed using docetaxel at a dose of 100 mg/m² given over 1 hour every 3 weeks [50-71]. The 1 hour schedule and the relatively small difference in doses used in phase II studies probably account for the consistency of results observed throughout the various studies. With respect to docetaxel, 4 phase III monochemotherapy trials are published or reported to date [68-71]. In the first trial, docetaxel 100 mg/m² was compared to doxorubicin 75 mg/m² in first line metastatic after failure of alkylating agents [68]. Docetaxel induced more responses than doxorubicin (48% vs 33%, p=0.008), while median time to progression was longer with docetaxel (26 weeks vs 21 weeks, p=ns) although overall survival was identical in both treatment arms. The risk-benefit ratio appeared to favor docetaxel in this trial, suggesting that docetaxel may be more powerful than doxorubicin in first-line therapy of advanced breast cancer.

The 3 other phase III trials were performed in patients with metastatic breast cancer after failure of anthracyclines, comparing docetaxel 100 mg/m² given over 1 hour to various salvage regimens [69-71]. The largest study (392 patients) randomized patients between docetaxel and mitomycin-C (12 mg/m² q6 weeks) plus vinblastine (6 mg/m² q3weeks) [69]. Efficacy was significantly better for docetaxel with higher overall response rate (30% vs 12%, p=0.001), longer time to treatment failure (19
weeks vs 11 weeks, p=0.001) and most importantly longer overall survival (11.4 months vs 8.7 months, p=0.0097). The next study (283 patients) was performed by the Scandinavian group and compared docetaxel to methotrexate plus 5-fluorouracil (5-FU), respectively 200 mg/m² and 600 mg/m² day 1 and 8 q3weeks [68]. Again and confirming the previous trial, efficacy was significantly in favor of docetaxel with better response rates (42% vs 21%, p=0.0001) and longer time to progression (27 weeks vs 13 weeks, p=0.0001). Survival was similar in both arms, possibly related to the built-in crossover. Finally, the last trial studied, in 172 patients, docetaxel vs NAF (vinorelbine 25 mg/m² day 1 and 5 q3weeks plus continuous

infusion 5-FU 750 mg/m² over days 1 through 5 q3weeks). Response rates were 43% for docetaxel and 39% for NAF (p=ns). Time to progression and overall survival were longer with docetaxel (respectively 28 weeks vs 22 weeks and 19.1 months vs 13.9 months), but did not reach statistical significance [71].

All these phase III data are suggesting that docetaxel represents potentially the single most active chemotherapeutic agent for the treatment of breast cancer.

2.4.2 Platinum Based Chemotherapies

The platinum co-ordination complexes are broadly active in human cancers. They are a crucial component of therapy for germ-cell tumours, lung and ovarian cancers. They also have substantial activity in metastatic breast cancer, but are seldom used in routine clinical practice. When used as first-line chemotherapy for metastases, objective response rates in two phase II trials of cisplatin were 52% and 47% [76]. Platinum compounds however are much less active in the salvage setting [75-77].

Carboplatin is a platinum co-ordination complex with a similar spectrum of activity, but with a different toxicity profile from the parent compound cisplatin. At routine doses carboplatin produces substantially less nephrotoxicity, neurotoxicity and nausea than does cisplatin. Myelotoxicity, and especially thrombocytopenia, are more prominent with carboplatin than with cisplatin. Carboplatin is active in metastatic breast cancer. In an early study, Kolaric et al reported a 20% rate of objective response among 20 previously untreated patients [74]. Martin et al treated 34 patients without prior chemotherapy for metastases and reported a 35% rate of response [74]. O'Brien and colleagues performed a phase II evaluation in which patients with metastatic disease were treated with a dose of carboplatin which was predicted to achieve an area under the time-versus-concentration curve of 7 mg/min/ml. In this study, objective responses were achieved by 33% of 27 previously untreated patients, and by 8% of 13 patients with prior chemotherapy for metastases [77].

Platinum-based combination regimens in breast cancer: Cisplatin and etoposide produced response rates of 0- 50%[78]. Other doublet regimens which have been studied include cisplatin/cytosar (7% response rate) and Cisplatin/5-FU [79-83]. Vinorelbine has emerged as a highly active drug in the treatment of metastatic breast cancer. The combination of vinorelbine and cisplatin was reported by Shamseddine et al to produce a very impressive 61% rate of response in 25 pre-treated patients [83]. Similarly Ray-Coquand et al reported a 41% response rate in pre-treated patients [84]. The MVAC (methotrexate, vinblastine, doxorubicin and cisplatin) regimen produced responses in 57-80% of patients [85-88].

The combination of carboplatin and etoposide has been subjected to several evaluations. In three of these studies, carboplatin/etoposide was used as first chemotherapy for metastases. Van der Gast and colleagues reported a 27% rate of objective response in 34 patients [89], Crown et al 42% response in 19 previously untreated patients [90], and Deltetto 50% in 12 patients [91]. There has been a wide range of reported response rates (0-50%) for this combination in pre-treated patients [92-95].

Smith and colleagues at the Royal Marsden have conducted a series of innovative trials using long-term continuous infusion 5-fluorouracil together with three-weekly administrations of lower dose cisplatin and epirubicin. In an early study involving patients with locally advanced disease, a response rate of 98% was reported, among forty-nine patients, including 66% clinical complete remissions. In a subsequent study, carboplatin was substituted for cisplatin. Fifty-two patients with metastatic (36) or locally advanced (16) breast cancer were studied. The response rate was 81% with a complete response rate of 17% in patients with metastatic disease and 56% in patients with locally advanced cancer [96].

2.4.3 Docetaxel-based combinations

Developing combination chemotherapy in metastatic setting has been a necessary step before proceeding to the adjuvant setting. Given the high individual activity of docetaxel and doxorubicin as single agents in breast cancer, and their potential limited cross-resistance, evidenced by the confirmed activity of docetaxel in patients resistant to anthracyclines, the rationale for the development of combinations or sequence based upon these two agents was compelling. Furthermore, the extrahematological toxicity profile of the 2 agents, with limitation of overlapping toxicities, suggested the potentiality for exploitation of the maximum benefit from each agent, in particular in terms of cardiac toxicity.

Nabholtz and colleagues showed that the combination of docetaxel and doxorubicin was superior to doxorubicin and cyclophosphamide in patients with previously untreated MBC [97]. These results also confirm the findings from phase II trials confirming the lack of influence of docetaxel on doxorubicin-induced cardiomyopathy.

Platinum Concerning Platinum-Taxane Combinations, docetaxel and cisplatin share different mechanisms of actions and resistance, and both have shown to have activity in advanced breast cancer. In phase I studies, the feasibility of administering docetaxel in combination with the platinums has been demonstrated in patients with lung and breast cancer. Phase I combination studies suggest that without growth factor support, 75 mg/m² of docetaxel followed immediately by 75 mg/m² of cisplatin, is a manageable regimen [97]. The dose limiting toxicities were hematologic. Approximately half of the cycles were followed by Grade 4 (< 0.5 x 10^9 cells/L) neutropenia, which was of brief duration, non-cumulative, and rarely complicated by febrile neutropenia. The most frequent non-hematologic toxicities were nausea/vomiting and diarrhea. Pharmacokinetics (PK) of docetaxel and cisplatin were performed during the first cycle of administration of this combination and were consistent with the published results from single-agent studies; suggesting no major pharmacokinetic interaction [98]. Cisplatin is known to cause a dose-dependent peripheral neuropathy. Single agent docetaxel has been reported to cause a mild neuropathy in Phase I and II. The combination of docetaxel and cisplatin induces a dose-dependent predominately sensory neuropathy. All patients enrolled into one Phase I study of locally advanced solid tumors had detailed neurological examinations. At cumulative doses of both cisplatin and docetaxel above 200 mg/m², 26 of 35 (74%) patients developed a neuropathy which was mild in 15, moderate in 10 and severe in one patient [99].

The cisplatin/docetaxel doublet is highly active in MBC. Investigators in Dublin and France, conducted a phase I-II evaluation of these drugs in patients with MBC and no prior chemotherapy for metastases. These patients had extensive exposure to adjuvant chemotherapy. Four dose levels were used: 75/75; 85/75; 85/100; 100/100 mg/m², of docetaxel and cisplatin respectively. Of the 38 patients treated, 69% had prior adjuvant chemotherapy, including anthracyclines in more than 40%. An overall response rate of 60% was reported, and among chemotherapy naïve patients, the response rate was approximately 80%. Doses of 75 mg/m² of each drug were well tolerated, with dose-limiting gastro-intestinal toxicity occurring at higher doses. Bernard et al treated 32 patients with prior anthracycline exposure with 100 mg/m² docetaxel followed by cisplatin 100 mg/m², repeated every 3 weeks for a maximum of 8 cycles [100]. The most common toxicity was grade 3-4 neutropenia which occurred in 17 patients (55%): 23 out of 32 patients required GCSF. Cutaneous and neurologic toxicities were frequent (78% and 75% of patients, respectively), but were moderate. 16 patients (50%) exhibited fluid retention (severe, 3 patients). No toxic death was noted. Twenty-one patients (65%) required a dose reduction. With a median follow-up of 10 months (1-14+), fifteen out of 30 evaluable patients (50%) had an objective response (2 CRs). The liver response was 50% and 9 of 17 anthracycline-resistant patients responded.

In a phase II study looking at anthracycline-pretreated metastatic breast cancer patients, Antoine et al enrolled 20 patients to receive 100 mg/m² of docetaxel followed by 100 mg/m² of cisplatin every 3 weeks for 8 cycles [101]. The most common toxicity seen was grade 4 neutropenia in 14 patients (70%) and 50% of cycles. Grade 3-4 thrombocytopenia was noted in 5 patients. Other toxicities were mild and of low grade. These included 5 patients with Grade 1-2 neurotoxicity, 7 patients with grade 1-2 cutaneous toxicities, 12 patients with grade 1-2 nausea/vomiting, and 2 patients with mild fluid retention. No toxic death was noted. One patient developed a grade 2 hearing loss and cisplatin was subsequently stopped after 3 cycles. At the time of these preliminary results, 13 patients were evaluable for response. Eight patients had a partial response (OR 61%) and responses occurred at any site. Three patients experienced disease stabilization and 2 patients progressed while on treatment. Response sites included liver, chest and soft tissue.

Llombart-Cussac et al enrolled 41 patients with anthracycline-resistant breast cancer in a phase I/II study looking at maximum tolerated dose and activity profile of docetaxel and cisplatin [102]. The dose-limiting toxicity was found to be febrile neutropenia seen in 2 of 9 patients at level 75/80 mg/m² of docetaxel and cisplatin, respectively, and 3 of 5 in the level 85/80 mg/m² docetaxel and cisplatin, respectively. Patients experiencing grade 3-4 toxicities were neutropenia 93% (febrile 20%), nausea/vomiting 17%, peripheral neuropathy 10%, stomatitis 7%, thrombocytopenia 5%, fluid retention 2%. One patient died of septic shock. Five patients had a CR (12%), and 18 patients a PR (44%) with liver responses in 14 (54%). Median response duration was 4.5 months and median overall survival in 13 months. Investigators in Florida have reported a 96% rate of objective response for the combination of cisplatin and docetaxel (70 mg/m² each) in patients with locally advanced breast cancer.

The combination of carboplatin and docetaxel has also been extensively studied in ovarian and lung cancer. Doses of carboplatin of approximately AUC 5-6 mg/mL/min can readily be combined with docetaxel at a dose of 75 mg/m². At higher doses, myelosuppression, especially thrombocytopenia, becomes dose-limiting. Severe neurotoxicity, or other non-hematopoietic toxicities are rare.

2.4.4 Docetaxel, Herceptin® clinical experience (TH)

Docetaxel and Herceptin® in combination have been studied in 4 phase II trials [103-106]. In the phase II study conducted by Burris et al, docetaxel was delivered at a dose of 75 mg/m² for 6 3-weekly cycles while Herceptin® was initiated as a 4mg/kg on day 1 (90-min IV infusion) followed by 2mg/kg weekly (30-min IV infusion) until disease progression. Patients with metastatic breast cancer overexpressing HER2 (IHC 2+/3+) were eligible for this trial. Twenty two patients have received 120 cycles of docetaxel (median 6 cycles) and more than 500 doses of Herceptin® (median 26 doses). Toxicity has been minimal with 1 episode of febrile neutropenia and 3 cases of dermatitis (2 patients with grade 2 and 1 patient with grade 3). No significant cardiac toxicity was observed. Antitumor activity assessment is reporting an overall response rate of 45% and clinical benefit of 65% so far. Patients with HER2 IHC 3+ status presented with major responses in 54% of cases and clinical benefit in 69% of treated patients [103].

The next 3 studies assessed weekly docetaxel with Herceptin® in patients with metastatic breast cancer. The first study [33] looked at weekly 35 mg/m² IV docetaxel (6 of 8) with weekly Herceptin® in 1st line metastatic breast cancer patients with no prior exposure to taxanes. Twenty-one patients have received 69 cycles (1 cycle = 8 weeks). Grade 3 or greater toxicities included 1 patient with neutropenia, 3 patients with fatigue, 3 patients with diarrhea, 1 patient with nausea and vomiting, 1 with neuropathy, 1 with dyspepsia/ulcer, and 2 patients with hypersensitivity reactions. Preliminary data showed 2 patients having a CR (11%) and 10 patients having a PR (52 %) in 19 evaluable patients, with an overall response rate of 63%. Median TTP was 12 months and median survival is 18.3 months [33].

The 2nd study looked at the same weekly dosing of docetaxel and Herceptin® as described above, but allowed for previous metastatic chemotherapy [105]. Of 12 patients evaluable for response and toxicity, 8 patients had received one metastatic chemotherapy regimen and 3 had received 2 prior metastatic regimens. Seventy-six doses of docetaxel and 80 doses of Herceptin® had been administered. No grade 3 or 4 toxicity was observed. The most frequent non-hematologic toxicities observed were fatigue (4 patients), dyspepsia (4 patients), diarrhea (4 patients), nausea (4 patients). Six patients achieved a PR (50%), 5 patients had stable disease, and 1 patient had disease progression. Median duration of response was 3.8 months.

The final study looked at weekly docetaxel and Herceptin® in metastatic HER2 2+, HER2 3+ or FISH positive patients [37]. Of 20 patients, 17 had received prior chemotherapy, whether as adjuvant or metastatic. One cycle is defined as 3 weekly administrations of docetaxel and Herceptin® followed by 1 week of rest. A median of 7 cycles was given. Grade 3, 4 neutropenia was observed in 6 patients. No febrile neutropenia, anemia or thrombocytopenia was observed. Grade 3 or 4 non-hematologic toxicity included 1 patient with catheter-related bacteremia, 1 patient with diarrhea, 1 patient with neuropathy, 5 patients with fatigue, 1 patient with pleural effusion, 1 patient with asymptomatic transitory decrease in LVEF below 50%, and 1 patient with congestive heart failure. Twelve patients (60%) had a PR, 5 patients had stable disease for at least 4 months. Median TTP is 7 months.

2.4.5 Docetaxel, Platinum salt (carboplatin or cisplatin) and Herceptin® Multicentric Pilot Phase II Trials (TCH)

Based on biologic data demonstrating pharmacologic synergy between Herceptin® and both docetaxel and platinum analogs in terms of antitumor activity, and on the cardiac toxicity associated with anthracycline-Herceptin® based combination regimens, BCIRG has proceeded with 2 pilot TCH phase II trials, one combining docetaxel/Herceptin® and carboplatin (TCH1 or BCIRG 102) and one combining docetaxel/Herceptin® and cisplatin (TCH2 or BCIRG 101).

The primary objectives of these pilot studies [107] were to evaluate the efficacy and safety of TCH as therapy for patients with HER2-positive advanced breast cancer. Secondary objectives were duration of response, time to disease progression and survival. A total of 120 HER2-positive (immunohistochemistry and/or fluorescence in situ hybridization [FISH]) stage III/IV breast cancer patients were planned to be treated (60 patients per trial). Eligible patients were treated with Herceptin® 4mg/kg on day 1 (90-min IV infusion) followed by 2mg/kg weekly (30-min IV infusion) plus docetaxel 75mg/m² (1-h IV infusion) and either cisplatin 75mg/m² (1-h IV infusion) or carboplatin (AUC of 6 mg/mL/min) on day 1 every 3 weeks (for 6-8 cycles). All patients are to continue weekly Herceptin® until disease relapse. A total of 62 patients were registered into the BCIRG 101 pilot (docetaxel, cisplatin and Herceptin®), with results currently available for 62 patients for safety and efficacy. A total of 62 patients were registered into the docetaxel, carboplatin and Herceptin® pilot (BCIRG 102) with results currently available on 58 patients for efficacy and 62 patients for safety [Baseline patient and tumor characteristics are found in Table 2. Data presented is effective as of October 2001.

Table 2 Patient and Tumor Baseline Characteristics, TCH Pilots

	BCIRG 101 Pilot T Cisplatin H	BCIRG 102 Pilot T Carboplatin H
N	62	62
Median age (range)	52 (29 – 76)	54 (31 – 76)
ECOG PS 0 1 2	40 (65%) 20 (32%) 2 (3%)	36 (58%) 25 (40%) 1 (2%)
Organs Involved 2 ≥ 3 organs	 40 (65 %) 22 (35 %)	 42 (68 %) 20 (32 %)
Organ Involvement Visceral - Liver - Lung Brain Bone Metastases Bone Lytic Only	 43(69 %) 24 (39 %) 25 (40 %) 1 (2%) 29 (47 %) 4 (6 %)	 43 (70 %) 16 (26 %) 31 (51 %) 3 (5%) 28(46 %) 5 8 %)
Prior Adjuvant Chemotherapy - anthracycline CT - taxane CT	36 (58 %) 20 (32 %) 0	35 (56 %) 28 (45 %) 9 (15 %)
Prior Metastatic Chemotherapy	0	3 (5 %)

Of note, patients treated with TCarboH had a higher percentage of lung metastases, prior adjuvant treatment with taxanes and anthracycline, prior chemotherapy for metastatic disease, and presence of brain metastases than patients treated with TCisH. On the other hand, patients treated with TCisH had a higher percentage of liver metastases.

Overall, both treatment regimens were feasible and well tolerated. A total of 389 cycles of chemotherapy (docetaxel-cisplatin) and 1,937 doses of Herceptin® were administered to 62 patients for the TCisH pilot population. In the TCisH pilot, the median number of cycles given was 6 (range 3-8). Two patients had received ≤ 3 cycles, 3 patients had received 4 cycles, 4 had received 5 cycles, 34 (55%) had received 6 cycles, and 19 (31%) had received > 6 cycles. There were 9 (15 %) chemotherapy discontinuations, 2 patients for progressive disease, 3 for adverse events (2 patients for grade 3 neurosensory at cycle 5 and 1 patient who developed congestive heart failure at cycle 4), 3 patients went to surgery, and one patient withdrawal. Of the 389 cycles of chemotherapy given, there were 8 dose reductions with docetaxel (4 for non-hematological toxicity, 1 with hematological toxicity only, and 3 with both hematological and non-hematological toxicities), and 11 dose reductions with cisplatin, 7 of which were for non-hematological toxicity, 3 of which were for both hematological and non-hematological reasons, and 1 which was not study drug related.

A total of 385 cycles of chemotherapy (docetaxel-carboplatin) and 1, 956 doses of Herceptin® have been administered in 62 patients in the TCarboH pilot population. Median number of cycles given was 6 (range 2-13). Of 62 patients, six patients had ≤ 3 cycles, 2 had 4 cycles, 2 had 5 cycles, 35 (56%) had 6 cycles, and 16 (26%) patients had > 6 cycles. There were 10 discontinuations (16%) in the TCarboH pilot, 6 with progressive disease, 1 patient withdrawal without experiencing any severe toxicity, and 3 patients with adverse events (1 patient with grade 3 diarrhea and edema at cycle 2, 1 patient with cardiac tamponade probably related to tumor progression as cytologic examination pericardial effusion was positive for malignancy, 1 patient with pancytopenia and electrolyte imbalance). Of the 385 cycles of chemotherapy given, there were 18 dose reductions with docetaxel (10 for hematological toxicity, 6 for non-hematological toxicity, 2 for both), and 16 dose reductions with carboplatin (10 for hematological, 4 for non-hematological toxicities and 2 for both hematological and non-hematological toxicities).

Of the 62 patients in the TCisH pilot, 8 (13%) had a febrile neutropenia event, compared to 10 of 62 patients evaluable for safety (16%) in the TCarboH pilot. There were 2 (3%) infectious episodes in the TCisH pilot, and no infectious episode in the TCarboH population. There were no septic deaths in either pilot. There were 6 grade 3 / 4 anemia events (9%) in the TCisH pilot and 4 events (6%) in the TCarboH pilot. No patient developed a grade 3 or 4 thrombocytopenia in the TCisH pilot compared to 7 events (12%) in the TCarboH pilot.

Non-hematological toxicities are found in Table 3. Although the TCisH regimen showed a less favorable overall non-hematological toxicity profile compared to that of the TCarboH, there was no substantial difference between the 2 regimens when considering grade 3 / 4 toxicities.

Table 3 Non-Hematological Toxicity

	BCIRG 101 TCisH		BCIRG 102 TCarboH	
N	62		62	
	Overall	Grade 3 / 4	Overall	Grade 3 / 4
Alopecia	58 (94 %)	n/a	43 (69 %)	n /a
Asthenia	58 (94 %)	11 (18 %)	50 (81 %)	11 (18 %)
Gastrointestinal				
- Nausea	56 (90 %)	11 (18 %)	43 (69 %)	7 (11 %)
- Vomiting	45 (69 %)	7 (11 %)	26 (42 %)	5 (8 %)
- Diarrhea	45 (73 %)	7 (11 %)	32 (52 %)	3 (5 %)
- Stomatitis	29 (47 %)	2 (3 %)	31 (50 %)	2 (3 %)
- Constipation	16 (26 %)	0	18 (29 %)	0
Myalgia / arthralgia	18 (29 %)	0	14 (23 %)	3 (5 %)
Nail Changes	17 (27 %)	0	9 (15 %)	0
Neurologic				
- Sensory	37 (60 %)	2 (3 %)	26 (42 %)	0
- Motor	7 (11 %)	1 (2%)	9 (15 %)	1 (2%)
Ototoxicity	23 (37 %)	1 (2%)	2 (3%)	0
Peripheral Edema	25 (40 %)	1 (2%)	20 (32 %)	1 (2 %)
Renal	25 (40 %)	2 (3%)	1 (2%)	0
Skin rash / erythema	17 (27 %)	1 (2%)	18 (29 %)	1 (2 %)

Left ventricular ejection fraction (LVEF) by MUGA or echo was required at baseline, every 12 weeks, at completion of chemotherapy and during Herceptin® therapy at any suspected change (TCarboH) or every 3 months in follow-up (TCisH). In addition to LVEF monitoring, cardiac toxicity was recorded using the NCI Common Toxicity Criteria, version 1.0. In the TCisH population, one 60-year-old patient developed congestive heart failure, with an absolute LVEF decrease of ≥ 15 points from baseline and below the institution's lower limit of normal. Of note, this patient had a prior history of coronary disease and had received radiation therapy to the left chest wall. Ten patients had asymptomatic decreases of LVEF. Four of these patients had an absolute decline by more than 20 points from baseline, 4 had a decline of ≥ 15 points from baseline and below the lower limit of normal, and 2 had asymptomatic decreases ≥ 10 points from baseline and below the institution's lower limit of normal. Two patients developed a grade 1 and grade 2-dysrhythmia toxicity, respectively.

In the TCarboH pilot, 1 patient developed congestive heart failure. She had no prior history of cardiac disease. Seven patients had asymptomatic decreases of LVEF. Of these 7 patients, two had an absolute decline by more than 20 points from baseline and below the institution's lower limit of normal. Three of these 7 patients had declines of ≥ 15 points from baseline and below the institution's lower limit of normal, and the remaining 2 had declines of ≥ 10 absolute points, respectively. One patient developed a grade 3 dysrhythmia toxicity.

The TCH regimen avoids the potential cardiac toxicity when Herceptin® is used with, or after, anthracycline based regimens. As noted earlier in the H0648g, study patients treated with concurrent administration of Herceptin® and AC had an increased risk of class III/IV cardiac dysfunction (16%) compared to patients treated with doxorubicin and cyclophosphamide alone (3%). Only 1 incident of Grade 3 cardiac dysfunction was found in each of the TCH studies and appears favorable when compared with the H0648g results [(Table 4).

Table 4 H0648g and TCH Pilots: Severe Toxicities

	H0648g		TCH Pilots	
	AC+H	Taxol®+H	TCisH	TCarboH
Febrile Neutropenia	NA	NA	13 %	16 %
Infection	2 %	1%	3 %	0
Nausea	6 %	3 %	18 %	11 %
Vomiting	3 %	9 %	11 %	8 %
Stomatitis	1 %	0	3 %	3 %
Neurosensory	0	2 %	3 %	0
Neuromotor	NA	NA	2 %	2 %
Arthralgia / Myalgia	< 1 %	9 %	0	5 %
Asthenia	7 %	8 %	18 %	18 %
Class III/IV Cardiac dysfunction	16%	2 %	*1.6 %*	*1.6 %*

AC= anthracycline and cyclophosphamide; H=Herceptin®; T=docetaxel; Cis=Cisplatin; Carbo=Carboplatin

Preliminary response data are found in Tables 5 and 6, respectively. In the TCarboH (Table 5) pilot, three patients had received prior treatment for metastatic disease. Of the 59 patients who were treated as first line in the pilot trial, 55 were evaluable for efficacy and had a FISH result available. In the TCisH (Table 6) pilot, of the 62 patients evaluable for efficacy, 54 had a FISH result available only.

Table 5 TCarboH Pilot Preliminary Response Data in Metastatic Breast Cancer Patients

	1st and 2nd line patients			1st line patients only		
	FISH Positive	FISH Negative	Fish Pending	FISH Positive	FISH Negative	Fish Pending*
CR	*7*	*1*	*0*	7	1	0
PR	*16*	*6*	*1*	16	6	1
MR	2	1	1	2	1	1
SD	*7*	*7*	*0*	6	7	0
PD	*7*	*2*	*0*	5	2	0
ORR	*23/39 (59%)*	*7/17 (41%)*	*1/2 (50%)*	23/36 (64%)	7/17 (41%)	1/2 (50%)
(Not evaluable)	2	2	0	2	2	0

*FISH testing is still ongoing

Table 6 TCisH Preliminary Response Data in 1st Line Metastatic Breast Cancer Patients

	FISH Positive	FISH Negative	FISH Pending	TOTAL
CR	2	1	0	3
PR	25	15	6	46
SD	8	2	2	12
PD	0	1	0	1
ORR	27/35 (77%)	16/19 (84%)	6/8 (75%)	49/62 (79%)

Response rates in the H0648g study are lower than seen in the available TCH pilot results in a very similar patient population. See Table 7. Efficacy data based on those patients who are FISH positive is also presented. The TCH regimens show promising response activity. Of note, the difference is more evident in the FISH positive subpopulation.

Table 7 Efficacy Data, HO648g Pivotal Study versus TCH Pilots in 1st Line Metastatic Breast Cancer

	H0648g AC – H	H0648g Taxol® - H	TCisH Pilot	TCarboH Pilot
Pts IHC 2+ or 3+	143	92	62	54
ORR	56 %	41%	84%	57%
95% CI	[48-64]	[31-51]	[72-92]	[43-71]
Pts Fish Positive	107	69	35	36
ORR	57%	49%	77%	64%
95% CI	[48-67]	[38-61]	[59-90]	[46-79]
Pts IHC 2+ or 3+	143	92	62	59
TTP	7.8	6.9	9.9	12.0
95% CI	7.3-9.4	5.3-9.9	8.3-13.1	7.4-16.3
Pts Fish positive	107	69	35	38
TTP	7.6	7.1	12.7	17.0
95% CI	7.1-9.4	3.9-14.1	9.2-13.1	9.1-NE*

***NE: Not Estimate**

^ TCarboH Pilot: data only from the 1st line metastatic patients is presented in this table.

In conclusion, the TCisH and TCarboH phase II multicenter studies in this metastatic HER2 positive patient population have shown that these regimens are feasible, very active, and safe without any enhancement of the expected toxicity of the individual agents. TCH avoids the potential cardiac toxicity when Herceptin® is used in combination with anthracycine.

2.5 Rationale for the Present Adjuvant Trial, the BCIRG 006 Trial

2.5.1 Rationale, General

The HER2 alteration has been identified in 15-30% of human breast cancers. Indeed a number of prospective and retrospective studies are being conducted using the presence or absence of the HER2 alteration for segregating patient groups for analysis and/or treatment. It is now clear that the presence of this alteration plays an important role in the pathogenesis of this disease. As a result, therapies targeting this molecular change are actively being pursued.

Substantial data now exist that anti HER2 antibodies (Herceptin®/Trastuzumab) can yield a significant and important therapeutic benefit to patients whose malignancies contain the alteration, including improvements in response rate, response duration, time to disease progression and most importantly, overall survival. The most significant potential benefit for the use of Herceptin® may be in the adjuvant setting. This significant improvement in efficacy comes with minimal significant toxicity with the sole exception of an increased incidence of cardiotoxicity, especially when this drug is used in combination with anthracycline based therapy. The molecular and preclinical data from studies evaluating the effects of this alteration demonstrate that important synergistic alterations occur between Herceptin® and the platinum compounds (cisplatin or carboplatin) as well as with docetaxel. As a result, the current study is designed to test the potential improved efficacy of this combination (Herceptin®/platinum/docetaxel). This combination has the additional advantage of potentially significantly lowering the occurrence of cardiotoxicity in patients receiving Herceptin®. The proposed BCIRG 006 study contains several unique features including:

1) Evaluation of a combination of drugs based on non-empiric approaches but rather sound biologic and molecular principles which have been confirmed in published literature.

2) Inclusion of node negative patients who contain the HER2 alteration into an adjuvant program. These patients are known to have a worse prognosis than node positive patients (1-3 nodes) who do not have the alteration.

3) Use of a more sensitive and specific method of identification of patients whose malignancies contain the alteration

thus circumventing the problem of inclusion of patients without a HER2 change as well as exclusion of patients who have the change.

2.5.2 Justification of the experimental arms

The results generated by Herceptin® in breast cancer patients have generated significant enthusiasm in North America and in Europe and several trials in combination with taxanes are being performed at the present time in the metastatic setting. Additionally, several cooperative groups have decided to proceed swiftly to the adjuvant setting and integrate Herceptin® in second generation taxane phase III trials (taxane-containing regimen vs taxane-containing regimen plus Herceptin®). However, the unexpected cardiac toxicity seen while using doxorubicin and Herceptin® in the metastatic setting creates potential difficulties. Consequently, two types of trials have been designed at the present time.

The first type is based upon the concept of adding Herceptin® to the present anthracycline-containing adjuvant strategies.

Considering the risk of using Herceptin® in combination with doxorubicin and despite a potential risk of recall phenomenon of cardiac toxicity when using a short interval sequence between anthracycline-Herceptin®, two American cooperative groups (the
Intergroup and NSABP B31) have embarked on the concept of adding Herceptin® to their sequential chemotherapy in adjuvant setting: Phase III trials comparing AC followed by paclitaxel (various schedules, weekly or 3-weekly) with or without Herceptin® in the adjuvant treatment of patients with breast cancer overexpressing the HER2 protein or containing amplification of the HER2 gene.

The present trial is following an alternative strategy consisting of

1 Building a combination program around Herceptin® following data of synergism between Herceptin® and chemotherapy in order to get a biologically-oriented regimen and
2 Avoiding the potential cardiac toxicity seen with Herceptin® and doxorubicin. This led to the choice of the TCH arm as a significant experimental arm.

In addition, we decided to add a third arm (so called “American arm” in reference to the 2 American cooperative groups conducting them) using the addition of Herceptin® to the sequence AC followed by docetaxel. This would allow us to compare prospectively the 2 philosophies of integration of Herceptin® in the adjuvant setting and in the event of cardiac toxicity seen with the sequence AC followed by TH, to keep the TCH arm as a strategy for Herceptin® in the adjuvant setting.

For sake of consistency for assessing the cardiac safety of the addition of Herceptin® to the control arm AC→T, we have used some of the guidelines for the management of Herceptin® and for repeat LVEFs for patients who develop asymptomatic decreases in left ventricular ejection fraction from the NSABP-B31 protocol. For the purpose of future comparison between the 2 American studies mentioned above and the BCIRG 006 study, we have chosen to remain as consistent as possible with the NSABP-B31 cardiac safety evaluation. BCIRG wishes to acknowledge the NSABP for the development of these guidelines, and for granting permission for their use.

As a result, this adjuvant trial represents a unique design based on molecular, biologic and clinical data rather than the historic approach of adding a new therapeutic to what is already used regardless of the concern or potential contradictions. If cardiotoxicity risks are limiting for the combination of Herceptin® and anthracycline based therapies, the NSABP B31 and the Intergroup Study could significantly delay the entry of Herceptin® into the adjuvant setting in breast cancer. The current BCIRG study will clearly circumvent this possibility. It has the additional advantage of evaluating a combination which may have superior efficacy.

2.5.3 Platinum salt selection in the TCH arm

Preclinical data suggests that cisplatin and carboplatin are equally active and synergistic when combined with Herceptin® (see section 2.3). The efficacy data of the pilot phase II studies described in section 2.4.5, show that both regimens

(TCarboH) and (TCisH) have a similar activity in HER2+ metastatic breast cancer patients, particularly in term of Time to Progression. On the other hand, although both regimens are safe and feasible (more than 80% of patients in both regimens received at least 6 cycles), the safety profile of TcarboH was better than that of TcisH: TcarboH showed less nausea, vomiting, diarrhea and no oto and renal toxicity in comparison to TcisH.
In addition, it should be noted that more than 80% of the participating centers have chosen carboplatin. Therefore, the selection of carboplatin in the TCH arm will definitely homogenize TCH regimen and will make the analysis easier to interpret.

The centers that have started treating patients with Cisplatin in the TCH combination prior the protocol being amended (Amendment #3, April 10, 2002) will treat those specific patients with Cisplatin for their remaining cycles.

2.5.4 Justification of qweekly and q3weekly Herceptin® administration

Two studies are looking at the safety, tolerability and pharmacokinetics of Herceptin® when administered every 3 weeks in patients with HER 2 positive (by immunohistochemistry or FISH) metastatic breast cancer [44]. The data indicates that the higher doses of Herceptin® are well tolerated and the q3weekly administration of Herceptin® is feasible. Despite the fact that the peak serum concentrations and AUC of the q3weekly schedule were higher than the peak of serum concentration and AUC of the weekly schedule (see section 2.3.3), the safety profile of the q3weekly Herceptin® (as monotherapy or in combination with paclitaxel) did not differ from that seen with the weekly schedule (see section 2.3.2.). Therefore it seems that the peak of serum concentration and the AUC at the doses explored with the two different schedules do not have a significant impact on the safety of Herceptin®. In addition, a recent study, conducted in 114 patients with HER2 overexpression metastatic breast cancer, comparing two different doses of Herceptin® (i.e. loading dose of 4 mg/kg followed by 2 mg/kg weekly versus loading dose of 8 mg/kg followed by 4 mg/kg weekly) did not show a clear evidence of a dose response relationship for adverse events including cardiac toxicity, as well as reponse or survival. It is interesting to note that the half-life of Herceptin® is similar in the two schedules and that the predose concentrations as week 3 with the q3weekly schedule were lower than seen with the weekly schedule. Response rates from the paclitaxel and Herceptin® study were also very similar. We thus propose to administer Herceptin® every 3 weeks instead of every week during the follow-up period starting 3 weeks after the last chemotherapy infusion, in order to reduce the frequency of visits and improve the compliance with Herceptin® administration. On the other hand, weekly schedule of Herceptin® will be maintained during chemotherapy since there are no efficacy and safety phase II data with q3 weeks Herceptin® in combination with Taxotere and Platinum salts.

2.5.5 Justification of the Comparator Arm AC → Taxotere

The comparator arm of the BCIRG 006 study is AC→T, which consists of 4 cycles of doxorubicin and cyclophosphamide (60, 600 mg/m²) followed by four cycles of docetaxel single agent 100 mg/m². The rationale for the selection of this comparator is as follows:

1. AC→Taxol® has recently been approved by the FDA in adjuvant breast cancer based on the preliminary results from the CALGB 9344 [18] study previously mentioned (see section 2.2.1).

2. Although the results from the NSABP-B28 and the 52-month follow-up results from the CALGB 9344 do not permit definitive recommendations regarding the impact of taxanes on either relapse-free or overall survival. AC→Taxol® is a valid adjuvant breast cancer regimen with a clear advantage in ER/PgR negative patients.

3. Single-agent docetaxel appears to be more active than paclitaxel in metastatic breast cancer. The following large phase III multicentric studies are available in metastatic breast cancer to illustrate this.

   Metastatic breast cancer patients (n= 326) resistant to alkylating agents were randomized to receive either docetaxel 100 mg/m² IV over 1 hour every 3 weeks or doxorubicin 75 mg/m² IV over 15 minutes every 3 weeks. [68]. Docetaxel induced more responses than doxorubicin (48% vs 33%, p=0.008), and median time to progression was longer with docetaxel (26 weeks vs 21 weeks, p=ns). Overall survival was identical in both treatment arms.

However, in a study of 331 metastatic breast cancer patients randomized to receive either paclitaxel 200 mg/m² versus doxorubicin 75 mg/m², results strongly favor doxorubicin with a response rate of 41% versus 25% (p=0.003) and with a significantly longer time to progression (7.5 months versus 4.2 months, p=0.001). There was no survival difference between the two arms, which could be related to the prospective crossover built into the study. Paclitaxel was thus inferior to doxorubicin in terms of RR and TTP [110].

This was illustrated again in the Intergroup trial where 739 patients were randomized to receive either paclitaxel 175 mg/m² or doxorubicin 60 mg/m² or the combination (175/60 mg/m²) as first-line metastatic treatment. Response rates were, respectively, 33%, 34% and 46% (p=ns) and median time to treatment failure was 5.9, 6.2 and 8 months, respectively, (p<0.05), and no significant difference in survival was detected between the three arms [111].

A phase III trial comparing AC (60/600 mg/m²) or AT (50 mg/m² doxorubicin and 75 mg/m² docetaxel), maximum 8 cycles as first-line treatment for patients with metastatic breast cancer were recently presented. A total of 429 patients without prior anthracycline exposure were randomized to a maximum of eight cycles of AC (n=215) or AT (n=214). The overall response rate was statistically significantly higher than patients treated with AC (60% for AT and 47% for AC, p=0.012). Patients treated with AT also experienced a significantly longer time to disease progression (p=0.01) and time to treatment failure (p=0.02) than those treated with AC [112].

However, in a large study that compared AC to AT (Taxol®) as first-line metastatic treatment in 275 women, no advantage in terms of response rate and progression free survival was shown with AT over AC at a median follow-up of 19 months [113].

4. A large program with docetaxel both in sequence and in combination with doxorubicin in early breast cancer patients is currently ongoing. Results will be available at the end of 2001.

5 AC→T (docetaxel) is already being used as an experimental arm in early breast cancer studies.

a) The NSABP-B27 study in neo-adjuvant and adjuvant breast cancer is comparing AC followed by surgery to AC followed by docetaxel followed by surgery to AC followed by surgery followed by docetaxel. More than 2200 patients have been randomized and no major safety concerns have been reported.
b) The BCIRG 005 adjuvant study in HER2 negative (FISH) patients is comparing TAC (docetaxel / doxorubicin / cyclophosphamide) to AC→T (docetaxel). The study is currently ongoing and accruing. Of note, TAC is the experimental arm in the BCIRG 001 (TAX 316) study comparing TAC versus FAC. The BCIRG 001 (TAX 316) is closed, and data will be available in the 4th quarter of 2001. The TAC arm in the BCIRG 005 arm will thus be validated.
c) The NSABP-B30 study is comparing AC x 4 (60/600)→T x 4 (100) to AT x 4 (50/75) to TAC x 4(75/50/500).

It is important to note that when the efficacy results from the BCIRG 006 become available, the results from the NSABP-B27, the BCIRG 001, the BCIRG 005 and the NSABP-B28 will also be available to the community.

Considering all of the above, we believe that AC→T (docetaxel) can be considered an effective and safe comparator arm for use in the present study.

2.5.6 Justification of Node Negative HER2 Positive Population

In a study whose results were recently published [31] comparing FISH versus immunohistochemistry, proportional hazards regression analysis was conducted to compare FISH-determined HER2 amplification as well as IHC-determined HER2 protein levels with the most commonly used clinical prognostic markers, including grade, histopathologic type, absence of ER and PR receptors, nodal involvement and S phase. In the node negative patient group, HER2 positive patients (> four HER2 signals/cell) had a statistically significant decrease of OS (RR=1.54) independent of tumor size and ER status when

compared to node negative patients with ≤ four signals/cell. Because of the poor prognosis associated with node negative HER2 positive patients, we have decided to include high risk node negative patients in the study. Patients with operable breast cancer with either positive or high risk negative nodal status and amplifying the oncogene HER2 will thus be eligible for this trial. High risk node negative status will be defined as node negative patients with at least one of the following: tumor size > 2 cm, hormonal receptor status negative, histologic and/or nuclear grade 2-3, age < 35 years [115].

2.5.7 Central Pathology Review (mandatory)

A central pathology review will be performed on sections derived from the paraffin block submitted for FISH testing. We will confirm the baseline tumor characteristics of all patients randomized into the study in a single central laboratory to avoid issues of inter-laboratory comparability. Standard prognostic histo-pathologic features such as grade, histologic subtype and vascular invasion as well as immunohistochemical markers for hormone receptors, and proliferation index (MIB-1) will be recorded. This review will take place after randomization.
Blocks will also be requested for confirmation of the histopathologic diagnosis of a new breast malignancy or second primary cancer.

2.5.8 Optional tests on Protein Expression Studies

Some markers may be considered predictors of response to certain anticancer agents (see Appendix 3). By predetermining the biological characteristics of a patient's tumor, therapies may be specifically targeted to those patients whose tumor has a characteristic that predicts an increased response to the anticancer agent. The FISH test is able to predict the type of tumor that will benefit from Herceptin® (see section 2.5.5).

Additional testing for such markers on the tumor sample is proposed. These markers include: p53, members of the bcl family (Bcl-2, bax, Bcl-X and Bag-1), MUC1 and Tubulin isoforms (particularly II, III, IV and Tau). Those factors, which are proven to have predictive utility in that trial, will be tested in the current trial in order to ensure comparability between groups.

The area of research into the identification of tumor markers and biological processes / targets to aid in the identification of clinical benefit in certain subsets of populations or even in the identification of anticancer therapies to target the marker, is rapidly growing. Following the mandatory FISH testing and central pathology review for the study, BCIRG wishes to store the tumor sample for future testing. As more development and information is revealed to us in the future, we would like to use these blocks for measurement of the new markers. The blocks will be stored in the central laboratory until future use is required. BCIRG may collaborate in the future with experts in the field, and the blocks (or portions thereof) may be shared with other researchers. A current example of this is the collaboration with Dr. J.C. Reed from the Burnham Institute who is researching the blocks for the Bcl family.

Although the tumor block is mandatory for the FISH and central pathology review, testing for the designated tumor markers and future testing is not mandatory. Refusal to grant permission for further testing will not affect the quality of care the participant is to receive.

2.5.9 Serum sample for Detection of shed HER2 Extracellular Domain (ECD) (optional)

Serum assays for HER2 are currently being investigated. This may prove to be a more practical means of testing for HER2 and thus targeting those patients who will benefit most from Herceptin®. Serum samples will be collected to mesure peripheral levels of shed HER2 ECD at baseline, at different time points during study and at time of relapse in order to

- determine whether peripheral levels of shed HER2 ECD prior to treatment are prognostic for desease free survival and survival,
- determine whether peripheral levels of shed HER2 ECD measured at different time points are predictive of response to the treatment,
- determine whether peripheral levels of shed HER2 ECD are increased at the time of relapse relative to their pretreatment levels.

Serum samples will be collected at baseline, at end of chemotherapy for AC→TH and AC→T arms or 6 weeks after the end of chemotherapy visit for TCH arm (FUp1a), and then every 6 months during the five first years of follow-up or until disease relapse, withdrawn consent or death whichever comes first and, at time of relapse.

Additional testing of molecular markers and assays for HER2 may be performed on the serum sample in the future as new developments become available.

Provision of the serum sample to the central laboratory is not mandatory. Refusal to grant permission for collection and release of the serum samples for the above mentioned purpose will not affect the quality of care the participant is to receive.

2.5.10 Cardiovascular Substudies (optional) – *(not applicable for France)*

This trial affords a unique opportunity to prospectively follow and evaluate the cardiovascular effects of three chemotherapeutic regimens, two of which are combinations that include Herceptin®. Analysis of outcomes in this large clinical trial holds the potential for the future development of guidelines to prevent the development of symptomatic heart failure from chemotherapeutic agents and, in particular, regimens containing Herceptin®. Initial steps in prevention of cardiotoxicity include:

1) identification of patients at high risk of developing heart failure who may not be good candidates for Herceptin® therapy, and
2) early detection of ventricular dysfunction, before clinical heart failure develops. Early detection of ventricular impairment may open the window for early treatment, which has been previously shown to improve outcomes in heart failure.

In addition to the serial monitoring of left ventricular ejection fraction by echocardiography or radionuclide imaging, we propose the examination of genetic and biochemical markers as an adjunct to the serial LVEF determinations.
The objectives of the cardiovascular substudy includes:

1) the potential for development of pretreatment screening tools to identify patients at high risk of developing LV dysfunction through assessment of genetic markers, and
2) the potential for early detection of ventricular dysfunction, before clinical heart failure develops through serial measurements of Troponin T and brain natriuretic peptide (BNP).

These strategies would allow for the safe identification of patients best suited for Herceptin® treatment and tailored treatment strategies.
Provision of the blood and plasma samples to the central laboratory is not mandatory. Refusal to grant permission for collection and release of the blood/plasma samples for the above mentioned purpose will not affect the quality of care the patient is to receive.

2.5.10.1 Genetic Markers

The human population is biologically diverse and genetically heterogeneous. Therefore, it is not surprising that differences in susceptibility to disease among individuals exist. The etiologies of many diseases including heart failure are due to a combination of factors, including genetic susceptibility and environmental exposures. Subtle differences in the genes that regulate cellular growth and development, DNA replication and repair, the metabolism of endogenous agents in the body, and the metabolism and excretion of exogenous agents that the body comes in contact with contribute to the risk of developing a disease. Identification and characterization of human genetic variation is providing new risk factors for disease in the form of DNA sequence variation. Single-nucleotide polymorphisms (SNPs) are common variations among the DNA of individuals, which can increase the risk of developing disease. Identifying these SNPs and the genes in which they reside is an important area in human genomics.

Recently genetic polymorphisms involving pathways mediating cardiac function, such as the β-adrenergic, angiotensin, and endothelin receptors, have been identified, which are associated with increased risk of cardiomyopathy and heart failure death [118-120]. The results of these studies are summarized in Table: Proposed Polymorphisms. Polymorphisms in a host of other genes including likely candidates such as BNP, IL-10, TNFα, TGFβ1, NOS3, Endothelin 1, Endothelin B receptor have been tested but have not demonstrated any association with heart failure incidence or prognosis. Likewise, polymorphisms in the HER2 signaling pathway itself may predispose to the development of cardiac dysfunction in response to Herceptin®. It has been shown that polymorphisms in HER2 are linked to the risk of breast cancer although its effects on cardiac function are unknown [121].

Table: Proposed Polymorphisms

Polymorphism	Type of Mutation	Effects on CHF Mortality or Morbidity	Reference
Angiotensin Converting Enzyme			
I/D	Insertion/Deletion	↑	[119]
β2-adrenoreceptor			
Gly16/Gln27	SNP	↓	[122]
Ile164	SNP	↑	[118]
ET Receptor A			
ETA C1363T	SNP	↑	[120]
HER2	SNP	?	[121]

SNP= single nucleotide polymorphism

Analyzing these genetic polymorphisms and correlating them with the development of LV dysfunction in the study population will allow identification patients at high risk of developing cardiac dysfunction secondary to chemotherapy with or without Herceptin® . To identify patients susceptible to Herceptin®-induced left ventricular dysfunction, we will examine polymorphisms previously associated with heart failure (ACE, β2-adrenoreceptor receptor, and the Endothelin A receptor) and HER2 polymorphism. A sample of whole blood will be requested prior to treatment administration. UCLA will isolate genomic DNA from these samples. UCLA will assay each subject's DNA for the five genetic polymorphisms described above using conventional PCR or a TaqMan assay. The SNP alleles will be analyzed for association with susceptibility to develop cardiomyopathy in response to Herceptin® and anthracyclines.

2.5.10.2 Biochemical Markers

2.5.10.2.1 Brain Natriuretic Peptide

Natriuretic peptides, brain natriuretic peptide (BNP) and atrial natriuretic peptide (ANP) are released in response to myocardial stretch and injury. ANP and BNP, promote natriuresis and vasodilation as well as suppressing the renin-angiotensin system, attenuating the hemodynamic disturbances seen in heart failure. The levels of these peptides are elevated in both patients with heart failure and patients with asymptomatic cardiac dysfunction [123]. Indeed, BNP is elevated before heart failure symptoms develop and may permit early detection of early cardiac dysfunction. In a comparative study, levels of ANP, N-terminal ANP, and BNP were measured in patients referred to a heart failure clinic. Levels of all three peptides were significantly higher in patients found to have heart failure by clinical assessment and echocardiography. However, BNP was reported to have the highest sensitivity, specificity, and positive predictive value compared to ANP and N-terminal ANP for the diagnosis of heart failure. BNP also has been used to predict the development of heart failure in healthy subjects [124].

Preliminary studies have looked at the utility of the BNP assay in detection of chemotherapeutic cardiotoxicity. A study from Japan measured several biochemical markers before and after anthracycline chemotherapy including BNP, ANP, norepinephrine, aldosterone, angiotensin II and CK-MB. Only BNP had significantly elevated post-chemotherapy values [125]. Elevations in BNP levels have also been correlated with subclinical myocardial dysfunction [126].

2.5.10.2.2 Cardiac Enzymes

Troponins are proteins involved in the regulation of both cardiac and skeletal muscle contraction, via their calcium-mediated interaction with actin and myosin. Cardiac troponins are encoded by different genes than skeletal troponins and thus have unique amino acid sequences easily distinguished by immunologic assays. Troponins are not normally found in the circulation and their presence in the sera is indicative of myocardial injury and loss of cell membrane integrity. Circulating levels of the Troponin T isoform are specific indicators of myocardial damage, more sensitive than the creatinine phosphokinase MB isoenzyme. The troponin assay is widely used to aid in diagnosis of myocardial ischemia and holds prognostic value. Elevated levels of circulating Troponin T are independent predictors of both short- and long-term mortality.

Recent studies have demonstrated that circulating troponin levels are associated with the presence and severity of heart failure of both ischemic and nonischemic etiology [127]. Mechanisms of release of troponins from the myocytes into the circulation are unknown but may include recurrent ischemia, impaired subendocardial perfusion, apoptotic cell death, and myocardial remodeling. Furthermore, there is a recently evolving role for troponins in heart failure prognosis. Cardiac troponins have been shown to predict the development of myocardial dysfunction and heart failure in patients receiving chemotherapeutic agents that are potentially cardiotoxic. A 1999 study demonstrated elevated troponin T in close to 30% of children receiving anthracycline based chemotherapy for ALL. Higher troponin elevation correlated with higher cumulative anthracycline dose and future elevated troponin values, suggesting chronically active cardiac injury [128]. Initial studies in rats show troponin T to be a sensitive means of predicting anthracycline cardiotoxicity in rats [129]. A recent study from Italy demonstrated troponin T to be a significant predictor of future development of significant and prolonged cardiac dysfunction. Patients with positive troponin values had significantly decreased mean left ventricular ejection fraction at end of 7 month follow up compared to those with negative troponin values who did not. Furthermore, a LVEF of less than 50% was seen during follow up in 30% of positive troponin patients but none of troponin negative patients [130]. As levels of troponin T remain elevated in serum longer than troponin I (12 vs. 7 days), troponin T is more suited to lab draw schedule of the BCIRG study protocol.

Studies to date have shown elevations in BNP and troponin T to be predictors of the later development of anthracycline-induced cardiac dysfunction. To determine prospectively if these markers are useful for the early detection of LV dysfunction, we propose to make serial measurements throughout the study. The assays are also to be drawn at time of any clinical evidence of cardiac failure (chest pain, dyspnea at rest, fluid overload, sinus tachycardia).

Plasma samples are to be collected at baseline, cycle 4, end of chemotherapy for AC→TH and AC→T arms or 6 weeks after the end of chemotherapy visit for TCH arm (FUp1a) and then every 6 months during the first 3 years of follow-up, at year 5 of follow-up (FU #14) and at any time of clinical evidence of cardiac failure. If the 60 months FUP visit has already happened for a patient when amendment 5 becomes available, plasma sample should be scheduled as soon as the patient has signed her informed consent, at time of LEVF8.

III STUDY OBJECTIVES

OBJECTIVES

Primary Objective

To compare disease-free survival after treatment with doxorubicin and cyclophosphamide followed by docetaxel (Taxotere®) (AC→T) with doxorubicin and cyclophosphamide followed by docetaxel and trastuzumab (Herceptin®) (AC→TH) and with docetaxel in combination with carboplatin and Herceptin® (TCH) in the adjuvant treatment of node positive and high risk node negative patients with operable breast cancer containing the HER2 alteration.

Secondary Objectives

- *For all patients*

To compare overall survival between the 3 above mentioned arms.

To compare cardiac toxicity between the 3 above mentioned arms.

To compare toxicity and quality of life between the 3 above mentioned arms.

To evaluate pathologic and molecular markers for predicting efficacy in these patient groups.

In addition, an independent socio-economic study will be conducted in parallel with the clinical study.

- *For patients having consented to the optional substudies*

To correlate baseline peripheral levels of the shed HER2 extracellular domain (ECD) with baseline FISH results and to determine whether peripheral levels of HER2 ECD measured at different timepoints constitute a prognostic and/or predictive factor of disease free survival and survival.

To evaluate genetic and biochemical markers for predicting risk of developing cardiac dysfunction and later cardiac events in these patient groups.

IV PATIENT DEFINITION

4.1 Number of Patients/Enrollment Period/Follow-up Period

This is a multi-center, international study involving 3,150 patients (1,050 patients per arm). Enrollment starts in March 2001 and stops in March 2004 with a clinical follow-up period of 10 years. Interim analyses are planned when 300, 450 and 650 events have been observed. Main analysis is planned when 900 events have been observed. The first follow-up analysis is planned for 3 years after the main analysis, and the second follow-up analysis planned for 5 years after the main analysis.

4.2 Duration of Treatment

All included patients in each arm will receive a fixed number of cycles of treatment.

AC→T: AC q3weeks x 4 cycles followed by docetaxel single agent q3weeks x 4 cycles.

AC→TH: AC q3weeks x 4 cycles followed by docetaxel single agent q3weeks x 4 cycles. Herceptin® will be administered weekly during treatment with docetaxel. Three weeks after the last infusion of chemotherapy, Herceptin® will then be administered every 3 weeks.

TCH: Docetaxel/carboplatin q3weeks x 6 cycles . Herceptin® will be administered weekly during treatment with chemotherapy. Three weeks after the last infusion of chemotherapy, Herceptin® will then be administered every 3 weeks.

4.3 Inclusion Criteria

1 Written informed consent prior to beginning specific protocol procedures, including expected cooperation of the patients for the treatment and follow-up, must be obtained and documented according to local regulatory requirements.

2. Histologically proven breast cancer with an interval between definitive surgery that includes axillary lymph node involvement assessment and registration of less than or equal to 60 days. If the definite surgery and the axillary node dissection are performed in two different days, both days should be within the 60 days window. A central pathology review may be performed post randomization for confirmation of diagnosis and molecular studies. The same block used for HER2 determination prior to randomization may be used for the central pathology review. See Appendix 3* for details on this process.

3 Definitive surgical treatment must be either mastectomy **with** axillary lymph node involvement assessment, or breast conserving surgery **with** axillary lymph node involvement assessment for operable breast cancer. Margins of resected specimen from definitive surgery must be histologically free of invasive adenocarcinoma and/or ductal carcinoma in situ (DCIS). The finding of lobular carcinoma in-situ will not be scored as a positive margin.

4 Patients must be either lymph node positive or high risk node negative. Lymph node positive patients will be defined as patients having invasive adenocarcinoma with at least one axillary lymph node (pN1) showing evidence of tumor among a minimum of six resected lymph nodes.

High risk lymph node negative patients will be defined as patients having invasive adenocarcinoma with either 0 (pN0) among a minimum of 6 resected lymph nodes or negative sentinel node biopsy (pN0) AND at least one of the following factors: tumor size > 2 cm, ER and PR status is negative, histologic and/or nuclear grade 2-3, or age < 35 years.

5 Tumor must show the presence of the HER2 gene amplification by Fluorescence In-Situ Hybridization (FISH analysis) in a designated central laboratory (see Appendix 3 for complete details).

6. Estrogen and/or progesterone receptor analysis performed on the primary tumor prior to randomization. Results must be known at the time of randomization.

(Note: Patients whose tumor is estrogen receptor negative with progesterone receptor status unknown or undetermined, MUST have the PR assayed in order to determine hormonal receptor status. Patients whose tumor is progesterone receptor negative with estrogen receptor status unknown or undetermined, MUST have the ER assayed in order to determine hormonal receptor status.)

7 Age ≥ 18 years and age ≤ 70 years. The upper age limit is not meant to be exclusionary but rather is based on the lack of safety data for the TCH regimen in women >70 years of age.

8 Karnofsky Performance status index ≥ 80%.

9 Normal cardiac function must be confirmed by LVEF (echocardiography or MUGA scan) and ECG within 3 months prior to registration. The result of the echocardiography or MUGA must be equal to or above the lower limit of normal for the institution.

10 Laboratory requirements: (within 14 days prior to registration)

a) Hematology:
   i) Neutrophils ≥ 2.0 10^9/L
   ii) Platelets ≥ 100 10^9/L
   iii) Hemoglobin ≥ 10 g/dL

b) Hepatic function:
   i) Total bilirubin ≤ 1 UNL

ii) ASAT (SGOT) and ALAT (SGPT) ≤ 2.5 UNL
iii) Alkaline phosphatase ≤ 5 UNL
iv) Patients with ASAT and/or ALAT > 1.5 x UNL **associated** with alkaline phosphatase > 2.5 x UNL are not eligible for the study.

c) Renal function:
i) Creatinine ≤ 175 µmol/L (2 mg/dL)
ii) If at upper normal limit the calculated creatinine clearance should be ≥ 60 mL/min.

11 Complete staging work-up within 3 months prior to registration. All patients will have contralateral mammography and/or ultrasound (mammogram is preferred), chest X-ray (PA and lateral) and/or CT and/or MRI, abdominal ultrasound and/or CT scan and/or MRI, and bone scan. In cases of positive bone scans, bone X-ray or bone MRI evaluation is mandatory to rule out the possibility of metastatic bone scan positivity. Other tests may be performed as clinically indicated (see appendix 5).

12 Patients must be accessible for treatment and follow-up. Patients registered on this trial must be treated and followed at participating centers which will include Principal or Co-investigator's sites.

13 Negative pregnancy test (urine or serum) within 7 days prior to registration for all women of childbearing potential.

4.4 Exclusion Criteria

1 Prior systemic anticancer therapy for breast cancer (immunotherapy, hormonotherapy, chemotherapy).

2 Prior anthracycline therapy, taxoids (paclitaxel, docetaxel) or platinum salts for any malignancy.

3 Prior radiation therapy for breast cancer.

4 Bilateral invasive breast cancer.

5 Pregnant, or lactating patients. Patients of childbearing potential must implement adequate non-hormonal contraceptive measures during study treatment (chemotherapy, Herceptin® and tamoxifen therapy) and must have negative urine or serum pregnancy test within 7 days prior to registration.

6 Any T4 or N2 or known N3 or M1 breast cancer.

7 Pre-existing motor or sensory neurotoxicity of a severity ≥ grade 2 by NCI criteria.

8 Cardiac disease that would preclude the use of doxorubicin, docetaxel and Herceptin®.
a) any documented myocardial infarction
b) angina pectoris that requires the use of antianginal medication
c) any history of documented congestive heart failure
d) Grade 3 or Grade 4 cardiac arrhythmia (NCI CTC, version 2.0)
e) clinically significant valvular heart disease
f) patients with cardiomegaly on chest x-ray or ventricular hypertrophy on ECG, unless they demonstrate by echocardiography or MUGA scan within the past 3 months that the LVEF is ≥ the lower limit of normal for the radiology facility
g) patients with poorly controlled hypertension i.e. diastolic greater than 100 mm/Hg. (Patients who are well controlled on medication are eligible for entry)
h) patients who currently receive medications (digitalis, beta-blockers, calcium channel-blockers, etc) that alter cardiac conduction, if these medications are administered for cardiac arrhythmia, angina or congestive heart failure. If these medications are administered for other reasons (ie hypertension), the patient will be eligible.

9 Other serious illness or medical condition:
   a) history of significant neurologic or psychiatric disorders including psychotic disorders, dementia or seizures that would prohibit the understanding and giving of informed consent
   b) active uncontrolled infection
   c) active peptic ulcer, unstable diabetes mellitus
   d) patients with symptomatic, intrinsic lung disease resulting in dyspnea

10 Past or current history of neoplasm other than breast carcinoma, except for:
   a) curatively treated non-melanoma skin cancer
   b) in situ carcinoma of the cervix
   c) other cancer curatively treated and with no evidence of disease for at least 10 years
   d) ipsilateral ductal carcinoma in-situ (DCIS) of the breast
   e) lobular carcinoma in-situ (LCIS) of the breast
   f) DCIS involving the contralateral breast removed by mastectomy

11 Current therapy with any hormonal agent such as raloxifene, tamoxifen, or other selective estrogen receptor modulators (SERMs), either for osteoporosis or prevention. Patients must have discontinued these agents prior to randomization.

12 Chronic treatment with corticosteroids **unless** initiated > 6 months prior to study entry **and** at low dose (≤ 20 mg methylprednisolone or equivalent).

13 Concurrent treatment with ovarian hormonal replacement therapy. Prior treatment must be stopped prior to randomization.

14 Definite contraindications for the use of corticosteroids.

15 Concurrent treatment with other experimental drugs. Participation in another clinical trial with any investigational not marketed drug within 30 days prior to study entry.

16 Concurrent treatment with any other anti-cancer therapy.

17 Male patients, as no clinical efficacy or safety data are available from phase I-II studies.

V PLAN OF THE STUDY

5.1 General

5.1.1 Surgery; Axillary Lymph Node Involvement Assessment

Definitive surgical treatment must be either mastectomy with axillary lymph node involvement assessment or breast conserving surgery with axillary lymph node involvement assessment, for operable breast cancer (T1-3, clinical N0-1, M0). Margins of resected specimen from definitive surgery must be histologically free of invasive carcinoma and/or ductal carcinoma in situ. The finding of lobular carcinoma in-situ will not be scored as a positive margin. For positive lymph node involvement assessment, axillary lymph node dissection must be performed with at least one axillary lymph node (pN1) among a minimum of 6 resected lymph nodes. For negative lymph node involvement assessment, 0 (pN0) among a minimum of 6 resected nodes OR a negative sentinel node biopsy (pN0) may be used for the axillary lymph node assessment. Only centers with established experience in sentinel lymph node biopsy will be allowed to use such a procedure for the purpose of this study.

5.1.2 High Risk Node Negative Patients

High risk node negative will be defined as a patient with pN0 lymph node involvement (as above), AND at least 1 of the following factors:
Tumor size > 2 cm, estrogen receptor and progesterone receptor status is negative, histologic and/or nuclear grade 2-3, or age < 35 years.

5.1.3 Estrogen and/or Progesterone Receptor Status

Patients must have an analysis of estrogen and/or progesterone receptor on the primary tumor sample. Results must be known prior to randomization. ER-positive tumors can be defined as positive by the Dextran-coated charcoal or sucrose-density gradient method, or positive (using individual laboratory criteria) by the enzyme immunoassay method (EIA), or by immunocytochemical assay. Those not definitively negative i.e "borderline", etc, will also be considered positive.

Patients whose tumor is estrogen receptor negative with progesterone receptor status unknown or undetermined, MUST have the PR assayed in order to determine hormonal receptor status.

Patients whose tumor is progesterone receptor negative with estrogen receptor status unknown or undetermined, MUST have the ER assayed in order to determine hormonal receptor status.

5.1.4 General Study Plan

This is a prospective, *non-blinded, randomized*, phase III trial. Patients will be post surgically stratified at inclusion into 3 groups according to institution, the number of no axillary lymph nodes involved (N0), 1 to 3 (N1-3), 4 or more axillary lymph nodes involved (N4+), and by estrogen and/or progesterone receptor status (positive or negative) and will be randomly assigned to receive adjuvant treatment with either:

AC→T: **AC followed by T:** Doxorubicin 60 mg/m² as an IV bolus in combination with cyclophosphamide 600 mg/m² IV followed by docetaxel 100 mg/m² as 1 hour IV infusion on day 1 every 3 weeks (see 5.2.1 for administration schedule).

AC→TH: **AC followed by T with Herceptin®:** Doxorubicin 60 mg/m² IV in combination with cyclophosphamide 600 mg/m² IV on an every 3 week basis for 4 cycles. Three weeks after the last cycle of AC, Herceptin® 4 mg/kg loading dose by IV infusion over 90 minutes on Day 1 of Cycle 5 will be administered, followed by Herceptin® 2 mg/kg by IV infusion over 30 minutes weekly starting Day 8; and docetaxel 100 mg/m² administered by IV infusion over 1 hour on Day 2 of Cycle 5, then on day 1 on an every 3 week basis for all subsequent cycles (total 4 cycles of docetaxel). Beginning three weeks after the last cycle of chemotherapy, Herceptin® 6 mg/kg by IV infusion over 30 minutes will be given every 3 weeks. Herceptin® will continue for 1 year from date of first administration i.e. Herceptin® administration will stop at 1 year from its initiation, regardless of the number of doses received or missed. For those cycles where docetaxel and Herceptin® are due to be administered on the same day, docetaxel to be administered first, except for the first cycle, when Herceptin® loading dose is given on day 1 and docetaxel on day 2.

TCH: **Docetaxel / Carboplatin / Herceptin®:** Herceptin® 4 mg/kg loading dose by IV infusion over 90 minutes on Day 1 of Cycle 1 only, followed by Herceptin® 2 mg/kg by IV infusion over 30 minutes weekly starting on Day 8 until three weeks after the last cycle of chemotherapy. Beginning three weeks after the last cycle of chemotherapy, Herceptin® 6 mg/kg by IV infusion over 30 minutes will be given every 3 weeks. Docetaxel 75 mg/m² will be administered on Day 2 of Cycle 1, then on Day 1 of all subsequent cycles by IV infusion over 1 hour followed by carboplatin at target AUC = 6 mg/mL/min administered by IV infusion over 30-60 minutes repeated every 3 weeks. A total of six cycles of docetaxel and carboplatin will be administered every 3 weeks. Herceptin® will continue for 1 year from date of first administration i.e. Herceptin® administration will stop at 1 year from its initiation, regardless of the number of doses received or missed. For those cycles where chemotherapy and Herceptin® are due to be administered on the same day, docetaxel will be

administered first, followed by carboplatin followed by Herceptin® except for the first cycle, when Herceptin® loading dose is given on day 1 and docetaxel/carboplatin on day 2.

The chemotherapy doses will be calculated according to baseline body surface area (BSA) for all cycles. If there is a 10% or greater decrease in body weight compared to baseline, the BSA will be recalculated.

If the calculated BSA of the patient is > 2.2 m², the dose to be given to the patient will be calculated according to BSA = 2.2 m². No ideal body weight should be used for the calculation of BSA.

Herceptin® dosing will be based on the baseline weight. Weekly weight measurements will be required for those patients receiving Herceptin® during chemotherapy and q3weekly Herceptin®during the follow-up phase. In case of a 10 % increase or decrease in weight, Herceptin® dose should be recalculated using the new weight.

Dose reduction and/or treatment delay and treatment discontinuation are planned for each arm in case of severe hematological and/or non-hematological toxicities. See Section 5.3.

- **Indication for Hormonal therapy**:

  Tamoxifen (20 mg p.o. daily) for 5 years will be administered starting 3 to 4 weeks after the last course of chemotherapy for patients with positive estrogen and/or progesterone receptors.

  Post menopausal patients are allowed to switch to Anastrozole in case of Tamoxifen related severe toxicities (e.g. hot flushes, vaginal bleeding, vaginal discharge, thromboembolic events). Anastrazole will be given at the dose of 1 mg daily. Of note, the total duration of the hormonal therapy, i.e. Tamoxifen followed by Anastrozole, should not exceed 5 years. Only post-menopausal patients are eligible to receive Anastrozole.

  Post menopausal women without contraindications to the use of Tamoxifen and who have already started Tamoxifen, can receive a sequential therapy consisting of Tamoxifen for 2 to 3 years followed by Anastrozole or Exemestane for a maximum of 5 years of hormonal therapy.

  Post menopausal patients who have not yet started hormonal therapy can receive 5 years of Anastrozole or sequential therapy consisting of Tamoxifen for 2 to 3 years followed by Anastrozole or Exemestane for a maximum of 5 years of hormonal therapy.

  Post menopausal patients who have completed 5 years Tamoxifen are allowed to continue the hormonal treatment with letrozole for a maximum of 3 years.

  Note: - the use of AIs should be accurately documented and reported on page FU6 of the CRF,

  - the same approach is followed for all patients treated in the BCIRG 006/TAX GMA 302 study in order to avoid treatment imbalance between the 3 arms.
  - Each center will be requested to provide BCIRG data center with their institution guideline related to the use of aromatase inhibitor by completing a questionnaire.

- **Radiation Indication - Each Arm**: Patients treated with lumpectomy will undergo postoperative radiation therapy after completion of chemotherapy and resolution of any side effect. Postmastectomy radiation therapy, and ipsilateral nodal radiation therapy, may be used at the discretion of the treating radiation oncologist. This will be done in a consistent manner according to the guidelines at each institution. Radiation guidelines will be requested from each institution prior to study start at the institution.

- **Each Arm:** No more than 8 days should elapse between the date of randomization and the start date of the first cycle of adjuvant chemotherapy.

5.1.5 Study Medication

For the purpose of this study, study medication will be defined as the combination of chemotherapy in each of the study arms for the duration of the active treatment. For the two arms containing Herceptin®, Herceptin® will be included in the definition of study medication. For those patients receiving hormonal therapy as per protocol, such treatment will not be considered in the definition of study medication.

5.2 Study Treatment

5.2.1 AC→T

AC→T will consist of 4 cycles of AC followed by 4 cycles of docetaxel:

AC Segment:

Doxorubicin will be given first

Dose: 60 mg/m², day 1
Route: 5 - 15 minute intravenous bolus injection (as per hospital policy)
Schedule: every 3 weeks

followed by

Cyclophosphamide

Dose: 600 mg/m², day 1
Route: 5 to 60 minutes intravenous bolus injection (as per hospital policy)
Schedule: every 3 weeks

This is called *a cycle of treatment and is to be given 4 times*.

Docetaxel Segment

Three weeks after the last course of AC, docetaxel will be given

Dose: 100 mg/m², day 1
Route: 1 hour intravenous infusion. During the first 5 minutes, the infusion must be done drop by drop in order to reduce the incidence of acute hypersensitivity reaction (AHSR).
Schedule: every 3 weeks

This is called *a cycle of* treatment and is to be given *4 times*.

5.2.2 AC→TH

AC→TH will consist of 4 cycles of AC followed by 4 cycles of docetaxel followed by Herceptin® for 1 year from the date of first Herceptin® administration i.e. Herceptin® administration will stop at 1 year from its initiation, regardless of the number of doses received or missed.

AC Segment:

Doxorubicin will be given first

Dose: 60 mg/m², day 1
Route: 5 - 15 minute intravenous bolus injection (as per hospital policy)
Schedule: every 3 weeks

followed by

Cyclophosphamide

Dose: 600 mg/m², day 1
Route: 5 to 60 minutes intravenous bolus injection (as per hospital policy)
Schedule: every 3 weeks

This is called *a cycle of treatment* and *is to be given 4 times*. Following completion of 4 cycles, the patient will enter the TH Segment for this arm of treatment.

TH Segment

FIRST CYCLE OF DOCETAXEL / HERCEPTIN® ONLY

Day 1: Herceptin®* 4 mg/kg administered by IV infusion over 90 minutes.

Day 2: Docetaxel 100 mg/m² by IV infusion (in minimum 250 mL of normal saline or dextrose 5% solution) over 1 hour.

Day 8: Herceptin®* 2 mg/kg administered by IV infusion over 30 minutes.

Day 15: Herceptin®* 2 mg/kg administered by IV infusion over 30 minutes.

***See Table 7 for Herceptin® infusion times and post-infusion observation periods**.

SUBSEQUENT CYCLES (to start 3 weeks after Day 1 of 1st cycle of the TH segment. The patient will receive a total of 4 cycles of Docetaxel with Herceptin® and then receive Herceptin® as monotherapy for one year).

Day 1: Docetaxel 100 mg/m² as 1 hour IV infusion every 3 weeks followed by Herceptin®* 2 mg/kg IV infusion over 30 minutes.

Day 8: Herceptin®* 2 mg/kg administered by IV infusion over 30 minutes.

Day 15: Herceptin®* 2 mg/kg administered by IV infusion over 30 minutes.

***See Table 7 for Herceptin® infusion times and post-infusion observation periods**.

LAST CYCLE

Day 1: Docetaxel 100 mg/m² as 1 hour IV infusion every 3 weeks followed by Herceptin®* 2 mg/kg IV infusion over 30 minutes.

Day 8: Herceptin®* 2 mg/kg administered by IV infusion over 30 minutes.

Day 15: Herceptin®* 2 mg/kg administered by IV infusion over 30 minutes.

Day 22: (=EOC) Herceptin®* 6 mg/kg administered by IV infusion over 30 minutes.

Herceptin®* will then be administered at 6 mg/kg by IV infusion over 30 minutes every 3 weeks. Initiation of the q3weekly infusion of Herceptin® will correspond to the end of chemotherapy (EOC) visit.

***See Table 7 for Herceptin® infusion times and post-infusion observation periods**.*

Treatment will continue until one-year Herceptin® treatment completion, disease relapse, withdrawn consent or unacceptable toxicity.

TABLE 7 Herceptin® Infusion Times and Post-Infusion Observation Period

Infusion of Herceptin®	Herceptin® Dose *Do not administer as i.v push or bolus*	Infusion Time (minutes)	Total Observation Period starting at the time of the infusion[b].
During Chemotherapy Cycles: (weekly infusions)			According to local prescribing information.
• First Infusion	4 mg / kg	90	For example: • European Union: 6 hours • US: 2 ½ hours
• Second Infusion	2 mg/kg	30 [a]	According to local prescribing information. For example: • European Union: 2 hours • US: 1 hour
• Subsequent Infusions	2 mg / kg	30 [a]	According to local prescribing information. For example: • European Union: 2 hours • US: 30 minutes[c]
During Follow-Up Visits (q3weekly infusion)	6 mg / kg	30 [a]	According to local prescribing information. For example: • European Union: 2 hours • US: None [c]

[a]Only if previous dose was well tolerated. In the first cycle, the docetaxel infusion should be started only after all acute toxicities from Herceptin® infusion have resolved.

[b] For those countries where longer observation periods are required, investigators should continue to monitor their patients according to local prescribing information - i.e: After receiving her first infusion patient should stay at Hospital for an additional period to fit to the local prescribing information.

[c] Only if previous dose was well tolerated

5.2.3 TCH

A total of 6 cycles of docetaxel and carboplatin will be administered every 3 weeks. Herceptin® will be administered weekly during treatment with chemotherapy and then every three weeks in the follow-up period. Herceptin® will continue for 1 year from date of first administration i.e. Herceptin® administration will stop at 1 year from its initiation, regardless of the number of doses received or missed. Herceptin® will continue until 1 year completion, disease relapse, withdrawn consent or unacceptable toxicity, whichever comes first.

FIRST CYCLE

Day 1: Herceptin®* 4 mg/kg loading dose administered by IV infusion over 90 minutes.

Day 2: Docetaxel 75 mg/m² by IV infusion over 1 hour followed by carboplatin at target AUC = 6 mg/mL/min administered by IV infusion over 30 – 60 minutes minutes

Day 8: Herceptin®* 2 mg/kg administered by IV infusion over 30 minutes.

Day 15: Herceptin®* 2 mg/kg administered by IV infusion over 30 minutes.

***See Table 7 for Herceptin® infusion times and post-infusion observation periods**.

SUBSEQUENT CYCLES (to start 3 weeks after Day 1 of 1st cycle)

Day 1: Docetaxel 75 mg/m² by IV infusion over 1 hour followed by carboplatin at target AUC = 6 mg/mL/min administered by IV infusion 30 – 60 minutes every 3 weeks followed by Herceptin®* 2 mg/kg administered by IV infusion over 30 minutes.

Day 8: Herceptin®* 2 mg/kg administered by IV infusion over 30 minutes.

Day 15: Herceptin®* 2 mg/kg administered by IV infusion over 30 minutes.

***See Table 7 for Herceptin® infusion times and post-infusion observation periods**.

LAST CYCLE

Day 1: Docetaxel 75 mg/m² by IV infusion over 1 hour followed by carboplatin at target AUC = 6 mg/mL/min administered by IV infusion 30 – 60 minutes followed by Herceptin®* 2 mg/kg administered by IV infusion over 30 minutes.

Day 8: Herceptin®* 2 mg/kg administered by IV infusion over 30 minutes.

Day 15: Herceptin®* 2 mg/kg administered by IV infusion over 30 minutes.

Day 22: (=EOC) Herceptin®* 6 mg/kg administered by IV infusion over 30 minutes.

Herceptin®* will then be administered at 6 mg/kg by IV infusion over 30 minutes every 3 weeks. Initiation of the q3weekly infusion of Herceptin®* will correspond to the end of chemotherapy (EOC) visit.

***See Table 7 for Herceptin® infusion times and post-infusion observation periods**.*

Herceptin® treatment will continue until 1-year Herceptin® treatment completion, disease relapse, withdrawn consent or unacceptable toxicity Herceptin® or Herceptin®

5.2.4 Carboplatin Dose

Carboplatin dose is calculated using a modified Calvert formula (creatinine clearance is substituted for GFR) as follows:

Total dose (mg) = (Target AUC) x (Creatinine Clearance + 25)

Note: 1. Carboplatin dose is calculated in mg, not mg/m²

2. Target AUC = 6 mg/mL/min, initially. It may be decreased due to toxicity as per section 5.4

3. Creatinine clearance can either be measured or estimated using the Cockroft-Gault formula, as follows:

$$\text{Creatinine Clearance (mL/min)} = \frac{(140 - \text{age}) \times (\text{weight in kg})}{72 \times \text{serum creatinine in mg/dL}} \times 0.85$$

For the dosing of the carboplatin using the modified Calvert forumula, the calculation of the creatinine clearance will be done according to baseline weight. If there is a 10% or greater decrease in body weight compared to baseline, the calculation should be revised according to the new actual weight.

In case of serum creatinine value $\leq$ 0.9 mg/dL, it is strongly advise to measure the creatinine clearance. If the creatinine clearance value cannot be measured, the serum creatinine value can be adjusted to 1mg/dl in the Cockroft-Gault formula, if the investigator considers that there is a risk of over dosing the patient by using her actual serum creatinine value.

If a patient has a BSA more than 2.2, the weight to be used to calculate the estimated creatinine clearance in the Cockroft-Gault formula should be the weight which results in a BSA of 2.2.

5.2.5 Prophylactic Premedication Regimen for Docetaxel-Related Hypersensitivity Reactions and Fluid Retention

The following premedication regimen must be administered for all patients treated with docetaxel.

Dexamethasone 8 mg p.o. for total of 6 doses.

1. night before chemotherapy
2. immediately upon waking the morning of chemotherapy
3. one hour before infusion of docetaxel (may be given oral or intravenous)
4. night of chemotherapy
5. morning the day after chemotherapy
6. evening the day after chemotherapy

Deaxamethasone can be administered i.v the morning of chemotherapy.

Dexamethasone 8 mg equivalent may be used
Dexamethasone 8 mg = Methylprednisolone 40 mg = Prednisone 50 mg = Prednisolone 50 mg

5.2.6 Use of Prophylactic Antibiotics

Primary prophylactic use of antibiotics is not allowed in any arm. Prophylactic use of antibiotics will be used in subsequent chemotherapy cycles for those patients who have experienced a serious or life-threatening infection only (see Section 5.4).

5.2.7 Use of Prophylactic G-CSF

G-CSF as primary prophylaxis (i.e. from 1st cycle onwards) is not mandatory but may be used at the discretion of the investigator.

5.2.8 Use of Prophylactic Antiemetics

Antiemetic prophylaxis is mandatory for all patients. Selection of antiemetics is at the discretion of the investigator.

5.3 Treatment Delays and Dose Reduction / Modification

5.3.1 Treatment Delays

Treatment with chemotherapy may be delayed no more than 2 weeks (up to Day 35) to allow recovery from acute toxicity.

Herceptin® treatment may continue while chemotherapy is being withheld due to chemotherapy-related toxicity at investigator discretion. The exception is for the asymptomatic decreases in left ventricular ejection fraction, where specific rules are outlined in Section VI.

If a patient misses a Herceptin® dose for any reason, the missed dose may be rescheduled later in that same calendar week or not given that week at all. A patient may not be given 2 Herceptin® doses in the same calendar week.

Clarification on the meaning of the calendar week.
If Herceptin® administration is due on a Monday but for any reason cannot be performed that day, the infusion can be made up to the Sunday of the same week (same calendar week) and the date of the following administration can be back to the initial schedule (Monday the week after).

Example: Infusion #1 planned on Monday Feb 4, 2002 delayed due to logistic issue
Infusion #1 administered on Sunday Feb 10, 2002 (same calendar week)
Infusion #2 can be administered on Monday Feb 11, 2002 (next calendar week)

If the patient does not suffer from any Herceptin®-related events that may cause Herceptin® to be stopped permanently, *Herceptin® administration is to stop at 1 year from its initiation, regardless of the number of doses of Herceptin® the patient has received or missed.* The exception is for those patients in the cardiac safety analysis who may have to permanently stop Herceptin® under conditions outlined in Section VI.

Regardless of Herceptin® treatment period, Herceptin® should be held for grade 3 or 4 non-hematologic toxicity which is related to Herceptin® until recovery to grade ≤ 2, excluding cardiac dysfunction (refer to cardiac dysfunction guideline in part 6). If patients experience non-hematologic Herceptin® related toxicity that does not resolve (to grade 1 or 2), continuing Herceptin administration® will be left at the discretion of the investigator. Uncertain casesshould be discussed with the sponsor. If the same non-hematologic toxicity recurs at a grade of 3 or 4, treatment should be permanently discontinued. The Herceptin® dose should not be held for hematologic toxicity.

Should the patient relapses, patient will be discontinued from the study treatment including Herceptin®.

If a patient misses a dose by less than a week, give the usual dose (2mg/kg for the weekly schedule, 6mg/kg for the 3 weekly schedule) as soon as possible (don't wait until the next planned cycle). Carry on the maintenance doses (2mg/kg for the weekly schedule, 6mg/kg for the 3 weekly schedule) according to the original schedule.

If a patient misses a dose by more than a week, re-start the treatment as if she were a new patient. The treatment should be re-started as soon as possible (don't wait until the next planned cycle) with a loading dose (4mg/kg for the weekly schedule, 8mg/kg for the 3 weekly schedule) given over 90 minutes. The usual maintenance schedule (2mg/kg for the weekly schedule, 6mg/kg for the 3 weekly schedule) should follow, as for a new patient (weekly starting one week after loading for the weekly schedule; 3 weekly, starting 3 weeks after loading for the 3-weekly schedule).

5.3.2 Treatment Dose Adjustments

Chemotherapy dose adjustments are to be made according to the system showing the greatest degree of toxicity. Toxicities will be graded using the NCI Common Toxicity Criteria, version 2.0 (see Appendix 12).

Dose reduction is planned for each arm in case of severe hematological and/or non-hematological toxicities.

Doses which have been reduced for toxicity must not be re-escalated with the exception of liver function tests that improve within ranges given.

There will be no dose modifications of Herceptin. If Herceptin is resumed following missed dose(s), refer to section 5.3.1 Treatment Delays for the rules that apply in case of Herceptin dose missed.

IF A PATIENT EXPERIENCES SEVERAL TOXICITIES AND THERE ARE CONFLICTING RECOMMENDATIONS, PLEASE FOLLOW THE MOST CONSERVATIVE DOSE ADJUSTMENT RECOMMENDED.

5.4 Toxicity Related Guidelines for Dose Reduction and Dose Modification

5.4.1 Hematological Toxicities

5.4.1.1 Febrile Neutropenia

Febrile neutropenia shall be defined as oral or tympanic fever of ≥ 38.5° C or 101.3 ° F in the presence of neutropenia (where neutropenia is defined as ANC < 1.0 x 10^9 /L). See NCI CTC, version 2.0 (Appendix 12).

A therapeutic intervention should proceed immediately following the diagnosis of febrile neutropenia. Therapeutic interventions can be as per the institution's guidelines, or may include

- hospital admission
- pre-antibiotic evaluation
- CBC with differential and blood culture should be performed
- start of an empirical antibiotic therapy

In case of febrile neutropenia, blood counts must be done every 2 days until recovery of ANC ≥ 1.0 or oral temperature < 38.5°C. This must be documented in the CRF Form for Febrile Neutropenia.

For all subsequent chemotherapy cycles, prophylactic G-CSF will be added as per guidelines outlined in section 5.4.1.5. Antibiotics are not allowed as prophylaxis for febrile neutropenia.

5.4.1.2 Infection With (or Without) Neutropenia

For severe (Grade 3) or life-threatening (Grade 4) infection during chemotherapy, with or without neutropenia, prophylactic G-CSF and prophylactic antibiotics will be added to all remaining cycles.

Ciprofloxacin is recommended at 500 mg orally twice daily for 10 days starting day 5 of each cycle for remaining chemotherapy cycles. If ciprofloxacin is not available or not tolerated, another oral antibiotic **must** be used. The choice of antibiotic is at the discretion of the investigator.

G-CSF will be added to all subsequent chemotherapy cycles as per guidelines outlined in section 5.4.1.5.

5.4.1.3 2nd Febrile Neutropenia and 2nd Infection Event

In the case of a second febrile neutropenia or infection event, patient will continue with the prophylactic G-CSF for all subsequent cycles. In addition, all chemotherapeutic drug doses will be reduced as follows for all remaining cycles. Herceptin® dose will not be adjusted.

Chemotherapy dose reductions are as follows:

Docetaxel Single Agent: from 100 mg/m² to 75 mg/m²
Docetaxel in Combination: from 75 mg/m² to 60 mg/m²
Doxorubicin: from 60 mg/m² to 50 mg/m²
Cyclophosphamide: from 600 mg/m² to 500 mg/m²
Carboplatin: from AUC 6 mg/mL/min to AUC of 5 mg/mL/min

In the case of a 3rd event, there will be no further dose reduction. Patient will go off chemotherapy (into regular follow-up). Herceptin® may continue at discretion of investigator.

5.4.1.4 Delayed ANC Recovery on Day 21

BLOOD COUNTS ON DAY 21

Neutrophils (x 10^9/L)	Action to be taken
≥ 1.5	Treat on time
< 1.5	1. CBC should be repeated every other day till day 35 Proceed with full dose chemotherapy as soon as ANC ≥ 1.5. Consider curative treatment with G-CSF. Add G-CSF in remaining cycles if recovery occurred after day 28. 2. If there is no recovery on day 35, (ANC < 1.5 x 10^9/L), the patient will go off chemotherapy. In arms with Herceptin®, Herceptin® may continue at the discretion of the investigator.

5.4.1.5 Use of Recombinant Granulocyte Colony Stimulating Factor (G-CSF/ Granocyte®/ Neupogen® /Neulasta®)

a) G-CSF Indications

The use of G-CSF is permitted only:

- As curative treatment in case of febrile neutropenia or infection.
- As prophylactic treatment in patients with a prior episode of febrile neutropenia or infection in earlier cycle (see dose modification section 5.4.1.2).
- As treatment for delayed recovery of absolute neutrophil count at day 21 and as prophylactic treatment for subsequent cycles if recovery occurred after day 28 (see section 5.4.1.4).

- If the investigator wishes to use G-CSF as primary prophylaxis, i.e. from 1st cycle onwards, he/she may do so. **Its use as primary prophylaxis is not MANDATORY, but is an option for investigators to use at their discretion.**

b) Dose and Schedule for G-CSF Prophylaxis

	Granocyte®	Neupogen®	Neulasta®
Dose	150 μg (19.2 MIU)/m²/day	5μg/kg/day	6 mg / cycle
Route	Subcutaneous	Subcutaneous	Subcutaneous
Schedule	Day 4 to Day 11* At Day 11 if ANC < 1.0 X 10^9 / L continue until Day 13 (10 days in total)	Day 4 to Day 11* At Day 11 if ANC < 1.0 X 10^9 / L continue until Day 13 (10 days in total)	One single injection per cycle on Day 2*.

* Day 1 being the day of the infusion, day 4 means 72 h after the day of the infusion.

5.4.1.6 Prophylactic Antibiotics

a) Indication for Prophylactic Antibiotics

No primary prophylactic administration (from first cycle) is permitted.

The use of prophylactic antibiotics is permitted only in patients with a prior episode of infection (grade 3 or grade 4) in an earlier chemotherapy cycle

NOTE : Prophylactic antibiotics will **not** be used in subsequent cycles for patients who have had a prior episode of febrile neutropenia.

b) Dose and Schedule for Prophylactic Antibiotic Prophylaxis

For patients where a serious or life-threatening infection has occurred, a prophylactic antibiotic is required for all subsequent chemotherapy cycles. Ciprofloxacin is recommended at 500 mg orally twice daily for 10 days starting day 5 of each cycle for remaining chemotherapy cycles. If ciprofloxacin is not available or not tolerated, another oral antibiotic **must** be used. The choice of antibiotic is at the discretion of the investigator.

5.4.1.7 Thrombocytopenia

The following dose adjustments are based on the hematologic counts **on the day of or day prior to chemotherapy** treatment.

Platelet Count (cells/μL)	**AC→T or AC→TH or TCH**
> 100,000	No change
< 100, 000	Hold for a maximum of 2 weeks. If during AC, reduce doxorubicin from 60 to 50 mg/m² If during docetaxel, reduce docetaxel from 100 to 75 mg/m² If during TCH, decrease carboplatin to AUC of 5 mg/mL/min and docetaxel from 75 to 60 mg/m².

If after 2 weeks, and no recovery above 50,000, all chemotherapy is permanently discontinued.
If after 2 weeks, recovery above 50,000, treat with dose reduction above for all subsequent doses.

Herceptin® may continue in the Herceptin®-containing arms in all cases above.

5.4.1.8 Anemia

In case of ≥ grade 2 decrease in hemoglobin, treatment with blood transfusion or erythropoietin should be given.
The use of prophylactic erythropoetin for < grade 2 anemia is not allowed.
The choice of the type of erythropoetin used (long action or regular) is at the inestigator's discretion.

In the case where the next cycle of chemotherapy is due, chemotherapy is to be administered if hemoglobin is > 10 g/dL.

In case of ≥ grade 3 or 4 decrease in hemoglobin, treatment with blood transfusion or erythropoetin should be given and doses should be reduced as follows:

With docetaxel as single agent in AC→T and AC→TH, docetaxel dose to be decreased from 100 mg/m² to 75 mg/m².
If during TCH, docetaxel to be reduced from 75 mg/m² to 60 mg/m² and carboplatin reduced from an AUC of 6 mg/mL/min to an AUC of 5 mg/mL/min.

Herceptin® may continue in the Herceptin®-containing arms in all cases above.

5.4.2 Nausea and Vomiting

Antiemetic prophylaxis is mandatory for all patients. Selection of antiemetics is at the discretion of the investigator.

Acute episodes of nausea and vomiting should be controlled with adequate antiemetics. In case of grade 4 vomiting that persists despite antiemetics, patient will go off chemotherapy.

Herceptin® may continue in the Herceptin®-containing arms in all cases above.

5.4.3 Diarrhea

No primary prophylactic treatment for diarrhea is recommended. However, in case of grade 2 to 3 diarrhea, the patient should be treated with loperamide. For subsequent cycles, give loperamide the day of the first episode of diarrhea, including grade 1. If despite this treatment, patient still experiences grade 3 or more diarrhea, the following dose reductions are to be performed.

AC Segment in AC→T and AC→TH
In case of diarrhea ≥ grade 3 in the AC segment of AC→T or AC → TH, reduce the dose of doxorubicin from 60 to 50 mg/m² in the subsequent cycles. If despite dose reduction, diarrhea still occurs at grade ≥ 3, the patient may go off chemotherapy as per investigator discretion.

Docetaxel (T) Segment in AC→T, AC→TH, TCH
In case of diarrhea ≥ grade 3 during treatment with docetaxel, reduce the dose of docetaxel from 75 to 60 mg/m² (TCH) or from 100 to 75 mg/m² (AC→T or AC →TH) in the subsequent cycles. If despite dose reduction diarrhea still occurs at grade ≥3, the patient may go off chemotherapy as per investigator discretion.

Herceptin® may continue in the Herceptin®-containing arms in all cases above.

5.4.4 Stomatitis

In case of grade 3 stomatitis (and/or oesophagitis):

AC→T or AC→TH

During AC Segment

Doxorubicin will be reduced from 60 to 50 mg/m². If despite dose reduction, stomatitis still occurs at grade ≥3, doxorubicin will be reduced from 50 to 40 mg/m². No further dose reduction is planned.

During Docetaxel (+/- Herceptin®) Segment

Docetaxel will be reduced from 100 to 75 mg/m². If despite dose reduction, stomatitis still occurs at grade ≥3, docetaxel will be further reduced from 75 to 60 mg/m². No further dose reduction is planned.

TCH

Docetaxel will be reduced from 75 to 60 mg/m². If despite dose reduction, stomatitis still occurs at grade ≥ 3, docetaxel will be further reduced from 60 to 50 mg/m². No further dose reduction is planned.

Herceptin® may continue in the Herceptin®-containing arms in all cases above.

5.4.5 Bilirubin and impaired liver function tests

Docetaxel and doxorubicin doses shall be modified for hepatic toxicity. If docetaxel is delayed due to hepatic toxicity, other drugs being used in combination at that time shall also be delayed and administered when docetaxel is resumed. The same applies for delays with doxorubicin i.e. other drugs being used in combination with doxorubicin will also be delayed until doxorubicin is resumed.

Since no data in patients with abnormal bilirubin level treated with lower dose of docetaxel are available, in the event that bilirubin levels are abnormal during the study, the next cycle will be delayed by a maximum of two weeks. If no recovery, the patient should be taken off chemotherapy. Herceptin® may continue in the Herceptin®-containing arms.

In the event that ASAT and/or ALAT and/or alkaline phosphatase levels are abnormal in the absence of relapse, the following dose modifications should apply:

ASAT / ALAT Values	Alkaline PhosphataseValues	Dose Modification
≤ 1.5 x UNL	≤ 5 x UNL	no dose modification
> 1.5 x UNL to ≤2.5 x UNL	≤ 2.5 x UNL	no dose modification
> 2.5 x UNL to ≤5 x UNL	≤ 2.5 x UNL	**TCH:** Reduce dose of docetaxel from 75 to 60 mg/m² **AC→T & AC→TH** Reduce dose of doxorubicin from 60 to 50 mg/m² Reduce dose of docetaxel from 100 to 75 mg/m²
> 1.5 x UNL to ≤ 5 x UNL	> 2.5 x UNL to ≤ 5 x UNL	**TCH:** Reduce dose of docetaxel from 75 to 60 mg/m² **AC→T & AC→TH** Reduce dose of doxorubicin from 60 to 50 mg/m² Reduce dose of docetaxel from 100 to 75 mg/m²
> 5 x UNL	> 5 x UNL	**All Arms:** Dose delay by a maximum of 2 weeks. If no recovery to the above figures, patient should go off chemotherapy.

Once the dose is reduced due to impaired liver function, no further dose reduction is recommended if no worsening of the parameters is observed.

In case of recovery of liver function tests on the following cycle, the dose should be re-escalated to the previous dose-level.

5.4.6 Peripheral neuropathy

In case of symptoms or signs experienced by the patient, dose modifications of docetaxel should be performed as follows:

Grade 0,1
Each Arm: no change

Grade 2:

TCH

Delay carboplatin and docetaxel treatment by maximum of two weeks.
As soon as patient recovers, treatment should continue with the following dose recommendations:

If patient recovers to Grade 1 toxicity, dose of docetaxel will be decreased from 75 to 60 mg/m².
If grade ≥ 2 persists for > 2 weeks, patient will either go off chemotherapy or continue with carboplatin and Herceptin® only.

In case of 2nd episode, reduce docetaxel dose from 60 to 50 mg/m².

No further dose reduction is planned.

AC→T and AC→TH:

Delay docetaxel treatment by maximum of two weeks.
As soon as patient recovers, treatment should continue with the following dose recommendations:
If patient recovers to Grade 1 toxicity, dose of docetaxel will be decreased from 100 to 75 mg/m² . If patient not recovered to Grade 1 in two weeks, patient will go off chemotherapy.

In case of 2nd episode, reduce dose of docetaxel from 75 to 60 mg/m²

No further dose reduction is planned.

Grade 3: patient will go off chemotherapy.

The same guideline also applies for patients with grade 1 neuropathy at baseline.

- Herceptin® may continue in the Herceptin®-containing arms in all cases above.

5.4.7 Cutaneous reactions

Grade 0, 1, 2
Each Arm: no change

Grade 3: delay maximum two weeks until ≤ grade 1 then for subsequent cycles of
TCH:
Reduce dose of docetaxel from 75 to 60 mg/m² ;
Second reduction allowed of docetaxel from 60 to 50 mg/m²
AC→T and AC→TH:
Reduce dose of docetaxel 100 to 75 mg/m²
Second reduction allowed of docetaxel from 75 to 60 mg/m²
If no recovery to ≤ grade 1 within two weeks delay, patient will go off chemotherapy.

Herceptin® may continue in the Herceptin®-containing arms in all cases above.

5.4.8 Docetaxel Anaphylactoid Type and Hypersensitivity Reactions

In the event that a hypersensitivity reaction occurs despite premedication, it is then very likely to occur within few minutes of start of the first or of the second infusion of docetaxel. Therefore, during the 1st and the 2nd infusions, the infusion must be given drop by drop for the first 5 minutes, and a careful evaluation of general sense of well being and whenever possible blood pressure and heart rate monitoring will be performed so that immediate intervention would occur in response to symptoms of an untoward reaction.

Facilities and equipment for resuscitation will be immediately available: antihistamine, corticosteroids, aminophylline, epinephrine.

If a reaction occurs, the specific treatment that can be medically indicated for a given symptom (e.g. epinephrine in case of anaphylactic shock, aminophylline in case of bronchospasm, etc.) will be instituted. In addition, it is recommended to take the measures listed below:

Mild symptoms: localized cutaneous reaction, such as: pruritus, flushing, rash	¨ Consider decreasing the rate of infusion until recovery of symptoms, stay at bedside. ¨ Then, complete docetaxel infusion at the initial planned rate. ¨ At subsequent cycles use the same premedication outlined in section 5.1.
Moderate symptoms: any symptom not listed above (mild symptoms) or below (severe symptoms), such as generalized pruritus, flushing, rash, dyspnea, hypotension with systolic blood pressure (BP) > 80 mm Hg	¨ Stop docetaxel infusion. ¨ Give i.v. dexamethasone 10 mg (or equivalent) and i.v. diphenhydramine 50 mg (or equivalent). ¨ Resume docetaxel infusion after recovery of symptoms. ¨ At subsequent cycles, give i.v. dexamethasone 10 mg (or equivalent) and i.v. diphenhydramine 50 mg (or equivalent) one hour before infusion, in addition to the premedication planned in section 5.1.
Severe symptoms: such as bronchospasm, generalized urticaria, hypotension with systolic BP ≤ 80 mm Hg, angioedema	¨ Stop docetaxel infusion. ¨ Give i.v. dexamethasone 10 mg (or equivalent) and i.v. diphenhydramine 50 mg (or equivalent), add epinephrine as needed. ¨ Whenever possible resume docetaxel infusion within 3 hours after recovery or reinfuse the patient within 72 hours using i.v. dexamethasone 20 mg (or equivalent) and i.v. diphenhydramine 50 mg (or equivalent) one hour prior to resumption of infusion. ¨ At the subsequent cycles, dexamethasone (or equivalent) will be given at 20 mg orally the evening before chemotherapy, the morning of chemotherapy and one hour before docetaxel infusion. Additionally diphenhydramine (or equivalent) will be given at 50 mg i.v. 1 hour before docetaxel infusion. ¨ If a severe reaction recurs, patient will go off chemotherapy.
Anaphylaxis (NCI grade 4 reaction)	NO FURTHER STUDY DRUG THERAPY

5.4.9 Herceptin® Infusion-Associated Reactions

During the first infusion with Herceptin®, chills and/or fever are commonly observed in patients. Other signs and/or symptoms may include nausea, vomiting, pain, rigors, headache, cough, dizziness, rash, and asthenia. These symptoms are usually mild to moderate in severity and occur infrequently with subsequent Herceptin® infusions. These symptoms can be treated with an analgesic/antipyretic such as meperidine/pethidine or acetaminophen/paracetamol, or an antihistamine such as diphenhydramine. Some adverse reactions to Herceptin® infusion, including dyspnea, hypotension, wheezing, bronchospasm, tachycardia, reduced oxygen saturation, and respiratory distress, can be serious and potentially fatal (see Appendix 10). The majority of these events occur during or shortly after of the start of the first infusion. Patients that experience severe or moderate infusions symptoms may be managed by slowing or stopping the Herceptin® infusion and supportive therapy with oxygen, beta agonists, antihistamines, corticosteroids. The patient must be monitored for a minimum of X hours after the infusion is stopped until resolution of any observed symptoms. If the patient is an outpatient, it is strongly recommended she be admitted to the hospital for monitoring if the toxicity does not resolve within 3 hours. If a grade 3 or 4 toxicity occurs during the post-infusion observation period, the patient must be evaluated for a minimum of 1 hour from the time the toxicity was first noticed until resolution of any observed severe symptoms. If the patient is being treated as an outpatient, she must be admitted to the hospital for monitoring if the toxicity does not resolve during that hour. Sometimes patients will experience symptoms after the infusion is complete. Patients should be warned of the possibility of delayed reactions and should be instructed to contact their physicians if they experience any symptoms throughout the study.

Prior to readministration of Herceptin®, patients should be prophylactically treated with pre-medications including antihistamines and/or corticosteroids.

5.4.10 Docetaxel Related Fluid Retention (peripheral edemas and/or effusions)

In case fluid retention occurs during the treatment with docetaxel, the signs and symptoms should be graded as mild or moderate or severe as recommended in appendix 4.
NO DOSE REDUCTION IS PLANNED.
The weight will be recorded and followed as frequently as possible to document any weight gain which could be related to edema.

Recommended curative treatment for fluid retention

Curative treatment should commence when signs and/or symptoms of fluid retention are observed, including weight gain from baseline ≥ grade 1 not otherwise explained.
The following treatment is recommended in case fluid retention occurs:

* Furosemide 20 mg p.o. o.d.

If the symptoms cannot be controlled adequately, i.e. worsening of the fluid retention or spread to another area, the dose of furosemide should be increased to 40 mg. The addition of metolazone p.o. at the recommended dose together with potassium ± magnesium supplement may be useful.

The clinical tolerance of the patient and the medical judgment of the investigator will determine if it is in the patient's best interest to continue or to discontinue the study drug. It is recommended, however, that patients with fluid retention of grade 3 severity (appendix 4) should be withdrawn from chemotherapy.

In case of difficulty to make a judgment whether an effusion would be disease related or study drug related, the treatment should be continued until progressive disease in other organs is documented.

5.4.11 Renal Toxicity

Carboplatin dose shall be modified for renal toxicity. Dose modifications are based on test results at the time of planned treatment (i.e. Day 1) of each cycle. No dose reduction for docetaxel, cyclophosphamide, doxorubicin or Herceptin® will be made for renal toxicity. However, drugs may be delayed if the creatinine is > 2 mg/dL (> 175 µmol/L).

Dose Modifications

Creatinine Clearance mL/min	Carboplatin Dose to be Administered
≥ 50 mL/min	AUC 6 mg/mL/min (regular dose as in protocol)
49 – 31 mL/min	AUC 5 mg/mL/min
≤ 30 mL/min	Delay until > 30 mL/min (max 2 weeks) then treat with AUC 5 mg/mL/min

Creatinine clearance can either be measured or estimated using the Cockroft-Gault formula, as follows:

Calculated creatinine clearance (Cockroft-Gault formula) for dose reduction:

Creatinine Clearance (mL/min) = $\frac{(140 - \text{age in years}) \times (\text{weight in kg})}{72 \times \text{serum creatinine in mg/dL}} \times 0.85$

Carboplatin dose is calculated using a modified Calvert formula (creatinine clearance is substituted for GFR) as follows:

Total dose (mg) = (Target AUC) x (Creatinine Clearance + 25)

If carboplatin is delayed for any length of time (1-2) weeks, the next dose should be decreased.

Herceptin® may continue in the Herceptin®-containing arms in all cases above.

5.4.12 Cardiotoxicity

For management of breast cancer treatment in case of cardiac toxicity, please refer to section VI.

5.4.14 Other Toxic Effects

Nail changes will not motivate dose-modification.

Other toxic effects should be managed symptomatically if possible.

- For grade 3 toxicities (see appendix 12), in general drug should be held for a maximum of two weeks from the planned date of reinfusion until resolution to ≤ grade 1, then reinstituted, if medically appropriate. A dose reduction will be discussed between the investigator and sponsor.
- If grade 4 toxicity occurs, except anemia, the patient will go off chemotherapy.

5.5 Treatment Duration and Follow-up

5.5.1 Treatment Duration (Chemotherapy and Herceptin®)

If a patient is randomized to the AC → T arm, 4 cycles of AC every 3 weeks followed by 4 cycles of single agent docetaxel every 3 weeks should be administered.
If a patient is randomized to the AC → TH arm, 4 cycles of AC every 3 weeks followed by 4 cycles of single agent docetaxel every 3 weeks with weekly Herceptin® during chemotherapy treatment and q3weekly during follow-up for 1 year from the time of 1st dose of Herceptin®, should be administered.

If a patient is randomized to the TCH arm, 6 cycles of TC every 3 weeks are to be administered with weekly Herceptin® during chemotherapy treatment and q3weekly during follow-up for 1 year from the time of 1st dose of Herceptin®, should be administered.

In the event of relapse during treatment (see section 7.2 for efficacy definitions), unacceptable toxicities, withdrawn consent, treatment shall finish earlier.

5.5.2 End of Chemotherapy (EOC) Definition

End of chemotherapy (EOC) is defined as 21 days post the last infusion of chemotherapy.

In the case of the arms with Herceptin® infusions continuing for 1 year, EOC will be defined as 21 days post the last infusion of docetaxel or docetaxel/platinum, respectively.

Patients will be observed 3 weeks after last study drug infusion until end of study to document outcome of ongoing side effects (see section X). Clinical adverse experiences requiring further ongoing evaluation include:

- Relevant non cancer related signs and symptoms occurring after completion of chemotherapy (i.e. congestive heart failure, toxicities related to tamoxifen and/or radiotherapy...).
- Reporting of serious adverse event (SAE) will continue for the entire duration of the Herceptin® administration (up to 1 year) for arms AC→TH and TCH.

5.5.3 Follow-Up Duration

Patients will be followed every 3 months for the first two years, every 6 months for years 3 – 5, and then once a year for ten
years or until relapse to document:

- Disease-free survival
- Survival
- Further therapy
- Quality of life (at 6, 12 and 24 months of follow-up)
- Socio-economic data (at 6, 12 and 24 months of follow-up) in USA, Canada and Germany only
- Late side effects, including all cardiac adverse experiences, regardless of grade, seriousness and relation to study medication, including cardiac events (see section 6.1.1.1) This should include chemotherapy, Herceptin®, Tamoxifen and adjuvant radiotherapy related toxicities.
- 2nd primary malignancy

In case of administration of any systemic therapy (chemotherapy, hormonotherapy or immunotherapy) given for disease relapse or 2nd primary malignancy other than the drugs outlined in the protocol, patients will be followed in an abbreviated follow-up for:

- Survival
- All symptomatic cardiac events (as per protocol definition see section 6.1.1.1)

(See Appendix 6) The abbreviated follow-up visits will be done yearly at the anniversary date of EOC until year 10 after EOC or until death.

5.5.4 Definition of First Follow-Up Visit

Because of the difference in duration of chemotherapy treatments between the three arms, we will need to balance the timing of the follow-up assessments in order to assess efficacy at equivalent intervals.

Follow-up Visit # 1 for

AC→ T will be 3 months after EOC

AC→ TH will be 3 months after EOC

TCH will be 4 ½ months after EOC, respectively.
An extra follow-up visit (FUp1a) is planned 6 weeks after EOC for the TCH arm. The timing of this extra follow-up visit occurs at the timing of the EOC for the AC→T and AC→TH arms. Physical exam, quality of life, socio-economic data (only in USA, Canada and Germany), assessment of adverse effects, and LVEF 4 will be performed at this visit.

5.6 Concomitant Treatment During Chemotherapy and Herceptin®

Allowed:

1 G-CSF [treatment of febrile neutropenia/infection, prophylactic use following a febrile neutropenia or infectious episode for all subsequent cycles, delayed neutrophil counts, and primary prophylaxis (at investigator discretion)] (see sections 5.4.1.5)
2 Antiemetics (section 5.2.9)
3 Antiallergic measures (section 5.4.8)
4 Antibiotics (section 5.4.1.6)
- oral prophylactic in case of prior infectious episode
- IV curative in case of febrile neutropenia or documented infection.

Ancillary treatments will be given as medically indicated. They must be specified in the Case Report Form.

Not permitted:

1. The patients will not receive other investigational drugs and anticancer treatment while on study (till relapse or up to 10 years).
2. Corticosteroids are not allowed, except as outlined in previous sections as premedication, antiemetics, and acute hypersensitivity reaction during the course of active treatment with chemotherapy and Herceptin® and except in cases of chronic treatment at a low dose initiated at least 6 months prior to study entry.
3. Concomitant treatment with bisphosphonates will not be allowed during the course of active treatment with chemotherapy and Herceptin®. Subsequently, bisphosphonates may be used only for non-oncologic indications.
4. Concomitant treatment with amifostine (Ethyol®) will not be allowed during the course of active treatment with chemotherapy and Herceptin®
5. Concomitant treatment with cardioprotectors (e.g. Dextrazoxane®) will not be allowed during the course of active treatment with chemotherapy and Herceptin®.

5.7 Reasons for Discontinuation or Withdrawal From Chemotherapy or Herceptin®

Reasons for premature withdrawal or discontinuation criteria include

1. Unacceptable Toxicity (see Section 5.4 Toxicity Related Guidelines for Dose Reduction and Dose Modification, 6.4 Management of Treatment Arms with Symptomatic Cardiac Toxicities)
2. Withdrawn Consent (see Informed Consent Appendix 7, page 8 of 9, Withdrawal From Study)
3. Relapse, second primary malignancy (with the exception of curatively treated non-melanoma skin cancer or in situ carcinoma of the cervix – see Exclusion Criteria 10a, 10b), death or administration of other systemic cancer treatment other than study drug or tamoxifen as per protocol (Section 5.5 Treatment Duration and Follow-up, Section 5.8.3 Therapy After Protocol Treatment is Discontinued)

The reason and date of chemotherapy discontinuation for all patients will be documented in the case report form (e.g. completed study, adverse event, lost to follow-up, etc.).

The investigator will attempt to complete all discharge procedures at the time a patient is discontinued from the study.

For patients in the Herceptin® containing arms who drop out because of chemotherapy related toxicities, Herceptin® may continue until completion of 1 year, or relapse, or Herceptin® related toxicity (including cardiac safety analysis), whichever comes first. These patients are to be followed in regular follow-up.

Patients who stop chemotherapy for any reason OTHER THAN having been administered systemic anticancer therapy for disease relapse or 2nd primary malignancy must be followed in a regular follow-up.

5.8 Post Chemotherapy Treatment

5.8.1 Indication for Hormonal therapy

Tamoxifen (20 mg p.o. daily) for 5 years will be administered starting 3 to 4 weeks after the last course of chemotherapy for patients with positive estrogen and/or progesterone receptors.

Post menopausal patients are allowed to switch to Anastrozole in case of Tamoxifen related severe toxicities (e.g. hot flushes, vaginal bleeding, vaginal discharge, thromboembolic events). Anastrazole will be given at the dose of 1 mg daily. Of note, the total duration of the hormonal therapy, i.e. Tamoxifen followed by Anastrozole, should not exceed 5 years. Only post-menopausal patients are eligible to receive Anastrozole.

Post menopausal women without contraindications to the use of Tamoxifen and who have already started Tamoxifen, can receive a sequential therapy consisting of Tamoxifen for 2 to 3 years followed by Anastrozole or Exemestane for a maximum of 5 years of hormonal therapy.

Post menopausal patients who have not yet started hormonal therapy can receive 5 years of Anastrozole or sequential therapy consisting of Tamoxifen for 2 to 3 years followed by Anastrozole or Exemestane for a maximum of 5 years of hormonal therapy.

Post menopausal patients who have completed 5 years Tamoxifen are allowed to continue the hormonal treatment with letrozole for a maximum of 3 years.

Note: - the use of AIs should be accurately documented and reported on page FU6 of the CRF,

- the same approach is followed for all patients treated in the BCIRG 006/TAX GMA 302 study in order to avoid treatment imbalance between the 3 arms.
- Each center will be requested to provide BCIRG data center with their institution guideline related to the use of aromatase inhibitor by completing a questionnaire.

5.8.2 Radiation Indication

5.8.2.1 Radiation Therapy

Treatment will begin 3 to 8 weeks after the chemotherapy is completed.

For those arms with Herceptin®, radiation is to proceed while patient is receiving Herceptin®.

For those patients who will receive tamoxifen (hormonal receptor status is positive), radiation is to proceed during tamoxifen therapy.

Advised indications are as follows:

Radiotherapy will be mandatory in case of breast conserving surgery. It will be allowed, but not mandatory, in case of mastectomy, according to the policy in use at each participating center. If radiotherapy is indicated, the center will follow the policy in use in the institution and will provide a copy of this policy to the BCIRG. Boost radiation therapy will be left at the discretion of the investigator.

Radiation therapy in internal mammary chain is prohibited for patients having developed a congestive heart failure during study chemotherapy and/or patients who have held Herceptin® administration at least once for asymptomatic decreases in left ventricular ejection fraction or for a symptomatic cardiac event (see Table 10 of the protocol). Otherwise, radiation therapy in internal mammary chain is left at the discretion of the investigator.

5.8.2.2 Radiation Guidelines

Radiation therapy will be indicated according to the guidelines of each institution. Each institution's guidelines will be collected prior to study start at that particular center. Each patient's type and dose of radiation therapy will be documented in the CRF.

5.8.3 Therapy after protocol treatment is discontinued

Except for study chemotherapy, hormonal therapy, radiotherapy and Herceptin® as per protocol, no further antitumor therapy is allowed (surgery, chemotherapy, immunotherapy, etc.) before tumor relapse is documented.

If patients are removed from the study because of disease relapse, further treatment is at the discretion of the investigator. The metastatic regimen (s) used will be collected in the Case Report Form.

NOTE:
In case of disease relapse or second primary malignancy (with the exception of curatively treated non-melanoma skin cancer or in situ carcinoma of the cervix – see Exclusion Criteria 10a, 10b) or administration of other systemic cancer treatment other than study drug and tamoxifen as per protocol, Herceptin® administration should be discontinued.
Since the half-life of Herceptin® is now estimated to be approximately 28.5 days (95% confidence interval, 25.5-32.8 days) (see section 2.3.3), rather than 5-6 days, Herceptin® may be present in the circulation for up to 24 weeks (range 18-24 weeks) after stopping Herceptin® treatment. It is already known that, when used in combination, Herceptin® and anthracyclines are associated with an increased risk of cardiotoxicity. Considering the above mentioned new pharmacokinetic data, the use of anthracyclines after stopping Herceptin® may carry a higher risk of cardiac toxicity. If possible, physicians should avoid anthracycline base therapy for up to 24 weeks after stopping Herceptin® therapy. If anthracyclines are used, the patient's cardiac function should be monitored carefully.

5.9 Study Evaluations

5.9.1 Prestudy Screen

	INVESTIGATIONS	TIMING within (time) prior to registration
1 Patient informed consent	Obtained	**Before study entry***
2 History and physical exam	**History** - including: diagnosis of breast adenocarcinoma, prior antitumor therapy and outcome, menopausal status, receptor status at diagnosis, general medical history including cardiac history and allergy, concurrent illness and existing signs and symptoms Concomitant medications, and their indication, used within one month prior to study entry. **Physical Exam** – complete physical examination including: height and weight, Karnofsky index for performance status/vital signs.	14 days
3 Hematology **	Hemoglobin WBC and neutrophil count Platelet count	14 days
4 Biochemistry ** **5 HER2 Assessment (mandatory)**	**Liver function:** • Alkaline phosphatase, • ASAT (SGOT), ALAT (SGPT), • Bilirubin **Renal function:** • serum creatinine, • creatinine clearance (if indicated) Positive FISH test (BCIRG central lab confirmation) (see appendix 3A)	14 days Liver function tests are to be repeated within 3 days, if abnormal results. Prerandomization
6 Serum sample (optional)	For HER2 extracellular domain (ECD) (see appendix 3C)	Prior to first drug administration (At time of hematology tests)
7 Blood Sample (optional) *(not applicable for France)*	For cardiac genetic marker (see appendix 3E)	Prior to first drug administration (At time of hematology tests)
8 Plasma Sample (optional) *(not applicable for France)*	For cardiac biochemical markers (see appendix 3D)	Prior to first drug administration (At time of hematology tests)

	INVESTIGATIONS	TIMING within (time) prior to registration
9 ER/PR status	√	Prerandomization
10 Pregnancy test	urine or serum (if applicable)	7 days
11 Imaging* **	**mandatory for all patients:** • contralateral mammography and/or ultrasound (mammogram is preferred), where applicable • chest-X-Ray (PA and lateral), CT or MRI • abdominal ultrasound and/or CT scan and/or MRI • bone scan and bone X-ray in case of hot spots in bone scan Other instrumental examinations as indicated.	3 months
12 ECG	ECG	3 months
13 LVEF**	Echocardiography or MUGA scan	3 months
14 Quality of life	QLQ–C30, BR23 & Euroquol questionnaires (see appendix 11)	14 days
15 Socio-economic data (only in Canada, Germany and US centers)	Productivity and time losses questionnaires	14 days
16 Other Investigations	as clinically indicated	3 months

* Informed Consent should be obtained prior to any tests specified in this clinical protocol that are not part of the patient's routine care
** Laboratory assessments will be performed whenever possible by the same laboratory throughout the study.
*** **Every effort will be made to use the same instrumental examination from baseline through follow-up.**

5.9.2 Study Entry – Registration

All eligible patients must be registered with the coordinators of the study based in Paris, for all the countries outside Canada and USA, or Los Angeles, for all Canadian and US centers, prior to start of treatment.
A patient who has not been registered before the first treatment administration will not be accepted for the study at a later date.

The registration forms should be faxed to the coordinators of the study. A registration package outlining the exact process for registering a patient and the registration forms will be forwarded and reviewed to all sites at the initiation site visit, by the site CRA.
Registration Fax#: +33 (1) 58 10 09 10 or 33 (1) 58 10 09 11 **for all countries outside Canada and USA**
310 478 8025 or 310 479 5745 **for Canada and USA**

Registration can be made once eligibility of the patient is checked (including laboratory, HER2 status, and radiological results).

The following information will be requested:

1 Protocol number

2 Institution name

3 Caller's name

4 Investigator's name

5 Patient's identifiers

6 Patient's birth date (day/month/year)

7 Date treatment planned.

8 Verification of selected inclusion and exclusion criteria as identified in the patient registration form.

9 Date when the paraffin block was sent to the designated BCIRG laboratory for HER2 determination*.

* HER2 screening will be performed by regional BCIRG designated central laboratories. The result of the test will be sent directly from the central lab to the BCIRG Paris office (within a maximum of 5 working days), who will determine patient eligibility based on the registration form received from the clinical site and from the result of the HER2 test, received from the designated lab. See Appendix 3A for more details on the process.

Each eligible patient will be randomized according to a center specific randomization block to receive either doxorubicin and cyclophosphamide followed by docetaxel (AC→T) or doxorubicin and cyclophosphamide followed by docetaxel with Herceptin® (AC→TH) or docetaxel and carboplatin with Herceptin® (TCH).

Investigators will be notified by fax immediately after ALL information has been received from the site as per the Registration Form. This fax will contain the patient's study number, the strata and the randomly allocated treatment group.

Note 1: Interval between definitive surgery that includes axillary lymph node assessment and registration is less than or equal to 60 days. If 61days has passed from the time of definitive surgery and registration, patient is not eligible.

Note 2: If further tests or results are needed to be performed at the time of registration, we will allow a maximum of 5 working days from the time of registration to the time of randomization.

Note 3: Treatment must start within 8 days from the time of randomization.

5.9.3 Evaluation During Chemotherapy

All patients during the study must be evaluated according to the schedule outlined in Appendix 5 until they come off chemotherapy.

Schema during chemotherapy	INVESTIGATIONS	TIMING
1 History and physical Exam	**Clinical History since previous infusion** **Physical Exam -** including: Weight, Karnofsky index for performance status Clinical tumor assessment	every 3 weeks (day 1 or day -1 of each cycle before chemotherapy)
2 Hematology	Hemoglobin, WBC, neutrophils, and platelets count.	every 3 weeks (day 1 or day -1 of each cycle before chemotherapy)
3 Biochemistry	Alkaline phosphatase, ASAT (SGOT), ALAT (SGPT), bilirubin, serum creatinine, creatinine clearance (if indicated)	every 3 weeks (within 3 days prior to chemotherapy)
4 LVEF	Echocardiography or MUGA	See Section VI
5 Quality of Life	QLQ C30, QLQ BR23, Euroquol questionnaires	See Section IX
6 Socio-economic data (only in USA, Canada, Gemany)	Productivity and time losse questionnaires	See Section X
7 Other Investigations		As clinically indicated
8 Adverse events(*) including cardiac	Investigations as indicated	Serious Adverse Events should be reported within 24 hours anytime
9 Serum sample (optional)	For HER2 extracellular domain (ECD) (see appendix 3C)	At time of recurrence if occurs earlier than EOC
10 Plasma Sample (optional) *(not applicable for France)*	For cardiac biochemical markers (see appendix 3D)	End of Cycle 4, At any time of clinical evidence of cardiac failure
(*) Toxicities will be recorded and graded according to the NCI - CTC criteria, version 2.0 (Appendix 12). In case NCI-CTC criteria are not applicable the event should be defined as 1 = mild, 2 = moderate, 3 = severe and 4 = life-threatening.		
Laboratory assessments will be performed whenever possible by the same laboratory throughout the study. **Every effort will be made to use the instrumental examination from baseline through follow-up.**		

5.9.4 Evaluation at End of Chemotherapy (EOC)

To be performed 21 days after the last treatment as summarized in Appendix 5: work-up will include = **physical examination with Karnofsky, hematology, biochemistry, record of toxicity, quality of life, socio-economic data (only in USA, Canada and Germany) and LVEF (echocardiography or MUGA).**
Serum samples for HER2 ECD and plasma samples for cardiac biochemical markers will be collected at end of chemotherapy visit for AC→ TH and AC→ T arms. They will be collected at FU1 a visit, 6 weeks after the end of chemotherapy visit, for arm TCH.

5.9.5 Evaluation in Follow-up After End of Chemotherapy

Timing of follow-up visits is based on EOC for arms AC→T and AC→TH or follow-up 1a (FUp1a: 6 weeks after EOC) for arm TCH and will be performed according to the following schedule (see Section 5.5.4 Definition of First Follow-Up Visit and Appendix 6). Clinical follow-up may be more frequent according to the standard of practice at the participating center and at the discretion of the investigator.

NOTE 1 LVEF REQUIREMENTS ARE OUTLINED IN SECTION VI
NOTE 2 Physical Examination in Follow-up include Karnofsky Performance Status

First 2 years ALL PATIENTS

every 3 months	physical examination, reporting of cardiac adverse experiences
every 6 months	physical examination, reporting of cardiac adverse experiences serum sample for HER2 shed ECD (optional) , plasma samples for cardiac biochemical markers (optional) - *(not applicable for France)* .
every 12 months	mammography and chest X-ray in addition to physical examination, reporting of cardiac experiences

Quality of Life and socio-economic data (only in USA, Canada and Germany) will be collected at 6, 12 and 24 months of follow-up and then at time of relapse.

The following additional exams and visits will be added in the first 18 month follow-up period for all patients until the cardiac safety analysis has been completed.

1. For patients in the TCH arm:
   A follow-up visit #1 (FUp1a) is planned 6 weeks after EOC. The timing of this extra Follow-Up occurs at the timing of the EOC for the AC→T and AC→TH arms. Physical exam, quality of life, assessment of adverse effects, and LVEF 4 will be performed at this visit. Serum samples for HER2 extacellular tests and plasma samples for cardiac biochemical markers will be also collected (this is optional).

2. At Follow-Up #1 (corresponds to 3 months after EOC of AC→T and AC→TH arms, and 4 ½ months after TCH) all arms
   LVEF assessment (LVEF 5) will also be performed (see Section 6.2.2).

3. At Follow-Up #4 (corresponds to 12 months after EOC of AC→T and AC→TH arms, and 13 ½ months after TCH from randomization) all arms (see section 6.2.2). LVEF assessment (LVEF 6) will also be performed

4. At Follow-up # 10 (corresponds to 36 months after EOC of AC→T and AC→TH arms and 37 ½ months after EOC of TCH). See section 6.2.2.
   LVEF assessment (LVEF 7) will also be performed.

Years 3 to 5 ALL PATIENTS

Every 6 months	physical examination, reporting of cardiac experiences, Serum sample for HER2 shed ECD until 60 months of follow-up (optional). Plasma samples for cardiac biochemical markers until 36 months of follow-up and one additional draw at 60 months (optional) - (not applicable for France).
Every 12 months	mammography in addition to physical examination, reporting of cardiac experiences.

Years 6 to 10 ALL PATIENTS

Every 12 months physical examination, mammography, reporting of cardiac experiences

During the follow-up period

Other diagnostic tests (i.e.: abdominal ultrasound and/or CT scan, bone scan) should be performed only in presence of signs and/or symptoms suggestive of cancer recurrence.

- Patients who develop congestive heart failure at any time during the study (either during active treatment or in follow-up), repeated LVEF will be required in follow-up every 3 months for the first year and every year until the end of follow-up or otherwise as clinically indicated.

- Patients who develop grade 3 or 4 arrhythmias will be required to have an ECG repeated during the follow-up every 3 months for the first year, and every year until the end of follow-up or otherwise as clinically indicated.

- Patients who develop grade 3 or 4 ischemia/infarction, will be required to have a LVEF and ECG repeated during the follow-up every 3 months for the first year, and every year until the end of follow-up or otherwise as clinically indicated.

Echocardiography or MUGA requirements will be the same for all patients according to section 6.2.2 of the protocol. A baseline LVEF evaluation will be required as per inclusion criteria #9. Further assessment of LVEF during active treatment, at completion of chemotherapy or during the follow-up will be left to the discretion of the investigator.

Serum sample for shed HER2 extracellular domain will also be requested at the time of relapse (see Appendix 3C) (optional).
An extra plasma sample will be collected at any time of clinical evidence of cardiac failure for the cardiac biochemistry marker substudy - *(not applicable for France)*.

Clinical adverse experiences requiring further ongoing evaluation include:
- Ongoing clinical adverse experiences possibly or probably related to study drug at the time of End of Chemotherapy
- Cardiac adverse experiences ongoing at the End of Chemotherapy regardless of relation to study medication
- Relevant non cancer related signs and symptoms occuring after completion of chemotherapy (i.e. congestive heart failure, toxicities only if related to Herceptin, Tamoxifen and/or radiotherapy...).
- Serious adverse event (SAE) and toxicities only if related to study chemotherapy will continue for the entire duration of the Herceptin administration (up to 1 year) for arms AC→TH and TCH.
- All cardiac adverse experiences, regardless of grade, seriousness and relation to study medication, including cardiac events *, chemotherapy, Herceptin®, Tamoxifen and adjuvant radiotherapy related toxicities should be reported.

VI CARDIAC SAFETY MONITORING

6.1 Cardiac Safety Evaluation

A secondary endpoint of the study is to compare the cardiac safety of the arms containing Herceptin® to the control arm of AC → T.

6.1.1 Definitions of Cardiac Toxicity

6.1.1.1 Symptomatic Cardiac Event

A cardiac event has occurred if a patient has had a cardiac death, congestive heart failure, grade 3 or grade 4 arrhythmias, or grade 3 or grade 4 ischemia/infarction.

Cardiac Death
Cardiac death will be defined as death due to one of the following

- confirmed congestive heart failure
- myocardial infarction
- documented primary arrhythmia
- probable cardiac death i.e. sudden death without documented etiology

An autopsy is preferred in cases where cause of death has a cardiac etiology.

Congestive Heart Failure (CHF)
Clinical signs and symptoms suggesting congestive heart failure (dyspnea, tachycardia, cough, neck vein distention, cardiomegaly, hepatomegaly, paroxysmal nocturnal dyspnea, orthopnea, peripheral edema, etc) must be investigated.

The suspicion of congestive heart failure, based on the signs and symptoms mentioned above, must be confirmed by a LVEF decrease in echocardiography or MUGA, with a chest X-ray. All CHF events and associated reports will be reviewed by an independent team of cardiologists.

LVEF assessment should be repeated 4 to 7 days afterwards to confirm a diagnosis of congestive heart failure.

Cardiac Arrythmias, Grade 3 or Grade 4
The NCI Common Toxicity Criteria, version 2.0 (see Appendix 12) will be used to classify an arrhythmia as grade 3, which is symptomatic and requiring treatment, or grade 4 which is an arrhythmia considered to be life-threatening e.g. an arrhythmia associated with CHF, hypotension, syncope, shock.

Cardiac Ischemia / Infarction, Grade 3 or Grade 4
The NCI Common Toxicity Criteria, version 2.0 (see Appendix 12) will be used to classify the severity of cardiac ischemia/infarction. Grade 3 ischemia is defined as angina without evidence of infarction. Grade 4 is defined as an acute myocardial infarction.

6.1.1.2 Asymptomatic Cardiac Abnormality

Asymptomatic decreases in left ventricular ejection fraction (LVEF) will be classified as an asymptomatic cardiac abnormality. Clinically significant asymptomatic cardiac abnormality is an absolute decline of LVEF of > 15 % points from baseline and a value below LLN.

A specific monitoring plan was devised for data collection of asymptomatic decreases in left ventricular ejection fraction. Data on the incidence and degree of LVEF decrease in the first 1,500 patients randomized was collected at scheduled time points. This data were collected and reviewed as defined below (Section 6.1.2). The results of these different analyses were reviewed by the IDMC, that did not express any concern regarding the cardiac toxicity observed in the study and that recommended the cardiac of the asymptomatic decreases in left ventricular ejection fraction to continue with all patients as for the first 1,500.

For patients with an asymptomatic decrease in LVEF, the treatment decision with respect to Herceptin® and repeat LVEF determinations will be defined by the measured left ventricular ejection fraction as it relates to the radiology facility's lower limit of normal and the change in LVEF from baseline.

Determination of left ventricular ejection fraction to be performed is outlined in section 6.2

6.1.2 Evaluable Patients for the Cardiac Safety Evaluation

All patients randomized to the study must have a normal baseline LVEF to be eligible.

Scheduled echocardiography or MUGAs have initially been planned for the first 1,500 randomized patients in order to evaluate asymptomatic changes in left ventricular ejection fraction from baseline; the IDMC recommended the same schedule to be followed for all patients randomized in the study (see section 6.2).

Timing of Analyses: Analyses of cardiac toxicity will take place when 100 randomized patients per arm (total 300 patients), 300 randomized patients per arm (total 900 patients), 500 randomized patients per arm (total 1,500 patients), and all patients randomized, respectively and on an intent-to-treat basis, have been followed up to and including the timing of LVEF 5. LVEF 5 corresponds to the timing of Follow-Up #1, which occurs 3 months after the EOC visit in the AC→T and AC→TH arms, and 4 ½ months after the EOC visit in the TCH arm (see Figure 1 in Section 6.2 for more details on the timing of LVEF measurements).
At each of these analyses, cardiac deaths, congestive heart failure events, grade 3 or 4 arrhythmias, grade 3 or 4 cardiac ischemia / infarction events, and asymptomatic left ventricular decreases will be reviewed and assessed as outlined in the statistical section 8.4.

6.1.3 Cardiac Safety Analysis

For more details, see the statistical analysis plan (section VIII.)

At each of the analyses mentioned above, the incidence of cardiac events (cardiac deaths, CHF, Grade 3 or Grade 4 ischemia/infarction, Grade 3 or Grade 4 arrhythmias) will be considered unacceptable when an absolute difference of > 4% between the AC→T arm and either of the Herceptin®-containing arms, AC→TH and TCH, has occurred.

At each of the analyses mentioned above, asymptomatic cardiac abnormalities (asymptomatic decreases in LVEF) will be reviewed and assessed as outlined in section 8.4. Clinically significant asymptomatic cardiac abnormality is an absolute decline of LVEF of > 15 % points from baseline and a value below LLN.

At either of these analyses, should one of the treatment arms have an unacceptably high incidence of cardiac toxicity or an unacceptable LVEF decrease profile (see Statistical Analysis Plan section VIII for more detail), this group will be discontinued following review by the Independent Data Monitoring Committee.

6.1.4 Following Completion of the Cardiac Safety Analysis

Patients will continue with the LVEF schedule defined below until the results of the cardiac safety analysis are complete. At this time, an amendment will be issued to decrease the echocardiography or MUGA requirements for patients randomized in the future. A baseline LVEF evaluation will be required as per inclusion criteria #9, and further assessment of LVEF during active treatment, at completion of chemotherapy or during the follow-up will be left to the discretion of the investigator.

6.2 Asymptomatic Decreases in Left Ventricular Ejection Fraction (Echocardiography or MUGA)

6.2.1 Determination of LVEF Status by MUGA or Echocardiography

Measurements of left ventricular ejection fraction as outlined in the protocol will be performed by echocardiography or MUGA. **The same method of assessment must be used throughout the study. We strongly advise that all echocardiography or MUGAs be determined at the same radiology facility used at baseline.**

NOTE: ECHOCARDIOGRAPHY WILL BE PREFERRED, AS IT WILL ALLOW US TO COLLECT DETAILED INFORMATION ON THE CARDIAC CHANGES THAT MAY OCCUR.

Additional assessments of LVEF over and above the echocardiography or MUGA scans required by the protocol, are left to the discretion of the investigator.

In cases where a delay in chemotherapy has occurred and an echocardiography or MUGA scan is due, the test may be rescheduled up to 3 weeks following the last dose of chemotherapy. LVEF visits in follow-up will be scheduled according to the timing from the end of chemotherapy visit (See Diagram 2 in Section 6.2.2 Timing of LVEF Evaluations in Follow-up)

Patients who develop an LVEF decrease resulting in Herceptin® to be either discontinued or held (with or without chemotherapy), will continue with the LVEF schedule outlined below. If LVEF has not recovered by 36 months (Follow-up Visit #10), patient must have LVEF determination annually until recovery or end of follow-up, whichever comes first.

6.2.1.1 Echocardiography Guidelines (for those sites using echocardiography)

Elements necessary for this assessment include:

1) Standard echocardiographic equipment including a machine capable of 2 dimensional imaging and spectral Doppler imaging,
2) Sonographer trained in standard adult transthoracic imaging,
3) Cardiologist with experience in interpretation of 2D and Doppler imaging,
4) VHS or digital format on echo machine and tape.

2D Imaging should include:

1. Parasternal Long Axis View: assessment of left ventricular end systolic and end diastolic dimensions
2. Parasternal Short Axis View: (at the papillary muscle level) assessment of the left ventricular dimensions at end systole and end diastole and assessment of overall left ventricular systolic function (LVEF)
3. In the apical 4 chamber view, an assessment of left ventricular systolic function

The software available on echocardiography machines usually performs the calculation of LVEF.

Calculations: Ejection fraction (EF%) = (EDV – ESV) / EDV x 100

Doppler Imaging should include:
In the apical 4 chamber view and the apical 2 chamber view spectral Doppler assessment of mitral valve inflows including IVRT (isovolumic relaxation time), DT (deceleration time), E/A ratios

Other abnormalities noted during this limited echo (e.g. pericardial effusions, significant mitral regurgitation, etc), should be reported in the patient's chart and CRF.

All echos are to be reported by VHS or digital and must be made available for review on request by BCIRG.

6.2.2 Timing of LVEF Evaluations

As part of the assessment of cardiac safety of each treatment arm, echocardiography or MUGAs are scheduled as follows for each arm.

6.2.2.1 Timing of LVEF Evaluations During Chemotherapy

LVEF B1 Pre-Randomization, Baseline LVEF evaluation.
Will occur within 3 months prior to randomization.

LVEF 2 The 2nd LVEF will occur after the 4th cycle of chemotherapy in each arm.
LVEF 2 should be scheduled no sooner than 2 weeks after the last cycle of chemotherapy, but before the next cycle of chemotherapy.

LVEF 2 corresponds to the LVEF determination after completion of AC in the AC→T and AC→TH arm, and after 4 cycles of TC with Herceptin® in TCH arm.

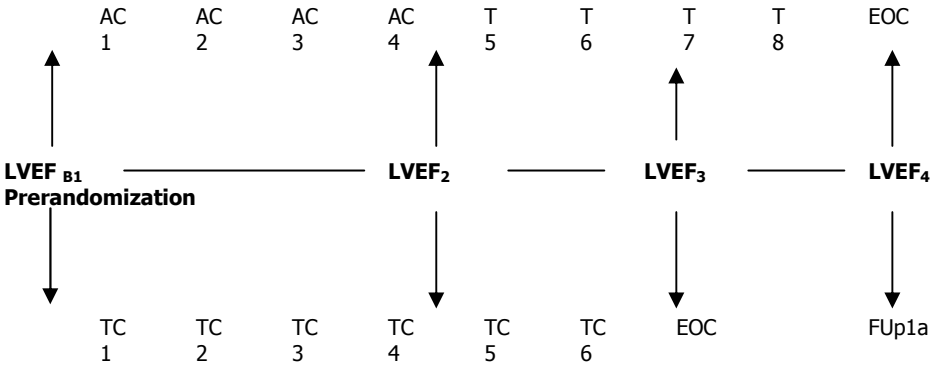
LVEF 3 The 3rd LVEF will occur after the 6th cycle of chemotherapy in each arm.
LVEF 3 corresponds to the End of Chemotherapy visit evaluation in the TCH arm.
This echocardiography or MUGA in either of the AC→T or AC→ TH arms must be scheduled at 2 ½ - 3 weeks after the last cycle, but before the next cycle of chemotherapy.

LVEF 4 LVEF 4 corresponds to the End of Chemotherapy visit in the AC→T and AC→TH arms
LVEF 4 corresponds to an extra follow-up visit (FUp1a) 6 weeks after the EOC for the TCH arm.

Diagram 1: Timing of LVEF Evaluations From Baseline to End of Chemotherapy

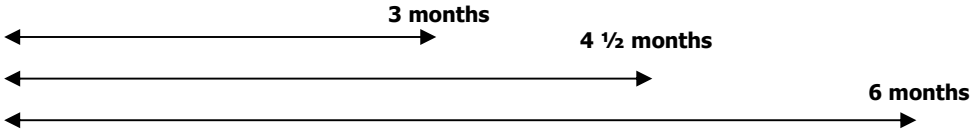
AC → T Arm

AC → TH Arm (Herceptin® x 1 year starting at cycle 5)



TCH x 6 Arm (Herceptin® x 1 year starting at cycle 1)

LVEF Timelines from Baseline (these are reflective of a timeline that has had no delays in chemotherapy)



6.2.2.2 Timing of LVEF Evaluations During Follow-up

Timing of follow-up visits is based on EOC. As part of the assessment of cardiac safety of each treatment arm, echocardiography or MUGAs are scheduled as follows in the follow-up segment.

LVEF 5 Corresponds to follow-up visit # 1.
The timing occurs 3 months after the EOC in the AC→T and AC→TH arms, and 4 ½ months after the EOC in the TCH arm.

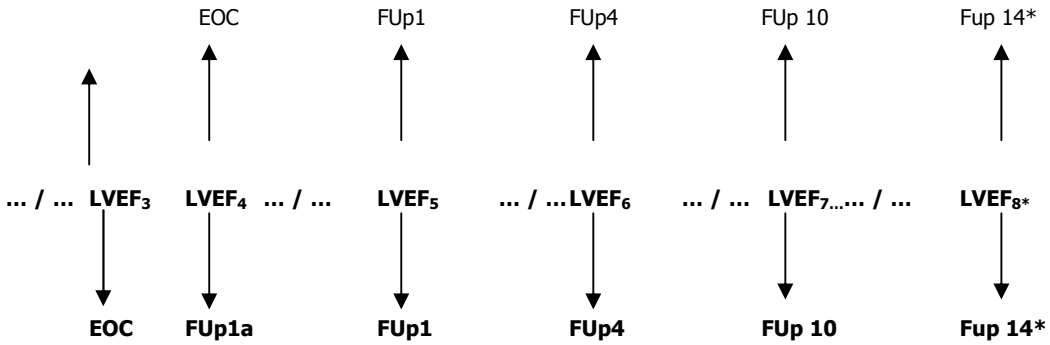
LVEF 6 Corresponds to Follow-up Visit # 4 for all arms (9 months after Follow-up Visit #1).
The timing occurs 12 months after the EOC visit in the AC→T and AC→TH arms, and 13 ½ months after the EOC visit for the TCH arm.

LVEF 7 Corresponds to the follow-up visit #10
The timing occurs 36 months after the EOC visit in the AC→T and AC→TH arms, and 37 ½ months after the EOC visit in the TCH arm.

LVEF 8 Corresponds to the follow-up visit #14
The timing occurs 60 months after the EOC visit in the AC→T and AC→TH arms, and 61 ½ months after the EOC visit in the TCH arm.
Note: If the 60 months FUP visit has already happen for a patient when this amendment becomes available, a LEVF evaluation should be scheduled as soon as the patient has signed her inform consent

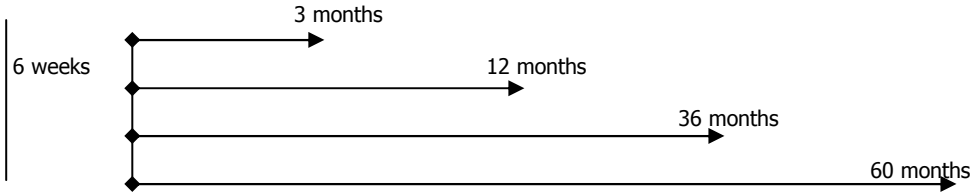
Diagram 2: Timing of LVEF Evaluations During Follow-up

AC → T Arm
AC → TH Arm (Herceptin® x 1 year starting at cycle 5)



TCH x 6 Arm (Herceptin® x 1 year starting at cycle 1)

LVEF Timelines from EOC for AC→T and AC→TH Arms



EOC EOC
TCH AC→T
AC→TH

*** If the 60 months FUP visit has already happened for a patient when amendment 5 becomes available, a LEVF evaluation should be scheduled as soon as the patient has signed her informed consent**

6.3 Guidelines for Herceptin® Use and Repeat LVEFs with Asymtomatic Decreases in LVEF

6.3.1 AC→T Arm

For patients randomized to the AC→T arm who develop an asymptomatic decrease in LVEF while on active treatment, docetaxel will continue to be given as per protocol regardless of the decrease.

Guidelines for performing repeat echocardiography or MUGA scan for AC→T patients who have an asymptomatic decrease in LVEF from baseline are as per TABLE 8.

TABLE 8 is also used for any follow-up echocardiography or MUGA in the AC→TH and TCH arms where Herceptin® is not or no longer being administered (See example 2).

Additional echocardiography or MUGA scans following a repeat of LVEF and prior to the next scheduled LVEF scan may be performed at the investigator's discretion.

Summary: Table 8 is used for all AC→T LVEFs AND for any LVEF in the AC→TH and TCH arms where Herceptin® is Not or NO LONGER being administered

TABLE 8 Repeat LVEF Schedule for Asymptomatic Decreases in LVEF

Asymptomatic decrease LVEF percentage points from baseline

RELATIONSHIP OF LVEF TO THE LOWER LIMIT OF NORMAL (LLN)	ABSOLUTE DECREASE OF < 10 PERCENTAGE POINTS	ABSOLUTE DECREASE OF 10 TO 15 PERCENTAGE POINTS	ABSOLUTE DECREASE OF ≥ 16 PERCENTAGE POINTS
Within radiology facility's normal limits	No action required	No action required	Repeat LVEF after 4 weeks
1 to 5 percentage points below the LLN	No action required	Repeat LVEF after 4 weeks	Repeat LVEF after 4 weeks
≥ 6 percentage points below the LLN	Repeat LVEF after 4 weeks	Repeat LVEF after 4 weeks	Repeat LVEF after 4 weeks

Ref: NSABP-B31 (adapted)

Example 1:	Patient randomized to AC→T	
	Institution's Lower Limit of Normal	50%
	Patient's Baseline LVEF	60%
	LVEF 3 Result	47%
	Treatment Decision (use Table 8)	Continue docetaxel as per protocol Repeat LVEF after 4 weeks.

Example 2:	Patient randomized to AC→TH	
	Institution's Lower Limit of Normal	50%
	Patient's Baseline LVEF	60%
	LVEF 2, LVEF 3, LVEF 4, LVEF 5	58%
	LVEF 7	46%
	Treatment Decision (use Table 8)	Repeat LVEF after 4 weeks.

In the case the LVEF value is repeated two times, is stable but sill in a category where it has to be repeated after 4 weeks as per Table 8, an exception will be made in order not to continue repeating LVEF every 4 weeks. The next LVEF due will be the one of Diagram 1 or 2.

6.3.2 AC→ TH Arm

Results from LVEF 2 will determine whether Herceptin® is to commence in the AC→TH arm. See Table 9 Guidelines for initiation of Herceptin® in the AC→TH arm.

LVEF 2 for the AC→TH arm must occur at least 2 weeks after the last cycle of AC, but before the start of docetaxel and Herceptin®.

If Herceptin® is prohibited in an asymptomatic patient as per Table 9, continuation of chemotherapy with docetaxel is allowed at the discretion of the investigator.

SUMMARY: Table 9 is used ONLY for the AC→TH arm, at LVEF 2 for the decision to initiate Herceptin® or not.

TABLE 9 Guidelines for Initiation of Herceptin® in AC→ TH at LVEF 2

ABSOLUTE CHANGE IN LVEF BETWEEN BASELINE AND 3 WEEKS AFTER LAST AC CYCLE (LVEF 2)	DECISION REGARDING INITIATION OF HERCEPTIN® TREATMENT
Increase or no change	Initiate Herceptin®
Decrease of ≤ 15 percentage points but at or above the radiology facility's lower limit of normal	Initiate Herceptin®
Decrease of ≤ 15 percentage points and below the radiology facility's lower limit of normal	No administration of Herceptin® Repeat LVEF after at least 2 weeks and before the 2nd planned cycle of Taxotere in combination with Herceptin® and then follow guidelines of table10 (in table10, read "initiate" instead of "continue Herceptin®").
Decrease of 16 or more percentage points(regardless of the radiology's facility's lower limit of normal)	No administration of Herceptin® Repeat LVEF after at least 2 weeks and before the 2nd planned cycle of Taxotere in combination with Herceptin® and then follow guidelines of table10 (in table10, read "initiate" instead of "continue Herceptin®").

Ref: NSABP-B31

For the remaining LVEF evaluations during **active treatment of AC→TH**, (LVEF 3, LVEF 4, LVEF 5) the decision to continue, hold, or stop Herceptin® and when to repeat the LVEF will be based on the guidelines outlined in Table 10.

If Herceptin® is held or stopped in an asymptomatic patient, continuation of chemotherapy with docetaxel is at the discretion of the investigator.

Repeat LVEFs in the follow-up period after Herceptin® has been completed (ex. LVEF 6, LVEF 7) will be as per the guidelines in Table 8.

SUMMARY: Table 10 and associated "rules" are used for LVEFs 3, 4, 5 in the AC→TH arm, and LVEFs 2, 3, 4, 5 in the TCH arm .

TABLE 10 Guidelines for Performing Echocardiography or MUGA Scan and Management of Herceptin® in Patients Who Have an Asymptomatic Decrease in LVEF From Baseline (for AC→TH and TCH)

Asymptomatic Decrease LVEF Percentage Points From Baseline

RELATIONSHIP OF LVEF TO THE LOWER LIMIT OF NORMAL (LLN)	ABSOLUTE DECREASE OF < 10 PERCENTAGE POINTS	ABSOLUTE DECREASE OF 10 TO 15 PERCENTAGE POINTS	ABSOLUTE DECREASE OF ≥ 16 PERCENTAGE POINTS
Within radiology facility's normal limits	Continue Herceptin®	Continue Herceptin®	Hold Herceptin® and repeat LVEF after 4 weeks
1 to 5 percentage points below the LLN	Continue Herceptin®	Hold Herceptin® and repeat LVEF after 4 weeks	Hold Herceptin® and repeat LVEF after 4 weeks
≥ 6 percentage points below the LLN	Hold Herceptin® and repeat LVEF after 4 weeks	Hold Herceptin® and repeat LVEF after 4 weeks	Hold Herceptin® and repeat LVEF after 4 weeks

Ref: NSABP-B31 (adapted)

LES for Table 10: Interpreting and Applying "repeat" LVEF Results

1. **Herceptin® must be permanently discontinued following two consecutive "hold" categories**
2. **Herceptin® must be permanently discontinued following three intermittent "hold" categories. At the investigator's discretion, Herceptin® may also be permanently discontinued prior to the occurrence of three intermittent "hold" categories.**

Example 3: Patient randomized to AC→ TH

LVEF Institution LLN	50%
Baseline LVEF	60%
LVEF 2	47%
Treatment Decision (use Table 9)	No administration of Herceptin®
	Docetaxel at Discretion of Investigator
	Repeat LVEF after at least 2 weeks and before the 2nd planned cycle of Taxotere in combination with Herceptin®

Repeat LVEF RESULT 48%

Treatment decision (use Table 10)	Hold Herceptin®
	Repeat LVEF after 4 weeks

Repeat LVEF result	55%
Treatment decision (use table 10)	Initiate Herceptin®

Example 4: Patient randomized to AC→ TH

LVEF Institution LLN	50%
Baseline LVEF	60%
LVEF 2	55%
Treatment Decision (use Table 9)	Herceptin® to be initiated
	Docetaxel to be initiated.

LVEF 3	48%
Treatment Decision (use Table 10)	Hold Herceptin®
	Repeat LVEF after 4 weeks

Repeat LVEF Result	47%
Treatment Decision (use Table 10)	Herceptin® is held again, for the second time (as per Table 10), which means 2 consecutive "holds". Herceptin® is thus discontinued.
	Docetaxel at discretion of investigator

6.3.3 TCH Arm

For patients in the TCH arm who have an asymptomatic decrease in LVEF **during active treatment with Herceptin®** (LVEF 2, LVEF 3, LVEF 4, LVEF 5) the management of Herceptin® and guidelines for performing repeat LVEFs are defined in TABLE 10.

If Herceptin® is held or discontinued during therapy with docetaxel and platinum, docetaxel and platinum may be continued at the investigator's discretion.

Guidelines for repeat LVEFs in the follow-up period after Herceptin® has been completed (LVEF 6, LVEF 7) are as per Table 8.

Example 5: Patient randomized to TCH

LVEF Institution LLN	50%
Baseline LVEF	60%
LVEF 2	48%

Treatment Decision (use Table 10)	Hold Herceptin® Continue with docetaxel and carboplatin Repeat LVEF after 4 weeks
Repeat LVEF Treatment Decision (use Table 10)	51% Restart Herceptin® Continue with docetaxel and carboplatin
LVEF 3 Treatment Decision (use Table 10)	49% Hold Herceptin® Repeat LVEF after 4 weeks
Repeat LVEF Treatment Decision (use Table 10)	53% Restart Herceptin®
LVEF 4 Treatment Decision (use Table 10)	51% Continue with Herceptin®
LVEF 5 Treatment Decision (use Table 10)	45% Permanently discontinue Herceptin® (3 intermittent "hold" as per Table 10 and Associated Rules)

6.4 Cardiac Genetic and Biochemical Marker Studies (optional) - *(not applicable for France)*

As outlined in Section 2.5.9, we propose extra collection of blood and plasma samples for the purpose of assessing a patient's genetic predisposition to the development of cardiac dysfunction and to assay specific biochemical markers through serial assays that may potentially detect early ventricular dysfunction. The cardiac genetic and biochemical marker study is not mandatory. Refusal to grant permission for further testing will not affect the quality of care the participant is to receive.

6.4.1 Cardiac Genetic Markers (optional)

A sample of whole blood will be collected prior to study start in order to analyze the following polymorphisms associated with heart failure: angiotensin converting enzyme (I/D), β2-adrenoreceptor (Gly16/Gln27 and Ile164), ET Receptor A (ETA C1363 T) and HER2. See Appendix 3E for details on the collection, storage and shipment procedure.

6.4.2 Biochemical Markers (optional)

lasma samples for the serial analysis of troponin T and brain natriuretic peptide (BNP) will be collected at the following times during follow-up:

- Baseline
- End of Cycle 4
- End of Chemotherapy visit for arm AC→TH and AC→T
  Follow-up visit 1a for arm TCH (6 weeks after end of chemotherapy)

  This corresponds to the start time of follow-up period.
- Every 6 months for the first 3 years of follow-up.
- At year 5 of follow-up (Follow up #14)

- At any time of clinical evidence of cardiac failure.

See appendix 3D for details on the collection, storage and shipment procedures.

6.5 Management of Treatment Arms with Symptomatic Cardiac Toxicities

Patients who develop a symptomatic cardiac event will be removed from the above LVEF schedule and will be followed as below:

Patients who develop congestive heart failure at any time during the study (both during active treatment and in follow-up), repeated LVEF will be required in follow-up every 3 months for the first year and every year until the end of follow-up or otherwise as clinically indicated. See Table 12 for management of study treatment.

For patients who develop grade 3 or grade 4 arrhythmias at any time during the study (both during active treatment and in follow-up), repeat ECG will be required in follow-up every 3 months for the first year, and every year until the end of follow-up or otherwise as clinically indicated. See Table 11 for management of study treatment.

For patients who develop grade 3 or grade 4 ischemia/infarction at any time during the study (both during active treatment and in follow-up), repeat LVEF and ECG will be required in follow-up every 3 months for the first year, and every year until the end of follow-up or otherwise as clinically indicated**.**

Patients who do develop a symptomatic cardiac toxicity while on active treatment (receiving chemotherapy and/or Herceptin®) and must have their treatment discontinued, will go into the regular follow-up schedule i.e. they do NOT go into an abbreviated follow-up.

Serious adverse events (SAEs) relating to symptomatic cardiovascular disease must be reported within the time lines established for SAE reporting under section X.

6.5.1 Cardiac Arrhythmias

For management of treatment arms in case of cardiac arrhythmias, see Table 11. Please refer to the NCI Common Toxicity Criteria, version 2.0 (Appendix 12) for grading.

TABLE 11 Management of Treatment Arms in case of Cardiac Arrhythmia

Treatment Arm	Grade 1	Grade 2	Grade 3 / Grade 4
AC → T *During AC*	Not necessary to permanently stop doxorubicin if acute dysrhythmias occurring during and shortly after doxorubicin infusion. Monitor frequently. Docetaxel may be given.	Hold AC and conduct cardiac evaluation. Based on results, continue AC at investigator discretion. Docetaxel at discretion of investigator.	Discontinue AC. Docetaxel at discretion of investigator if recovery.
During Docetaxel	Stop or slow docetaxel infusion. Subsequent cycles to be done under continuous cardiac monitoring	Hold docetaxel and conduct cardiac evaluation. Based on results, continuation of docetaxel at investigator's discretion.	Discontinue docetaxel .
AC → TH *During AC*	Not necessary to permanently stop doxorubicin if acute dysrhythmias occurring during and shortly after doxorubicin infusion. Monitor frequently docertaxel +/- Herceptin® may be given.	Hold AC and conduct cardiac evaluation. Based on results, continue of AC at investigator discretion. docetaxel +/- Herceptin® at discretion of investigator.	Discontinue AC. Herceptin® not permitted. Docetaxel at discretion of investigator if recovery.
During Docetaxel and Herceptin®	If during either docetaxel or Herceptin®, slow or stop infusion. Subsequent cycles to be done under continuous cardiac monitoring.	Hold docetaxel and Herceptin®. Conduct cardiac evaluation. Continuation of Docetaxel +/- Herceptin® at investigator's discretion.	Discontinue docetaxel and Herceptin®
TCH	If during either docetaxel or Herceptin®, slow or stop infusion. Subsequent cycles to be done under continuous cardiac monitoring.	Hold TCH and conduct cardiac evaluation. Based on results, continuation of docetaxel/platinum +/- Herceptin® at discretion of investigator.	Discontinue TCH

For patients who develop grade 3 or grade 4 arrhythmias at any time during the study (both during active treatment and in follow-up), repeat ECG will be required in follow-up every 3 months for the first year, and every year until the end of follow-up or otherwise as clinically indicated**.**

6.5.2 Symptomatic Cardiac Left Ventricular Function

Clinical Signs and symptoms suggesting congestive heart failure (dyspnea, tachycardia, cough, neck vein distention, cardiomegaly, hepatomegaly, paroxysmal nocturnal dyspnea, orthopnea, peripheral edema, etc) must be investigated.

The suspicion of congestive heart failure, based on the signs and symptoms mentioned above, must be confirmed by a decrease in LVEF and a chest X-ray. All CHF events and associated reports will be reviewed by an independent team of cardiologists.

LVEF assessment should be repeated 4 to 7 days afterwards to confirm a diagnosis of congestive heart failure before considering the patient to come off treatment as per the guidelines below in Table 12.

Patients who develop congestive heart failure at any time during the study (both during active treatment and in follow-up), repeated LVEF will be required in follow-up every 3 months for the first year and every year until the end of follow-up or otherwise as clinically indicated.

Congestive Heart Failure (CHF) is considered as a Protocol Defined Serious Adverse Event for this study. CHF should be reported as a serious adverse event regardless of causality during the observation period and if in the investigator's opinion it is study drug related, or medically significant during the follow-up period.

TABLE 12 Management of Treatment Arm in case of Congestive Heart Failure

Treatment Arm	Grade 3 or 4
AC → T *During AC*	Discontinue AC when symptoms of heart failure present and diagnosis of CHF confirmed. Docetaxel at discretion of investigator if heart failure adequately controlled.
During Docetaxel	Docetaxel to continue at discretion of investigator.
AC → TH *During AC*	Discontinue AC when symptoms of heart failure present and diagnosis of CHF confirmed. Herceptin® not permitted. Docetaxel at discretion of investigator if heart failure adequately controlled.
During Docetaxel and Herceptin®	Discontinue Herceptin. Docetaxel at discretion of investigator.
TCH	Discontinue Herceptin®. Docetaxel / platinum at discretion of investigator.

6.5.3 Cardiac Ischemia / Infarction

For patients who develop grade 3 or grade 4 ischemia/infarction at any time during the study (both during active treatment and in follow-up), repeat LVEF and ECG will be required in follow-up every 3 months for the first year, and every year until the end of follow-up or otherwise as clinically indicated.

Management of treatment arms in case of cardiac ischemia or infarction are as in Table 13.

TABLE 13 Management of Treatment Arm in case of Cardiac Ischemia / Infarction

Treatment Arm	Grade 1 or 2	Grade 3 or 4
AC → T *During AC*	Continue AC with frequent monitoring. Docetaxel may be given with frequent monitoring.	Discontinue AC. Docetaxel at discretion of investigator.
During Docetaxel	Continue docetaxel with frequent monitoring.	Docetaxel to continue at investigator discretion.
AC → TH *During AC*	Continue AC with frequent monitoring. Docetaxel +/- Herceptin® may be given with frequent monitoring.	Discontinue AC. Herceptin® not permitted. Docetaxel at discretion of investigator.
During Docetaxel and Herceptin®	If during either taxotere or Herceptin®, slow or stop infusion. Docetaxel +/- Herceptin® to continue with frequent monitoring.	Discontinue Herceptin®. Docetaxel to continue at investigator discretion.
TCH	If during either docetaxel or Herceptin®, slow or stop infusion. TCH to continue with frequent monitoring.	Discontinue Herceptin®. Docetaxel / platinum to continue at investigator discretion.

6.6 Reporting of Cardiac Toxicities

A specific "Cardiac Toxicity Monitoring Form" has been designed to capture the following information:

- LVEF assessments as outlined in the cardiac monitoring section (see Section 6.2.2 Timing of LVEF Evaluations and Section 6.3 Guidelines for Herceptin® Use and Repeat LVEFs with Asymptomatic Decrease in LVEF)
- Cardiac Adverse Experiences, regardless of grade, seriousness and relation to study medication

The information captured on this form is to be faxed to the BCIRG Data Management Center within 14 days of the LVEF evaluation or as soon as a cardiac event is known (FAX # 780 702 0188). Copies of supporting documentation may be requested and are to be provided by the center.

Absolute LVEF determinations will be reported in the Cardiac Toxicity Monitroing Form of the CRF. Management of absolute asymptomatic decreases must follow the directions outlined in Section 6.3 Guidelines for Herceptin® Use and Repeat LVEFs with Asymptomatic Decrease in LVEF.

In addition, serious adverse events (SAEs) relating to symptomatic cardiovascular disease must be reported within the time lines established for SAE reporting under section XI (Adverse Events / Toxicity) to the BCIRG safety officer.

Follow-up on cardiac status will be collected on the baseline registration form prior to randomization, the cardiac adverse experiences will be collected on the cardiac toxicity monitoring form for all patients in the cardiac safety analysis during the cycles, then at every follow-up until year 10.

VII SAFETY AND EFFICACY PARAMETERS

7.1 Safety Evaluations

7.1.1 Cardiac Safety Evaluation

A secondary endpoint of the study is to compare the cardiac safety of the arms containing Herceptin® to the control arm of AC → T.

All patients randomized to the study must have a normal baseline LVEF to be eligible. All patients randomized to the study, with the required normal baseline LVEF, will be considered evaluable for the cardiac safety evaluation on an intent-to-treat basis.

Each will be followed for symptomatic cardiac events and asymptomatic cardiac abnormality (see section 6.1.1 for definitions). A cardiac event has occurred if a patient has had a cardiac death, congestive heart failure, grade 3 or grade 4 arrhythmias, or grade 3 or grade 4 ischemia/infarction. Scheduled LVEF tests will be performed at specified time intervals to monitor for asymptomatic decreases in left ventricular ejection fraction (asymptomatic cardiac abnormality). All patients randomized will continue with the LVEF schedule until the results of the cardiac safety analysis are known.

Timing of Analyses: Analyses will take place when 100 randomized patients per arm (total 300 patients), 300 randomized patients per arm (total 900 patients), and 500 randomized patients per arm (total 1,500 patients), as well as all patients randomized in the study, respectively and on an intent-to-treat basis, have been followed up to and including the timing of LVEF 5. LVEF 5 corresponds to follow-up # 1 (3 months following the end of chemotherapy visit for the AC→T and AC→TH arms, and 4 ½ months after the EOC visit for the TCH arm. .

At each of these analyses, cardiac deaths, congestive heart failure events, grade 3 or 4 arrhythmias, grade 3 or 4 cardiac ischemia / infarction events will be reviewed and assessed as outlined in the statistical section 8.4. At each of these analyses, data on asymptomatic decreases in left ventricular ejection fraction will be reviewed and assessed as outlined in the statistical section 8.4. Clinically significant asymptomatic cardiac abnormality is an absolute decline of LVEF of > 15 % points from baseline and a value below LLN.

At either of these analyses, should one of the treatment arms have an unacceptably high incidence of cardiac toxicity or an unacceptable asymptomatic LVEF decrease profile, this group will be terminated after data review by the Independent Data Monitoring Committee and as per outlined in Section 8.4.

In addition, there will be an on-going safety evaluation through the Serious Adverse Event reporting system. If an unexpected high incidence of cardiotoxicity is observed prior to the analyses mentioned above, an IDMC meeting will be called to evaluate.

7.1.2 Clinical Safety

The following tests will be performed prior to and/or on specified days during and following therapy:

- Complete history of malignant and non-malignant diseases including known hypersensitivity reactions and cardiac history.
- Full clinical examination, vital signs (note: routine blood pressure, heart rate, temperature will not be captured in the CRF), height, weight, assessment of any residual toxicity due to previous therapy, assessment of performance status according to Karnofsky Index. The Karnofsky Index Score will be considered as part of the physical examination.
- Electrocardiogram (ECG), left ventricular ejection fraction (LVEF) as per protocol
- Imaging studies as per protocol

- Adverse events: each patient will be assessed regularly for potential adverse events according to the NCI CTC, version 2.0 (Appendix 12).

  Toxicities which cannot be graded using the NCI common toxicity criteria will be graded as followed:
  - mild (asymptomatic)
  - moderate (symptomatic but not interfering significantly with function)
  - severe (causing significant interference with function)
  - life threatening

7.1.3 Laboratory Determinations

The following tests will be performed prior to and on specified days during and following therapy:

- Hematology: WBC, neutrophils and platelet count, hemoglobin
- Biochemistry: total bilirubin, alkaline phosphatase, SGOT (ASAT), SGPT (ALAT) creatinine, creatinine clearance (as indicated)
- Pregnancy test: urine or serum (if applicable)
- ER status and/or PR status: results must be known prior to randomization
- HER2 determination (FISH): results must be known prior to randomization
- HER2 shed extracellular domain (ECD): This is not mandatory but preferred for comparison of levels throughout patient's course.
- Cardiac Genetic Markers: This is not mandatory.
- Cardiac Biochemical Markers: This is not mandatory

7.2 Efficacy Evaluations

All randomized patients will be included in an intent-to-treat analysis.

If one or more study chemotherapy drug(s) is discontinued (whatever the reason), the patient will be analyzed in the Disease Free Survival and Survival analysis according to the intent-to-treat analysis.

7.2.1 Objective Relapse

Any clinical or radiologic evidence of tumor relapse including the central nervous system. Obtain histologic or cytologic proof of failure, if feasible. Detail on flow sheets the appearance of any evidence of malignant disease. Follow for survival.

7.2.1.1 Local relapse

Defined as evidence of tumor in the breast surgical scar, ipsilateral breast (conservative surgery), or evidence of tumor in the ipsilateral anterior chest wall (mastectomy) or skin or soft tissues within the local area.

Histologic or cytologic proof is mandatory.

7.2.1.2 Regional relapse

Defined as evidence of tumor in the axillary scar, ipsilateral nodal areas (axillary, internal mammary, and infraclavicular) as well as skin or soft tissues within the regional area.

Histologic or cytologic proof is mandatory. .

7.2.1.3 Distant relapse

Defined as evidence of tumor beyond the local-regional level as previously defined.

This includes the following:

1) lymph nodes not included in the areas defined above (i.e. supraclavicular, contralateral axilla, paratracheal, etc.)
2) skin not included in the areas defined above
3) liver
4) lung
5) bone
6) central nervous system
7) contralateral breast
8) other sites not defined above

Histologic or cytologic proof is preferred especially in solitary lesions.

Positive bone scans must be correlated with bone X-ray.

Multiple pulmonary nodules on chest X-ray, multiple liver nodules on liver ultrasound or CT-scan or MRI, multiple lytic or blastic bone lesions or multiple hot spots on the bone scan will be acceptable without pathologic correlation.

Any new breast malignancy must be biopsied if possible and blocks must be sent to the central operational office for confirmation of primary or metastatic status along with pathologic and molecular studies.

7.2.1.4 Other circumstances

The following do not constitute relapse, however, they should initiate a new evaluation for extent of disease:

10% or more decrease in baseline Karnofsky performance status

A single new lesion on bone scan without evidence of lytic disease by radiography or bone scan.

Elevation of serum markers such as CEA or CA15-3 by themselves will not constitute evidence of relapse without other objective evidence of relapse. These studies are not recommended.

7.2.2 Second Primary Cancer

Defined as any other histopathologically proven cancer including second invasive primary breast cancer in ipsilateral or contralateral breast.

7.2.3 Disease-Free Survival

Disease-Free Survival (DFS) will be calculated from the date of randomization up to the first date of local, regional, or distant relapse, second primary cancer (with the exception of curatively treated non-melanoma skin cancer or in situ carcinoma of the cervix – see Exclusion Criteria 10a, 10b), or death.

7.2.4 Survival

Survival will be measured from the date of randomization up to the date of death of any cause.

VIII DATA ANALYSIS / STATISTICAL CONSIDERATIONS

8.1 Sample Size Determination

The sample size determination was done based on the following assumptions:
The primary objective of this trial is to show that the treatments under investigation differ in terms of disease-free survival (DFS).
The following assumptions were initially made:

- the proportions of patients who have no axillary lymph nodes involved (N0), 1 to 3 axillary lymph nodes involved (N1-3) and 4 or more axillary lymph nodes involved trial (N4+) will be, respectively, 20%, 50% and 30%
- the DFS at 5 years of patients receiving AC→T in these strata are, respectively, equal to about 67%, 57% and 42%
- the overall DFS of all patients receiving AC→T will be equal to about 55% (20% x 67% + 50% x 57% + 30% x 42%)
- it is of clinical interest to detect a 7% improvement in 5-year DFS (i.e. an increase from 55% to 62%)
- the overall error rate for a false positive outcome (α) is set to 5%, using two-sided significance tests. Since the three pairwise treatment comparisons will be of interest in the final analysis, the error rate for each comparison is set at a conservative level of 0.017
- the error rate for a false negative outcome (β) is set to 20% i.e the power of the trial is set to 80% for the difference of clinical interest

A total of 3,150 patients are necessary to have sufficient power to compare AC→T with AC→TH with TCH for all randomized patients, assuming an anticipated ineligible rate of 3%. 1, 050 patients will be necessary in each treatment group. This sample size calculation takes into account the fact that 1 interim analysis will be performed when 50% of the events (654 events) have been observed and the final analysis will take place when a total of 1,308 events have taken place among all patients.

The randomization will be centralized and stratified at time of inclusion for institution, for node status: node negative, node positive 1-3 nodes and node positive ≥ 4 or more, and for hormonal receptor status (estrogen and/or progesterone receptor positive versus negative)

The group sequential design, according to Peto's method, will be used to a significance level of 0.001 for the interim analysis. This allows the use of an unadjusted level of 0.05 for the final analysis.

With the sample size as stated in the primary endpoint, this trial has 80% power (at a significance level of 0.05) to detect an absolute difference in overall survival of approximately 5%. If overall survival at 5 years in the control arm is between 70% and 80%, the detectable difference in one of the experimental arms is between 5.5% and 4.7%, respectively. This translates into a reduction in relative risk between 21% and 26%.

Updated data on DFS from BCIRG 001 study (TAC arm) have been used to modify the number of events required for interim and final analyses, by assuming 7% absolute advantage in 5 years DFS in favor of one of the Herceptin containing regimen with a power of 80% and $\alpha = 0.05$ as originally planned.

Among the node positive and Her 2 neu positive by FISH patients treated with TAC in the BCIRG 001 trial, 73% of patients were disease free at a median follow-up of 55 months which translates into an estimated 5 year Disease Free Survival of 70% among the patients randomized to the arm without Herceptin, i.e., AC followed by T. As a consequence, the revised calculations are based on a presumed 5 year DFS of 70% in AC-T treated patients.

Additional interim analyses have also been added and a "step-down" testing procedure has been proposed in order to compare the control arm (AC-T) to each Herceptin-containing arm (AC-TH and TCH) at a level equal to α / 2 to account for multiple testing, if both of these comparisons reach statistical significance, then compare the two Herceptin-containing arms at level α, otherwise stop.

The revised schedule for conducting the analyses is the following: three interim analyses will be conducted when respectively 300, 450 and 650 events have been observed; and the main analysis will take place when 900 events are observed. In order to avoid any confusion, the previously called "final" analysis is now called the "main" analysis, to reflect the fact that two follow-up analyses will be performed after this analysis.

With the revised assumptions as well as the final number of randomized patients of 3,222, the trial is powered to detect a 7% difference between the control arm and the Herceptin-containing arm, assuming the 5-year DFS in the control arm (AC-T) is 70% [i.e. a 23.7% reduction in relative risk].

With the sample size as stated in the primary endpoint, this trial has 80% power (at a significance level of 0.05) to detect an absolute difference in the 5 years overall survival of approximately 5%. If overall survival at 5 years in the control arm is between 70% and 80%, the detectable difference in one of the experimental arms is between 5.5% and 4.7%, respectively. This translates into a reduction in the relative risk between 21% and 26%.
If the overall survival at 5 years in the control arm is between 80 and 90%, the detectable difference in one of the experimental arm is between 4.7% and 3.4%, respectively. This translates into a reduction in the relative risk between 26% and 35%.

8.1.1 In the Case of Closing a Treatment Arm

Should one of the treatment arms have an unacceptably high incidence of cardiac toxicities, the Independent Data Monitoring Committee will recommend the Steering Committee to discontinue it. In this case, the trial will continue until accrual reaches the target sample size of 1, 050 patients in each of the remaining two other treatment arms. Since in this case there will be
no need to carry out pairwise comparisons, the final analysis will use an unadjusted level of significance of 5% and will have a power of 90% (instead of 80%) to detect the clinically relevant difference of 7% between the two treatment arms being compared.

8.2 Randomization

The treatment assigned will be based on a dynamic minimization procedure using center, status of axillary lymph nodes (N0 versus N1-3 versus N4+), and hormonal receptor status (estrogen and/or progesterone receptor positive versus negative) as factors in the minimization algorithm, which will use a stochastic treatment allocation algorithm based on the variance method.

8.3 Efficacy evaluation

8.3.1 Efficacy Parameters

Primary

The primary efficacy parameter will be 5 year Disease-Free Survival (DFS). The DFS is defined as the interval from the date of randomization to the date of local, regional or metastatic relapse or the date of second primary cancer (with the exception of curatively treated non-melanoma skin cancer or in situ carcinoma of the cervix – see Exclusion Criteria 10a, 10b) or death from any cause whichever occurs first.

Secondary

Secondary efficacy parameters will consist of the comparison between groups based on Overall Survival (OS) and quality of life and on the pathologic and molecular markers for predicting efficacy.

8.3.2 Populations to be Analyzed

The analysis of DFS and of OS will be performed on the **Intent-to-Treat (I.T.T.) population**, defined as the population of all randomized patients analyzed in the treatment group they were assigned to. Randomized patients who did not receive chemotherapy will be analyzed in their group of randomization. The analysis of DFS and OS will also be performed on the **eligible patients populations**, defined as the ITT population patients less patients who were randomized but were not eligible for the trial according to the inclusion and exclusion criteria.

8.3.3 Statistical Methods

The Kaplan-Meier product limit method will be used to estimate the DFS and the OS. The log rank test, stratified for nodal status (N0 versus N1-3 versus N4+), for hormonal receptor status (estrogen and/or progesterone receptor positive versus negative), will be used to perform all pair wise comparisons between the three treatment arms with respect to DFS and OS. All tests of hypotheses will be two-sided. Confidence intervals of the median survival will be calculated using the method of Simon.

Cox's proportional hazards regression analysis will be performed for DFS and OS in order to adjust the treatment comparison for the major prognostic factors. These factors include age, menopausal status, type of surgery, histopathological findings, tumor size, pathological markers and molecular markers. Such adjusted analyses, for instance by nodal status (N0 versus N1-3 versus N4+) will be reported with appropriate caveats.

In the statistical analysis, a center will correspond to a participating institution. It is expected to have at the end of the study a large number of centers with few patients per center. It is consequently not planned to include any center effect in the analyses.

However, should there be centers with a large recruitment, it is planned to compare the consistence of the results between this (these) large center(s) and the entire study results, in terms of major baseline characteristics and primary endpoint.

8.3.4 Interim Analysis and Follow-up Analyses

Three interim analyses will be conducted after 300, 450 and 650 events are observed. A pragmatic group sequential design, as suggested by O'Brien-Fleming, will be used with overall significance levels of 0.0002, 0.0030, 0.0111, respectively, for the interim analyses, and an overall significance level of 0.0461 for the main analysis.

The main analysis will take place when a total of 900 events have occurred among all patients.
Some patients are expected to have a very long disease free survival. Consequently, a 10-year clinical follow-up has been planned. Two confirmatory analyses will be performed: at 3 years and 5 years after the main analysis. The purpose of these follow-up analyses is to update the DFS and OS estimates. All randomized patients will be followed until death or up to 5 years after the main analysis whichever occur first.

8.4 Cardiac Safety Analysis

One of the secondary endpoints of the study is to compare the cardiac safety of the three treatment arms.

At each of the cardiac analyses, cardiac deaths, congestive heart failure events, grade 3 or 4 arrhythmias, grade 3 or 4 cardiac ischemia / infarction events, and asymptomatic left ventricular decreases will be reviewed and assessed.

Cardiac events which encompass cardiac deaths, congestive heart failure, grade 3 or 4 ischemia, grade 3 or 4arrhythmias will be considered in the statistical analyses below. The following assumptions have been made.

1. The baseline incidence of events (cardiac deaths and congestive heart failure, grade 3 or 4 ischemia, grade 3 or 4 arrythmias) in the AC→T arm is expected to be 1%
2. A difference of > 4% between the AC→T arm and either of the Herceptin®-containing arms, AC→TH and TCH, respectively, will be considered unacceptable.

At each analysis, the two-tailed significance level of each interim analyses will be set at 0.05. This level of significance is not adjusted to take repeated analyses into account, and hence it will be merely indicative of a potential increase in incidence that needs to be scrutinized by the IDMC. Assuming a baseline incidence of cardiac deaths and symptomatic cardiac events of 1% in the control arm, the analyses will have approximately the following power to detect a difference of at least 4% in either treatment arm: 40% with 300 patients, 80% with 900 patients, 95% with 1,500 patients and >99.9% with all patients randomized in the study (3,222 patients) . The statistical power to detect a 4% difference would be slightly higher than these figures should the baseline incidence be lower than 1%, and slightly lower than these figures should the baseline incidence be higher than 1%.

Should one of the treatment arms have an unacceptably high incidence of cardiac toxicity, the Independent Data Monitoring Committee will recommend the Steering Committee to terminate it.

Asymptomatic cardiac abnormalities i.e. asymptomatic decreases in left ventricular ejection fraction will be part of the cardiac monitoring within the cardiac safety evaluation plan, but will not be evaluated within the cardiac safety analysis above. Clinically significant asymptomatic cardiac abnormality is an absolute decline of LVEF of > 15 % points from baseline and a value below LLN. The asymptomatic decreases will be evaluated as follows:

The four interim cardiac toxicity analyses will take place around 9 months after the 100th patient per arm, the 300th patient per arm and the 500th patient per arm and all patients randomized in the study, respectively, have been recruited and the results of the 5th LVEF evaluation among these patients have been collected and validated. Of note, the interval time of 9 months from randomization corresponds to the time of the 5th LVEF measurement in a patient who has had no delays or early discontinuation of chemotherapy.

Because of lack of data with respect to the significance of an asymptomatic decrease and its relation to the development of clinical congestive heart failure, no unacceptable number will be defined up front. The IDMC will be responsible for determining when the incidence of and/or the degree of asymptomatic decreases has become unacceptable and make the recommendation to the Steering Committee to discontinue one of the treatment arms.

After the review of the results of the first three cardiac analyses, the IDMC made the recommendation to continue the study as planned and to keep the same schedule of cardiac monitoring for all patients as for the first 1,500 patients.

8.5 Overall Safety Evaluation

8.5.1 Grading of adverse events

The National Cancer Institute Common Toxicity Criteria (NCI-CTC), version 2.0 and the corresponding grading system will be used to grade adverse events for recording in the CRF. For all adverse events not classified by the NCI-CTC a COSTART grading classification (FDA 1989) will be performed (severity as 1: mild, 2: moderate, 3: severe, and 4 life threatening).

Please refer to Appendix 13 for the standard ranges that will be used to analyse the hematological parameters for the study.

8.5.2 Populations to be analyzed

The safety analysis will be conducted on all patients who started at least one infusion of the study treatment.

8.5.3 Statistical Methods

Adverse events will be compared using two-tailed χ^2 tests or, when expected counts are low, Fisher's exact test or one of its generalizations. In view of the anticipated large number of statistical tests, p-values will not be interpreted in the usual sense but will be used as a "flagging device" to highlight differences worth further attention.

Descriptive statistics will be given on the number of patients in whom the study medication had to be replaced, delayed or permanently stopped.

8.6 Independent Data Monitoring Committee (IDMC)

8.6.1 Composition and mission of the IDMC

The Independent Data Monitoring Committee (IDMC) will be composed of three medical oncologists, one statistician, and two cardiologists. These members will be independent of the trial and familiar with the methodology of oncology trials. They must be aware of the dangers of conclusions based on immature data and agree with the design and the goals of this protocol.

The mission of the IDMC will be to ensure the ethical conduct of the trial and to protect the safety interests of patients in this study. This committee ensures the feasibility and progress of the trial. The IDMC will be responsible for both review of trial efficacy and safety.

8.6.2 Meetings of the IDMC

In the absence of any major event requiring the meeting of the IDMC members, an annual meeting of the IDMC will be held. The first review of the trial by the IDMC will take place one year after the first randomization. The committee will meet annually to assess the incidence and severity of each serious adverse event and adverse event. In the case where an unanticipated serious event or incidence is reported prior to the scheduled meeting, a meeting will be called immediately to address and assure the safety of the patients in the study. The latter may also include data that has come in from other studies but involves the same agents being administered in this study. The latter may also include newly presented efficacy data from another relevant study whose data in some way may influence the current BCIRG 006 study. In addition to the IDMC meetings scheduled above, the IDMC will meet after 300, 900 and 1500 patients, respectively, have been accrued and followed until 3 months after end of chemotherapy in AC → T, AC → TH arms, 4.5 months after end of chemotherapy in TCH arm, i.e around 9 months after the randomization, in a patient whos has had no delays or early discontinuation of chemotherapy. The IDMC will have written operating procedures and will maintain records of all its meetings.

With the exception of the above analyses for the cardiac safety evaluation, before any meeting of the IDMC, the Data Center will provide the IDMC with at least the following key documents:

- eligibility data
- on study protocol deviations (i.e. error in treatment allocation, early discontinuation of chemotherapy without any reason, unacceptable concomitant treatment, etc.)
- patient accrual
- lost to follow-up patients
- summary of patient and tumor characteristics
- summary of drug delivery
- toxicity and safety data
- and any other major problems encountered.

All data will be broken down by treatment arm and participating institution (whenever necessary). In addition, the Data Center will provide the IDMC with efficacy data at the time of the interim analysis. All results are confidential and must not be divulged to nonmembers of the Independent Data Monitoring Committee.

8.6.3 Recommendations of the IDMC

After each meeting, the IDMC will provide the Steering Committee with a written recommendation to either modify the trial (with reasons), or make the interim trial results of the trial public (with reasons), or continue the trial unchanged. Recommendations from the IDMC following each of these cardiac safety meetings will be to either close an arm, close the study, or continue the study without modifications. General safety reports will also be provided at the timing of these meetings and will be assessed by the IDMC. The final decision to amend the protocol or to discontinue the trial will be taken by the Steering Committee.

IX QUALITY OF LIFE EVALUATION

(see Appendix 11)

A quality of life assessment for each arm is a secondary endpoint of the study. Centers participating in the analysis will need to be defined prior to initiation of the study at their center. Some countries may be unable to participate due to the unavailability of the tools in the patient's first language.

The EORTC cancer-specific and EUROQUOL (ED-5D) general health indexes were chosen in this comparative study.

The QLQ-30 (v.3.0) profile questionnaire and the BR-23 module specific to breast cancer are, respectively, 30 and 23 items in a questionnaire format. The EUROQUOL ED-5D is a five question format in addition to a visual analog scale. They will be self-administered by the patient (Appendix 11) and should be completed in accordance with the following schedules.

	AC→T - AC→TH	TCH
Baseline	Within 14 days prior to randomization	Within 14 days prior to randomization
Cycle 4	day –1 to day 1 (before chemotherapy)	day –1 to day 1 (before chemotherapy)
Cycle 7 AC→T Cycle 7 AC→TH EOC* TCH	day –1 to day 1 (before chemotherapy)	EOC* Visit
EOC* AC→T EOC* AC→TH FUp1a TCH	EOC* Visit	6 weeks after the EOC visit FUp1a visit
Follow-Up	At 6, 12 and 24 months of follow-up after planned EOC	6, 12 and 24 months of follow-up after planned FUp1a
At Relapse	At Relapse Visit	At Relapse Visit

EOC* is the End of Chemotherapy Visit (3 weeks post last infusion of chemotherapy)

This schedule has been established to best assess potential differences in the longitudinal effects on quality of life and other socio-economic parameters between the two arms, particularly given the different lengths of treatment with each regimen. The patient should complete the questionnaire by herself at the center prior to physician assessment and prior to receiving treatment.

It is recommended that a key person (e.g research nurse) at each center be responsible for questionnaire data collection in order to optimize the compliance of the patient and to ensure the completeness of the data.

X SOCIO-ECONOMIC EVALUATION

A socio-economic comparison between the three arms will be also performed. The productivity and time losses questionnaires will only be given and analysed in Canada, Germany and USA. The productivity and time loss questionnaires will be self-administered by the patient and should be completed in accordance with the following schedules.

	AC→T AC→TH	TCH
Baseline	Within 14 days prior to randomization	Within 14 days prior to randomization
Cycle 4	day –1 to day 1 (before chemotherapy)	day –1 to day 1 (before chemotherapy)
Cycle 7 AC→T, AC→TH EOC* TCH	day –1 to day 1 (before chemotherapy)	EOC* Visit
EOC* AC→T, AC→TH FUp1a TCH♦	EOC* Visit	6 weeks after the EOC visit FUp1a visit
Follow-Up	Follow-up Visit at 6, 12 and 24 months after planned EOC	Follow-up Visit at 6, 12 and 24 months after planned FUp1a visit
At Relapse	At Relapse Visit	At Relapse Visit

* EOC is the End of Chemotherapy Visit (3 weeks post last infusion of chemotherapy – See Section 5.5.2 End of Chemotherapy Definition)

♦ FUp1a visit is an extra visit planned for the TCH arm 7 weeks after EOC. This corresponds to the EOC visit in the AC→T and AC→TH arms.

This schedule has been established to best assess potential differences in the longitudinal effects on socio-economic parameters between the two arms, particularly given the different lengths of treatment with each regimen. The patient should complete the questionnaire by herself at the center prior to physician assessment and prior to receiving treatment.

It is recommended that a key person (e.g. a research nurse) at each center should be responsible for questionnaire data collection in order to optimize the compliance of the patient and to ensure the completeness of the data.

XI ADVERSE EVENTS / TOXICITY

11.1 Definitions

11.1.1 Adverse Event

The term **adverse event** covers any sign, symptom, syndrome, or illness that appears or worsens in a subject during the period of observation in the clinical study and that may impair the well being of the subject. The term also covers laboratory findings or results of other diagnostic procedures that are considered to be clinically relevant (e.g., that require unscheduled diagnostic procedures or treatment measures, or result in withdrawal from the study).

The adverse event may be:
- A new illness
- Worsening of a concomitant illness
- An effect of the study medication, including comparator agents
- A combination of two or more of these factors.

No causal relationship with the study medication or with the clinical study itself is implied by the use of the term "adverse event".

Adverse events fall into the categories "non serious" and "serious" (see Section 11.1.2 Serious adverse event).

Surgical procedures themselves are not adverse events; they are therapeutic measures for conditions that require surgery. The condition for which the surgery is required may be an adverse event. Planned surgical measures permitted by the clinical study protocol and the condition(s) leading to these measures are not adverse events.

Worsening of a sign or symptom of the condition under treatment will normally be measured by efficacy parameters and should only be recorded as an Adverse Event if the outcome is serious (see section 11.1.2 Serious adverse event).

11.1.2 Serious Adverse Event

A serious adverse event is one that at any dose:
- Results in death
- Is life-threatening[1]
- Requires inpatient hospitalization or prolongation of existing hospitalization,
- Results in persistent or significant disability/incapacity[2]
- Is a congenital anomaly/birth defect
- Is an important medical event[3].

[1]"Life-threatening" means that the subject was at immediate risk of death at the time of the serious adverse event; it does not refer to a serious adverse event that hypothetically might have caused death if it were more severe.

[2]"Persistent or significant disability or incapacity" means that there is a substantial disruption of a person's ability to carry out normal life functions.

[3] Medical and scientific judgement should be exercised in deciding whether expedited reporting is appropriate in situations where none of the outcomes listed above occurred. Important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the patient or may require intervention to prevent one of the other outcomes listed in the definition above should also usually be considered serious. Examples of such events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse. The List of Critical Terms (1998 adaptation of WHO Adverse Reaction Terminology Critical Terms List) should be used as guidance for adverse events that

may be considered serious because they are medically important. (The List of Critical Terms can be found in the "Instructions for reporting serious adverse events (SAEs) occurring in clinical trials".

Cases of overdose with an adverse event that meets one of the criteria given above should of course be reported as "serious".

Congestive heart failure (CHF) is considered as a Protocol Defined Serious Adverse Event for this study. CHF should be reported as a serious adverse event regardless of causality and time of occurrence.

Clarification of the difference in meaning between "severe" and "serious":
The term "severe" is often used to describe the intensity (severity) of a specific event (as in mild, moderate, or severe myocardial infarction); the event itself, however, may be of relatively minor medical significance (such as severe headache). This is not the same as "serious", which is based on the outcome or action criteria usually associated with events that pose a threat to life or functioning. Seriousness (not severity) serves as a guide for defining regulatory reporting obligations. Severity of the adverse events should be graded according to the NCI Common Toxicity Criteria, version 2.0 (see Appendix 12).

11.1.3 Alert Terms and Other Reasons for Expedited Reporting to Pharmacovigilance.

No special events are subject to reporting as alert terms in this study.

Cases in which a "significant overdose" was taken and a non-serious adverse event or no adverse event occurred are to be reported to the sponsor in an expedited manner on a serious adverse event form, "Serious Adverse Event/Expedited Report from a Clinical Trial" form.

A "significant overdose" includes any overdose in which either a serious adverse event, a non-serious adverse event, or no adverse event occurs and is considered by the investigator as clinically relevant, i.e. poses an actual or potential risk to the subject's well being.

11.2 Period of Observation

For the purposes of this study, the period of observation for collection of Adverse Events extends from the time the subject starts treatment with the study medication until 30 days after the last infusion of study treatment (chemotherapy or Herceptin®).

Note since the half-life of Herceptin® is estimated to be approximately 28.5 days (95% confidence interval, 25.5-32.8 days), Herceptin® may be present in the circulation for up to 24 weeks (range 18-24 weeks) after stopping Herceptin ®treatment.

Cancer relapse (defined as component of the clinical efficacy endpoint) will not be reported as serious adverse event unless it satisfies the Serious Adverse Event definition during the period of observation (refer to section 11.1.2). After the end of the observation period, cancer relapse will not be reported as a serious adverse event.

Second primary malignancies, that satisfy the Serious Adverse Event definition (refer to section 11.1.2) will be reported as a serious adverse event regardless of causality and regardless of time of occurrence (during or after the period of observation).

Death from any cause that satisfy the Serious Adverse Event definition (refer to section 11.1.2) will be reported as a serious adverse event during the observation period. They will not be reported as a serious adverse event after the observational period unless study drug related.

11.3 Documentation and Reporting of Adverse Events by Investigator

All adverse events that occur after the start of the observation period set in this protocol (see Section 11.2 - Period of observation) must be documented on the pages provided in the case report form in accordance with the "Instructions for the completion of adverse event reports in clinical studies". These instructions are provided in the investigator's study file and in the case report form itself.

The following approach will be taken for documentation:

- **All adverse events** (whether serious or non serious, or considered as an alert term) must be documented on the "Adverse event" page of the case report form.

If the adverse event is serious (see Section 11.1.2 - Serious adverse event), the investigator must complete, in addition to the "Adverse Event" page in the case report form, a **"**Serious adverse event/ Expedited report from a clinical trial" form at the time the serious adverse event is detected. This form must be sent to the sponsor's representative:

For all countries outside Canada and USA
BCIRG Safety Manager
13, rue Martin Bernard
75013 Paris FRANCE
Tel: 33 (0) 1 58 10 08 98 or 33 (0) 1 58 10 08 80
Fax: 33 (0) 1 58 10 09 05

For Canada and USA
Fax: 310 478 7085

who will forward it to Aventis.

In the situation when a "significant overdose" had occurred without any adverse event (see Section 11.1.3 -Alert terms and other reasons for expedited reporting to Pharmacovigilance), the investigator should only complete a **"**Serious adverse event/ Expedited report from a clinical trial" form. This form must be sent to the sponsor's representative – BCIRG Safety Manager. In this situation, there is no need to complete the "Adverse event" page in the case report form.

Every attempt should be made to describe the adverse event in terms of a diagnosis. If appropriate, component symptoms should also be listed below the diagnosis. If only non-specific signs or symptoms are present, then these should be recorded as a diagnosis.

All subjects who have adverse events, whether considered associated with the use of the study medication or not, must be monitored to determine the outcome. The clinical course of the adverse event will be followed up according to accepted standards of medical practice, even after the end of the period of observation, until a satisfactory explanation is found or the investigator considers it medically justifiable to terminate follow-up. Should the adverse event result in death, a full pathologist's report should be supplied, if possible.

All questions on the completion and supply of adverse event report forms and any further forms issued to the investigator at a later date to clarify unresolved issues should be addressed to the sponsor's representative – the BCIRG Safety Manager.

11.4 Immediate Reporting to BCIRG

Serious adverse events and adverse events that fulfill a reason for expedited reporting to Pharmacovigilance (alert term and/or "significant overdose", as defined in Section 11.1.3 - Alert terms and other reasons for expedited reporting to Pharmacovigilance) must be documented on the **"**Serious adverse event/ Expedited report from a clinical trial" form as attached to this protocol as Appendix 8 in accordance with the "Instructions for reporting serious adverse events (SAEs) occurring in clinical trials". This form must be completed and supplied to the BCIRG Safety Manager within 24 hours, or at

the latest on the following working day. The "Serious adverse event/ Expedited report from a clinical trial" form is also provided in the investigator's study file together with the instructions to fill out such form.

The BCIRG Safety Manager will then report the adverse events to the sponsor (Aventis) as provided for in the Agreement for Clinical Research Management Services between the sponsor, Aventis and BCIRG. The Sponsor will then report the adverse events to the responsible health authorities in compliance to all legal and reporting requirements and as provided for in aforementioned Agreement.

The sponsor will ensure that all legal reporting requirements are met.

The initial report must be as complete as possible, including details of the current illness and (serious) adverse event, and an assessment of the causal relationship between the event and the study medication.

Information not available at the time of the initial report (e.g., an end date for the adverse event or laboratory values received after the report) must be documented on a follow-up "Serious adverse event/ Expedited report from a clinical trial" form.

The "Instructions for the completion of adverse event reports in clinical studies" give more detailed guidance on the reporting of serious adverse events, adverse events that comply with alert terms, and adverse events initially reported as non serious that become serious. In the latter situation, when a non-serious event becomes serious, details must be forwarded immediately to the sponsor's representative – the BCIRG Safety Manager on a "Serious adverse event in a clinical trial" form.

XII STUDY MEDICATION

12.1 Drug Packaging, labeling, dispensing and storage

12.1.1 Packaging and Labeling

A) DOCETAXEL (see Appendix 9 for detailed information)

Docetaxel will be supplied by Aventis.

Packaging:

Docetaxel will be provided as a sterile concentrate for infusion (concentration = 40 mg/mL).
The appropriate solvent for diluting the docetaxel concentrate for infusion will also be provided.
Vials are intended for single administration only.

Labeling:

The label attached to the Taxotere® (docetaxel) will contain the following information:

- Manufacturer's name and address
- Sponsor's name and address
- Product name
- Study code number
- Contents
- Directions for Use
- Storage Conditions
- Batch number and packaging numbers
- Legal requirements (expiry date)

B) HERCEPTIN® (see Appendix 10 for detailed information)

Herceptin® will be provided for the purpose of this study.

Packaging:

There are 2 preparations available for Herceptin®, the 440 mg multi dose vials, and the 150 mg unit dose vials. The preparation available for your site will vary depending on the country. See Appendix 10 for more details.

Labeling:

The label attached to the Herceptin® will contain the following information:

- Manufacturer's name and address
- Product name
- Contents
- Directions for Use
- Storage Conditions
- Batch number and packaging numbers
- Legal requirements (expiry date)

12.1.2 Dispensing and Storage

12.1.2.1 Docetaxel (see Appendix 9)

For preparation of the docetaxel solution and storage of the vials, please refer to Appendix 9.

Docetaxel will be administered to the patient as a one hour IV infusion. Use of a peristaltic *infusion pump* is recommended. Docetaxel should be given drop by drop for the first 5 minutes of the first 2 infusions to prevent an acute hypersensitivity reaction.

12.1.2.2 Doxorubicin

See preparation instructions on the package insert.

12.1.2.3 Cyclophosphamide

See preparation instructions on the package insert.

12.1.2.4 Carboplatin

See preparation instructions on the package insert.

12.1.2.5 Herceptin®

For preparation of the Herceptin® solution and storage of the vials, please refer to Appendix 10.

See section 5.2 for administration guidelines.

12.2 Drug Accountability

The person responsible for drug dispensing is required to maintain adequate records of all study drugs (docetaxel, Herceptin®, doxorubicin, cyclophosphamide). These records (e.g., drug movement form) include the dates the study medications are received from the manufacturer, the dates dispensed for the individual patient and the dates destroyed at the site as per each country's policy and guidelines (or returned to manufacturer). Patient number, date of infusion, investigator name, lot number, expiry date of the study medication must be documented in the CRF.

The person responsible for drug administration to the patient will record precisely the date and the time the drug is administered to the patient. In case the drug infusion has to be stopped, the exact date and time that the infusion has been stopped and restarted will be carefully recorded.

XIII ADMINISTRATIVE ASPECTS

13.1 Monitoring, Auditing, and Inspecting

The study will be monitored by regular site visits and telephone calls to the investigator by members of or personnel designated by the BCIRG Clinical Operations Department. During site visits, the monitor should review original patient records, drug accountability records and document retention. Additionally, the monitor should observe study procedures and will discuss any problems with the investigator. During the course of the study BCIRG and/or Aventis, or any third party entitled by BCIRG and Aventis jointly, may conduct site audits. The investigator will provide direct access to source data/documents for trial related monitoring, audits, IRB/EC review and regulatory inspections.

13.2 Patient Identification

All patients screened for the study will have their initials and birth date entered chronologically on the patient log at the initial visit. In the event a patient is excluded from study participation, the reason is to be documented in the space provided on the patient log.

Each patient will be assigned a Patient Randomization Number on registration. This number and the patient initials are to

be entered on the Case Report Form.

13.3 Recording of Data

The study will be conducted using paper based CRFs. NCR™ Case Report Forms will be supplied by BCIRG providing a white original and colored copies. These forms must be typewritten or *PRINTED LEGIBLY* using black ball-point pen when prepared for submission to BCIRG.

The forms should be verified against all original records (and workbooks, if applicable) by the BCIRG Clinical Monitor before submission. The bottom copy will be retained in the investigator's files, and all other copies will be returned to the BCIRG. Central Operational Office in Edmonton Canada. No case report forms are to be mailed to the BCIRG without specific authorization. Case Report Forms and all original data should be readily available for review during scheduled monitoring visits. Any data to be recorded directly on the Case Report Forms will be considered to be source data.

13.4 Record Retention

1. Copies of all pertinent information will be retained by the investigator for a period of at least 15 years from study completion. Additional considerations must be made about complying with applicable local laws, guidelines, etc.
2. A study document binder will be provided by BCIRG for all required study documents.

13.5 Confidential Follow-up

The investigator will be responsible for retaining sufficient information about each patient (e.g. name, address, phone number, social security number/identity number, and identity in the study) so that regulatory agencies, BCIRG, or Aventis may access
this information should the need to do so arise. These records should be retained in a confidential manner for as long as legally mandated according to local requirements.

13.6 Patient Informed Consent (Appendix 7)

Prior to the screening evaluation, the patient will be informed of the nature of the study drug and will be given pertinent information as to the intended purpose, possible benefits, and possible adverse experiences. The procedures and possible hazards to which the patient will be exposed will be explained.

An approved informed consent statement will then be read and signed by the patient, and, when required, a witness, and the investigator. The patient will be provided with a copy of the signed informed consent statement. The patient may withdraw from the study at anytime without prejudicing future medical treatment.

13.7 Ethics Committee/Institutional Review Board

The final approved protocol and the informed consent statement will be reviewed by a properly constituted Ethics Committee/IRB. The Ethics Committee's/Board's decision concerning the conduct of the study will be made in writing to the investigator and a copy of this decision will be provided to BCIRG and Aventis.

Particular attention is drawn to the FDA's regulation regarding the IRBs. By signing the "Statement of Investigator" form (Form 1572), the investigator provides BCIRG and Aventis with the necessary assurance that an IRB is responsible for the initial and continuing review and approval of the proposed clinical study in accordance with these regulations.

The investigator will agree to make required progress reports to the Ethics committee/IRB, as well as report any serious adverse events, life-threatening problems or deaths. The investigator will also inform the Ethics Committee/IRB of reports of serious adverse events (provided to him/her by Aventis) in other clinical studies conducted with the study drug. The

Ethics Committee/IRB must be informed by the investigator of the termination of the study.

13.8 Declaration of Helsinki

This study is to be performed in accordance with the Declaration of Helsinki (Edinburgh, Scotland, Amendment October 2000 Note of Clarification on paragraph 29 added by the WMA General Assembly, Washington 2002, Note of Clarification on Paragraph 30 added by the WMA General Assembly, Tokyo 2004), as described in Appendix 1.

13.9 Insurance of Liabilities

If required, the investigator may forward the Ethics Committee/IRB a copy of the Insurance that the sponsor has to take out covering his and any other participating parties liabilities.

13.10 Modification of the Protocol

Any modifications to the protocol which may impact on the conduct of the study, potential benefit of the patient or may affect patient safety, including changes of study objectives, study design, patient population, sample sizes, study procedures, or significant administrative aspects will require a formal amendment to the protocol. Such amendment will be agreed upon by BCIRG and Aventis, and approved by the Ethics Committee/IRB prior to implementation and notified to the health authorities in accordance with local regulations.

Administrative changes of the protocol are minor corrections and/or clarifications that have no effect on the way the study is to be conducted. These administrative changes will be agreed upon by BCIRG and Aventis, and will be documented in a memorandum. The Ethics Committee/IRB may be notified of administrative changes at the discretion of BCIRG.

13.11 Use of Information and Publication

All information concerning the study drug supplied by Aventis in connection with this study and/or by any other party collaborating with BCIRG and Aventis within this Study, and not previously published, is considered confidential and proprietary information. This information includes the Investigator's Brochures, clinical protocol, workbooks if applicable, Case Report Forms, assay methods, BCIRG technical methodology, and basic scientific data. This confidential information shall remain the sole property of BCIRG and Aventis or the respective collaborating party and, shall not be disclosed to others without prior written consent from BCIRG and Aventis and shall not be used except in the performance of this study.

To allow for the use of the information derived from this clinical study and to insure compliance to current regulations, the investigator is obliged to provide BCIRG with complete test results and all data developed in this study. Only BCIRG and Aventis, or any third party entitled by BCIRG and Aventis jointly, may make information obtained during this study available to the physicians and to regulatory agencies, except as required by regulation.

No publication, abstract or presentation of the study will be made without approval of the advisory board of the BCIRG and Aventis. BCIRG and Aventis will review the manuscript to prevent forfeiture of patent rights to data not in the public domain. The authorship list will be agreed by the investigators prior to publication. The names on the author list will be given according to the participation in the design of the protocol as well as taking into consideration the input of the number of eligible and evaluable patients accrued by the investigators in each center. The study will only be published once it is completed and the final analysis has been performed by BCIRG and Aventis. Interim abstracts will be presented according to the statistical plan and in agreement with BCIRG and Aventis.

In the event BCIRG and Aventis choose to publish the data from this study, BCIRG and Aventis may provide the advisory board of the study with a manuscript at least 30 days prior to the expected date of submission to the intended publisher.

XIV INVESTIGATOR'S AGREEMENT

I have read the preceding protocol

BCIRG 006 (TAX GMA 302)

A MULTICENTER PHASE III RANDOMIZED TRIAL COMPARING DOXORUBICIN AND CYCLOPHOSPHAMIDE FOLLOWED BY DOCETAXEL (AC→T) WITH DOXORUBICIN AND CYCLOPHOSPHAMIDE FOLLOWED BY DOCETAXEL AND TRASTUZUMAB (AC→TH) AND WITH DOCETAXEL, CARBOPLATIN AND TRASTUZUMAB (TCH) IN THE ADJUVANT TREATMENT OF NODE POSITIVE AND HIGH RISK NODE NEGATIVE PATIENTS WITH OPERABLE BREAST CANCER CONTAINING THE HER2 ALTERATION.

1. and agree that it contains all necessary details for conducting this study. I will conduct the study as outlined in the preceding protocol and in compliance with GCPs. I will provide copies of the protocol and all drug information relating to preclinical and prior clinical experience furnished to me by Aventis and BCIRG, to all physicians responsible to me who participate in this study. I will discuss this material with them to assure that they are fully informed regarding the drug and the conduct of the study. I agree to keep records on all patient information (case report forms and patient's informed consent statement), drug shipment and return forms, and all other information collected during the study in accordance with legal regulations

_____
Investigator (PRINT NAME)

_____
Investigator Signature Date

_____
Global Project and Medical Director
BCIRG Date

_____ Date
Medical Director, GMA Aventis

XV REFERENCES

1. CA - A Cancer Journal for Clinicians, Cancer Statistics 2000, American Cancer Society, Inc., Atlanta, USA, Lippincott-Raven Publishers, Vol 50.No 1, 2000

2. National Cancer Institute of Canada: Canadian Cancer Statistics 2000, Toronto, Canada, 2000.

3. Jensen OM, Estève J, Moller H, et al. Cancer in the European Community and its members states. Eur J Cancer 26 : 1167-1256, 1993

4. Henderson IC : Chemotherapy for metastatic disease, in : Harris JR, Hellman S, Henderson IC, Kinne DW eds : Breast diseases. Philadelphia : J.B. Lippincott company, 2nd edition, 1991 : pp. 604-665

5. Bonadonna G. and Valagussa P. : Current status of adjuvant chemotherapy for breast cancer. Sem. Oncol. 14 : 8-22, 1987

6. NIH Consensus Development Conference Statement, Adjuvant therapy for breast cancer (draft), 2000

7. Early Breast Cancer Trialists' Collaborative Group: Polychemotherapy for early breast cancer: an overview of the randomized trials. The Lancet, 1998; 352; 930: 942.

8. Osborne CK, Clark GM, Ravdin PM: Adjuvant systemic therapy of primary breast cancer. In Disease of the Breast, Harris JR, Lippman ME, Morrow M, Hellman S, Eds., Lippincot-Raven, Philadelphia, New-York. 1996, 548.

9. Bonadonna G, Brusamolino E, Valagussa P et al: Combination chemotherapy as an adjuvant treatment in operable breast cancer. N Engl J Med 1976; 294: 405.

10. Fisher B, Brown AM, Dimitrov et al: Two months of doxorubicin-cyclophosphamide with and without interval reinduction therapy compared with 6 months of cyclophosphamide, methotrexate, and fluorouracil in positive-node breast cancer patients with tamoxifen-non-responsive tumors: results from NSABP B-15. J Clin Oncol 1990; 8: 1483.

11. Carpenter JT, Velez-Garcia E, Aron BS et al: Five-year results of a randomized comparison of cyclophosphamide, doxorubicin (Adriamycin) and fluorouracil (CAF) vs. cyclophosphamide, methotrexate and flurouracil (CMF) for node positive breast cancer: a Southeastern Cancer Study Group Study. Proc ASCO 1994; 13: 66.

12. Buzzoni R, Bonadonna G, Valagussa P et al: Adjuvant chemotherapy with doxorubicin plus cyclophosphamide, methotrexate, and fluorouracil in the treatment of resectable breast cancer with more than three positive axillary nodes. J Clin Oncol 1991; 9: 2134.

13. Coombes RC, Bliss JM, Marty M et al: A randomized trial comparing adjuvant FEC with CMF in premenopausal patients with positive resectable breast cancer. Proc AmSoc Clin Onc 1991; 10 : 41.

14. Misset JL, Gil-Dalgado M, Chollet Ph et al: Ten-year results of the French trial comparing Adriamycin, vincristine, 5-fluorouracil and cyclophosphamide to standard CMF as adjuvant therapy for node positive breast cancer. Proc Am Soc Clin Onc 1992; 11 : 54.

15. Goldhirsch A, Glick J, Gelber R et al: Meeting Highlights: International Consensus Panel on the Treatment of Primary Breast Cancer. J. Natl. Cancer Inst. 1998; 90(21): 1601-1608.

16. Gianni L, Munzone E, Capri G et al. Paclitaxel by 3-hour infusion in combination with bolus doxorubicin in women with untreated metastatic breast cancer: High antitumor efficacy and cardiac effects in a dose-sequence-finding study. J Clin Onc 1995; 13:2688-99.

17. Gehl J, Boesgaard M, Paaske T et al. Combinated doxorubicin and paclitaxel in advanced breast cancer : Effective and cardiotoxic. Ann Oncol 1996; 7:687-93.

18. Henderson IC, Berry D, Demetri G, et al. Improved disease-free (DFS) and overall survival (OS) from the addition of sequential paclitaxel (T) but not from the escalation of doxorubicin (A) dose level in the adjuvant chemotherapy of patients (PTS) with node-positive primary breast cancer (BC). Proc. Am. Soc. Clin. Oncol. 1998 17:390a.

19. Thomas E, Buzdar A, Theriault R et al: Role of Paclitaxel in Adjuvant Therapy of Operable Breast Cancer: Preliminary Results of Prospective Ranomized Clinical Trial. Proc Am Soc Clin Onc. 2000 19:285a.

20. Cross M. Dexter TM. Growth factors in development, transformation, tumorigenese. Cell 1991; 64:271-280.

21. Slamon DJ, Clark GM, Wong SG, et al. Human breast cancer: correlation of relapse and survival with amplification of the HER2/proto-ongene. Science 1987; 235:177-182.

22. Slamon DJ, Godolphin W, Jones LA, et al. Studies of the HER2/protoco-oncogene proto-oncogene in human breast and ovarian cancer. Science 1989; 244: 707-712.

23. Press MF, Pike MC, Hung G, et a, Amplification and overexpression of HER2 in carcinomas of the salivary gland: correlation with poor prognosis. Cancer Res 1994;54(21):5675-82

24. Fendly BM, Winget M, Hudziak RM, et al. Characterization of murine monoclonal antibodies reactive to either the human epidermal growth factor receptor or HER2/protoco-oncogene gene product. Cancer Res 1990; 50:1550-1558.

25. Shepard HM, Lewis GD, Sarup JC, et al. Monoclonal antibody therapy of human cancer: taking the HER2 protocon-oncogene to the clinic. J Clin Immunol 1991; 11: 117-127

26. Chazin, V.R., Kaleko, M., Miller, A.D., Slamon, D.J. Transformation Mediated by the Human HER2 Gene Independent of the Epidermal Growth Factor Receptor. Oncogene. 7:1859-66, 1992

27. Lewis GD, Figari I, Fendly B, et al. Differential responses of human tumour cell lines to anti-p185 HER2 monoclonal antibodies. Cancer Immunol Immunother 1993; 37:255-263.

28. Pietras RI, Fendly BM, Chazin VR, et al. Antibody to HER2 receptor blocks DNA repair after cisplatin in human breast and ovarian cancer cells. Oncogene 1994 Jul; 9(7): 1829-1838

29. Pegram MD, Hsu S, Pietras R, et al Inhibitory effects of combination of HER2 antibody and chemotherapeutic agents used for treatment of human breast cancers. Oncogene 1998 18,2241-2251, 1999

30. Pegram, M.D., Finn, R.S., Arzoo, K., Beryt, M., Pietras, R.J., Slamon D.J, The Effect of HER2 Overexpression on Chemotherapeutic Drug Sensitivity in Human Breast and Ovarian Cancer Cells. Oncogene, 15. 537-547. 1997

31. Pauletti, G, Dandekar, S., Rong, H-M, Ramos, L, Peng, H-J M.D., Seshadri, R,. Slamon, D.J. Assessment of Methods for Tissue Based Detection of the HER2 /neu Alteration in Human Breast Cancer: A direct Comparison of FISH and Immunhostochemistry. J Clin.Onc 18: 3651-3664, 2000

32. Pauletti, G., Godolphin, W., Press, M.F., Slamon, D.J. Detection and Quantitation of HER2 /neu Gene Amplification in Human Breast Cancer Archival Material Using Fluorescence *In Situ* Hybridization, Oncogene 13, 63-72. 1996

33. Mass RD, Sanders C, Charlene K et al The Concordance between the Clinical Trials Assay (CTA) and Fluorescence in situ hybridization in the Herceptin® Pivotal Trials. Proc Am Soc Clin Onc 2000; 29 291a.

34. Mass RD, Press M, Anderson S, et al. Improved Survival Benefit From Hercpetin (Trastuzumab) in Patients Selected by Fluorescence in Situ Hybridization (FISH). Proc Am Soc Clin Onc 2001; 20; 22a; 85 abs

35. Vogel CL, Cobleigh M, Tripathy D, et al. Superior Outcomes with Herceptin (Trastuzumab) in Fluorescence in Situ Hybridization (FISH)-Selected Patients. Proc Am Soc Clin Onc 2001; 20; 22a; 86 abs

36. Vogel CL, Cobleigh M, Tripathy D, et al. Superior Outcomes with Herceptin (Trastuzumab) in Fluorescence in Situ Hybridization (FISH)-Selected Patients. Proc Am Soc Clin Onc 2001; 20; 22a; 86 abs

37. Esteva FJ, Valero V, Booser D, et al. A Phase II Trial of Weekly Docetaxel (D) and Herceptin (H) for Patients with HER2

Overexpressing (HER 2+) Metastatic Breast Cancer (MBC): Safety, Efficacy and Correlation with Serum HER2 (sHER2) Levels. Proc Am Soc Clin Onc 2001; 20; 68b; 2019 abs

38. Slamon DJ, et al., unpublished

39. Baselga J, Tripathy D, Mendelsohn J, et al. Phase II study of weekly intravenous recombinant humanized anti-p185 HER2 monoclonal antibody in patients with HER2 overexpressing metastatic breast cancer. J. Clin. Oncol, 1996; 14(3): 737-44

40. Cobleigh M, Vogel CL, Tripathy D, et al. Efficacy and safety of Herceptin® (humanized anti-HER2 antibody) as a single agent in 222 women with HER2 overexpression who relapsed following chemotherapy with breast cancer. Proceedings ASCO 1998. Vol 17, abs 376, page 97a.

41. Pegram MD, Lipton A, Hayes D, et al. Phase II study of receptor enhanced chemosensitivity using recombinant humanized anti-p 185 HER2 monoclonal antibody (rhuMab HER2) plus cisplatin in patients with HER2 overexpressing metastatic breast cancer refractory to chemotherapy treatment. J Clin Oncol 1998 (in press)

42. Slamon D, Leyland-Jones B, Shak S, et al. Addition of Herceptin® (humanized anti-HER2 antibody) to first line chemotherapy for HER2 overexpressing metastatic breast cancer (HER2+/MBC) markedly increases anticancer activity: a randomized, multinational controlled
phase III trial. Proceedings ASCO 1998. Vol 17, abs 377, page 98a.

43. Norton L, Slamon D, Leyland-Jones B et al. Overall Survival Advantage to Simultaneous Chemotherapy Plus the Humanized anti-HER2 monoclonal antibody Herceptin® in HER2-overexpressing (HER2+) metastatic breast cancer. Proc Am Soc Clin Onc 1999. Vol 18; 483a.

44. Gelmon K, Arnold A, Verma S, et al. Pharmacokinetics (PK) and Safety of Trastuzumab (Herceptin®) when Administered every 3 Weeks to Women with Metastatic Breast Cancer. Proc Am Soc Clin Onc 2001; 20: 271a

45. Herceptin® investigator brochure-Genentech,Inc./F. Hoffmann-La Roche Ltd. Version date :3 may 2001

46. Hancock MD, Langton BC,Chan T et al. A monoclonal antibody against the c-erB-2 protein enhances the cytotoxicity of cisdiamminedichloroplatinum against human breast and ovarian tumor cell lines. Cancer Res 1991 Sep 1;51 (17): 4575-4580.

47. Arteaga CL, Winnier AR, Poirier MC, et al. p185c-erB-2 signal enhances cisplatin-induced cytotoxicity in human breast carcinoma cells; association between an oncogenic receptor tyrosine kinase and drug-induced DNA repair. Cancer res 1994 Jul 15; 54 (14): 3758-3765

48. Baselga J, Seidman AD, Rosen, et al. HER2 overexpression and paclitaxel sensitivity in breast cancer: Therapeutic implications. Oncology (Hunting) 1997; 11 (3suppl 2): 43-48.

49. Yen L, Nie ZR, You XL, et al. Regulation of cellular response to cisplatin-induced DNA damage and DNA repair in cells overexpressing p 185 (erB-2) is dependent on the ras signaling pathway. Oncogene 1997 Apr 17; 14 (15); 1827-1835.

50. Trudeau ME, Eisenhauer EA, Higgins BP, et al: Docetaxel in patients with metastatic breast cancer: A phase II study of the National Cancer Institute of Canada Clinical Trials Group. J Clin Oncol (1996) 14: 422-428.

51. Ten Bokkel Huinink WW, Prove AM, Piccart M, et al: A phase II trial with docetaxel (Taxotere) in second line treatment with chemotherapy for advanced breast cancer. A study of the EORTC-ECTG. Ann Oncol (1994) 5: 527-532.

52. Hudis CA, Seidman AD, Crown JPA, et al: Phase II pharmacologic study of docetaxel as initial chemotherapy for metastatic breast cancer. J Clin Oncol (1996) 14: 58-65.

53. Chevallier B, Fumoleau P, Kerbrat P, et al: Docetaxel is a major cytotoxic drug for the treatment of advanced breast cancer: A phase II trial of the Clinical Screening Co-operative Group of the EORTC. J Clin Oncol (1995) 13: 314-322.

54. Fumoleau P, Chevalier B, Kerbrat P, et al: A multicentre phase II study of the efficacy and safety of docetaxel as first-line treatment of advanced breast cancer: A report of the Clinical Screening Group of the EORTC. Ann Oncol (1996) 7: 165-171.

55. Van Oosterom AT, Dieras V, Tubiana-Hulin M, et al: Taxotere in previously treated patients with metastatic breast carcinoma (MBC) stratification for anthracycline resistance. Proc Am Soc Clin Oncol (1996) 15: 141 (abstr. 231).

56. Alexopoulos CG, Rigatas G, Efremidou A, et al: Phase II study of Taxotee monotherapy in previously treated patients with advanced breast cancer. Eur J Cancer (1997) 33(suppl 8): S153 (abstr. 680).

57. Vorobiof DA, Chasen MR, Moeken R: Phase II trial of single agent docetaxel in previously treated patients with advanced breast cancer (ABC). Proc Am Soc Clin Oncol (1996) 15: 130 (abstr. 185).

58. Terzoli E, Nistico C, Garufi C, et al: Docetaxel in advanced breast carcinoma (MBC) patients (pts) pretreated with anthracyclines. Pro Am Soc Clin Oncol (1998) 17: 177a (abstr. 682).

59. Adachi I, Wantabe T, Takashima S, et al: A late phase II study of RP 56976 (docetaxel) in patients with advanced or recurrent breast cancer. Br J Cancer (1996) 73: 210-216.

60. Taguchi T, Hiarata K, Kunii Y, et al: An early phase II study of RP 56976 (docetaxel) in patients with breast cancer. Gan To Kagaku Ryoho (1994) 21: 2453-2460.

61. Taguchi T, Mori S, Abe R, et al: Late phase II clinical study of RP 56976 (docetaxel) in patients with advanced/recurrent breast cancer. Gan To Kagaku Ryoho (1994) 21: 2625-2632.

62. Dieras V, Chevallier B, Kerbrat P, et al: A multicentre phase II study of docetaxel 75 mg/m² as first-line chemotherapy for patients with advanced breast cancer: A report of the Clinical Screening Group of the EORTC. Br J Cancer (1996) 74:650-656.

63. Leonard RC, O'Brien M, Barrett-Lee P, et al: A prospective analysis of 390 advanced breast cancer patients treated with Taxotere throughout the UK. Ann Oncol (1996) 7(suppl 5): 19 (abstr. 810).

64. Shapiro JD, Michael M, Millward MJ, et al: Activity and toxicity of docetaxel (Taxotere) in women with previously treated metastatic breast cancer. Aust N Z J Med (1997) 27: 40-44.

65. Valero V, Holmes F, Walters RS, et al: Phase II trial of docetaxel: A new, highly effective antineoplastic agent in the management of patients with anthracycline-resistant metastatic breast cancer. J Clin Oncol (1995) 13: 2886-2894.

66. Ravdin PM, Burris HA, Cook G, et al: Phase II trial of docetaxel in advanced anthracycline-resistant or anthracenedione-resistant breast cancer. J Clin Oncol (1995) 13: 2879-2885.

67. Bonneterre J, Guastalla JP, Fumoleau P, et al: A phase II trial of docetaxel in patients with anthracycline resistant metastatic breast cancer. Breast Cancer Res Treat (1995) 37: 89 (abstr. 305).

68. Chan S, Friedrichs K, Noel D, et al: Prospective randomized trial of docetaxel versus doxorubicin in patients with metastatic breast cancer. J. Clin. Oncol. (1999) 17(8): 2341-2354.

69. Nabholtz J-M, Senn HJ, Bezwoda WR, et al. Prospective randomized trial of docetaxel versus mitomycin C plus vinblastine in patients with metastatic breast cancer progressing despite previous anthracycline-containing chemotherapy. J. Clin. Oncol. (1999) 17(5): 1413-1424.

70. Sjostrom J, Mouridsen H, Pluzanska A et al. Taxotere versus methotrexate-5-fluorouracil in patients with metastatic breast cancer as 2nd line chemotherapy: A phase III study. Proc. Am. Soc. Clin. Oncol. (1998) 17: 111a (abstr).

71. Bonneterre J, Monnier A, Roche H et al. Taxotere versus 5-fluorouracil + navelbine in patients with metastatic breast cancer as 2nd line chemotherapy: A phase III study. Breast Cancer Res. Treat. (1997) 50: 261 (Abstr.)

72. Sledge GW Jr, Loehrer PJ Sr, Roth BJ, et al. Cisplatin as first-line therapy for metastatic breast cancer. J Clin Oncol 1988 Dec;6(12):1811-4

73. Yap HY, Salem P, Hortobagyi GN, Bodey GP Sr, et al. Phase II study of cis-dichlorodiammineplatinum(II) in advanced breast cancer. Cancer Treat Rep 1978 Mar;62(3):405-8

74. Forastiere AA, Hakes TB, Wittes JT, et al Cisplatin in the treatment of metastatic breast carcinoma:A prospective randomized trial of two dosage schedules. Am J Clin Oncol 1982 Jun;5(3):243-7

75. Martino S, Samal BA, Singhakowinta A, et al. Phase II study of cis-diamminedichloroplatinum II for advanced breast cancer. Two

dose schedules. J Cancer Res Clin Oncol 1984;108(3):354-6

76. Kolaric K, Vukas D Carboplatin activity in untreated metastatic breast cancer patients--results of a phase II study. Cancer Chemother Pharmacol 1991;27(5):409-12

77. O'Brien ME, Talbot DC, Smith IE. Carboplatin in the treatment of advanced breast cancer: aphase II study using a pharmacokinetically guided doseschedule. J Clin Oncol 1993 Nov;11(11):2112-7

78. Franciosi V, Cocconi G, Michiara M, et al. Front-line chemotherapy with cisplatin and etoposide for patients with brain metastases from breast carcinoma, nonsmall cell lung carcinoma, or malignant melanoma: a prospective study. Cancer 1999 Apr 1;85(7):1599-605

79. Cocconi G, Bisagni G, Bacchi M, et al. Cisplatin and etoposide as first-line chemotherapy for metastatic breast carcinoma: a prospective randomizedtrial of the Italian Oncology Group for Clinical Research. J Clin Oncol 1991 Apr;9(4):664-9

80. Oster MW, Schilsky RL, Faraggi D, et al. Cytosine arabinoside and cisplatin for advanced breast cancer. A phase II study of the Cancer and Leukemia Group B. Cancer 1991 Oct 15;68(8):1696-8

81. Kucuk O, Pandya KJ, Skeel RT, et al. Phase II study of cisplatin and 5-fluorouracil in previously treated metastatic breast cancer: an Eastern Cooperative Oncology Group study. Breast Cancer Res Treat 1999 Sep;57(2):201-6

82. Fernandez Hidalgo O, Gonzalez F, Gil A, Campbell W, et al. 120 hours simultaneous infusion of cisplatin andfluorouracil in metastatic breast cancer. Am J Clin Oncol 1989 Oct;12(5):397-401

83. Shamseddine AI, Taher A, Dabaja B, et al. Combination cisplatin-vinorelbine for relapsed and chemotherapy-pretreated metastatic breast cancer. Am J Clin Oncol 1999 Jun;22(3):298-302

84. Ray-Coquard I, Biron P, Bachelot T, et al. Vinorelbine and cisplatin (CIVIC regimen) for thetreatment of metastatic breast carcinoma after failure ofanthracycline- and/or paclitaxel-containing regimens. Cancer 1998 Jan 1;82(1):134-40

85. Madajewicz S, Matei D, Hentschel P, et al. Actual 5-year survival of patients with stage IIIB breast carcinoma: phase II trial of methotrexate, vinblastine, adriamycin, cisplatin, and folinic acid. Cancer Invest 1999;17(7):463-7

86. Langer CJ, Catalano R, Weiner LM, et al. Phase II evaluation of methotrexate, vinblastine,doxorubicin, and cisplatin (M-VAC) in advanced,measurable breast carcinoma. Cancer Invest 1995;13(2):150-9

87. Bisagni G, Cocconi G, Ceci G, et al. CM-VAC combination in locally advanced, locally recurrent or metastatic breast carcinoma. A phase II study. Ann Oncol 1994 Jan;5(1):93-4

88. Roth BJ, Sledge GW Jr, Williams SD, et al. Methotrexate, vinblastine, doxorubicin, and cisplatin in metastatic breast cancer. A phase II trial of the Hoosier Oncology Group. Cancer 1991 Jul 15;68(2):248-52

89. van der Gaast A, Bontenbal M, Planting AS, et al. Phase II study of carboplatin and etoposide as a first line regimen in patients with metastatic breast cancer. Ann Oncol 1994 Nov;5(9):858-860

90. Crown J, Hakes T, Reichman B, et al. Phase II trial of carboplatin and etoposide in metastatic breast cancer. Cancer 1993 Feb 15;71(4):1254-7

91. Deltetto F, Durando A, Camanni M, et al. Carboplatin plus etoposide regimen in advanced breast cancer. A phase II study. Eur J Gynaecol Oncol 1997;18(3):185

92. Vinolas N, Daniels M, Estape J, et al. Phase II trial of carboplatin and etoposide activity in pretreated breast cancer patients. Am J Clin Oncol 1992 Apr;15(2):160-2

93. Barker LJ, Jones SE, Savin MA, et al. Phase II evaluation of carboplatin and VP-16 for patients with metastatic breast cancer and only one prior chemotherapy regimen. Cancer 1993 Aug 1;72(3):771-3

94. Fountzilas G, Skarlos D, Theoharis D, et al. Carboplatin and oral etoposide in the treatment of patients with advanced breast cancer refractory to anthracyclines. Tumori 1993 Dec 31;79(6):389-92

95. Ron IG, Vishne TH, Kraminsky N, et al. Phase II study of carboplatin and etoposide as salvage treatment for patients with metastatic breast cancer. Am J Clin Oncol 1999 Feb;22(1):35-7

96. Smith IE, Walsh G, Jones A, et al. High complete remission rates with primary neoadjuvant infusional chemotherapy for large early breast cancer. J Clin Oncol 1995 Feb;13(2):424-9

97. Nabholtz JM, Falkson G, Campos D, et al. A phase III trial comparing doxorubicin (A) and docetaxel (T) (AT) to doxorubicin and cyclophosphamide (AC) as first line chemotherapy for MBC. Proc. Amer. Soc. Clin. Oncol. (1999) 18: 127a (Abstr 485).

98. Millward M, Zalcberg J, Bishop J, Webster L et al: Phase I trial of docetaxel and cisplatin in previously untreated patients with advanced non-small cell cancer. JCO 1997;15:750-758.

99. Hilkens P, Pronk L, Verweiji J et al: Peripheral neuropathy induced by combination chemotherapy of docetaxel and cisplatin. Br J Cancer 1997;75:417-422.

100. Bernard A et al. Docetaxel (T) and cisplatin (P) in anthracycline-pretreated advanced breast cancer (ABC): results of a phase II pilot study. Proc Am Soc Clin Oncol 1998;17:128A Abstract 491.

101. Antoine EC. Coeffic D. Gozy M. et al. A phase II study of docetaxel (T) and cisplatin (P) in anthracycline (A)-pretreated metastatic breast cancer (PMBC): preliminary results. Proc Am. Soc Clin Oncol. 1997, 16:A551.

102. Lombart-Cussac A. Spielmann M. Dohollou N. Le Cesne A. Rixe O. Cisplatin-Taxotere phase I/II trial in patients with anthracycline-resistant advanced breast cancer (ABC).Proc Am Soc Clin Oncol. 1997, 16:A629.

103. Burris HA, Kuzur ME, Albain KS et al: A phase II trial of docetaxel and Herceptin® for metastatic breast cancer overexpressing HER2. Breast Cancer Res Treat 2000: 64; 24abs.

104. Uber KA, Nicholson BP, Thor AD, et al: A phase II Trial of Weekly Docetaxel (D) and Hercpetin (H) as First or Second-line Treatment in HER2 over-expressing metastatic breast cancer. Proc Am Soc Clin Oncol 2001; 20: 50b, abs 1949.

105. Meden H, BenekeA, Hesse T, et al. Weekly intravenous recombinant humanized anti-HER2 monoclonal antibody (Trastuzumab) plus docetaxel in patients with metastatic breast cancer (MBC): a pilot study. Proc Am Soc Clin Oncol 2001; 20: 60b, abs 1987.

106. Pienkowski T, Fumoleau P, Eiermann W, et al. Taxotere, Cisplatin and Herceptin (TCH) in First-Line HER2 Positive Metastatic Breast Cancer (MBC) Patients, a Phase II Pilot Study by the Breast Cancer International Research Group (BCIRG). Proc Am Soc Clin Onc 2001: 20: 70b.

107. Nabholtz JM, Crown J, Yonemoto L, et al. Results of two open-lael multicenter pilot phase II trails with Herceptin® in combination with docetaxel and platinum salts (cis- or carboplatinum) (TCH) as therapy for advanced breast cancer in women overexpressing HER2. Breast Cancer Res Treat 2000: 64; 327abs.

108. Slamon DJ, Patel R, Northfelt M, et al. Phase II Pilot Study of Herceptin Combined with Taxotere and Carboplatin (TCH) in Metastatic Breast Cancer (MBC) Patients Overexpressing the HER2 Proto-Oncogene. A Pilot Study of the UCLA Network. Proc Am Soc Clin Onc 2001: 20: 49a (abst 193).

109. Slamon D, Leyland-Jones B, Shak S, et al. Use of chemotherapy plus a monoclonal antibody against Her2 for metastatic breast cancer that overexpresses HER2. N Engl J Med 2001; 344: 783 – 92.

110. Gamucci T, Piccart M, Brunning P et al. Single agent Taxol®versus doxorubicin as first- and second-line chemotherapy in advanced breast cancer: final results of an EORTC randomized study with crossover. Proc Am Soc Clin Onc 1998; 17:428a

111. Sledge GW, Neuberg D, Ingle J et al. Phase III trial of doxorubicin versus paclitaxel versus doxorubicin + paclitaxel as first-line therapy for metastatic breast cancer; an intergroup trial. Am Soc Clin Onc 1997; 16 1a (abst 2)

112. Nabholtz JM, Falkson C, Campos D et al. A phase III trial comparing doxorubicin and docetaxel to doxorubicin and cyclophosphamide as first line chemotherapy for MBC. Proc Am Soc Clin Onc 1999; 18: 485a

113. L Biganzoli, Cufer T, Bruning R et al. Doxorubicin / Taxol® versus Doxorubicin / Cyclophosphamide as First-Line Chemotherapy in Metastatic Breast Cancer (MBC); A phase III Study. Proc Am Soc Clin Onc 2000: 19: 282a

114. Pauletti, G, Dandekar, S., Rong, H-M, Ramos, L, Peng,H-J M.D., Seshadri, R,. Slamon, D.J. Assessment of Methods for Tissue Based Detection of the HER2 Alteration in Human Breast Cancer: A direct Comparison of FISH and Immunhostochemistry. J Clin.Onc 18: 3651-3664, 2000

115. Goldhirsch A, Glick JH, Gelber R et al. Meeting Highlights: International Consensus Panel on the Treatment of Primary Breast Cancer. J NCI; 1998; vol 90: 21;1601-1608.

116. Pauletti, G., Godolphin, W., Press, M.F., Slamon, D.J. Detection and Quantitation of HER2 Gene Amplification in Human Breast Cancer Archival Material Using Fluorescence *In Situ* Hybridization, Oncogene 13, 63-72. 1996

117. Mass RD, Sanders C, Charlene K et al. The Concordance between the Clinical Trials Assay (CTA) and Fluorescence in situ hybridization in the Herceptin®^ Pivotal Trials. Proc Am Soc Clin Onc 2000; 29 291a.

118. Liggett SB, Wagoner LE, Craft LL, Hornung RW, Hoit BD, McIntosh TC, Walsh RA: The Ile164 beta2-adrenergic receptor polymorphism adversely affects the outcome of congestive heart failure. *J.Clin.Invest.* 1998;102:1534-1539

119. Vancura V, Hubacek J, Malek I, Gebauerova M, Pitha J, Dorazilova Z, Langova M, Zelizko M, Poledne R: Does angiotensin-converting enzyme polymorphism influence the clinical manifestation and progression of heart failure in patients with dilated cardiomyopathy? *Am.J.Cardiol.* 1999;83:461-462.

120. Charron P, Tesson F, Poirier O, Nicaud V, Peuchmaurd M, Tiret L, Cambien F, Amouyel P, Dubourg O, Bouhour J, Millaire A, Juilliere Y, Bareiss P, Andre-Fouet X, Pouillart F, Arveiler D, Ferrieres J, Dorent R, Roizes G, Schwartz K, Desnos M, Komajda M: Identification of a genetic risk factor for idiopathic dilated cardiomyopathy. Involvement of a polymorphism in the endothelin receptor type A gene. CARDIGENE group. *Eur.Heart J.* 1999;20:1587-1591

121. Xie D, Shu XO, Deng Z, Wen WQ, Creek KE, Dai Q, Gao YT, Jin F, Zheng W: Population-based, case-control study of HER2 genetic polymorphism and breast cancer risk. *J.Natl.Cancer Inst.* 2000;92:412.-417.

122. Wagoner LE, Craft LL, Singh B, Suresh DP, Zengel PW, McGuire N, Abraham WT, Chenier TC, Dorn GW, Liggett SB: Polymorphisms of the beta(2)-adrenergic receptor determine exercise capacity in patients with heart failure. *Circ.Res.* 2000;86:834-840

123. Cowie MR, Struthers AD, Wood DA, Coats AJ, Thompson SG, P.A., Sutton GC: Value of natriuretic peptides in assessment of patients with possible new heart failure in primary care. *Lancet* 1997;350:1349-1353

124. Fox KF, Cowie MR, Wood DA, Coats AJ, P.A., Sutton GC: A Rapid Access Heart Failure Clinic provides a prompt diagnosis and appropriate management of new heart failure presenting in the community. *Eur.J.Heart Fail.* 2000;2:423-429

125. Suzuki T, Hayashi D, Yamazaki T, Mizuno T, Kanda Y, Komuro I, Kurabayashi M, Yamaoki K, Mitani K, Hirai H, Nagai R, Yazaki Y: Elevated B-type natriuretic peptide levels after anthracycline administration. *Am.Heart J.* 1998;136:362-363

126. Nousiainen T, Jantunen E, Vanninen E, Remes J, Puustinen J, Rantala A, Vuolteenaho O, Hartikainen J: Acute neurohumoral and cardiovascular effects of idarubicin in leukemia patients. *Eur.J.Haematol.* 1998;61:347-353

127. Del Carlo CH, O'Connor CM: Cardiac troponins in congestive heart failure. *Am.Heart J.* 1999;138:646-653

128. Lipshultz SE, Rifai N, Sallan SE, Lipsitz SR, Dalton V, Sacks DB, Ottlinger ME: Predictive value of cardiac troponin T in pediatric patients at risk for myocardial injury. *Circulation* 1997;96:2641-2648

129. Herman EH, Zhang J, Lipshultz SE, Rifai N, Chadwick D, Takeda K, Yu ZX, Ferrans VJ: Correlation between serum levels of cardiac troponin-T and the severity of the chronic cardiomyopathy induced by doxorubicin. *J.Clin.Oncol.* 1999;17:2237-2243

130. Cardinale D, Sandri MT, Martinoni A, Tricca A, Civelli M, Lamantia G, Cinieri S, Martinelli G, Cipolla CM, Fiorentini C: Left ventricular dysfunction predicted by early troponin I release after high-dose chemotherapy. *J.Am.Coll.Cardiol.* 2000;36:517-522

APPENDIX 1 - DECLARATION OF HELSINKI

WORLD MEDICAL ASSOCIATION DECLARATION OF HELSINKI
Ethical Principles for Medical Research Involving Human Subjects

Adopted by the 18th WMA General Assembly Helsinki, Finland, June 1964 and amended by the
29th WMA General Assembly, Tokyo, Japan, October 1975
35th WMA General Assembly, Venice, Italy, October 1983
41st WMA General Assembly, Hong Kong, September 1989
48th WMA General Assembly, Somerset West, Republic of South Africa, October 1996
and the 52nd WMA General Assembly, Edinburgh, Scotland, October 2000
Note of clarification on Paragraph 29 added by the WMA General Assembly, Washington 2002
Note of clarification on Paragraph 30 added by the WMA General Assembly, Tokyo 2004

A. INTRODUCTION

1. The World Medical Association has developed the Declaration of Helsinki as a statement of ethical principles to provide guidance to physicians and other participants in medical research involving human subjects**.** Medical research involving human subjects includes research on identifiable human material or identifiable data.

2. It is the duty of the physician to promote and safeguard the health of the people. The physician's knowledge and conscience are dedicated to the fulfillment of this duty.

3. The Declaration of Geneva of the World Medical Association binds the physician with the words, "The health of my patient will be my first consideration," and the International Code of Medical Ethics declares that, "A physician shall act only in the patient's interest when providing medical care which might have the effect of weakening the physical and mental condition of the patient."

4. Medical progress is based on research which ultimately must rest in part on experimentation involving human subjects.

5. In medical research on human subjects, considerations related to the well-being of the human subject should take precedence over the interests of science and society.

6. The primary purpose of medical research involving human subjects is to improve prophylactic, diagnostic and therapeutic procedures and the understanding of the aetiology and pathogenesis of disease. Even the best proven prophylactic, diagnostic, and therapeutic methods must continuously be challenged through research for their effectiveness, efficiency, accessibility and quality.

7. In current medical practice and in medical research, most prophylactic, diagnostic and therapeutic procedures involve risks and burdens.

8. Medical research is subject to ethical standards that promote respect for all human beings and protect their health and rights. Some research populations are vulnerable and need special protection. The particular needs of the economically and medically disadvantaged must be recognized. Special attention is also required for those who cannot give or refuse consent for themselves, for those who may be subject to giving consent under duress, for those who will not benefit personally from the research and for those for whom the research is combined with care.

9. Research Investigators should be aware of the ethical, legal and regulatory requirements for research on human subjects in their own countries as well as applicable international requirements. No national ethical, legal or regulatory requirement should be allowed to reduce or eliminate any of the protections for human subjects set forth in this Declaration.

B. BASIC PRINCIPLES FOR ALL MEDICAL RESEARCH

10. It is the duty of the physician in medical research to protect the life, health, privacy, and dignity of the human subject.

11. Medical research involving human subjects must conform to generally accepted scientific principles, be based on a

thorough knowledge of the scientific literature, other relevant sources of information, and on adequate laboratory and, where appropriate, animal experimentation.

12. Appropriate caution must be exercised in the conduct of research which may affect the environment, and the welfare of animals used for research must be respected.

13. The design and performance of each experimental procedure involving human subjects should be clearly formulated in an experimental protocol. This protocol should be submitted for consideration, comment, guidance, and where appropriate, approval to a specially appointed ethical review committee, which must be independent of the investigator, the sponsor or any other kind of undue influence. This independent committee should be in conformity with the laws and regulations of the country in which the research experiment is performed. The committee has the right to monitor ongoing trials. The researcher has the obligation to provide monitoring information to the committee, especially any serious adverse events. The researcher should also submit to the committee, for review, information regarding funding, sponsors, institutional affiliations, other potential conflicts of interest and incentives for subjects.

14. The research protocol should always contain a statement of the ethical considerations involved and should indicate that there is compliance with the principles enunciated in this Declaration.

15. Medical research involving human subjects should be conducted only by scientifically qualified persons and under the supervision of a clinically competent medical person. The responsibility for the human subject must always rest with a medically qualified person and never rest on the subject of the research, even though the subject has given consent.

16. Every medical research project involving human subjects should be preceded by careful assessment of predictable risks and burdens in comparison with foreseeable benefits to the subject or to others. This does not preclude the participation of healthy volunteers in medical research. The design of all studies should be publicly available.

17. Physicians should abstain from engaging in research projects involving human subjects unless they are confident that the risks involved have been adequately assessed and can be satisfactorily managed. Physicians should cease any investigation if the risks are found to outweigh the potential benefits or if there is conclusive proof of positive and beneficial results.

18. Medical research involving human subjects should only be conducted if the importance of the objective outweighs the inherent risks and burdens to the subject. This is especially important when the human subjects are healthy volunteers.

19. Medical research is only justified if there is a reasonable likelihood that the populations in which the research is carried out stand to benefit from the results of the research.

20. The subjects must be volunteers and informed participants in the research project.

21. The right of research subjects to safeguard their integrity must always be respected. Every precaution should be taken to respect the privacy of the subject, the confidentiality of the patient's information and to minimize the impact of the study on the subject's physical and mental integrity and on the personality of the subject.

22. In any research on human beings, each potential subject must be adequately informed of the aims, methods, sources of funding, any possible conflicts of interest, institutional affiliations of the researcher, the anticipated benefits and potential risks of the study and the discomfort it may entail. The subject should be informed of the right to abstain from participation in the study or to withdraw consent to participate at any time without reprisal. After ensuring that the subject has understood the information, the physician should then obtain the subject's freely given informed consent, preferably in writing. If the consent cannot be obtained in writing, the non-written consent must be formally documented and witnessed.

23. When obtaining informed consent for the research project the physician should be particularly cautious if the subject is in a dependent relationship with the physician or may consent under duress. In that case the informed consent should be obtained by a well-informed physician who is not engaged in the investigation and who is completely independent of this relationship.

24. For a research subject who is legally incompetent, physically or mentally incapable of giving consent or is a legally

incompetent minor, the investigator must obtain informed consent from the legally authorized representative in accordance with applicable law. These groups should not be included in research unless the research is necessary to promote the health of the population represented and this research cannot instead be performed on legally competent persons.

25. When a subject deemed legally incompetent, such as a minor child, is able to give assent to decisions about participation in research, the investigator must obtain that assent in addition to the consent of the legally authorized representative.

26. Research on individuals from whom it is not possible to obtain consent, including proxy or advance consent, should be done only if the physical/mental condition that prevents obtaining informed consent is a necessary characteristic of the research population. The specific reasons for involving research subjects with a condition that renders them unable to give informed consent should be stated in the experimental protocol for consideration and approval of the review committee. The protocol should state that consent to remain in the research should be obtained as soon as possible from the individual or a legally authorized surrogate.

27. Both authors and publishers have ethical obligations. In publication of the results of research, the investigators are obliged to preserve the accuracy of the results. Negative as well as positive results should be published or otherwise publicly available. Sources of funding, institutional affiliations and any possible conflicts of interest should be declared in the publication. Reports of experimentation not in accordance with the principles laid down in this Declaration should not be accepted for publication.

C. ADDITIONAL PRINCIPLES FOR MEDICAL RESEARCH COMBINED WITH MEDICAL CARE

28. The physician may combine medical research with medical care, only to the extent that the research is justified by its potential prophylactic, diagnostic or therapeutic value. When medical research is combined with medical care, additional standards apply to protect the patients who are research subjects.

29. The benefits, risks, burdens and effectiveness of a new method should be tested against those of the best current prophylactic, diagnostic, and therapeutic methods. This does not exclude the use of placebo, or no treatment, in studies where no proven prophylactic, diagnostic or therapeutic method exists.[1]

30. At the conclusion of the study, every patient entered into the study should be assured of access to the best-proven prophylactic, diagnostic and therapeutic methods identified by the study.[2]

31. The physician should fully inform the patient which aspects of the care are related to the research. The refusal of a patient to participate in a study must never interfere with the patient-physician relationship.

32. In the treatment of a patient, where proven prophylactic, diagnostic and therapeutic methods do not exist or have been ineffective, the physician, with informed consent from the patient, must be free to use unproven or new prophylactic, diagnostic and therapeutic measures, if in the physician's judgement it offers hope of saving life, re-establishing health or alleviating suffering. Where possible, these measures should be made the object of research, designed to evaluate their safety and efficacy. In all cases, new information should be recorded and, where appropriate, published. The other relevant guidelines of this Declaration should be followed.

[1]Note of clarification on paragraph 29 of the WMA Declaration of Helsinki

The WMA herby reaffirms its position that extreme care must be taken in making use of a placebo- controlled trial and that in general the methodology should only be used in the absence of existing proven therapy. However, a placebo-controlled trial may be ethically acceptable, even if proven therapy is available, under the following circumstances:

- Where for compelling and scientifically sound methodological reasons its use is necessary to determine the efficacy or safety of a prophylactic, diagnostic or therapeutic method; or

-Where a prophylactic, diagnostic or therapeutic method is being investigated for a minor condition and the patients who receive placebo will not be subject to any additional risk of serious or irreversible harm.

All other provisions of the Declaration of Helsinki must be adhered to, especially the need for appropriate ethical and scientific review.

[2]Note of clarification on paragraph 30 of the WMA Declaration of Helsinki

The WMA herby reaffirms its position that it is necessary during the study planning process to identify post-trial access by study participants to prophylactic, diagnostic and therapeutic procedures identified as beneficial in the study or access to other appropriate care. Post-trial access arrangements or other care must be described in the study protocol so the ethical review committee may consider such arrangement during its review.
09.10.2004

APPENDIX 2 - KARNOFSKY INDEX FOR PERFORMANCE STATUS

100	Normal, no complaints: no evidence of disease.
90	Able to carry on normal activity; minor signs or symptoms of disease.
80	Normal activity with effort, some signs or symptoms of disease.
70	Cares for self but unable to carry on normal activity or to do work.
60	Requires occasional assistance but is able to care for most of personal needs.
50	Requires considerable assistance and frequent medical care.
40	Disabled; requires special care and assistance.
30	Severely disabled; hospitalization is indicated although death not imminent.
20	Very ill; hospitalization and active supportive care necessary.
10	Moribund.
0	Dead.

APPENDIX 3 - PATHOLOGY REVIEW AND MOLECULAR MARKER TESTING

APPENDIX 3A - HER2 FISH TESTING; PATHOLOGY AND MOLECULAR MARKER REVIEW (MANDATORY)

1. HER2 FISH Testing

Human Epidermal Growth Factor Receptor 2 (HER2)

Abnormal expression of human epidermal growth factor receptor 2 (HER2) is frequently observed in a number of primary tumors, suggesting that the over expression of this growth factor receptor may contribute to transformation and tumorigenesis. In most cases, HER2 protein over expression is thought to result from gene amplification and has been correlated with poor clinical outcome in patients with breast and ovarian cancers that over express HER2. Approximately 25% to 30% of patients with breast and ovarian cancers over express HER2.

The BCIRG 006 study requires, as one of the patient eligibility criteria, HER2 amplification which represents ¼ to 1/3 of the breast cancer patient population. Approximately 12,000 patients are expected to be screened in order to identify 3,150 patients whose breast cancer amplifies the HER2 gene (BCIRG 006).
A representative paraffin block from potentially eligible women with breast cancer will be tested for c-erbB-2 status using Fluorescence In-Situ Hybridization (Vysis kit) in one of the BCIRG designated central laboratories.

Dr. Michael Press, University of Southern California, Los Angeles, U.S.A. will be responsible for the HER2 screening procedure. The two BCIRG central laboratories are as follows:

For Sites in North, South America, Australia
Dr. Michael F. Press
University of Southern California
Norris Cancer Centre
Suite # 5409 1441 Eastlake Avenue,
Los Angeles, CA, 90033
Telephone: (323) 865-0563

For All Other Countries
Dr. Guido Sauter
Institute of Pathology
University of Basel
Schonbeinstrasse 40
CH-4003 Basel, Switzerland
Telephone: (41 61 265 28 89)

1.1 Procedure:

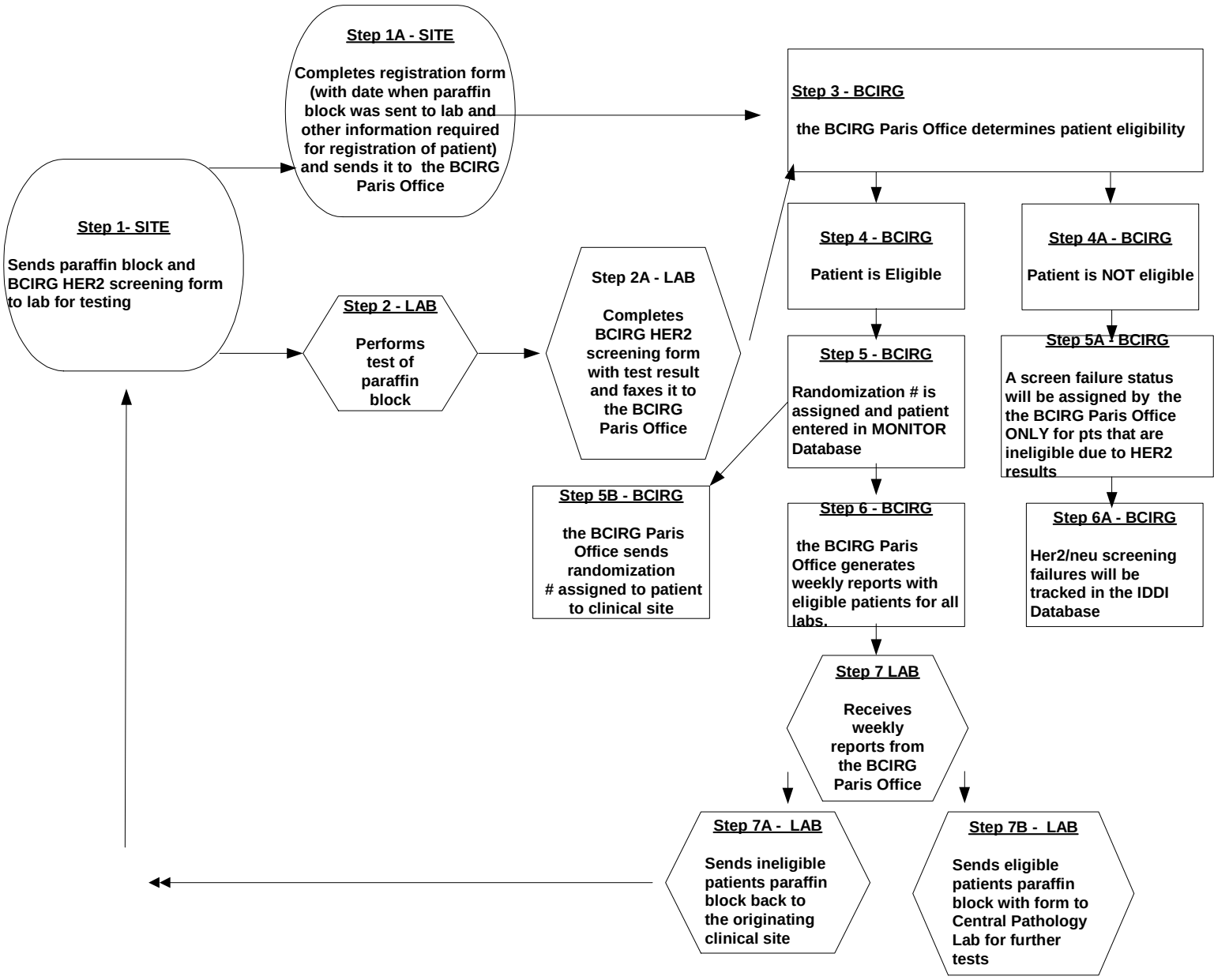
All patients identified by the participating center as potentially eligible will start the study screening procedures as per study protocol.
A representative tumor block will be sent by the clinical site to one of the BCIRG designated labs. The block will be sent accompanied by a HER2 screening form provided by BCIRG to the clinical site. This form will include a patient identifier such as patient initials, patient's date of birth and clinical site number.
The BCIRG laboratories will perform the *c-erb*B-2 analysis using the Vysis FISH kit and report the result to the BCIRG Office. The lab will then be informed by the BCIRG Paris if the patient was eligible and will either send the block back to the originating centre (if patient is not accrued to a BCIRG trial) or to the BCIRG designated Central Pathology laboratory (if the patient is accrued to the BCIRG trial).
See Appendix 3A.

1.2 Operating Principles for the Designated Labs:

1. Use of the Vysis kit (which will be supplied by the BCIRG).
2. Quality control and standardized procedure as determined by Dr. Michael Press.
3. Turn-around time of 5 working days for faxing results to the BCIRG Paris.
4. The disposal procedure for the blocks is as follows:
   - all cases not accrued to a BCIRG trial (as informed by the BCIRG Paris) should have their blocks returned directly to the contributing centre.
   - all cases enrolled post HER2 screening for BCIRG trials will have their blocks forwarded to the BCIRG designated Central Pathology Laboratory for molecular studies

Flow chart of HER2 screening and central pathology

BCIRG HER2 Flowchart



2.Pathology and Marker Review

The purpose of this investigation is to establish the baseline characteristics of the tumors and ensure comparability between the experimental arms. Each tumor will be tested for:

i. a number of accepted histo-pathologic and molecular marker prognostic factors and
ii. several other factors with proven utility in predicting response to Taxotere.

Due to the well described difficulty in obtaining inter-observer reproducibility in the assessment of pathologic factors, the assessments of these prognostic and predictive factors will be performed by a central lab.

Methodology

Hematoxylin and Eosin stained slides prepared from the paraffin block submitted for FISH testing will be assessed for Histologic subtype, grade and vascular invasion. Additional unstained slides from the same block will be assayed for *ER, PR, p53* and *MIB-1* using automated immunohistochemistry at the BCIRG designated Central Pathology Laboratory. Histopathologic and immunohistochemical assessment of these factors will be performed by a single reference pathologist with external review of 30% of the material by a second observer. If indicated, the investigation of the *Bcl* family (Bcl-2, bax, Bcl-X and Bag-1) will be done in collaboration with Dr. J. C. Reed MD, Ph.D., Scientific Director, The Burnham Institute (formerly, The La Jolla Cancer research Foundation), La Jolla, California. If indicated, the investigation of the tubulin isoforms (II, III, IV and Tau) will be done in collaboration with Drs. C. Dumontet and I. Treilleux in Lyon, France. If indicated, the investigation of MUC1 will also be performed at the Central Pathology Laboratory.

The pathology materials which must be supplied are:

1. one paraffin block from a representative area of the tumor. If insufficient material remains in this original block, the center will be contacted for an alternate block. The blocks will be kept in the central registry for the duration of the trial. They can be accessed during this period by the original lab. The block may be returned to the original pathology lab at the close of the trial if desired so by the patient. Blocks will otherwise be stored in a tumour bank for future investigations.
2. a copy of the pathology report for the specimen from which the block in part I is derived.

APPENDIX 3B – SUMMARY OF OPTIONAL ECD AND CARDIAC SUBSTUDIES PROCESS

	ECD substudy	**Biochemichal markers** *(not applicable for France)*	**Genetic markers** *(not applicable for France)*
Time of collection	-Baseline, EOC or FUp1a -Every 6 months during the first 5 years of follow-up -And at time of relapse	Baseline, cycle 4, EOC or FUp1a Every 6 months during the first 3 years of follow-up, At year 5 of follow-up At any clinical evidence of cardiac failure	Baseline
Number of time	12 and 1 for 50% of patients at relapse	9	1
Number of tubes/collection	2	2	1
Type of tubes	Red top tubes	Purple top tubes	Purple top tubes
Material	Serum	Plasma	Whole blood
Anticoagulants	None	EDTA	EDTA or Heparin
Storage/Shipment time	-20 during 4 months or -80 during 6 months	-20 during 4 months or -80 during 6 months	Fridge for a maximum of 1 month
Condition of shipment	Dry ice	Dry ice	Dry ice (Sample is to be placed into an insulating material placed itself in the Styrofoam lined shipping box containing dry ice)

APPENDIX 3C - SHED HER2 ECD SUBSTUDY (OPTIONAL)

Collection, storage and shipping instructions for serum for shed extracellular domain (ECD) of HER2 and serum bank specimen

This optional study will also ask for a serum sample to be provided to the central lab prior to study start, at the end of chemotherapy visit for AC→ TH and AC→ T arms and at follow-up visit 1a (6 weeks after End of Chemotherapy) for arm TCH, every 6 months during the five first years of follow-upand at the time of relapse. Serum assays for HER2 are currently being investigated. We wish to collect serum samples prior to treatment in order to review the levels of shed extracellular domain (ECD) of HER2 in peripheral blood for comparison with the FISH test as a predictive factor in a patient's outcome with Herceptin®-containing regimens. We will also ask that a serum/ sample be sent to our lab at the time of disease relapse for comparison of levels. Additional testing of molecular markers and assays for HER2 may be performed on the serum sample in the future. Refusal to grant permission for collection and release of the serum samples for the above mentioned purpose would not affect the quality of care the participant is to receive.

1 Procedures:

1. Specimens will be collected at baseline, at End of Chemotherapy for AC→ TH and AC→ T arms and at follow-up visit 1a (6 weeks after End of Chemotherapy) for arm TCH and then every 6 months during the first 5 years of follow-up.and at the time of recurrence.
2. Please note the expiry date on the kit box. The expiration date on the kit box corresponds to the expiry date on the red top tube. The red top tube tends to loose its vacuum after this date. It is safe to use the tube after its expiry date, however, it may not fill to the desired volume, resulting in the need to collect multiple tubes in order to obtain the required volume of serum.
3. Obtain the 2 10mL red top tubes (with serum separator gel) to draw the blood specimens.
4. Label the red top tubes with the patient identification labels provided by BCIRG in the kit box. (BCIRG patient number, patient initials, and date and time sample is collected).
5. Label the plastic transport vial with the patient identification labels provided by BCIRG in the kit box. (BCIRG patient number, patient initials, and date and time sample is collected) Secure these labels onto the tubes with transparent tape, by wrapping the tape once around the label. This will prevent the label from falling off in the freezer.
6. Attach a label containing the patient BCIRG information (patient number, patient initials, date and time sample is collected) to the "Serum Collection Form" (Please refer to next page for an example).
7. Draw the blood from the patient into the 2 10mL red top tubes. Allow the tube to fill completely.
8. Let sample sit for 20 minutes at room temperature. This allows the sample to clot.
9. Centrifuge the sample at 1000 – 1200 g for 15 minutes. Ensure the centrifuge is balanced.
10. Carefully remove stopper from the red top tube.
11. Using a pipette, aliquot the serum into the plastic transport vial. Vial should contain 2 – 7 mL of serum.
12. Ensure that the lid on the transport vial is secure.
13. Discard the red top tube containing residual serum and cells into a biohazard waste container.
14. Place the transport vial into the biohazard bag (ziplock bag). Please note this bag has an absorbent sheet that must remain in the bag. This absorbent sheet will absorb any serum that leaks from the transport vial (**Please note - The most common cause for leakage is unsecured caps on the transport vials**).
15. Ensure that the "Serum Collection Form" is completed and placed in the side pouch of the biohazard bag.
16. Place the biohazard bag containing the transport vial in the freezer, until read to ship to the central laboratory (see address hereafter).
17. When you are ready to ship the samples, remove the biohazard bag containing the transport vial from the freezer.
18. Fill half of the Styrofoam lined shipping box with dry ice. Place the biohazard bag containing transport vials and the "Serum Collection Form" on the dry ice. Fill the rest of the Styrofoam lined shipping box with the remaining dry ice.
19. Place the Styrofoam lid securely on the Styrofoam lined shipping box. Do not secure this lid with tape. The dry ice needs to sublimate.
20. Close the corrugated cardboard lid of the shipping box and secure this with packing tape.

21. Attach the shipping label (Please note – dry ice is considered a dangerous goods shipment) and ship this box via the deisgnated courier company to:

For all sites :
Lilian RAMOS
UCLA
Dpt of Medicine – Dr Slamon's Research Laboratory
675 Charles E.Young Drive South, Room 5/535, Building MRL
Los Angeles, CA 90095
USA
Phone: 1 310 206 1408 / Fax: 1 310 825 3761

2 .Serum sample supplies

You will be provided with a serum sample kit box (1 kit box required for 1 sample collected) and an additional collection of extra supplies (1 extra collection of supplies will be provided for every 3 samples collected).

Serum Collection Forms will be provided to you at the time of the study initiation visit.

If supplies are needed, please fill out the BCIRG 006 Study Supply Order Form. If you have any questions regarding the supplies, please contact your BCIRG study monitor, or the BCIRG general office number at 780 702 0200.

Each Kit Box includes:

Two - 10 mL evacuated red-top vacutainer tube
Plastic Transport Vial
Pipette
Alcohol, cotton swabs, sterilized plastic strip
21-gauge vacutainer needle, 1-1/2 " (multiple-sampling)

Patient Identification Labels
Biohazard Ziplock Bag (with absorbent sheet inside)

Bag of Extra Collection Supplies includes:

10 mL evacuated red-top vacutainer tube
Plastic Transport Vial
Pipette
Alcohol, cotton swabs, sterilized plastic strip
21-gauge vacutainer needle , 1-1/2 " (multiple-sampling)
Biohazard Ziplock Bag (with absorbent sheet inside)
Tourniquet
Yellow Vacutainer Holder

Also Supplied:

One Styrofoam Lined Shipping Box

Site (or Lab) Must Have:

Disposable gloves
Refrigerator, Freezer
Centrifuge capable of accommodating 10-mL vacutainer tubes

Needle disposal Container
Biohazard Container
Packing Material (for shipment of sample)
Packing Tape

Appendix 3D - CARDIAC BIOCHEMICAL SUBSTUDIES (optional) – *(not applicable for France)*

Collection, storage and shipping instructions for plasma samples for cardiac biochemical markers

This optional study will also ask for a plasma sample to be provided to the central lab prior to study start and specified time intervals outlined in the first step of this procedure. Plasma assays for Cardiac Biochemical Markers, BNP and Troponin T, are currently being investigated. We wish to collect plasma samples prior to treatment in order to review the levels BNP and Troponin T in peripheral blood as a predictive factor in a patient's cardiac health. We will also ask that a plasma sample be sent to our lab at the defined intervals as a comparison of levels. Refusal to grant permission for collection and release of the blood samples for the above-mentioned purpose, will not affect the quality of care the participant is to receive.

1 Procedures:

1. Specimens will be collected at Baseline, Cycle 4, End of Chemotherapy for AC→ TH and AC→ T arms and at follow-up visit 1a (6 weeks after End of Chemotherapy for arm TCH), every 6 months for the first 3 years of follow-up and at any time of clinical evidence of cardiac dysfunction
2. Please note the expiry date on the kit box. The expiry date on the kit box corresponds to the expiry date of the EDTA (anti-coagulant) in the purple top tube. Please do not use an EDTA, purple top tube past it's expiry date.
3. Obtain the two 5mL (EDTA) purple top tubes to draw the plasma specimens.
4. Label the purple top tubes with the patient identification labels provided by BCIRG in the kit box. (BCIRG patient number, patient initials, and date and time sample is collected).
5. Label the plastic transport vials with the patient identification labels provided by BCIRG in the kit box. (BCIRG patient number, patient initials, and date and time sample is collected) Secure these labels onto the tubes with transparent tape, by wrapping the tape once around the label. This will prevent the label from falling off in the freezer.
6. Attach a label containing the patient BCIRG information (patient number, patient initials, date and time sample is collected) to the "Plasma Collection Form" (Please refer to next page for an example).
7. Draw the blood from the patient into the two 5mL purple top tubes. Allow the tube to fill completely.
8. Gently invert the purple top tubes 4 to 5 times, to allow the EDTA (anti-coagulant) and blood to mix. Do not vigorously shake the tubes. This is cause the red cells to lyse and will result in a hemolytic sample.
9. Centrifuge the sample at 1000 – 1200 g for 15 minutes. Ensure the centrifuge is balanced.
10. Carefully remove stopper from the purple top tubes.
11. Using a pipette, aliquot the plasma into the plastic transport vials. Vials should contain 1 – 3 mL of plasma. (If there is no cell contamination, proceed to step 12) When transferring the sample ensure that you do not shake or disturb the packed red cells with the pipette. This will cause red cell contamination of your plasma sample. If the cells have contaminated the plasma, securely recap the sample with the purple stopper and centrifuge the sample once more (Return to step 9).
12. Ensure that the lids on the transport vials are secure.
13. Discard the purple top tube containing residual plasma and cells into a biohazard waste container.
14. Place the transport vial into the biohazard bag (ziplock bag). Please note this bag has an absorbent sheet that must remain in the bag. This absorbent sheet will absorb any serum that leaks from the transport vial (**Please note - The most common cause for leakage is unsecured caps on the transport vials**).
15. Ensure that the "Plasma Collection Form" is completed and placed in the side pouch of the biohazard bag.
16. Place the biohazard bag containing the transport vials in a freezer, until ready to ship to the central laboratory (see address hereafter).
17. When you are ready to ship the samples, remove the biohazard bag containing the transport vial from the freezer.
18. Fill half of the Styrofoam lined shipping box with dry ice. Place the biohazard bag containing transport vials and the "Blood Collection Form" on the dry ice. Fill the rest of the Styrofoam lined shipping box with the remaining dry ice.
19. Place the Styrofoam lid securely on the Styrofoam lined shipping box. Do not secure this lid with tape. The dry ice needs to sublimate.
20. Close the corrugated cardboard lid of the shipping box and secure the cardboard lid with packing tape.
21. Attach the shipping label (Please note – dry ice is considered a dangerous goods shipment) and ship this box via the designated courier company to:

For all sites :

Lilian RAMOS
UCLA
Dpt of Medicine – Dr Slamon's Research Laboratory
675 Charles E.Young Drive South, Room 5/535, Building MRL
Los Angeles, CA 90095
USA
Phone: 1 310 206 1408 / Fax: 1 310 825 3761

2 Plasma sample supplies

You will be provided with a plasma sample kit box (1 kit box required for 1 sample collected) and an additional collection of extra supplies (1 extra collection of supplies will be provided for every 3 samples collected).

Plasma Collection Forms will be provided to you at the time of the study initiation visit.

If supplies are needed, please fill out the BCIRG 006 Study Supply Order Form. If you have any questions regarding the supplies, please contact your BCIRG study monitor, or the BCIRG general office number at 780 702 0200.

Each Kit Box includes:

Two - 5 mL (EDTA) purple-top vacutainer tube
Two Plastic Transport Vial with purple stripe
Pipette

Alcohol, cotton swabs, sterilized plastic strip
21-gauge vacutainer needle, 1-1/2 " (multiple-sampling)

Patient Identification Labels
Biohazard Ziplock Bag (with absorbent sheet inside)

Bag of Extra Collection Supplies includes:

Two - 5 mL (EDTA) purple-top vacutainer tube
Plastic Transport Vial
Pipette
Alcohol, cotton swabs, sterilized plastic strip
21-gauge vacutainer needle, 1-1/2 " (multiple-sampling)
Biohazard Ziplock Bag (with absorbent sheet inside)
Tourniquet
Yellow Vacutainer Holder

Also Supplied:
One Styrofoam Lined Shipping Box

Site (or Lab) Must Have:
Disposable gloves
Refrigerator, Freezer -20 or -80ºC
Centrifuge capable of accommodating 5-mL vacutainer tubes
Needle disposal Container
Biohazard Container
Packing Material (for shipment of sample)
Packing Tape
Dry Ice for shipping

Appendix 3E - CARDIAC GENETIC SUBSTUDY (optional) – *(not applicable for France)*

Collection, storage and shipping instructions for whole blood for the cardiac genetic markers

This optional study will also ask for a whole blood sample to be provided to the central lab prior to study start at Baseline only. Whole blood assays for Cardiac Genetic Markers, to identify patients susceptible to Herceptin®-induced LV dysfunction is currently being investigated. We wish to collect a whole blood prior to treatment in order to review the levels of ACE, 2-adrenergic receptor, and the Endothelin A receptor in blood cells as a predictive factor in a patient's cardiac health. Refusal to grant permission for collection and release of the blood samples for the above-mentioned purpose, will not affect the quality of care the participant is to receive.

1 Procedure for Baseline Collection:

1. Specimens will be collected at Baseline.
2. Please note the expiry date on the kit box. The expiry date on the kit box corresponds to the expiry date on the EDTA (anticoagulant) in the purple top tube. Please do not use an EDTA, purple top tube past its expiry date.
3. Obtain the one 5mL (EDTA) purple top tubes to draw the blood specimen.
4. Label the purple top tube with the patient identification labels provided by BCIRG in the kit box (BCIRG patient number, patient initials, and date and time sample is collected).
5. Attach a label containing the patient BCIRG information (patient number, patient initials, date and time sample is collected) to the "Blood Collection Form" (Please refer to next page for an example)
6. Draw the blood from the patient into the 5mL purple top tube. Allow the tube to fill completely.
7. Gently invert the purple top tube 4 to 5 times, to allow the EDTA (anti-coagulant) and blood to mix. Do not vigorously shake the tubes. This is cause the red cells to lyse and will result in a hemolytic sample.
8. ***Do not centrifuge this purple top tube**.
9. Place this purple top EDTA tube, into the bubble pack provided.
10. Place the sample into an insulating material (such as cardboard, polystyrene…..)
11. Place the bubble pack containing the purple top EDTA tube into the biohazard bag (ziplock bag). Please note this bag has an absorbent sheet that must remain in the bag. This absorbent sheet will absorb any blood that leaks from the tube.
12. Ensure that the "Blood Collection Form" is completed and placed in the side pouch of the biohazard bag.
13. Fill half of the Styrofoam lined shipping box with dry ice. Place the biohazard bag containing the sample and its insulating material and the "Blood Collection Form" on the dry ice. Fill the rest of the Styrofoam lined shipping box with the remaining dry ice.
14. Close the kit box and secure it with packing tape to ensure nothing falls out.
15. Ship this box via the designqted courier company within 1 month after the blood collection to:

For all sites
Lilian RAMOS
UCLA
Dpt of Medicine – Dr Slamon's Research Laboratory
675 Charles E.Young Drive South, Room 5/535, Building MRL
Los Angeles, CA 90095
USA
Phone: 1 310 206 1408 / Fax: 1 310 825 3761

2 Whole Blood sample supplies

You will be provided with a blood sample kit box (1 kit box required for 1 sample collected) and an additional collection of extra supplies (1 extra collection of supplies will be provided for every 3 samples collected).

Blood Collection Forms will be provided to you at the time of the study initiation visit.

If supplies are needed, please fill out the BCIRG 006 Study Supply Order Form. If you have any questions regarding the supplies, please contact your BCIRG study monitor, or the BCIRG general office number at 780 702 0200.

Each Kit Box includes:

One - 5 mL EDTA purple-top vacutainer tube
Elastic Band

Alcohol, cotton swabs, sterilized plastic strip
21-gauge vacutainer needle , 1-1/2 " (multiple-sampling)

Patient Identification Labels
Bubble Pack
U Tek Refrigerant Pack
Biohazard Ziplock Bag (with absorbent sheet inside)

Bag of Extra Collection Supplies includes:

5 mL EDTA purple-top vacutainer tube
Elastic Band
Alcohol, cotton swabs, sterilized plastic strip
21-gauge vacutainer needle, 1-1/2 " (multiple-sampling)
Biohazard Ziplock Bag (with absorbent sheet inside)
Bubble Pack
Tourniquet
Yellow Vacutainer Holder

Site (or Lab) Must Have:

Disposable gloves
Refrigerator, Gel cold pack should be cold not frozen.
Needle disposal Container
Biohazard Container
Packing Material (for shipment of sample)
Packing Tape

APPENDIX 4 - FLUID RETENTION SEVERITY GRADING

EDEMA	SEVERITY GRADING	EFFUSION
• Asymptomatic *and/or* • Very well tolerated *and/or* • Dependent in evening only	MILD 1	• Asymptomatic • No intervention required
• Moderate functional impairment *and/or* • Pronounced and well tolerated *and/or* • Dependent throughout day	MODERATE 2	• Symptomatic : - exertional dyspnea *and/or* - chest pain *and/or* • ECG changes *and/or* • Abdominal distention • Drainage may be required
• Significant impairment of function *and/or* • Pronounced and not well tolerated *and/or* • Generalized anasarca	SEVERE 3	• Symptomatic effusion - dyspnea at rest *and/or* - tamponade *and/or* - pronounced abdominal distention • Drainage urgently required

⇩

FLUID RETENTION
grading
[MILD, MODERATE, SEVERE]
Reporting the highest grade of edema or effusion

APPENDIX 5 - FLOW CHART OF EXAMINATION

Examination	PRESTUDY SCREEN		DURING THERAPY	End of Chemo-therapy	Follow-up*****
	Completed no more than (time) prior to registration		**Every 3 weeks**	****	
Patient informed consent	before study entry	X			
History	14 days	X			
Physical examination Weight Performance Status	14 days	X	X*	X	
Signs and symptoms**	14 days	X	X	X	
Adverse events, including cardiac **Concomitant medication****	14 days	X	X X	X X	
Hematology Hemoglobin, WBC, neutrophils, platelets	14 days	X	X[1]	X	
Biochemistry **Liver function** ASAT/ ALAT alkaline phosphatase Bilirubin	14 days (Liver function tests repeated within 3 days if abnormal)	X	X (within 3 days prior to chemotherapy)	X	
Renal function creatinine creatinine clearance (if indicated)	14 days	X	X		
FISH TEST (positive) (see appendix 3A)	before study entry	X X			
Serum Sample (for shed ECD) – optional **(see appendix 3C)**	At study entry	X		X+	Every 6 months during the first five years of follow-up or until time of relapse and at time of relapse

Blood Sample (Cardiac Genetic Markers)-optional – *(not applicable for France)* **(see appendix 3E)**	At study entry		X			
Examination	PRESTUDY SCREEN		DURING THERAPY	End of Chemo-therapy	Follow-up*****	
	Completed no more than (time) prior to registration		Every 3 weeks	****		
Plasma Sample (Cardiac Biochemical Markers) Optional – *(not applicable for France)* (see appendix 3D)	At study entry	X	End of Cycle 4	X+	-Every 6 months during the 3 first years of follow-up, - at year 5 of follow-up, -at time of any clinical evidence of cardiac failure	
ER Status / PR Status	before study entry	X				
Pregnancy test (urine or serum)	7 days	X				
ECG	3 months	X	As clinically indicated			
LVEF: MUGA scan or echocardiography	3 months	X	See Section VI			
Mammography and/or ultrasound (mammogram is preferred)	3 months	X				
Work up to rule out metastatic disease Chest-X-ray (PA and lateral) and/or chest CT scan and/or chest MRI Abdominal ultrasound or CT or MRI Bone scan and bone X-ray in case of hot spots in bone scan	 3 months 3 months 3 months	 X X X				
Quality of life	14 days	X	X++	X+	X 6, 12 and 24 months	
Socio-economic data (only for Canada, Germany and US)	14 days	x	X++	X+	X 6, 12 and 24 months	
Other investigations	as clinically indicated					

X[1] CBC and differential is to be done every three weeks prior to receiving chemotherapy (day –1 or day 1 of each cycle). In case of fever ≥ 38.1°C, the CBC and differential must be performed and repeated every 2 days until recovery with temperature < 38.1°C or absolute neutrophil count ≥ 1.5

*Physical exam will be performed at day 1 or –1 of the cycle.

** Signs and symptoms will be recorded for baseline in the appropriate CRFs and for ALL other visits in the Clinical Adverse Experience CRF.

***Concomitant medication will be recorded for baseline on the appropriate CRFs, and will include all medication used within one month prior to registration. For ALL other visits concomitant medication will be captured ONLY if related to adverse events.

**** The End of Chemotherapy evaluation will be performed at 21 after the last dose of chemotherapy (including patients that did not complete all cycles)

***** see Appendix 6 for follow up schedule

+ **End of chemotherapy for AC→ TH and AC→ T arms and at follow-up visit 1a, 6 weeks after End of Chemotherapy for arm TCH**

++ **Cycle 7 for AC→ TH and AC→ T arms and End of chemotherapy for TCH arm**

APPENDIX 6 - FOLLOW UP VISIT FLOW CHART

FUP timing		Required assessments			
AC→T & AC→TH Arms (after EOC)	TCH Arm (after EOC)	Physical exam	HER2 ECD[+] Cardiac Biochemical markers[++]	Mammography	QOL (for all countries) Socio-economic data (for USA, Canada, Germany only)
	1.5 months	X			X
3 months	4.5 months	X			
6 months	7.5 months	X	X		X
9 months	10.5 months	X			
12 months	13.5 months	X	X	X	X
15 months	16.5 months	X			
18 months	19.5 months	X	X		
21 months	22.5 months	X			
24 months	25.5 months	X	X	X	X
2. 5 years	31.5 months	X	X		
3 years	37.5 months	X	X	X	
3. 5 years	43.5 months	X	X		
4 years	49.5 months	X	X	X	
4. 5 years	55.5 months	X	X		
5 years	61.5 months	X	X	X	
6 years	73.5 months	X		X	
7 years	85.5 months	X		X	
8 years	97.5 months	X		X	
9 years	109.5 months	X		X	
10 years	121.5 months	X		X	

- [+]HER2 ECD: Serum samples will be collected every 6 months for the first 5 years of follow-up or until time of relapse included. This is an optional substudy.
- [++]Cardiac biochemical markers: Plasma samples will be collected every 6 months for the first 3 years of follow-up , at 5 years follow-up and at any clinical evidence of cardiac failure. This is an optional substudy – *(not applicable for France)*.
- *Clinical Adverse Experiences evaluated during the Follow up include
- - the ones possibly or probably related to the study drug at the End of Chemotherapy or cardiac adverse experiences ongoing at the End of Chemotherapy regardless of relation to study medication or relevant non cancer related signs and symptoms occuring after the completion of chemotherapy (i.e. toxicities related to Tamoxifen and/or radiotherapy).
- All cardiac adverse experiences, regardless of grade, seriousness and relation to study medication, including cardiac events See Section 6.5 Management of Treatment Arms with Symptomatic Cardiac Toxicities and section 6.6 Reporting of cardiac toxicities))

Note: Follow up visit date is calculated according to the date of the End of Chemotherapy visit.

LVEF Determinations in Follow-up

Patients who are in the cardiac safety analysis must follow LVEF schedule as outlined in Section VI.

ABBREVIATED FOLLOW UP

In case of administration of any systemic therapy (chemotherapy, hormonotherapy or immunotherapy) given for disease relapse or 2nd primary malignancy other than the drugs outlined in the protocol, patients will be followed in an abbreviated follow-up for

- Survival
- All cardiac events (per protocol definition see section 6.1.1.1) See Section 6.5 Management of Treatment Arms with Symptomatic Cardiac Toxicities and section 6.6 Reporting of cardiac toxicities)

APPENDIX 7 SAMPLE PATIENT INFORMED CONSENT

Note: These are samples patient informed consent. Each Ethics Committee or Institutional Review Board will revise and adapt according to their own institution's guidelines.

MULTICENTER PHASE III RANDOMIZED TRIAL COMPARING DOXORUBICIN AND CYCLOPHOSPHAMIDE FOLLOWED BY DOCETAXEL (AC→T) WITH DOXORUBICIN AND CYCLOPHOSPHAMIDE FOLLOWED BY DOCETAXEL AND TRASTUZUMAB (AC→TH) AND WITH DOCETAXEL, CARBOPLATIN AND TRASTUZUMAB (TCH) IN THE ADJUVANT TREATMENT OF NODE POSITIVE AND HIGH RISK NODE NEGATIVE PATIENTS WITH OPERABLE BREAST CANCER CONTAINING THE HER2 ALTERATION.

Study number: BCIRG 006 (TAX GMA 302)

Investigator name:
Address:

Consent Form:

This consent form is part of the informed consent process. It is designed to give you an idea of what this research study is about and what will happen to you if you choose to be in the study. If you would like to know more about something mentioned in this form, or have any questions regarding this research study, please be sure to ask your doctor or nurse. Read this form carefully to make sure you understand all the information it provides. You will get a copy of this form to keep. This study is sponsored by Aventis. Aventis is also the company that manufactures the drug Taxotere®. Aventis will supply this drug free of charge for the purpose of this study. Roche and Genentech are the companies that manufacture the drug Herceptin®. This drug will be provided free of charge for the purpose of this study. This study will take place at various centers throughout Canada, South America, Europe, US, Asia, South Africa and Australia. Approximately 3,150 subjects will be participating in this study. Your doctor, who is one of the researchers, will discuss the study with you. Your participation in this study is entirely voluntary. You do not have to take part in this study and your care does not depend on whether you take part or not. This study may not help you directly, but we hope that it will teach us something that will help others in the future.

BACKGROUND Information

Your doctor has explained to you that you have breast cancer with a risk of relapse (cancer may return or spread after it was treated). Breast cancer can be treated with a combination of chemotherapy agents, such as doxorubicin, cyclophosphamide followed by Taxotere® (AC→T). This particular chemotherapy regimen is one arm used in this study. The kind of cancer you have has receptors on it to which a drug called Herceptin® has been made to specifically target the cancer. Herceptin® has been given safely to many women with metastatic breast cancer and it was found to be effective and to improve survival in combination with chemotherapy as compared to chemotherapy alone. We are going to use this drug in combination with chemotherapy agents to see if it is better than a standard regimen that does not contain Herceptin®. Some preliminary studies have shown that the addition of Herceptin® to drugs such as Taxotere®, doxorubicin and a platinum salt, will result in improved activity against the kind of breast cancer you have. Taxotere® (docetaxel) has been administered in approximately 2,000 patients with advanced breast cancer in a clinical trial setting and has been approved for commercial use (as a single agent) for this indication. We are going to compare a standard regimen of four cycles of doxorubicin and cyclophosphamide followed by 4 cycles of Taxotere® (AC→T) to two other regimens. The first will be the same as the AC→T, but Herceptin® will be added for one year starting at the time when the Taxotere® will be given. This arm will be known as AC→TH. The other regimen will consist of giving 6 cycles of Taxotere® and a carboplatin every 3 weeks, with Herceptin® being given for 1 year (TCH). The reason why a carboplatin has been selected is because when doxorubicin was given with Herceptin® in advanced breast cancer patients, the incidence of heart failure was more than with doxorubicin alone. In addition, preclinical studies have shown that carboplatin (platinum salt) given with Herceptin® has an increased antitumor activity, better than each alone.

Patient Initials /
_____

1. Background information on Herceptin® (Trastuzumab)

Studies conducted with a drug called Herceptin® suggest that this drug can block the effect of the HER2 gene and slow down the growth of the cancer. The HER2 gene is found in larger amounts (overexpressed) in breast cancer cells than in normal cells. Approximately 25-30% of breast and ovarian cancers have this gene overexpressed. Former studies showed that the overexpression of the HER2 gene was associated with a more rapid growth of the cancer cell. These studies have been conducted in metastatic breast cancer only.

Breast cancers that are associated with a large overexpression of a gene called HER2 have been found to grow quickly and spread to other parts of a person's body (metastasize). Studies have also showed that when Herceptin® is combined with the chemotherapeutic drugs Taxotere® and carboplatin, the benefit of the combination is higher than if Herceptin® was given alone. It is also believed that Herceptin® may stimulate the body's own defense mechanism to attack the cancer cells.

Several studies in small groups of patients are currently being conducted in metastatic cancer looking at the combination of Herceptin®, Taxotere® and platinum salt, and the results have been encouraging so far. Based on the interaction between Herceptin® and chemotherapy, the combination of Herceptin®, Taxotere® and a platinum salt implemented in the earlier stages of breast cancer leads us to believe that the cancer growth can be slowed down and relapse delayed or even prevented.

STUDY PURPOSE

The aim of the study is to see how effective 3 different combinations of treatment are compared to each other. The first combination consists of doxorubicin and cyclophosphamide followed by Taxotere® (AC→T). The second combination consists of doxorubicin and cyclophosphamide followed by Taxotere® and Herceptin® (AC→TH). The third combination consists of Taxotere®, carboplatin and Herceptin® (TCH). You will have an equal chance of being treated either with AC→T, or AC→TH or TCH. The decision as to which treatment you receive will be made by chance.

If the tests and exams show that you can be in the study, you will be randomly assigned (randomized) to one of the three treatment groups: Group 1, Group 2 or Group 3. This means a computer program will put you into Group 1, or Group 2 or Group 3 by chance. Neither you nor your doctor will choose which group you are in. You will have an equal chance of being placed in either of the three groups.

ELIGIBILITY

Tissue from your tumor at the time of your surgery will need to be taken and tested by a central laboratory for the presence of human epidermal growth factor receptor 2 (HER2), a protein. If it is found that you are positive to this test, you will be eligible for this study.

STUDY DESIGN

If you choose to take part in this study, you have to come to the center every 3 weeks for 18 to 24 weeks. If you are randomized to a treatment arm with Herceptin®, you will have to come every week during your chemotherapy sessions and every 3 weeks after the end of your chemotherapy treatment for a total of one year, to receive your Herceptin® dose.

A cycle of therapy consists of 21 days (3 weeks). You will receive a maximum of 6 cycles if you are treated with the TCH combination and a maximum of 8 cycles (i.e.4 AC and 4T) if you are treated with the AC→T or AC→TH combination.

Patient Initials / _____

Chemotherapy Administration:

➢ **If you are treated with the AC→T combination, you will receive the following:**

- Doxorubicin 60mg/m² IV intravenous (into the vein) over 15 minutes
- Cyclophosphamide 600mg/m² IV over 5 - 60 minutes
- Every 3 weeks for 4 cycles

Three weeks after the 4th cycle, you will receive

- Taxotere® 100mg/m² over 1 hour every 3 weeks for another 4 cycles.

➢ **If you are treated with the AC→TH combination you will receive the following:**

- Doxorubicin 60mg/m² IV over 15 minutes.
- Cyclophosphamide 600mg/m² IV over 5 - 60 minutes
- Every 3 weeks for 4 cycles

Three weeks after the 4th cycle you will receive the TH segment:

First cycle of Taxotere®/ Herceptin®®:

Day 1: Herceptin® 4 mg/kg IV over 90 minutes. You will then be observed for X hours before you go home.
Day 2: Taxotere® 100 mg/m² IV over 1 hour.
Day 8: Herceptin® 2 mg/kg IV over 30 minutes. You will be observed for X hours before you go home.
Day 15: Herceptin® 2 mg/kg IV over 30 minutes. You will be observed for X hours before you go home.

Subsequent cycles:

Day 1: Taxotere® 100 mg/m² IV over 1 hour every 3 weeks followed by Herceptin® 2 mg/kg IV over 30 minutes.
Day 8: Herceptin® 2 mg/kg IV over 30 minutes. You will be observed for X hours before you go home.
Day 15: Herceptin® 2 mg/kg IV over 30 minutes. You will be observed for X hours before you go home.

Following completion of the 4 cycles of Taxotere®, Herceptin® 6 mg/kg over 30 minutes will continue every 3 weeks for a total time of one year. You will be observed for X minutes after each 6mg/kg Herceptin® infusion.

[Investigator Note: please adapt the Herceptin post-observation period to your local regulations].

➢ **If you are treated with the TCH combination you will receive the following:**

First cycle of TCH:

Day 1: Herceptin® 4 mg/kg IV over 90 minutes. You will then be observed for X hours before you go home.
Day 2: Taxotere® 75 mg/m² IV over 1 hour followed immediately by either carboplatin at target AUC = 6 mg/mL/min IV over 30-60 minutes.
Day 8: Herceptin® 2 mg/kg IV over 30 minutes. You will be observed for X hours before you go home.
Day 15: Herceptin® 2 mg/kg IV over 30 minutes. You will be observed for X hours before you go home

(Note: AUC stands for the term “area under the curve” and refers to a means of dosing certain drugs.)

Patient Initials / _____

Subsequent cycles:

Day 1: Taxotere® 75 mg/m² IV over 1 hour followed immediately by carboplatin at target AUC = 6 mg/mL/min IV over 30-60 minutes followed by Herceptin® 2 mg/kg IV over 30 minutes.
Day 8: Herceptin® 2 mg/kg IV over 30 minutes. You will be observed for X hours before you go home.
Day 15: Herceptin® 2 mg/kg IV over 30 minutes. You will be observed for X hours before you go home.

Following completion of the 6 cycles of Taxotere®/carboplatin, Herceptin® 6 mg/kg over 30 minutes will continue every 3 weeks for a total time of one year. You will be observed for X minutes after each 6mg/kg Herceptin® infusion Herceptin®
[Investigator Note: please adapt the Herceptin post-observation period to your local regulations].

Tamoxifen is a hormonal agent that has been shown to decrease the risk of a relapse in those patients whose tumor expresses estrogen and/or progesterone receptor when it is taken for 5 years. Clinical trials have clearly demonstrated that 5 years of Tamoxifen administration is better than a shorter administration time. Your tumor will be tested to check if it has estrogen and/or progesterone receptors. If it does, Tamoxifen will be prescribed at 20 mg daily for 5 years starting 3-4 weeks after the last course of chemotherapy for patients who have positive estrogen and/or progesterone receptors. Anastrozole (Arimidex®) is also a hormonal agent recently approved in USA by the Food and Drug Administration (FDA) as adjuvant treatment for post-menopausal patients who have positive estrogen and/or progesterone receptors. In a recently published trial (ATAC trial) which compared Tamoxifen with Anastrozole or combination therapy (Tamoxifen + Anastrozole) among 9366 patients, Anastrozole at 1 mg/daily by oral route was shown to decrease the breast cancer recurrence rate with a follow up of more than 5 years. It has however to be noted that in a subset analysis of patients who had had previous chemotherapy, the beneficial effect of Anastrozole versus Tamoxifen with respect to time to recurrence was not apparent.
If while on Tamoxifen, you experience severe hot flushes, vaginal bleeding, vaginal discharge or thromboembolic events, your doctor might choose to switch your treatment from Tamoxifen to Anastrozole.
For postmenopausal patients without contraindications to the use of Tamoxifen, your doctor is allowed to administer a sequential therapy consisting of Tamoxifen for 2 to 3 years followed by anastrozole or exemestane for a maximum of 5 years of hormonal therapy.
The total duration of the hormonal therapy, i.e. tamoxifen followed by anastrozole, should not exceed 5 years.
For postmenopausal patients who have completed 5 years of tamoxifen, your doctor is allowed to continue the hormonal treatment with letrozole for a maximum of 3 years.
If while on Anastrozole, you experience severe arthralgia, musculoskeletal disorders, your doctor might choose to switch your treatment from Anastrozole to Tamoxifen.
In addition, two large studies have recently shown a decrease of breast cancer recurrence in post-menopausal patients who switched to Anastrozole or exemestane, another hormonal agent belonging to the same family as Anastrozole, after 2 or 3 years of Tamoxifen for a total of 5 years of hormonal treatment in comparison to 5 years Tamoxifen.
Finally, a large study has also demonstrated a decrease of breast cancer recurrence in post-menopausal patients who continued the hormonal treatment with letrozole, a hormonal agent belonging to the same family as Anastrozole and exemestane, for 3 years after the completion of 5 years Tamoxifen.
Both exemestane and letrozole have been recently approved by the FDA for the adjuvant treatment of post-menopausal patients whose tumor is hormone receptor positive after 2-3 years or 5 years of Tamoxifen, respectively. According to local authorities, they were also approved for adjuvant and advanced treatment of post-menopausal women with Hormone receptor positive.
Your doctor will discuss with you which of the above-mentioned hormonal treatment is more suitable for you, based on your past medical history, as well as your current medical condition, and according to the treatment approved in your country.
If you are taking Anastrozole, the following side effects might occur: hot flushes, musculoskeletal disorder, fatigue, mood disturbances, nausea/vomiting, fractures, vaginal bleeding and vaginal discharge less frequent than under Tamoxifen.
With Letrozole, you may experience: hot flashes, loss of appetite, general pain, weakness, nausea, or diarrhea.
With Exemestane, you may experience: sweating, fatigue, swelling, hot flashes, mood alteration, abdominal pain, nausea or diarrhea.

Patient Initials / _____

Radiation therapy will be given only after the cycles of chemotherapy treatment have been completed, as prescribed by your doctor. It consists of radiation administration to the involved breast and the local region surrounding it. Radiation therapy has been shown to decrease the risk of local relapse in patients who have had a lump or lymph nodes removed from the axilla. Once the original tumor is removed by surgery, the remaining breast tissue will be irradiated in order to prevent a recurrence of local disease. On the other hand, if you have had the entire breast removed, radiation might also be given to you. Your doctor in this case will explain to you if radiation therapy should be considered.
In case you develop side effects from the chemotherapy your doctor may reduce the dose of the drugs and/or delay the cycle before stopping treatment. You will be offered an alternative treatment in case chemotherapy is stopped.

Oral medication will be given twice daily for 3 days starting the night before Taxotere® to prevent hypersensitivity reaction (allergy). You will also be given drugs to prevent nausea and vomiting as well as extra fluid given by vein in order to prevent kidney damage.

The addition of Herceptin® to combination chemotherapy can have an effect on your heart function. It may decrease the pumping action of your heart. Your heart function will be monitored closely with multi-gated angiogram (MUGA) scans or echocardiographies before starting the study, after the 4th and 6th cycle of chemotherapy, again in 6 weeks, and approximately 3 months, 12 months and 36 months after chemotherapy completion**.** A MUGA and echocardiography exam is a test that evaluates if the pumping of your heart is normal. Heart function may be monitored more frequently if the result is not normal. Your doctor can explain the cardiac monitoring in greater detail.

Interim analyses are planned throughout the study. Interim analyses are scheduled so that the data can be reviewed in order to see if the treatment arms are safe. An unacceptably high incidence of any side effect observed in one of the treatment arms, for example, decreased heart function, that treatment arm will be terminated and alternative treatment will be considered.

INVESTIGATIONS DURING THE STUDY

A blood test will be done before the study begins and before each course of chemotherapy to check your blood count and blood chemistry. Each sample of blood will be 2 to 3 teaspoons. These regular blood tests and other examinations will be performed to check that the drugs are not adversely affecting your bone marrow, kidneys, and liver. You will be monitored with blood tests regularly while being treated with chemotherapy.

A physical examination and a number of tests (scans and X-rays), including heart function tests (MUGA test or echocardiography and electrocardiogram "ECG") will be done before you start the study, during the study and the follow up period. A pregnancy test will be done before you receive any chemotherapy if there is a chance that you might become pregnant. Chemotherapy and tamoxifen therapy could affect an unborn child so it is very important that you do not get pregnant while receiving these treatments. Your doctor will talk with you about methods of birth control if you are of childbearing age. If you think that you might be pregnant, call the doctor or nurse whose phone numbers are on the last page of this form.

You will be asked to fill in a quality of life and a socio-economic (specific Canada, Germany, US) questionnaire before chemotherapy begins, at cycle 4 for the three treatment arms and once you have finished the chemotherapy. For the AC→T and AC→TH arm, an additional quality of life questionnaire will be completed at cycle 7. If you are treated with TCH, an additional questionnaire will be done at 6 weeks after the end of your chemotherapy treatment. If you were to have a relapse, you will be asked to complete a questionnaire at that time. The quality of life questionnaire will ask you how you feel regarding the treatment of your disease and help your doctor to judge the efficacy assessed by medical means against the benefit you feel.

Thereafter your physician will follow you in the same way as other breast cancer patients in order to confirm that the cancer has not relapsed. You will be prescribed hormonotherapy (tamoxifen) if needed. You will receive tamoxifen only if you will test positive for estrogen and/or progesterone receptors. This test will be done before you enter the study.

Patient Initials / _____

Please consult your study doctor and/or nurse before taking any new prescription or non-prescription medication while on study.

INVESTIGATIONS DURING FOLLOW UP

The follow up will take place at the end of the study (3 weeks after the last infusion) as well as every 3 months thereafter for the first 2 years, every 6 months years 3 to 5 and every year up to year 10.

For the first 2 years, you will have a physical exam every 3 months and you will be asked to complete a quality of life and socio-economic (specific Canada, Germany, US) questionnaire at 6, 12 and 24 months of follow-up. Every 12 months, mammography will be performed. To evaluate cardiac safety, additional exams (MUGA scan or echocardiography) and visits will be added at 6 weeks after the last course of chemotherapy for the TCH arm, and at 9 and 18 months after the start of the study for all three-treatment arms.
For years 3 to 5, you will be assessed every 6 months by physical exam, and every 12 months, mammography will be added.
For years 6 to 10, visits will be performed every 12 months, including physical exam and mammography.
If at any time you develop signs or symptoms that your doctor feels may be related to cancer, the tests may be performed sooner, and additional tests may be ordered.

SIDE EFFECTS

Every treatment can have side effects. Even the standard treatment has side effects, which your doctor will explain to you. It is important that you know the possible side effects of the treatments given in this study. The following are the side effects of each drug used in this study. These side effects may or may not be more severe when the drugs are taken together. These are the side effects we know about at present. However, since this is a study of new treatments there may be other side effects that we do not know about yet. It is therefore important that you report immediately to your study doctor/ nurse the occurrence of any unusual symptoms.

➢ With Taxotere®, you may also experience low white blood cell counts which may lead to an increased risk of developing fever and infection, mild to moderate nausea and/or vomiting, mouth irritation which may cause you some problems for food intake, diarrhea, fatigue, reversible pins and needles sensation in hands or feet, hair loss, skin reactions and low blood pressure which needs a close monitoring during infusion. All these side effects were experienced by patients from previous studies and you may also experience other ones, which are not predictable at the moment. There is a chance of developing shortness of breath or a drop in blood pressure with the use of Taxotere®. Rarely these reactions can be severe or life threatening. The center where you are being treated is equipped to deal with such events in this case. You may be asked to weigh yourself weekly to enable your doctor to assess early on if you are developing any fluid retention which can lead to swelling of limbs or fluid around the lungs or abdomen. The infusion itself may cause temporary local irritation and bruises if the drug is infused using a vein in your arm.

➢ With doxorubicin, you may have the following side effects: blood test changes which may render you more prone to infection and bruising, mouth irritation which may impair eating, hair loss, nausea and/or vomiting, diarrhea, loss of appetite and fever. If the drug comes in contact with your skin, there may be some skin damage but your doctor and nurses will be careful to avoid this. After a few infusions of doxorubicin, damage to your heart may occur, however your doctor will monitor you. The heart damage may result in a fatal event, in rare cases.

➢ With cyclophosphamide, you may experience nausea, vomiting, stopping of the menstrual periods, blood test changes which may render you more prone to infection and bruising. There is a very remote risk in developing a secondary leukemia with cyclophosphamide. However, it is believe that the benefit of this treatment outweighs the risk of developing leukemia.

➢With Herceptin® you may experience some side effects. Your doctor will carefully monitor your symptoms throughout the study. It is important for you to tell your doctor if you are experiencing any discomforts.

Herceptin® has been safely administered intravenously to patients. Some patients have experienced flu-like symptoms such as chills and a mild, brief fever, pain sometimes at tumors sites, diarrhea during or after receiving their first dose of Herceptin®. These symptoms were usually mild to moderate in severity and were treated with Tylenol, Benadryl or, rarely, Demerol. These symptoms occurred less often with later infusions. You may also experience low blood counts, an increased risk of infection, headache, dizziness, rash, and loss of appetite.

Infrequently, more severe infusion related or allergic reactions have been reported. The symptoms have included shortness of breath, low blood pressure, wheezing, constriction of the airways that lead to the lungs, rapid heart rate, abnormal fluid in the lungs, reduced oxygen in the blood, and breathing difficulties, including adult respiratory distress syndrome. Uncommonly, hypersensitivity or allergic reactions such as hives and swelling in the throat occurred. In rare cases these events were associated with a clinical course resulting in a fatal outcome, particularly in those who already had lung disease and shortness of breath. In the majority of the patients, the onset of symptoms was associated with the first infusion of Herceptin®. Serious reactions have been treated with supportive therapy.

Congestive heart failure is a serious reaction that some patients have experienced while receiving Herceptin® and can result in death. Symptoms of congestive heart failure include shortness of breath, swelling of the lower legs, and fatigue. Patients who have received chemotherapy seem to develop these symptoms more frequently than patients who have not received chemotherapy. Your doctor will monitor your heart function regularly throughout the study.
In addition, rare cases of myocardial infarction have been observed in patients exposed to Herceptin. However, the relationship with Herceptin has not yet been confirmed.
Herceptin® may be present in the circulation for up to 24 weeks after stopping Herceptin® treatment. The use of anthracyclines during these 24 weeks may carry a higher risk of cardiac toxicity. If your physician decides to use anthracyclines as the best treatment to treat your disease, your cardiac function will be monitored carefully.

➢ With carboplatin you may experience nausea and vomiting, low blood counts, stomatitis that may increase your risk of developing an infection and/or a hemorrhage, decreased appetite, hair loss.
These side effects may be a minor inconvenience or could be severe, but the physician in charge of you will watch you closely if any occurs and will decide to adjust your chemotherapy doses or to stop the treatment.
In case of fever or bruising after receiving any drug, you must contact doctors in the department.
If you have a fever and/or infection, your doctor will do some blood work and may prescribe an antibiotic (such as ciprofloxacin). If your white blood cells (cells responsible for fighting infection) are low at the time, your doctor may also prescribe a medication (G-CSF) to stimulate the production of your white blood cells. This would be given as a once daily needle injection. You may be asked to learn how to give yourself these injections.

If you are taking Tamoxifen, hot flashes and/or vaginal discharge are likely to occur. You may also experience constipation or pain with intercourse and problems controlling your bladder when you cough or sneeze. On rare occasions, serious side effects may occur. These can include uterine cancer or abnormal non-cancer cell growth in the pelvic area that may cause pain or bleeding; eye problems (including cataracts, a clouding of the lens inside the eye); liver cancer or changes in blood tests that show possible liver damage; stroke; and blood clots in areas such as the legs, the eyes, or the lungs that could be life-threatening.

If you receive radiation therapy, the most common side effects will be skin burns, skin tenderness, fibrosis in the area being irradiated. Fatigue is also common.

Patient Initials / _____

INTRAVENOUS NEEDLES AND BLOOD WORK

Some known risks, although rare, are associated with placing a needle into a vein or under the skin. These include discomfort, the possibility of infection, and may leave a temporary bruise, or swelling.

MANDATORY TESTING ON YOUR TISSUE SAMPLE

Tumor material taken at time of your recent surgery(ies) will be sent to a laboratory that Breast Cancer International Research Group (BCIRG) will designate in order to confirm that your cancer has the amplification of the HER2 gene. We also will repeat some standard tests already performed on your tumor at the site where you are being treated. These standard tests include histologic subtype, differentiation grade, vascular invasion, tumor size, estrogen and progesterone receptors.

ADDITIONAL TESTING ON YOUR TISSUE SAMPLE

We are now asking you for permission to store your tumor sample and use it to measure certain markers in the future. Markers are substances made by breast cancer cells. It has been found that there is a correlation between some markers in certain types of cancers and the treatment response. There will be a number of these measurements, which will be made on your tumor material in order to determine a possible correlation with the benefit that you will have from the treatment you receive during this study. The markers that will be tested are : p53, members of the Bcl family (Bcl-2, Bax, Bcl-x and Bag-1), MUC1, MIB1 and tubulin isoforms (particularly II, III, IV and Tau). It is possible that as more information about these research measurements is made available to us during this study, newer markers will also be measured on your tissue sample.

Should you (or your legally authorized representative) not wish the scientists to use your tumor sample, refusing to grant permission for testing of these other markers will not affect either your participation in BCIRG 006 trial or the quality of the care you will receive as a participant in this study in any way. Your tumor sample will be returned to your doctor once the mandatory testing for HER2 has been performed, and the repetition of the standard tests.

The results of the marker testing may not help you directly now, or in the future. The research will not have an effect on your care. There are no additional tests required for you to undertake as a result of giving us permission to use this tumor tissue. The BCIRG will keep the samples and use the material in future studies to learn more about breast cancer and other medical problems. The samples will be kept for 10 years. The tissue will be used only for research and will not be sold. Some new products could be made because of the results of the research that uses your samples. These products might be sold at some time in the future but you will not be paid. The results of the markers will not be given to you or your doctor during the course of your participation in this study unless specifically requested. This information will not be put in your health record.

You can request at any time that your tissue not be used any longer for research. You need only contact your study doctor to let him/her know that you do not want us to use your tissue. At this point, the tissue will no longer be used for research.

Do you agree to have your tumor sample stored and used to measure the markers currently identified in the study and for markers identified in the future? Please circle your answer.

YES **NO** **Patient Initials : _____**

ADDITIONAL TESTING FOR SERUM SAMPLE

In addition to the tissue sample, we are also asking for your permission to forward an extra sample of serum to the central laboratory at certain times throughout the course of this study. It will always be taken at the same time as the standard hematology tests, except at 1.5 months of follow-up for TCH arm. The first time will be prior to you starting the study, then again once you have finished the chemotherapy, then every 6 months during the first five years of your follow-up (in total 12 samples). If your disease comes back, we would also like a sample of blood at that time as well. We wish to use these serum samples to see if we can determine whether the type of cancer you have with the HER2 gene can be found through a marker in the serum. In the future, this will be useful in detecting your kind of cancer in patients with breast cancer. This sample will be used only for research and will not be sold. The research may be helpful to other patients in the future. Some new products could be made because of the results of the research that uses your sample. These products might be sold at some time in the future, but you will not be paid.

Should you not wish to provide the scientists with a serum sample at any time, refusing to grant permission for these additional tests will not affect your participation in BCIRG 006 trial or the quality of the care you will receive as a participant in this study in any way.

Do you agree to have an extra sample of serum to be forwarded to the central laboratory prior to you starting the study, once you have finished the chemotherapy and, every 6 months during your first five years of follow-up and at time your disease comes back? Please circle your answer.

YES **NO** **Patient Initials : _____**

ADDITIONAL TESTING FOR BLOOD AND PLASMA SAMPLES

In addition to the tissue and the serum sample, we are also asking for your permission to forward an extra sample of blood and an extra sample of plasma to the central laboratory at certain times throughout the course of this study. It will always be taken at the same time as the standard hematology tests, except at 1.5 months of follow-up for TCH arm.

The blood sample will be collected prior to you starting the study, at the same time as any other blood evaluation. We wish to use the blood sample to Identify and characterize human genetic variations as a new risk factor for cardiac dysfunction.

The plasma samples will be collected for the first time prior to you starting the study, then at cycle 4 of your chemotherapy treatment, once you have finished the chemotherapy and then every 6 months during the first 3 years of follow-up and also at any time of clinical evidence of cardiac failure (in total 9 samples). We wish to use these plasma samples to identify cardiac biochemical markers leading to an early dectection of cardiac dysfunction, before clinical heart failure.

Thee samples will be used only for research and will not be sold. The research may be helpful to other patients in the future. Some new products could be made because of the results of the research that uses your sample. These products might be sold at some time in the future, but you will not be paid.

Should you not wish to provide the scientists with a blood sample or plasma samples at any time, refusing to grant permission for these additional tests will not affect your participation in BCIRG 006 trial or the quality of the care you will receive as a participant in this study in any way.

Patient Initials / _____
Page 9 of 12

Do you agree to have an extra sample of blood to be forwarded to the central laboratory prior to you starting the study? Please circle your answer.

YES NO Patient Initials : _____

Do you agree to have extra plasma samples to be forwarded to the central laboratory prior to you starting the study, at end of cycle 4 of your chemotherapy treatment, once you have finished the chemotherapy and every 6 months during the 3 first years of follow-up and at any time of a clinical evidence of cardiac failure?
Please circle your answer.

YES NO Patient Initials : _____

STANDARD TREATMENTS

Your participation in this study is voluntary. If you decide to take part but later change your mind, you are free to do so and do not have to give any reason, however, you should advise your doctor of your decision so he can tell you the procedure to be followed with your medical condition to be properly evaluated and then to continue medical care. The level of care you receive from your doctor will not be affected.

If you do not wish to participate in this study, there are other treatments available to you. Your doctor will discuss with you other treatment options available to patients with your type of cancer and explain the risks and benefits of these options to you. Right now, the usual treatment is to use any or all of the standard therapies, other drugs and procedures or other investigational drugs. The doctors can provide detailed information about this and the benefits of various treatments available to you. Other therapies, which are optional to you, may not be curative but may control your symptoms. You may also choose to have no treatment in which case your tumor will be expected to grow.

POTENTIAL BENEFITS

Participation in this study may be of no personal benefit to you. However, based on the results of this study, it is hoped that, in the long-term, patient care can be improved.

WITHDRAWAL FROM STUDY

In discussion with you, your doctor at the center, either at his/her own initiative or at the request from the sponsor of this study, may withdraw you from the study at any time if it is in your best interests. You may also withdraw from the study at any time if you wish to do so. You will be informed of any significant new findings about the drugs used in the study. This new information may or may not affect your willingness to continue participating in the study.

COSTS

You will not have to pay for the treatment you receive in this study. If you are covered by a private insurance company, you will get some or all your money back, but if you do not have private insurance, the sponsors of this study will cover these costs. You may have to pay for the drugs you need for side effects, such as your anti-nausea medications. You will be coming to the cancer center more often than if you were not part of a study. There may be some extra costs, such as parking and meals that you will have to pay.

Patient Initials / _____

INJURY CLAUSE

It is important to note that nothing said in this consent form alters your legal rights to recover damages should injury be suffered as a result of participation in the study.

If you have any questions regarding a research-related injury or other medical concerns, or any further inquiries concerning the procedures of this study, you should contact:

Doctor name: _____ Telephone number: _____

Study Coordinator name: _____ Telephone number: _____

For information concerning research and your rights as a study participant, you should contact:

Name :_____ Telephone number: _____

Name and number of patient representative or ethics committee contact person:

Name: _____ Telephone number: _____

CONFIDENTIALITY

The information that we collect, as part of this study, will be shared with other researchers and doctors. However, you will not be identified in any of these reports. Data and materials collected as part of this study, and some information from your original medical records as it relates to this study, may need to be sent to the statistical headquarters of BCIRG. Strict confidentiality will be maintained and you will not be identified by name on any of the data and materials submitted.

We will keep all the material we collect for this study in a safe storage area.

Representatives from the government, Canadian Health Protection Branch or the Food and Drug Administration in the United States or other Regulatory authorities around the world, the Ethics Committee, BCIRG, the sponsor, and a third party designated by BCIRG and the sponsor, may want to look at your medical record as it relates to this study at the center. This is part of the process of quality control. Each person looking at your records will follow the relevant center's policies and procedures that control these actions.

Patient Initials / _____
Page 11 of 12

PATIENT CONSENT

I have been informed of the purpose, procedures and duration of the study (BCIRG 006) of its possible advantages and inconveniences and I agree to participate to this study conducted by Dr _____ .

A summary of the information has been given to me.

I know that I am free to refuse to participate and that I can withdraw my consent at any time during the study. I have been given a copy of this consent form to retain.

_____ _____

Name of Patient (Print) Signature of Patient

_____

Date

_____ _____

Name of Investigator (Print) Signature of Investigator:

_____

Date

Patient Initials / _____

Additional patient Information and Informed Consent Form

STUDY NUMBER BCIRG 006 (TAX GMA 302), amendment 5, dated 27 June 2008

TITLE: MULTICENTER PHASE III RANDOMIZED TRIAL COMPARING DOXORUBICIN AND CYCLOPHOSPHAMIDE FOLLOWED BY DOCETAXEL (AC→T) WITH DOXORUBICIN AND CYCLOPHOSPHAMIDE FOLLOWED BY DOCETAXEL AND TRASTUZUMAB (HERCEPTIN®) (AC→TH) AND WITH DOCETAXEL, CARBOPLATIN AND TRASTUZUMAB (TCH) IN THE ADJUVANT TREATMENT OF NODE POSITIVE AND HIGH RISK NODE NEGATIVE PATIENTS WITH OPERABLE BREAST CANCER CONTAINING THE HER2 ALTERATION.

ADDITIONAL CONSENT FORM

You already signed an inform consent for the above study. This additional consent form is part of the process of informed consent. It is designed to explain why we are asking you to perform an additional cardiac evaluation and if you would accept, to have an additional blood sample being taken. If you would like to know more about something mentioned in this form, or if you have any questions regarding this additional evaluation, please be sure to ask your doctor or your nurse. Read this form carefully to make sure you understand all the information it provides. You will get a copy of this form to keep. Your doctor, who is one of the researchers, will discuss the reason for this additional evaluation with you. This additional cardiac evaluation is now required by the trial you are participating to. These additional evaluations may not help you directly, but we hope that it will teach us something that will help others in the future.

BCIRG 006 REMINDER AND BACKGROUND:

Addition of Herceptin® to combination chemotherapy could have affected your heart function. In the study your heart function was closely monitored with multi-gated angiogram (MUGA) scans or echocardiographies, the last one had been scheduled approximately 36 months after chemotherapy completion. A MUGA and echocardiography exam is a test that evaluates if the pumping of your heart is normal

Additionally you had been proposed to have plasma sample forwarded to the central laboratory to identify cardiac biochemical markers leading to early detection of cardiac dysfunction.

It was planned to perform interim analysis so that data could be reviewed in order to see if the treatment arms were safe. If unacceptability high incidence of any side effect had been observed in one of the treatment arms, for example, decreases heart function, that treatment arm would have been terminated.

Two interim analyses have been performed so far and have been reviewed by an independent scientific committee, including cardiac safety of all patients included in the study. This independent scientific committee did not have any concern with cardiac safety in the study nevertheless they recommended to continue to check cardiac safety at approximately 60 months after end of your chemotherapy to have information on cardiac effect of Herceptin ® for a longer period.

ADDITIONAL EVALUATIONS DETAILS AND PURPOSE:

Theses additional evaluations do not require you to come back more often to the center where you have been treated. They are performed during a regular study follow-up visit, 60 months after the end of your chemotherapy. In case you already performed this visit, these additional evaluations will be performed as soon as you give your agreement to have it done.

They consist of:

- one additional heart function test (MUGA or echocardiography)
- the collection of one additional plasma sample if you have previously agreed to and still agree (collection of one additional plasma sample is optional).

The purpose of these additional evaluations is to follow the above independent scientific committee recommendation that is to have information on cardiac effect of Herceptin ® for approximately 60 months after end of chemotherapy.

Patient Initials / Page 1/3

ADDITIONAL HEART FUNCTION TEST:

The heart function test is the same as the one you already performed prior your entry in this study and 6 times thereafter and will be done during the visit planned approximately 60 months after end of your chemotherapy

ADDITIONAL SAMPLE TESTING FOR PLASMA SAMPLE:

We are also asking you specifically for your permission to forward an extra sample of plasma to the central laboratory approximately 60 months after you end of chemotherapy.

This additional plasma sample is to identify cardiac biochemical markers leading to an early detection of cardiac dysfunction, before clinical heart failure. This sample will be used only for research and will not be sold. The research may be helpful to other patients in the future. Some new products could be made because of the results of the research that uses your sample. These products might be sold at some time in the future, but you will not be paid.

Should you not wish to provide the scientists with this plasma sample, refusing to grant permission for this additional test will not affect your participation in BCIRG 006 trial or the quality of the care you will receive as a participant in this study in any way.

Do you agree to have this extra plasma sample being forwarded to the central laboratory approximately 60 months after you end of chemotherapy? Please circle your answer:

YES **NO** **Patient Initials :**_____

INTRAVENOUS NEEDLES AND BLOOD WORK

Some known risks, although rare, are associated with placing a needle into a vein or under the skin. These include discomfort, the possibility of infection, and may leave a temporary bruise, or swelling.

ALTERNATIVE

Your participation to trial remains voluntary If you decided to take part but later change your mind, you are free to do so and do not have to give any reason. The level of care you receive from your doctor will not be affected.

POTENTIAL BENEFITS

Participation in these additional evaluations may be of no personal benefit to you. However, based on the results of these additional evaluations, it is hoped that, in the long-term, patient care can be improved.

INJURY CLAUSE

It is important to note that nothing said in this additional consent form that alters your legal rights to recover damages. However, if you suffer an injury as a result of participating in this research because of the negligence of the Sponsor, the policy of the study Sponsor is to pay for all medical treatments (or services) recommended by your doctors that are not covered by health insurance (e.g., Medicare). While BCIRG makes no commitment to provide compensation beyond this point, you retain all your legal rights to pursue other possible avenues of compensation (e.g., legal action).

If you have any questions regarding a research-related injury or other medical concerns, or any further inquiries concerning the procedures of this study, you should contact:

Doctor name: _____ Telephone number: _____

Study Coordinator name: _____ Telephone number: _____

For information concerning research and your rights as a study participant, you should contact:

Name : _____ **Telephone number:**

Name and number of patient representative or ethics committee contact person:

Name: _____ Telephone number: _____

CONFIDENTIALITY

The information that we collect for these additional evaluations will be shared with other researchers and doctors. However, you will not be identified in any of these reports. Data and materials collected for these additional evaluations, and some information from your medical records as it relates to these additional evaluations of the study, may need to be sent to the statistical headquarters of BCIRG. Strict confidentiality will be maintained and you will not be identified by name on any of the data and materials submitted.

We will keep all the material we collect for these additional evaluations in a safe storage area. In the future, other researchers may want to use this material for new studies.

By agreeing to participate in this study, the following organizations may have confidential access to your medical records for research, quality assurance, and data analysis:

- the Cancer International Research Group (CIRG), including monitors and auditors;
- Sanofi-aventis (including auditors), a company that is providing support for the Trial;
- Genentech, Inc. (including auditors), a company that is providing support for the trial;
- the CIRG Central Laboratory;
- The Ethic Committee of this Institution a group of people who reviewed and approved the research study to protect your rights and
- government agencies including the Food and Drug Administration (FDA) , the Office for Human Research Protections (OHRP) and <Name of local organization to be reported>. These agencies may review the research to see that it is being done safely and correctly.

PATIENT CONSENT

I am signing this form to show that I have read the consent form. In no way does this waive my legal rights nor release the investigators, sponsors, or involved institutions from their legal and professional responsibilities. I can refuse to perform this/these additional evaluation(s) at any time without jeopardizing my health care. If I do not change my mind, I am to be kept as informed as my initial consent. I am free to ask for further explanations about these additional evaluations. I will get to keep a copy of this consent for information and for further references.

Name of Patient (print name)	Signature of Patient
	Date (day/month/year)[1]
Name of the person who conducted the inform consent discussion (print name)	Signature of the person who conducted the inform consent discussion
	Date (day/month/year) [1]

[1] Each person who signs the consent must personally enter the date for his/her signature

APPENDIX 8 – SERIOUS ADVERSE EVENT REPORT FORM

Aventis

SAE/ER* from a Clinical Trial

Study No.				Investigator's No.		Patient No.	Country
Patient Initials			Date of Birth			Age	Sex	Height (cm)	Weight (kg)
first	mid	Last	day	month	year		☐ male ☐ female		

Criteria for expedited reporting (tick all applicable)

Seriousness criteria:
- ☐ was life threatening
- ☐ required or prolonged inpatient hospitalization
- ☐ was persistently or significantly disabling/incapacitating
- ☐ is a congenital anomaly
- ☐ resulted in death
- ☐ is medically important

Alert Term ☐ No ☐ Yes
If the event is serious, make sure to tick the appropriate seriousness criterion

Did the patient take a **significant overdose?**
☐ No ☐ Yes

Date and type of report

Date of THIS report |__|__| |__|__| |__|__|__|__|
(day) / (month) / (year)

Date reported to Investigator |__|__| |__|__| |__|__|__|__|
(day) / (month) / (year)

Report type: ☐ initial ☐ follow up

For follow-up report, give only changes to previous

Adverse event number |__|__|__|__|
(AE# in study book for the same AE)

Was this serious adverse event first reported as a non-serious adverse event?
☐ No ☐ Yes

IF YES, Date event became serious |__|__| |__|__| |__|__|__|__|
(day) / (month) / (year)

If event started BEFORE first dose of study medication

Did this event continue and worsen in intensity (severity and/or frequency) after the first dose of study medication?
☐ No ☐ Yes

Description of adverse event and duration

Diagnosis/syndrome of AE _____

Component signs or symptoms (only if applicable) 1. _____ 2. _____
3. _____ 4. _____ 5. _____

Start of adverse event |__|__| |__|__| |__|__|__|__|
(day) / (month) / (year)

End of adverse event |__|__| |__|__| |__|__|__|__|
(day) / (month) / (year)

Duration if adverse event lasted less than 24 hours: |__|__|__| |__|__| |__|__|
(hours) / (minutes) / (sec)

Time since last dose of study medication |__|__|__| |__|__|
(hours) / (minutes)

Comments on adverse event:
_____

_____

This section continued on next page

*SAE = serious adverse event, ER = expedited report

Aventis

SAE/ER from a Clinical Trial

Comments continued _____

_____

_____

_____

_____

_____

Remedial measures

Study medication (select one)

- ☐ no change
- ☐ dosage increased due to event
- ☐ dosage decreased due to event
- ☐ discontinued and reintroduced, no event recurred
- ☐ discontinued and reintroduced, same event recurred
- ☐ discontinued and reintroduced, different event occurred
- ☐ discontinued due to event and subject withdrawn
- ☐ permanently discontinued for other reason
- ☐ other, *please specify* _____

_____

Were there other countermeasures? ☐ No ☐ Yes

***If YES,* Medication** ☐ No ☐ Yes

Other *(please specify)* _____

_____

Causal relationship

Is there a reasonable possibility that the adverse event is associated with the study medication?

☐ No ☐ Yes

*Whether **NO** or **YES**, an alternative explanation may be ticked and details given below:*

- ☐ underlying/concomitant illness
- ☐ possibly associated with concomitant drug(s)
- ☐ directly related to study procedures
- ☐ other known or suspected cause *(please comment)*

_____

_____

Intensity

- ☐ mild
- ☐ moderate
- ☐ severe

Outcome

- ☐ recovery without sequelae
- ☐ recovery with sequelae
- ☐ ongoing at time of report
- ☐ event not resolved, follow-up not deemed necessary by investigator
- ☐ subject died

Relevant medical history (e.g. previous diseases, surgery, allergies, pregnancy)

_____

Study medication: _____

(1) ☐ study medication (2) ☐ active comparator
(3) ☐ placebo (4) ☐ code not broken

Date started |__|__| |__|__| |__|__|__|__|
(day) / (month) / (year)

Indication _____

Date ended |__|__| |__|__| |__|__|__|__|
(day) / (month) / (year)

Daily dose _____ Unit _____ Route _____ Batch No. _____

*SAE = serious adverse event, ER = expedited report

Aventis

SAE/ER from a Clinical Trial

Concomitant medication
(List all drugs even if listed elsewhere in study book; exclude those given to treat adverse event)

Drug	Daily dose	Unit	Route	Indication	Date started day / month / year	Date stopped day / month / year	Ongoing No Yes	*Causal relationship No Yes
[1]								
[2]								
[3]								
[4]								
[5]								
[6]								
[7]								
(8]								
[9]								

**Is there a reasonable possibility that the adverse event is associated with the concomitant medication?*

In case of death

Date of death |__|__| |__|__| |__|__|__|__| (day) / (month) / (year)	Was an autopsy performed? ☐ No ☐ Yes ☐ planned *If YES, please provide a copy of the autopsy report*
Was the death related to the study medication?	☐ No ☐ Direct consequence ☐ Indirect consequence
Cause of death *(tick all applicable)*	**Cause(s) of death ranked in order of likelihood**

	No	Yes
Disease for which subject was enrolled into study	☐	☐
Other pre-existing condition(s)	☐	☐
Serious adverse event	☐	☐
Unknown	☐	☐

(Further information will be requested)

1. _____
2. _____
3. _____
4. _____

Investigator Name and address:	Investigator Date (day/month/year): Signature:
MON Date received (day/month/year): Name: Signature:	Affiliate Safety Officer Date received (day/month/year): Name: Signature:
International drug surveillance number (to be filled out by company)	Clintrace Entry Site Date report received:

*SAE = serious adverse event, ER = expedited report

GREGU-GPE-SD-07-01

Approval Date: October 25, 2000

"SAE/ER from a Clinical Trial" Form (version 2)

ADDITIONAL INFORMATION to SAE/ER from a Clinical Trial Form
BCIRG 006 Study

Study Medication:
Date Started (dd-mm-yy):
Date Ended (dd-mm-yy):
Dose:
Unit:
Frequency/Schedule:
Causal Relation to Event* (y/n)
Remedial Measures**:

Study Medication:
Date Started (dd-mm-yy):
Date Ended (dd-mm-yy):
Dose:
Unit:
Frequency/Schedule:
Causal Relation to Event* (y/n)
Remedial Measures**:

Study Medication:
Date Started (dd-mm-yy):
Date Ended (dd-mm-yy):
Dose:
Unit:
Frequency/Schedule:
Causal Relation to Event* (y/n)
Remedial Measures**:

Study Medication:
Date Started (dd-mm-yy):
Date Ended (dd-mm-yy):
Dose:
Unit:
Frequency/Schedule:
Causal Relation to Event* (y/n)
Remedial Measures**:

Comments:

* see Causal Relationship, on page 2 of 3 SAE/ER from a Clinical Trial
** see Remedial measures study medication on page 2 of 3 SAE/ER from a Clinical Trial

APPENDIX 9 - Implementation Of Taxotere® With New Storage Conditions

Implementation of the Taxotere with new storage conditions for clinical trials is following the implementation on the Market. First Market supplied is the European Community*, plus Norway and Switzerland.

In these first countries, for Clinical trials, Taxotere® stored between +2°C to +8°C will be replaced by Taxotere® stored between +2°C to + 25 °C. Locally, Taxotere® stored between +2°C to +8°C will be used until inventories are depleted.

Supplies for clinical trials of the Taxotere® with new storage conditions will be implemented progressively, depending on Local Approval, and Market Launch.

For a short period, inventories of Taxotere® stored between +2°C to +8°C and inventories of Taxotere® stored between +2°C to + 25 °C will be available at the same investigational sites. We suggest not to mix premix solution of Taxotere® stored between +2°C to +8°C, and premix solution of Taxotere® stored between +2°C to +25°C. In case it happens, the period for use of the premix and the infusion bags will be the shortest, corresponding to the instructions for use of Taxotere® stored between +2°C to +8°C.

* European Community

Austria
Belgium
Denmark
Finland
France
Germany
Greece
Ireland
Italy
Luxemburg
Portugal
Spain
Sweden
The Netherlands
United Kingdom

APPENDIX 9(I):

PREPARATION GUIDE FOR USE WITH TAXOTERE® CONCENTRATE AND SOLVENT FOR SOLUTION FOR INFUSION FOR TAXOTERE®

Storage Conditions + 2° C and +8 °C

1. Drug substance

- International non-proprietary name : docetaxel
- Code name : RP56976

2. Formulations

Taxotere® concentrate for solution for infusion is a clear viscous, yellow to brown-yellow solution containing 40 mg/ml docetaxel (anhydrous) in polysorbate 80. The Solvent for Taxotere® is a 13% w/w solution of ethanol in water for injection.

3. Presentation

3.1 Taxotere® 80 mg vial:

- The Taxotere® 80 mg vial is a 15 ml clear glass vial with a red flip-off cap.
- The labeled dosage strength is 80 mg docetaxel per vial.
- The labeled volume of one vial is 2 ml of a 40 mg/ml solution of docetaxel in polysorbate 80.
- **Practically Taxotere® 80 mg vial contains 2.36 ml of the 40 mg/ml solution of docetaxel equivalent to 94.4 mg docetaxel. This volume has been established and validated during the development of Taxotere® to compensate for liquid loss during preparation of the premix (see section 4) due to foaming, adhesion to the walls of the vial and "dead-volumes". This overfill ensures that there is a minimal extractable premix volume of 8 ml containing 10 mg/ml docetaxel which corresponds to the labeled amount of 80 mg per vial.**

3.2 Solvent for Taxotere® 80 mg vial:

- The Solvent for Taxotere® 80 mg vial is a 15 ml clear glass vial with a transparent colorless flip-off cap.
- The Solvent for Taxotere® composition is a 13% w/w solution of ethanol in water for injection
- The theoretical volume of one vial is 6 ml of Solvent for Taxotere®
- **Practically, a solvent for Taxotere® 80 mg vial contains 7.33 ml ± 5% of Solvent. This volume has been established and validated based on the practical content of the Taxotere® 80 mg vial and ensures a premix concentration of 10 mg/ml docetaxel.**

STORAGE CONDITIONS :

In a refrigerator, protected from bright light.

4. Preparation of the premix solution under aseptic conditions

4.1. Remove the required number of Taxotere® 80 mg vials and solvent for Taxotere® vials from the refrigerator and allow to stand at room temperature for 5 minutes.

4.2. For each Taxotere® 80 mg vial, using a syringe fitted with a needle, withdraw **THE ENTIRE CONTENTS** of the corresponding Solvent for Taxotere® 80 mg vial (7.33 ml ± 5% for Taxotere® 80mg vial) and inject it into the corresponding Taxotere® 80 mg vial.
The addition of **THE ENTIRE CONTENTS** of one Solvent for Taxotere® 80 mg vial to one Taxotere® 80 mg vial ensures a minimal extractable volume of the premix solution of 8 ml.

4.3. Remove the syringe and needle and shake the mixture manually for 15 seconds.

4.4. Allow the premix vial to stand for 5 minutes at room temperature and then check that the solution is homogenous and clear. (Foaming is normal even after 5 minutes due to the presence of polysorbate 80 in the formulation)
The premix solution contains 10 mg/ml docetaxel and should be used immediately to prepare the infusion solution.

5. Preparation of the infusion solution under aseptic conditions

5.1. More than one premix vial may be necessary to obtain the required dose for the patient. Based on the required dose for the patient expressed in mg, use graduated syringes fitted with a needle to withdraw the corresponding premix volume containing 10 mg/ml docetaxel from the appropriate number of premix vials. For example, a dose of 140 mg docetaxel would require 14 ml premix solution.

5.2. Inject the required premix volume into a 250 ml infusion bag or bottle containing either 5% glucose solution or 0.9% sodium chloride solution.

If a dose greater than 200 mg of docetaxel is required, use a larger volume of the infusion vehicle so that a concentration of 0.74 mg/ml docetaxel is not exceeded.

5.3. Mix infusion bag or bottle manually using a rocking motion.

The Taxotere® infusion solution should be administered intravenously within the four hours including a one hour infusion under room temperature and normal lighting conditions.

6. Visual inspection

As with all parenteral products, Taxotere® should be inspected visually for particulate matter or discoloration prior to administration whenever the solution and container permit. If Taxotere® premix solution or infusion solution is not clear or appears to have precipitation, the solution should be discarded.

7. Recommendations for the safe handling

Taxotere® is an antineoplastic agent and, as with other potentially toxic compounds, caution should be exercised when handling it and preparing Taxotere® solutions. The use of gloves is recommended.
If Taxotere® concentrate, premix solution or infusion solution should come into contact with skin, wash immediately and thoroughly with soap and water. If Taxotere® concentrate, premix solution or infusion solution should come into contact with mucous membranes, wash immediately and thoroughly with water.

* U.S.A.

APPENDIX 9(II): PREPARATION GUIDE FOR USE WITH TAXOTERE® CONCENTRATE AND SOLVENT FOR SOLUTION FOR INFUSION FOR TAXOTERE®

Storage Conditions + 2° C and +25 °C

1. Drug Substance

- International non-proprietary name : docetaxel
- Code name : RP56976

2. Formulations

Taxotere® concentrate for solution for infusion is a clear viscous, yellow to brown-yellow solution containing 40 mg/ml docetaxel (anhydrous) in polysorbate 80. The Solvent for Taxotere® is a 13% w/w solution of ethanol in water for injection.

3. Presentation

3.1 Taxotere® 80 mg vial:

- The Taxotere® 80 mg vial is a 15 ml clear glass vial with a red flip-off cap.
- The labeled dosage strength is 80 mg docetaxel per vial.
- The labeled volume of one vial is 2 ml of a 40 mg/ml solution of docetaxel in polysorbate 80.
- **Practically, Taxotere® 80 mg vial contains 2.36 ml of the 40 mg/ml solution of docetaxel equivalent to 94.4 mg docetaxel.**

  This volume has been established and validated during the development of Taxotere® to compensate for liquid loss during preparation of the premix (see section 4) due to foaming, adhesion to the walls of the vial and "dead-volumes". This overfill ensures that there is a minimal extractable premix volume of 8 ml containing 10 mg/ml docetaxel which corresponds to the labeled amount of 80 mg per vial.

3.2 Solvent for Taxotere® 80 mg vial:

- The Solvent for Taxotere® 80 mg vial is a 15 ml clear glass vial with a transparent colorless flip-off cap.
- The Solvent for Taxotere® composition is a 13% w/w solution of ethanol in water for injection
- The theoretical volume of one vial is 6 ml of Solvent for Taxotere®
- **Practically, a solvent for Taxotere® 80 mg vial contains 7.33 ml ± 5% of Solvent. This volume has been established and validated based on the practical content of the Taxotere® 80 mg vial and ensures a premix concentration of 10 mg/ml docetaxel.**

STORAGE CONDITIONS :

Vials should be stored between +2°C and +25°C and protected from bright light.

4. Preparation of the premix solution under aseptic conditions

4.1. Remove the required number of Taxotere® 80 mg vials and solvent for Taxotere® vials from the refrigerator and allow to stand at room temperature for 5 minutes.

4.2. For each Taxotere® 80 mg vial, using a syringe fitted with a needle, withdraw **THE ENTIRE CONTENTS** of the corresponding Solvent for Taxotere® 80 mg vial (7.33 ml ± 5% for Taxotere® 80mg vial) and inject it into the corresponding Taxotere® 80 mg vial.
The addition of **THE ENTIRE CONTENTS** of one Solvent for Taxotere® 80 mg vial to one Taxotere® 80 mg vial ensures a minimal extractable volume of the premix solution of 8 ml.

4.3. Remove the syringe and needle and shake the mixture manually for 15 seconds.

4.4. Allow the premix vial to stand for 5 minutes at room temperature and then check that the solution is homogenous and clear. (Foaming is normal even after 5 minutes due to the presence of polysorbate 80 in the formulation)
The premix solution contains 10 mg/ml docetaxel and should be used immediately to prepare the infusion

solution.However the chemical and physical stability of the premix solution has been demonstrated for 8 hours when stored either between +2°C and +8°C or at room temperature.

5. Preparation of the infusion solution under aseptic conditions

5.1. More than one premix vial may be necessary to obtain the required dose for the patient. Based on the required dose for the patient expressed in mg, use graduated syringes fitted with a needle to withdraw the corresponding premix volume containing 10 mg/ml docetaxel from the appropriate number of premix vials. For example, a dose of 140 mg docetaxel would require 14 ml premix solution.

5.2. Inject the required premix volume into a 250 ml infusion bag or bottle containing either 5% glucose solution or 0.9% sodium chloride solution.
If a dose greater than 200 mg of docetaxel is required, use a larger volume of the infusion vehicle so that a concentration of 0.74-mg/ml docetaxel is not exceeded.

5.3. Mix infusion bag or bottle manually using a rocking motion. The Taxotere® infusion solution should be used within 4 hours and should be aseptically administered as a 1 hour infusion under room temperature and normal lighting conditions.

The Taxotere® infusion solution should be administered intravenously within the four hours including a one hour infusion under room temperature and normal lighting conditions.

6. Visual inspection

As with all parenteral products, Taxotere® should be inspected visually for particulate matter or discoloration prior to administration whenever the solution and container permit. If Taxotere® premix solution or infusion solution is not clear or appears to have precipitation, the solution should be discarded.

7. Recommendations for the safe handling

Taxotere® is an antineoplastic agent and, as with other potentially toxic compounds, caution should be exercised when handling it and preparing Taxotere® solutions. The use of gloves is recommended.
If Taxotere® concentrate, premix solution or infusion solution should come into contact with skin, wash immediately and thoroughly with soap and water. If Taxotere® concentrate, premix solution or infusion solution should come into contact with mucous membranes, wash immediately and thoroughly with water.

POLYSORBATE 80 (TWEEN 80®) CONTAINING DRUGS

VEPESID®	ETOPOSIDE
DEPO-PROVERA®	MEDROXYPROGESTERONE ACETATE
DEPO-PRODASONE® 500 - 250mg	MEDROXYPROGESTERONE
DEXTANCYL®	DEXAMETHASONE
DIPROSTENE®	BETAMETHASONE
HYDROCORTANCYL®	PREDNISOLONE (ACETATE)
HYDROCORTANCYL (Roussel) ®	HYDROCORTISONE (ACETATE)
CORTISONE (Roussel) ® 25, 125 mg	CORTISONE (ACETATE)
ALTIM®	CORTIVASOL
TEDAROL® 50 mg	TRIAMCINOLONE
KENACORT - retard®	TRIAMCINOLONE
ARISTOPAN®	TRIAMCINOLONE
DURACILLIN A.S. ®	PENICILLINE
LIBRIUM®	CHLORODIAZEPOXIDE
E. FEROL®	VITAMINE E
CORBIONAX®	AMIODARONE
ACTILYSE® 20, 50 mg	T.P.A. (Activateur tissulaire du plasminogene)
DECAPEPTYL®	TRIPTORELINE
ORTHOCLONE OKT3®	
TERPONE®	ESSENCES TERPENIQUES
VACCIN GENHEVAC B® (Pasteur)	

APPENDIX 10 – Herceptin® Multi Dose and Single Dose vials

A. Single Dose Vials (SDV) DESCRIPTION

1.1. Formulation, Packaging, Labeling, Preparation and Administration

1.1.1 Formulation

Lyophilized Formulation: Herceptin® will be supplied as a freeze-dried preparation at a content of 150 mg per vial for parenteral administration. Herceptin® is formulated in histidine, trehalose, and polysorbate 20. Each vial is reconstituted with 7. 2 mL of Sterile Water for Injection (SWI), USP, yielding a solution of 21 mg/mL Herceptin®. Reconstituted Herceptin® will be added to 250 mL of 0.9% Sodium Chloride Injection, USP. This formulation does not contain a preservative (once the infusion is prepared it should be administered immediately) and is suitable for single use only. This formulation must be infused within 8 hours after reconstitution.

DO NOT FREEZE HERCEPTIN® THAT HAS BEEN RECONSTITUTED.

1.1.2 Drug Preparation

Appropriate aseptic technique should be used. Each vial of Herceptin® is reconstituted with 7. 2 mL of Sterile Water for Injection (SWI). Herceptin® should be carefully handled during reconstitution. Causing excessive foaming during reconstitution or shaking the reconstituted Herceptin® may result in problems with the amount of Herceptin® that can be withdrawn from the vial.

The following instructions have to be followed:

1) Using a sterile syringe, slowly inject 7.2 ml of sterile water for injections in the vial containing the lyophilised Herceptin®, directing the stream into the lyophilised cake.

2) Swirl vial gently to aid reconstitution. DO NOT SHAKE!

This yields a 7.4 ml solution for single-dose use, containing 21 mg/mL Herceptin®, at a pH of approximately 6.0.
Slight foaming of the product upon reconstitution is not unusual. Allow the vial to stand undisturbed for approximately 5 minutes. The reconstituted Herceptin® results in a colourless to pale yellow transparent solution and should be essentially free of visible particulates.

Determine the volume of the solution required based on a loading dose of 4 mg Herceptin®/kg body weight, or a maintenance dose of either 2 mg Herceptin®/kg body weight during chemotherapy or 6 mg Herceptin®/kg body weight during follow-up period:

$$\text{Volume (mL)} = \frac{\text{Body weight (kg)} \times \text{dose (4 mg/kg for loading or either 2 or 6 mg/kg for maintenance)}}{\text{21 (mg/mL, concentration of reconstituted solution)}}$$

The appropriate amount of solution should be withdrawn from the vial and added to an infusion bag containing 250 mL of 0.9% sodium chloride. Glucose-containing solution should not be used since this can cause aggregation of the protein. The bag should be gently inverted to mix the solution in order to avoid foaming. Parenteral drug products should be inspected visually for particulates and discoloration prior to administration. Once the infusion is prepared it should be administered immediately. If diluted aseptically, it may be stored for 24 hours when refrigerated at 2°C–8°C.

1.1.3 Dosage and Administration

Herceptin® is administered as a 90-minute intravenous infusion. Patients should be observed for at least six hours after the start of the first infusion and for two hours after the start of the subsequent infusions for symptoms like fever and chills or other infusion-related symptoms. Interruption of the infusion may help control such symptoms. The infusion may be resumed when symptoms abate.

.If the initial loading dose was well tolerated, the subsequent doses can be administered as a 30-minute infusion.
Emergency equipment must be available.

On very rare occasions, patients have experienced the onset of infusion symptoms or pulmonary symptoms more than six hours after the start of the Herceptin® infusion. Patients should be warned of the possibility of such a late onset and should

be instructed to contact their physician if these symptoms occur.

Do not administer as an intravenous push or bolus.

1.1.4 Storage Requirements

Vials of Herceptin® are shipped on wet ice at a temperature ranging from 2°C to 8°C (36°F to 46°F), and must be placed in a refrigerator (same temperature range) immediately upon receipt to ensure optimal retention of physical and biochemical integrity. Do not use beyond the expiration date stamped on the vial. Temperature logs must be maintained (in accordance with local pharmacy practice) on the refrigerator to ensure proper storage conditions. Do not use beyond the expiration date stamped on the vial.
DO NOT FREEZE. Herceptin® may be sensitive to shear-induced stress (e.g., agitation or rapid expulsion from a syringe).
DO NOT SHAKE. Vigorous handling of solutions of Herceptin® results in aggregation of the protein and may create cloudy solutions.

After reconstitution with sterile water for injections the reconstituted solution is physically and chemically stable for 48 hours at 2°C – 8°C. Any remaining reconstituted solution should be discarded.
Solutions of Herceptin® for infusion are physically and chemically stable in polyvinylchloride or polyethylene bags containing 0.9% sodium chloride for 24 hours at temperatures not exceeding 30°C.
From a microbiological point of view, the reconstituted solution and Herceptin® infusion solution should be used immediately.

B. Multi-Dose Vial (MDV) DESCRIPTION

1. Formulation, Packaging, Labeling, Preparation and Administration

1.1.1 Formulation

Lyophilized Formulation: Herceptin® will be supplied for use as a freeze-dried preparation at a nominal content of 440 mg per vial for parenteral administration. The drug is formulated in histidine, trehalose, and polysorbate 20.

For intravenous (IV) administration, each vial of Herceptin® is reconstituted with 20 mL sterile Bacteriostatic Water for Injection (BWFI), USP (containing 1.1% benzyl alcohol), which is supplied with each vial. The reconstituted solution contains 21 mg/mL Herceptin® and will be added to 250 mL of 0.9% Sodium Chloride Injection, USP. This formulation is designed for multiple uses and must be used within 28 days after reconstitution.

DO NOT FREEZE HERCEPTIN® THAT HAS BEEN RECONSTITUTED.

1.1.2 Drug Preparation

Appropriate aseptic technique should be used. Each vial of Herceptin® is reconstituted with 20 mL of Bacteriostatic Water for Injection, as supplied. Herceptin® should be carefully handled during reconstitution. Causing excessive foaming during reconstitution or shaking the reconstituted Herceptin® may result in problems with the amount of Herceptin® that can be withdrawn from the vial.
The following instructions have to be followed:
1) Using a sterile syringe, slowly inject 20 ml of sterile water for injections in the vial containing the lyophilised Herceptin®, directing the stream into the lyophilised cake.
2) Swirl vial gently to aid reconstitution. DO NOT SHAKE!

This yields a multi-dose solution, containing 21 mg/mL Herceptin®, at a pH of approximately 6.0.
Slight foaming of the product upon reconstitution is not unusual. Allow the vial to stand undisturbed for approximately 5 minutes. The reconstituted Herceptin® results in a colourless to pale yellow transparent solution and should be essentially free of visible particulates.

Determine the volume of the solution required based on a loading dose of 4 mg Herceptin®/kg body weight, or a maintenance dose of either 2 mg Herceptin®/kg body weight during chemotherapy or 6 mg Herceptin®/kg body weight during follow-up:

Volume (mL) = **Body weight** (kg) x **dose** (**4** mg/kg for loading or either **2 or 6** mg/kg for maintenance)
21 (mg/mL, concentration of reconstituted solution)

The appropriate amount of solution should be withdrawn from the vial and added to an infusion bag containing 250 mL of 0.9% sodium chloride. Glucose-containing solution should not be used since this can cause aggregation of the protein. The bag should be gently inverted to mix the solution in order to avoid foaming. Parenteral drug products should be inspected visually for particulates and discoloration prior to administration. Once the infusion is prepared it should be administered immediately. If diluted aseptically, it may be stored for 24 hours when refrigerated at 2°C–8°C.

1.1.3Dosage and Administration

Herceptin® is administered as a 90-minute intravenous infusion. Patients should be observed for at least six hours after the start of the first infusion and for two hours after the start of the subsequent infusions for symptoms like fever and chills or other infusion-related symptoms. Interruption of the infusion may help control such symptoms. The infusion may be resumed when symptoms abate.
If the initial loading dose was well tolerated, the subsequent doses can be administered as a 30-minute infusion. Emergency equipment must be available.
On very rare occasions, patients have experienced the onset of infusion symptoms or pulmonary symptoms more than six hours after the start of the Herceptin® infusion. Patients should be warned of the possibility of such a late onset and should be instructed to contact their physician if these symptoms occur.

Do not administer as an intravenous push or bolus.

1.1.4 Storage Requirements

Vials of Herceptin® are shipped on wet ice at a temperature ranging from 2°C to 8°C (36°F to 46°F), and must be placed in a refrigerator (same temperature range) immediately upon receipt to ensure optimal retention of physical and biochemical integrity. Do not use beyond the expiration date stamped on the vial. Temperature logs must be maintained (in accordance with local pharmacy practice) on the refrigerator to ensure proper storage conditions. Do not use beyond the expiration date stamped on the vial.

DO NOT FREEZE. Herceptin® may be sensitive to shear-induced stress (e.g., agitation or rapid expulsion from a syringe). **DO NOT SHAKE**. Vigorous handling of solutions of Herceptin® results in aggregation of the protein and may create cloudy solutions.

The reconstituted formulation (440 mg vial) with BWFI is designed for multiple uses. Unused drug may be stored for 28 days at 2°C-8°C (36°F-46°F). Discard any remaining multi-dose reconstituted solution after 28 days.
Solutions of Herceptin® for infusion are physically and chemically stable in polyvinylchloride or polyethylene bags containing 0.9% sodium chloride for 24 hours at temperatures not exceeding 30°C.
From a microbiological point of view, the Herceptin® infusion solution should be used immediately. The product is not intended to be stored after dilution.

GENERAL: Dosage and Administration

Herceptin® will be administered in an outpatient setting. Patients in IV infusion the test arm will receive a loading dose of 4 mg/kg Herceptin® at Day 1, followed by weekly administration of 2 mg/kg IV infusion of Herceptin® until three weeks after the last cycle of chemotherapy. Three weeks after the last cycle of chemotherapy, Herceptin® will be administered at a dose of 6 mg/kg by IV infusion every 3 weeks. Do not administer as an IV push or bolus. The initial total dose should be based on the baseline body weight and should not be changed unless the body weight changes by greater than 5 percent. The initial dose of Herceptin® will be administered over a 90-minute-period (see Table 7). If this first dose is well tolerated, subsequent infusion period may be shortened to 30 minutes. If the initial or subsequent doses are not well tolerated, (e.g., the patient experiences fever or chills), subsequent infusions may be shortened only after a dose is well tolerated. Patients

must remain under medical supervision for 4-hours 1/2 hour following completion of the initial 4 mg/kg-loading dose of Herceptin® (see Table 7). If no adverse events occur with the first infusion, the post-infusion observation period for the second infusion may be shortened to 1-hour 1/2 . During the follow-up visits Herceptin® will be administered over a 30-minutes infusion period with a post-infusion observation of 90 minutes (see Table 7).

APPENDIX 11 – Quality of Life

EORTC Quality of Life instruments
QLQ-C30 and QLQ-BR23

The English versions of the QLQ-C30 (version 3.0), QLQ-BR 23 (version 1.0) and the Euroquol follow.

Note: The questionnaires are available in the following languages.
Bulgarian
Chinese (Taiwanese)
Croatian
Czech
Danish
Dutch
English
Finnish
French
German
Hebrew
Hungarian
Indian (Hindi)
Indian (Gujarathi)
Indian (Marathi)
Iranian
Italian
Japanese
Lithuanian
Norwegian
Polish
Portugese
Russian
Serbian
Slovenian
Spanish
Swedish
Turkish

EORTC QLQ-C30 (version 3.0)

We are interested in some things about you and your health. Please answer all of the questions yourself by circling the number that best applies to you. There are no "right" or "wrong" answers. The information that you provide will remain strictly confidential.

Please fill in your initials: (first two letters of first name, first letter of surname)
Your birthdate: (Day, Month, Year):
Today's date: (Day, Month, Year):

		Not at All	A Little	Quite a Bit	Very Much
1.	Do you have any trouble doing strenuous activities, like carrying a heavy shopping bag or a suitcase?	1	2	3	4
2.	Do you have any trouble taking a long walk?	1	2	3	4
3.	Do you have any trouble taking a short walk outside of the house?	1	2	3	4
4.	Do you need to stay in bed or a chair during the day?	1	2	3	4
5.	Do you need help with eating, dressing, washing yourself or using the toilet?	1	2	3	4

During the past week:		Not at All	A Little	Quite a Bit	Very Much
6.	Were you limited in doing either your work or other daily activities?	1	2	3	4
7.	Were you limited in pursuing your hobbies or other leisure time activities?	1	2	3	4
8.	Were you short of breath?	1	2	3	4
9.	Have you had pain?	1	2	3	4
10.	Did you need to rest?	1	2	3	4
11.	Have you had trouble sleeping?	1	2	3	4
12.	Have you felt weak?	1	2	3	4
13.	Have you lacked appetite?	1	2	3	4
14.	Have you felt nauseated?	1	2	3	4
15.	Have you vomited?	1	2	3	4

Please go on to the next page

During the past week:		**Not at All**	**A Little**	**Quite a Bit**	**Very Much**
16.	Have you been constipated?	1	2	3	4
17.	Have you had diarrhea?	1	2	3	4
18.	Were you tired?	1	2	3	4
19.	Did pain interfere with your daily activities?	1	2	3	4
20.	Have you had difficulty in concentrating on things, like reading a newspaper or watching television?	1	2	3	4
21.	Did you feel tense?	1	2	3	4
22.	Did you worry?	1	2	3	4
23.	Did you feel irritable?	1	2	3	4
24.	Did you feel depressed?	1	2	3	4
25.	Have you had difficulty remembering things?	1	2	3	4
26.	Has your physical condition or medical treatment interfered with your family life?	1	2	3	4
27.	Has your physical condition or medical treatment interfered with your social activities?	1	2	3	4
28.	Has your physical condition or medical treatment caused you financial difficulties?	1	2	3	4

For the following questions please circle the number between 1 and 7 that best applies to you

29. How would you rate your overall health during the past week?

1 2 3 4 5 6 7

Very poor Excellent

30. How would you rate your overall quality of life during the past week?

1 2 3 4 5 6 7

Very poor Excellent

EORTC QLQ - BR23

Patients sometimes report that they have the following symptoms or problems. Please indicate the extent to which you have experienced these symptoms or problems during the past week.

During the past week:		**Not at All**	**A Little**	**Quite a Bit**	**Very Much**
31.	Did you have a dry mouth?	1	2	3	4
32.	Did food and drink taste different than usual?	1	2	3	4
33.	Were your eyes painful, irritated or watery?	1	2	3	4
34.	Have you lost any hair?	1	2	3	4
35.	Answer this question only if you had any hair loss: Were you upset by the loss of your hair?	1	2	3	4
36.	Did you feel ill or unwell?	1	2	3	4
37.	Did you have hot flushes?	1	2	3	4
38.	Did you have headaches?	1	2	3	4
39.	Have you felt physically less attractive as a result of your disease or treatment?	1	2	3	4
40.	Have you been feeling less feminine as a result of your disease or treatment?	1	2	3	4
41.	Did you find it difficult to look at yourself naked?	1	2	3	4
42.	Have you been dissatisfied with your body?	1	2	3	4
43.	Were you worried about your health in the future?	1	2	3	4

During the past <u>four</u> weeks:		**Not at All**	**A Little**	**Quite a Bit**	**Very Much**
44.	To what extent were you interested in sex?	1	2	3	4
45.	To what extent were you sexually active? (with or without intercourse)	1	2	3	4
46.	Answer this question only if you have been sexually active: To what extent was sex enjoyable for you?	1	2	3	4

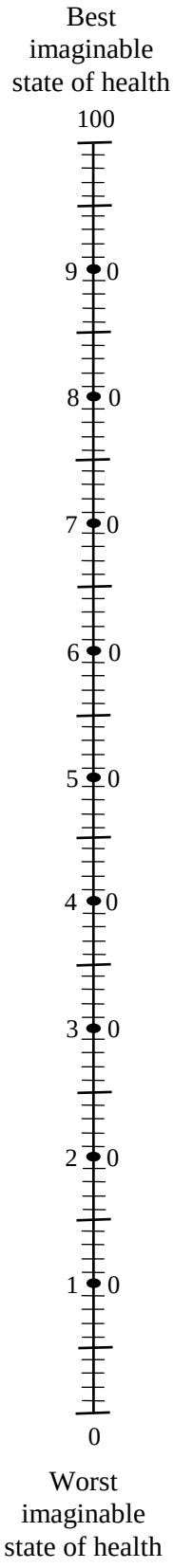
Please go on to the next page

During the past week:		**Not at All**	**A Little**	**Quite a Bit**	**Very Much**
47.	Did you have any pain in your arm or shoulder?	1	2	3	4
48.	Did you have a swollen arm or hand?	1	2	3	4
49.	Was it difficult to raise your arm or to move it sideways?	1	2	3	4
50.	Have you had any pain in the area of your affected breast?	1	2	3	4
51.	Was the area of your affected breast swollen?	1	2	3	4
52.	Was the area of your affected breast oversensitive?	1	2	3	4
53.	Have you had skin problems on or in the area of your affected breast (e.g., itchy, dry, flaky)?	1	2	3	4

To help people say how good or bad their state of health is, we have drawn a scale (rather like a thermometer) on which the best state you can imagine is marked 100 and the worst state you can imagine is marked 0.

We would like you to indicate on this scale how good or bad your own health is today, in your opinion. Please do this by drawing a line from the box below to whichever point on the scale indicates how good or bad your state of health is today.

Your own state of health today



By placing a check-mark in one box in each group below, please indicate which statements best describe your own state of health today.

Mobility

I have no problems in walking about ❑

I have some problems in walking about ❑

I am confined to bed ❑

Self-Care

I have no problems with self-care ❑

I have some problems washing or dressing myself ❑

I am unable to wash or dress myself ❑

Usual Activities (*e.g. work, study, housework, family or leisure activities)*

I have no problems with performing my usual activities ❑

I have some problems with performing my usual activities ❑

I am unable to perform my usual activities ❑

Pain/Discomfort

I have no pain or discomfort ❑

I have moderate pain or discomfort ❑

I have extreme pain or discomfort ❑

Anxiety/Depression

I am not anxious or depressed ❑

I am moderately anxious or depressed ❑

I am extremely anxious or depressed ❑

APPENDIX 12 – NCI COMMON TOXICITY CRITERIA (CTC)

Version 2.0

Grade

Adverse Event	0	1	2	3	4
ALLERGY/IMMUNOLOGY					
Allergic reaction/ hypersensitivity (including drug fever)	none	transient rash, drug fever < 38°C (<100.4°F)	urticaria, drug fever ≥ 38°C (≥100.4°F), and/or asymptomatic bronchospasm	symptomatic bronchospasm, requiring parenteral medication(s), with or without urticaria; allergy-related edema/angioedema	anaphylaxis
Note: Isolated urticaria, in the absence of other manifestations of an allergic or hypersensitivity reaction, is graded in the DERMATOLOGY/SKIN category.					
Allergic rhinitis (including sneezing, nasal stuffiness, postnasal drip)	none	mild, not requiring treatment	moderate, requiring treatment	-	-
Autoimmune reaction	none	serologic or other evidence of autoimmune reaction but patient is asymptomatic (e.g., vitiligo), all organ function is normal and no treatment is required	evidence of autoimmune reaction involving a non-essential organ or function (e.g., hypothyroidism), requiring treatment other than immunosuppressive drugs	reversible autoimmune reaction involving function of a major organ or other adverse event (e.g., transient colitis or anemia), requiring short-term immunosuppressive treatment	autoimmune reaction causing major grade 4 organ dysfunction; progressive and irreversible reaction; long-term administration of high-dose immuno-suppressive therapy required
Also consider Hypothyroidism, Colitis, Hemoglobin, Hemolysis.					
Serum sickness	none	-	-	present	-
Urticaria is graded in the DERMATOLOGY/SKIN category if it occurs as an isolated symptom. If it occurs with other manifestations of allergic or hypersensitivity reaction, grade as Allergic reaction/hypersensitivity above.					
Vasculitis	none	mild, not requiring treatment	symptomatic, requiring medication	requiring steroids	ischemic changes or requiring amputation
Allergy/Immunology-Other (Specify, _____)	none	mild	Moderate	severe	life-threatening or disabling
AUDITORY/HEARING					
Conductive hearing loss is graded as Middle ear/hearing in the AUDITORY/HEARING category.					
Earache is graded in the PAIN category.					
External auditory canal	normal	external otitis with erythema or dry desquamation	external otitis with moist desquamation	external otitis with discharge, mastoiditis	necrosis of the canal soft tissue or bone
Note: Changes associated with radiation to external ear (pinnae) are graded under Radiation dermatitis in the DERMATOLOGY/SKIN category.					
Inner ear/hearing	normal	hearing loss on audiometry only	tinnitus or hearing loss, not requiring hearing aid or treatment	tinnitus or hearing loss, correctable with hearing aid or treatment	severe unilateral or bilateral hearing loss (deafness), not correctable
Middle ear/hearing	normal	serous otitis without subjective decrease in hearing	serous otitis or infection requiring medical intervention; subjective decrease in hearing; rupture of tympanic membrane with discharge	otitis with discharge, mastoiditis or conductive hearing loss	necrosis of the canal soft tissue or bone
Auditory/Hearing-Other (Specify, _____)	normal	mild	Moderate	severe	life-threatening or disabling
BLOOD/BONE MARROW					

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Bone marrow cellularity	normal for age	mildly hypocellular or 25% reduction from normal cellularity for age	moderately hypocellular or >25 - ≤ 50% reduction from normal cellularity for age or >2 but <4 weeks to recovery of normal bone marrow cellularity	severely hypocellular or >50 - ≤ 75% reduction in cellularity for age or 4 - 6 weeks to recovery of normal bone marrow cellularity	aplasia or >6 weeks to recovery of normal bone marrow cellularity
Normal ranges:					
children (≤ 18 years)	90% cellularity average				
younger adults (19-59)	60-70% cellularity average				
older adults (≥ 60 years)	50% cellularity average				
Note: Grade Bone marrow cellularity only for changes related to treatment not disease.					
CD4 count	WNL	< LLN - 500/mm^3	200 - < 500/mm^3	50 - < 200/mm^3	< 50/mm^3
Haptoglobin	normal	decreased	-	absent	-
Hemoglobin (Hgb)	WNL	< LLN - 10.0 g/dl < LLN - 100 g/L < LLN - 6.2 mmol/L	8.0 - < 10.0 g/dL 80 - < 100 g/L 4.9 - < 6.2 mmol/L	6.5 - < 8.0 g/dL 65 - 80 g/L 4.0 - < 4.9 mmol/L	< 6.5 g/dL < 65 g/L < 4.0 mmol/L
Note: The following criteria may be used for leukemia studies or bone marrow infiltrative/myelophthisic process if the protocol so specifies.					
For leukemia studies or bone marrow infiltrative/ myelophthisic processes	WNL	10 - <25% decrease from pretreatment	25 - <50% decrease from pretreatment	50 - <75% decrease from pretreatment	≥75% decrease from pretreatment
Hemolysis (e.g., immune hemolytic anemia, drug-related hemolysis, other)	none	only laboratory evidence of hemolysis [e.g., direct antiglobulin test (DAT, Coombs') schistocytes]	evidence of red cell destruction and ≥ 2gm decrease in hemoglobin, no transfusion	requiring transfusion and/or medical intervention (e.g., steroids)	catastrophic consequences of hemolysis (e.g., renal failure, hypotension, bronchospasm, emergency splenectomy)
Also consider Haptoglobin, Hemoglobin.					
Leukocytes (total WBC)	WNL	< LLN - 3.0 x 10^9 /L < LLN - 3000/mm^3	≥2.0 - < 3.0 x 10^9 /L ≥2000 - < 3000/mm^3	≥1.0 - < 2.0 x 10^9 /L ≥1000 - < 2000/mm^3	< 1.0 x 10^9 /L < 1000/mm^3
For BMT studies:	WNL	≥2.0 - <3.0 X 10^9/L ≥2000 - <3000/mm^3	≥1.0 - <2.0 x 10^9 /L ≥1000 - <2000/mm^3	≥0.5 - <1.0 x 10^9 /L ≥500 - <1000/mm^3	<0.5 x 10^9 /L <500/mm^3
Note: The following criteria using age, race and sex normal values may be used for pediatric studies if the protocol so specifies.					
		≥75 - <100% LLN	*≥50 - <75% LLN*	*≥25 - 50% LLN*	*<25% LLN*
Lymphopenia	WNL	<LLN - 1.0 x 10^9 /L <LLN - 1000/mm^3	≥0.5 - <1.0 x 10^9 /L ≥500 - <1000/mm^3	<0.5 x 10^9 /L <500/mm^3	-
Note: The following criteria using age, race, and sex normal values may be used for pediatric studies if the protocol so specifies.					
		≥75-<100%LLN	*≥50-<75%LLN*	*≥25-<50%LLN*	*<25%LLN*
Neutrophils/granulocytes (ANC/AGC)	WNL	≥1.5 - <2.0 x 10^9 /L ≥1500 - <2000/mm^3	≥1.0 - <1.5 x 10^9 /L ≥1000 - <1500/mm^3	≥0.5 - <1.0 x 10^9 /L ≥500 - <1000/mm^3	< 0.5 x 10^9 /L < 500/mm^3
For BMT:	WNL	≥1.0 - <1.5 x 10^9 /L ≥1000 - <1500/mm^3	≥0.5 - <1.0 x 10^9 /L ≥500 - <1000/mm^3	≥0.1 - <0.5 x 10^9 /L ≥100 - <500/mm^3	<0.1 x 10^9 /L <100/mm^3

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Note: The following criteria may be used for leukemia studies or bone marrow infiltrative/myelophthisic process if the protocol so specifies.					
For leukemia studies or bone marrow infiltrative/ myelophthisic process	WNL	10 - <25% decrease from baseline	25 - <50% decrease from baseline	50 - <75% decrease from baseline	≥75% decrease from baseline
Platelets	WNL	< LLN - <75.0 x 10^9 /L < LLN - 75000/mm^3	≥50.0 - < 75.0 x 10^9 /L ≥50000 - < 75000/mm^3	≥10.0 - < 50.0 x 10^9 /L ≥10000 - < 50000/mm^3	< 10.0 x 10^9 /L < 10000/mm^3
For BMT:	WNL	≥50.0 - <75.0 x 10^9 /L ≥50000 - <75000/mm^3	≥20.0 - <50.0 x 10^9 /L ≥20000 - <50000/mm^3	≥10.0 - <20.0 x 10^9 /L ≥10000 - <20000/mm^3	<10.0 x 10^9 /L <10000/mm^3
Note: The following criteria may be used for leukemia studies or bone marrow infiltrative/myelophthisic process if the protocol so specifies.					
For leukemia studies or bone marrow infiltrative/ myelophthisic process	WNL	10 - <25% decrease from baseline	25 - <50% decrease from baseline	50 - <75% decrease from baseline	≥75% decrease from baseline
Transfusion: Platelets	none	-	-	yes	platelet transfusions and other measures required to improve platelet increment; platelet transfusion refractoriness associated with life-threatening bleeding. (e.g., HLA or cross matched platelet transfusions)
For BMT:	none	1 platelet transfusion in 24 hours	2 platelet transfusions in 24 hours	≥3 platelet transfusions in 24 hours	platelet transfusions and other measures required to improve platelet increment; platelet transfusion refractoriness associated with life-threatening bleeding. (e.g., HLA or cross matched platelet transfusions)
Also consider Platelets.					
Transfusion: pRBCs	none	-	-	Yes	-
For BMT:	none	≤2 u pRBC (≤*15mL/kg*) in 24 hours elective or planned	3 u pRBC (>*15* ≤*30mL/kg*) in 24 hours elective or planned	≥4 u pRBC (>*30mL/kg*) in 24 hours	hemorrhage or hemolysis associated with life-threatening anemia; medical intervention required to improve hemoglobin
Also consider Hemoglobin.					
Blood/Bone Marrow-Other (Specify, _____)	none	mild	Moderate	severe	life-threatening or disabling
CARDIOVASCULAR (ARRHYTHMIA)					

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Conduction abnormality/ Atrioventricular heart block	none	asymptomatic, not requiring treatment (e.g., Mobitz type I second-degree AV block, Wenckebach)	symptomatic, but not requiring treatment	symptomatic and requiring treatment (e.g., Mobitz type II second-degree AV block, third-degree AV block)	life-threatening (e.g., arrhythmia associated with CHF, hypotension, syncope, shock)
Nodal/junctional arrhythmia/dysrhythmia	none	asymptomatic, not requiring treatment	symptomatic, but not requiring treatment	symptomatic and requiring treatment	life-threatening (e.g., arrhythmia associated with CHF, hypotension, syncope, shock)
Palpitations	none	present	-	-	-
Note: Grade palpitations only in the absence of a documented arrhythmia.					
Prolonged QTc interval (QTc > 0.48 seconds)	none	asymptomatic, not requiring treatment	symptomatic, but not requiring treatment	symptomatic and requiring treatment	life-threatening (e.g., arrhythmia associated with CHF, hypotension, syncope, shock)
Sinus bradycardia	none	asymptomatic, not requiring treatment	symptomatic, but not requiring treatment	symptomatic and requiring treatment	life-threatening (e.g., arrhythmia associated with CHF, hypotension, syncope, shock)
Sinus tachycardia	none	asymptomatic, not requiring treatment	symptomatic, but not requiring treatment	symptomatic and requiring treatment of underlying cause	-
Supraventricular arrhythmias (SVT/atrial fibrillation/ flutter)	none	asymptomatic, not requiring treatment	symptomatic, but not requiring treatment	symptomatic and requiring treatment	life-threatening (e.g., arrhythmia associated with CHF, hypotension, syncope, shock)
Syncope (fainting) is graded in the NEUROLOGY category.					
Vasovagal episode	none	-	present without loss of consciousness	present with loss of consciousness	-
Ventricular arrhythmia (PVCs/bigeminy/trigeminy/ ventricular tachycardia)	none	asymptomatic, not requiring treatment	symptomatic, but not requiring treatment	symptomatic and requiring treatment	life-threatening (e.g., arrhythmia associated with CHF, hypotension, syncope, shock)
Cardiovascular/ Arrhythmia-Other (Specify, _____)	none	asymptomatic, not requiring treatment	symptomatic, but not requiring treatment	symptomatic, and requiring treatment of underlying cause	life-threatening (e.g., arrhythmia associated with CHF, hypotension, syncope, shock)
CARDIOVASCULAR (GENERAL)					
Acute vascular leak syndrome	absent	-	symptomatic, but not requiring fluid support	respiratory compromise or requiring fluids	life-threatening; requiring pressor support and/or ventilatory support
Cardiac-ischemia/infarction	none	non-specific T-wave flattening or changes	asymptomatic, ST- and T- wave changes suggesting ischemia	angina without evidence of infarction	acute myocardial infarction

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Cardiac left ventricular function	normal	asymptomatic decline of resting ejection fraction of ≥ 10% but < 20% of baseline value; shortening fraction ≥ 24% but < 30%	asymptomatic but resting ejection fraction below LLN for laboratory or decline of resting ejection fraction ≥ 20% of baseline value; < 24% shortening fraction	CHF responsive to treatment	severe or refractory CHF or requiring intubation
CNS cerebrovascular ischemia is graded in the NEUROLOGY category.					
Cardiac troponin I (cTnI)	normal	-	-	levels consistent with unstable angina as defined by the manufacturer	levels consistent with myocardial infarction as defined by the manufacturer
Cardiac troponin T (cTnT)	normal	≥ 0.03 - < 0.05 ng/mL	≥ 0.05 - < 0.1 ng/mL	≥ 0.1 - < 0.2 ng/mL	≥ 0.2 ng/mL
Edema	none	asymptomatic, not requiring therapy	symptomatic, requiring therapy	symptomatic edema limiting function and unresponsive to therapy or requiring drug discontinuation	anasarca (severe generalized edema)
Hypertension	none	asymptomatic, transient increase by >20 mmHg (diastolic) or to > 150/100* if previously WNL; not requiring treatment	recurrent or persistent or symptomatic increase by > 20 mmHg (diastolic) or to > 150/100* if previously WNL; not requiring treatment	requiring therapy or more intensive therapy than previously	hypertensive crisis
**Note: For pediatric patients, use age and sex appropriate normal values > 95th percentile ULN.*					
Hypotension	none	changes, but not requiring therapy (including transient orthostatic hypotension)	requiring brief fluid replacement or other therapy but not hospitalization; no physiologic consequences	requiring therapy and sustained medical attention, but resolves without persisting physiologic consequences	shock (associated with acidemia and impairing vital organ function due to tissue hypoperfusion)
Also consider Syncope (fainting).					
Note: Angina or MI is graded as Cardiac- ischemia/infarction in the CARDIOVASCULAR (GENERAL) category.					
For pediatric patients, systolic BP 65 mmHg or less in infants up to 1 year old and 70 mmHg or less in children older than 1 year of age, use two successive or three measurements in 24 hours.					
Myocarditis	none	-	-	CHF responsive to treatment	severe or refractory CHF
Operative injury of vein/artery	none	primary suture repair for injury, but not requiring transfusion	primary suture repair for injury, requiring transfusion	vascular occlusion requiring surgery or bypass for injury	myocardial infarction; resection of organ (e.g., bowel, limb)
Pericardial effusion/ pericarditis	none	asymptomatic effusion, not requiring treatment	pericarditis (rub, ECG changes, and/or chest pain)	With physiologic consequences	tamponade (drainage or pericardial window required)
Peripheral arterial ischemia	none	-	brief episode of ischemia managed non-surgically and without permanent deficit	requiring surgical intervention	life-threatening or with permanent functional deficit (e.g., amputation)
Phlebitis (superficial)	none	-	Present	-	-

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Note: Injection site reaction is graded in the DERMATOLOGY/SKIN category.					
Thrombosis/embolism is graded in the CARDIOVASCULAR (GENERAL) category.					
Syncope (fainting) is graded in the NEUROLOGY category.					
Thrombosis/embolism	none	-	deep vein thrombosis, not requiring anticoagulant	deep vein thrombosis, requiring anticoagulant therapy	embolic event including pulmonary embolism
Vein/artery operative injury is graded as Operative injury of vein/artery in the CARDIOVASCULAR (GENERAL) category.					
Visceral arterial ischemia (non-myocardial)	none	-	brief episode of ischemia managed non-surgically and without permanent deficit	requiring surgical intervention	life-threatening or with permanent functional deficit (e.g., resection of ileum)
Cardiovascular/ General-Other (Specify, _____)	none	mild	Moderate	severe	life-threatening or disabling
COAGULATION					
Note: See the HEMORRHAGE category for grading the severity of bleeding events.					
DIC (disseminated intravascular coagulation)	absent	-	-	laboratory findings present with <u>no</u> bleeding	laboratory findings <u>and</u> bleeding
Also grade Platelets.					
Note: Must have increased fibrin split products or D-dimer in order to grade as DIC.					
Fibrinogen	WNL	≥0.75 - <1.0 x LLN	≥0.5 - <0.75 x LLN	≥0.25 - <0.5 x LLN	<0.25 x LLN
Note: The following criteria may be used for leukemia studies or bone marrow infiltrative/myelophthisic process if the protocol so specifies.					
For leukemia studies:	WNL	<20% decrease from pretreatment value or LLN	≥20 - <40% decrease from pretreatment value or LLN	≥40 - <70% decrease from pretreatment value or LLN	<50 mg%
Partial thromboplastin time (PTT)	WNL	> ULN - ≤ 1.5 x ULN	> 1.5 - ≤ 2 x ULN	>2 x ULN	-
Phelbitis is graded in the CARDIOVASCULAR (GENERAL) category.					
Prothrombin time (PT)	WNL	> ULN - ≤ 1.5 x ULN	> 1.5 - ≤ 2 x ULN	>2 x ULN	-
Thrombosis/embolism is graded in the CARDIOVASCULAR (GENERAL) category.					
Thrombotic microangiopathy (e.g., thrombotic thrombocytopenic purpura/TTP or hemolytic uremic syndrome/HUS)	absent	-	-	laboratory findings present without clinical consequences	laboratory findings and clinical consequences, (e.g., CNS hemorrhage/ bleeding or thrombosis/ embolism or renal failure) requiring therapeutic intervention
For BMT:	-	evidence of RBC destruction (schistocytosis) without clinical consequences	evidence of RBC destruction with elevated creatinine (≤3 x ULN)	evidence of RBC destruction with creatinine (>3 x ULN) not requiring dialysis	evidence of RBC destruction with renal failure requiring dialysis and/or encephalopathy
Also consider Hemoglobin (Hgb), Platelets, Creatinine.					
Note: Must have microangiopathic changes on blood smear (e.g., schistocytes, helmet cells, red cell fragments).					
Coagulation-Other (Specify, _____)	none	mild	Moderate	severe	life-threatening or disabling

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
CONSTITUTIONAL SYMPTOMS					
Fatigue (lethargy, malaise, asthenia)	none	increased fatigue over baseline, but not altering normal activities	moderate (e.g., decrease in performance status by 1 ECOG level or 20% Karnofsky or *Lansky*) or causing difficulty performing some activities	severe (e.g., decrease in performance status by ≥2 ECOG levels or 40% Karnofsky or *Lansky*) or loss of ability to perform some activities	bedridden or disabling
Note: See Appendix III for performance status scales.					
Fever (in the absence of neutropenia, where neutropenia is defined as AGC < 1.0 x 10^9/L)	none	38.0 - 39.0°C (100.4 - 102.2°F)	39.1 - 40.0°C (102.3 - 104.0°F)	> 40.0°C (>104.0°F) for < 24hrs	> 40.0°C (>104.0°F) for > 24hrs
Also consider Allergic reaction/hypersensitivity.					
Note: The temperature measurements listed above are oral or tympanic.					
Hot flashes/flushes are graded in the ENDOCRINE category.					
Rigors, chills	none	mild, requiring symptomatic treatment (e.g., blanket) or non-narcotic medication	severe and/or prolonged, requiring narcotic medication	not responsive to narcotic medication	-
Sweating (diaphoresis)	normal	mild and occasional	frequent or drenching	-	-
Weight gain	< 5%	5 - <10%	10 - <20%	≥ 20%	-
Also consider Ascites, Edema, Pleural effusion.					
Weight gain - veno-occlusive disease (VOD)					
Note: The following criteria is to be used ONLY for weight gain associated with Veno-Occlusive Disease.					
	<2%	≥2 - <5%	≥5 - <10%	≥10% or as ascities	≥10% or fluid retention resulting in pulmonary failure
Weight loss	< 5%	5 - <10%	10 - <20%	≥20%	-
Also consider Vomiting, Dehydration, Diarrhea.					
Constitutional Symptoms-Other (Specify, _____)	none	mild	Moderate	severe	life-threatening or disabling
DERMATOLOGY/SKIN					
Alopecia	normal	mild hair loss	pronounced hair loss	-	-
Bruising (in absence of grade 3 or 4 thrombocytopenia)	none	localized or in dependent area	Generalized	-	-
Note: Bruising resulting from grade 3 or 4 thrombocytopenia is graded as Petechiae/purpura and Hemorrhage/bleeding with grade 3 or 4 thrombocytopenia in the HEMORRHAGE category, not in the DERMATOLOGY/SKIN category.					
Dermatitis, focal (associated with high-dose chemotherapy and bone marrow transplant)	none	faint erythema or dry desquamation	moderate to brisk erythema or a patchy moist desquamation, mostly confined to skin folds and creases; moderate edema	confluent moist desquamation, ≥1.5 cm diameter, not confined to skin folds; pitting edema	skin necrosis or ulceration of full thickness dermis; may include spontaneous bleeding not induced by minor trauma or abrasion

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Dry skin	normal	controlled with emollients	not controlled with emollients	-	-
Erythema multiforme (e.g., Stevens-Johnson syndrome, toxic epidermal necrolysis)	absent	-	scattered, but not generalized eruption	severe or requiring IV fluids (e.g., generalized rash or painful stomatitis)	life-threatening (e.g., exfoliative or ulcerating dermatitis or requiring enteral or parenteral nutritional support)
Flushing	absent	present	-	-	-
Hand-foot skin reaction	none	skin changes or dermatitis without pain (e.g., erythema, peeling)	skin changes with pain, not interfering with function	skin changes with pain, interfering with function	-
Injection site reaction	none	pain or itching or erythema	pain or swelling, with inflammation or phlebitis	ulceration or necrosis that is severe or prolonged, or requiring surgery	-
Nail changes	normal	discoloration or ridging (koilonychia) or pitting	partial or complete loss of nail(s) or pain in nailbeds	-	-
Petechiae is graded in the HEMORRHAGE category.					
Photosensitivity	none	painless erythema	painful erythema	erythema with desquamation	-
Pigmentation changes (e.g., vitiligo)	none	localized pigmentation changes	generalized pigmentation changes	-	-
Pruritus	none	mild or localized, relieved spontaneously or by local measures	intense or widespread, relieved spontaneously or by systemic measures	intense or widespread and poorly controlled despite treatment	-
Purpura is graded in the HEMORRHAGE category.					
Radiation dermatitis	none	faint erythema or dry desquamation	moderate to brisk erythema or a patchy moist desquamation, mostly confined to skin folds and creases; moderate edema	confluent moist desquamation, ≥1.5 cm diameter, not confined to skin folds; pitting edema	skin necrosis or ulceration of full thickness dermis; may include bleeding not induced by minor trauma or abrasion
Note: Pain associated with radiation dermatitis is graded separately in the PAIN category as Pain due to radiation.					
Radiation recall reaction (reaction following chemotherapy in the absence of additional radiation therapy that occurs in a previous radiation port)	none	faint erythema or dry desquamation	moderate to brisk erythema or a patchy moist desquamation, mostly confined to skin folds and creases; moderate edema	confluent moist desquamation, ≥1.5 cm diameter, not confined to skin folds; pitting edema	skin necrosis or ulceration of full thickness dermis; may include bleeding not induced by minor trauma or abrasion

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Rash/desquamation	none	macular or papular eruption or erythema without associated symptoms	macular or papular eruption or erythema with pruritus or other associated symptoms covering <50% of body surface or localized desquamation or other lesions covering <50% of body surface area	symptomatic generalized erythroderma or macular, papular or vesicular eruption or desquamation covering ≥50% of body surface area	generalized exfoliative dermatitis or ulcerative dermatitis
For BMT:	none	macular or papular eruption or erythema covering <25% of body surface area without associated symptoms	macular or papular eruption or erythema with pruritis or other associated symptoms covering ≥25 - <50% of body surface or localized desquamation or other lesions covering ≥25 - <50% of body surface area	symptomatic generalized erythroderma or symptomatic macular, papular or vesicular eruption, with bullous formation, or desquamation covering ≥50% of body surface area	generalized exfoliative dermatitis or ulcerative dermatitis or bullous formation
Also consider Allergic reaction/hypersensitivity.					
Note: Erythema multiforme (Stevens-Johnson syndrome) is graded separately as Erythema multiforme.					
Urticaria (hives, welts, wheals)	none	requiring no medication	requiring PO or topical treatment or IV medication or steroids for <24 hours	requiring IV medication or steroids for ≥24 hours	-
Wound- infectious	none	cellulitis	superficial infection	infection requiring IV antibiotics	necrotizing fascitis
Wound- non-infectious	none	incisional separation	incisional hernia	fascial disruption without evisceration	fascial disruption with evisceration
Dermatology/Skin- Other (Specify, _____)	none	mild	Moderate	severe	life-threatening or disabling
ENDOCRINE					
Cushingoid appearance (e.g., moon face with or without buffalo hump, centripetal obesity, cutaneous striae)	absent	-	Present	-	-
Also consider Hyperglycemia, Hypokalemia.					
Feminization of male	absent	-	-	present	-
Gynecomastia	none	mild	pronounced or painful	pronounced or painful and requiring surgery	-
Hot flashes/flushes	none	mild or no more than 1 per day	moderate and greater than 1 per day	-	-
Hypothyroidism	absent	asymptomatic,TSH elevated, no therapy given	symptomatic or thyroid replacement treatment given	patient hospitalized for manifestations of hypothyroidism	myxedema coma

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Masculinization of female	absent	-	-	present	-
SIADH (syndrome of inappropriate antidiuretic hormone)	absent	-	-	present	-
Endocrine-Other (Specify, _____)	none	mild	Moderate	severe	life-threatening or disabling
GASTROINTESTINAL					
Amylase is graded in the METABOLIC/LABORATORY category.					
Anorexia	none	loss of appetite	oral intake significantly decreased	requiring IV fluids	requiring feeding tube or parenteral nutrition
Ascites (non-malignant)	none	asymptomatic	symptomatic, requiring diuretics	symptomatic, requiring therapeutic paracentesis	life-threatening physiologic consequences
Colitis	none	-	abdominal pain with mucus and/or blood in stool	abdominal pain, fever, change in bowel habits with ileus or peritoneal signs, and radiographic or biopsy documentation	perforation or requiring surgery or toxic megacolon
Also consider Hemorrhage/bleeding with grade 3 or 4 thrombocytopenia, Hemorrhage/bleeding without grade 3 or 4 thrombocytopenia, Melena/GI bleeding, Rectal bleeding/hematochezia, Hypotension.					
Constipation	none	requiring stool softener or dietary modification	requiring laxatives	obstipation requiring manual evacuation or enema	obstruction or toxic megacolon
Dehydration	none	dry mucous membranes and/or diminished skin turgor	requiring IV fluid replacement (brief)	requiring IV fluid replacement (sustained)	physiologic consequences requiring intensive care; hemodynamic collapse
Also consider Hypotension, Diarrhea, Vomiting, Stomatitis/pharyngitis (oral/pharyngeal mucositis).					
Diarrhea Patients without colostomy:	none	increase of < 4 stools/day over pre-treatment	increase of 4-6 stools/day, or nocturnal stools	increase of ≥7 stools/day or incontinence; or need for parenteral support for dehydration	physiologic consequences requiring intensive care; or hemodynamic collapse
Patients with a colostomy:	none	mild increase in loose, watery colostomy output compared with pretreatment	moderate increase in loose, watery colostomy output compared with pretreatment, but not interfering with normal activity	severe increase in loose, watery colostomy output compared with pretreatment, interfering with normal activity	physiologic consequences, requiring intensive care; or hemodynamic collapse
For BMT	none	>500 - ≤1000mL of diarrhea/day	>1000 - ≤1500mL of diarrhea/day	>1500mL of diarrhea/day	severe abdominal pain with or without ileus
For Pediatric BMT:		*>5 - ≤10 mL/kg of diarrhea/day*	*>10 - ≤15 mL/kg of diarrhea/day*	*>15 mL/kg of diarrhea/day*	-
Also consider Hemorrhage/bleeding with grade 3 or 4 thrombocytopenia, Hemorrhage/bleeding without grade 3 or 4 thrombocytopenia, Pain, Dehydration, Hypotension.					

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Duodenal ulcer (requires radiographic or endoscopic documentation)	none	-	requiring medical management or non-surgical treatment	uncontrolled by outpatient medical management; requiring hospitalization	perforation or bleeding, requiring emergency surgery
Dyspepsia/heartburn	none	mild	Moderate	severe	-
Dysphagia, esophagitis, odynophagia (painful swallowing)	none	mild dysphagia, but can eat regular diet	dysphagia, requiring predominantly pureed, soft, or liquid diet	dysphagia, requiring IV hydration	complete obstruction (cannot swallow saliva) requiring enteral or parenteral nutritional support, or perforation
Note: If adverse event is radiation-related, grade either under Dysphagia- esophageal related to radiation or Dysphagia- pharyngeal related to radiation.					
Dysphagia- esophageal related to radiation	none	mild dysphagia, but can eat regular diet	dysphagia, requiring predominantly pureed, soft or liquid diet	dysphagia requiring feeding tube, IV hydration or hyperalimentation	complete obstruction (cannot swallow saliva); ulceration with bleeding not induced by minor trauma or abrasion or perforation
Also consider Pain due to radiation, Mucositis due to radiation.					
Note: Fistula is graded separately as Fistula- esophageal.					
Dysphagia - pharyngeal related to radiation	none	mild dysphagia, but can eat regular diet	dysphagia, requiring predominantly pureed, soft, or liquid diet	dysphagia, requiring feeding tube, IV hydration or hyperalimentation	Complete obstruction (cannot swallow saliva); ulceration with bleeding not induced by minor trauma or abrasion or perforation
Also consider Pain due to radiation, Mucositis due to radiation.					
Note: Fistula is graded separately as Fistula- pharyngeal.					
Fistula- esophageal	none	-	-	present	requiring surgery
Fistula- intestinal	none	-	-	present	requiring surgery
Fistula- pharyngeal	none	-	-	present	requiring surgery
Fistula- rectal/anal	none	-	-	present	requiring surgery
Flatulence	none	mild	Moderate	-	-
Gastric ulcer (requires radiographic or endoscopic documentation)	none	-	requiring medical management or non-surgical treatment	bleeding without perforation, uncontrolled by outpatient medical management; requiring hospitalization or surgery	perforation or bleeding, requiring emergency surgery
Also consider Hemorrhage/bleeding with grade 3 or 4 thrombocytopenia, Hemorrhage/bleeding without grade 3 or 4 thrombocytopenia.					
Gastritis	none	-	requiring medical management or non-surgical treatment	uncontrolled by out-patient medical management; requiring hospitalization or surgery	life-threatening bleeding, requiring emergency surgery
Also consider Hemorrhage/bleeding with grade 3 or 4 thrombocytopenia, Hemorrhage/bleeding without grade 3 or 4 thrombocytopenia.					

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Hematemesis is graded in the HEMORRHAGE category.					
Hematochezia is graded in the HEMORRHAGE category as Rectal bleeding/hematochezia.					
Ileus (or neuroconstipation)	none	-	intermittent, not requiring intervention	requiring non-surgical intervention	requiring surgery
Mouth dryness	normal	mild	Moderate	-	-
Mucositis					
Note: Mucositis not due to radiation is graded in the GASTROINTESTINAL category for specific sites: Colitis, Esophagitis, Gastritis, Stomatitis/pharyngitis (oral/pharyngeal mucositis), and Typhlitis; or the RENAL/GENITOURINARY category for Vaginititis.					
Radiation-related mucositis is graded as Mucositis due to radiation.					
Mucositis due to radiation	none	erythema of the mucosa	patchy pseudomembranous reaction (patches generally ≤ 1.5 cm in diameter and non-contiguous)	confluent pseudomembranous reaction (contiguous patches generally > 1.5 cm in diameter)	necrosis or deep ulceration; may include bleeding not induced by minor trauma or abrasion
Also consider Pain due to radiation.					
Note: Grade radiation mucositis of the larynx here.					
Dysphagia related to radiation is also graded as either Dysphagia- esophageal related to radiation or Dysphagia- pharyngeal related to radiation, depending on the site of treatment.					
Nausea	none	able to eat	oral intake significantly decreased	no significant intake, requiring IV fluids	-
Pancreatitis	none	-	-	abdominal pain with pancreatic enzyme elevation	complicated by shock (acute circulatory failure)
Also consider Hypotension.					
Note: Amylase are graded in the METABOLIC/LABORATORY category.					
Pharyngitis is graded in the GASTROINTESTINAL category as Stomatitis/pharyngitis (oral/pharyngeal mucositis).					
Proctitis	none	increased stool frequency, occasional blood-streaked stools, or rectal discomfort (including hemorrhoids), not requiring medication	increased stool frequency, bleeding, mucus discharge, or rectal discomfort requiring medication; anal fissure	increased stool frequency/diarrhea, requiring parenteral support; rectal bleeding, requiring transfusion; or persistent mucus discharge, necessitating pads	perforation, bleeding or necrosis or other life-threatening complication requiring surgical intervention (e.g., colostomy)
Also consider Hemorrhage/bleeding with grade 3 or 4 thrombocytopenia, Hemorrhage/bleeding without grade 3 or 4 thrombocytopenia, and Pain due to radiation.					
Note: Fistula is graded separately as Fistula- rectal/anal.					
Proctitis occurring more than 90 days after the start of radiation therapy is graded in the RTOG/EORTC Late Radiation Morbidity Scoring Scheme. (See Appendix IV)					
Salivary gland changes	none	slightly thickened saliva/may have slightly altered taste (e.g., metallic); additional fluids may be required	thick, ropy, sticky saliva; markedly altered taste; alteration in diet required	-	acute salivary gland necrosis

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Sense of smell	normal	slightly altered	markedly altered	-	-
Stomatitis/pharyngitis (oral/pharyngeal mucositis)	none	painless ulcers, erythema, or mild soreness in the absence of lesions	painful erythema, edema, or ulcers, but can eat or swallow	painful erythema, edema, or ulcers requiring IV hydration	severe ulceration or requires parenteral or enteral nutritional support or prophylatic intubation
For BMT:	none	painless ulcers, erythema, or mild soreness in the absence of lesions	painful erythema, edema or ulcers but can swallow	painful erythema, edema, or ulcers preventing swallowing or requiring hydration or parenteral (or enteral) nutritional support	severe ulceration requiring prophylactic intubation or resulting in documented aspiration pneumonia
Note: Radiation-related mucositis is graded as Mucositis due to radiation.					
Taste disturbance (dysgeusia)	normal	slightly altered	markedly altered	-	-
Typhlitis (inflammation of the cecum)	none	-	-	abdominal pain, diarrhea, fever, and radiographic or biopsy documentation	perforation, bleeding or necrosis or other life-threatening complication requiring surgical intervention (e.g., colostomy)
Also consider Hemorrhage/bleeding with grade 3 or 4 thrombocytopenia, Hemorrhage/bleeding without grade 3 or 4 thrombocytopenia, Hypotension, Febrile/neutropenia.					
Vomiting	none	1 episode in 24 hours over pretreatment	2-5 episodes in 24 hours over pretreatment	≥6 episodes in 24 hours over pretreatment; or need for IV fluids	Requiring parenteral nutrition; or physiologic consequences requiring intensive care; hemodynamic collapse
Also consider Dehydration.					
Weight gain is graded in the CONSTITUTIONAL SYMPTOMS category.					
Weight loss is graded in the CONSTITUTIONAL SYMPTOMS category.					
Gastrointestinal-Other (Specify, _____)	none	mild	Moderate	severe	life-threatening or disabling

HEMORRHAGE

Note: Transfusion in this section refers to pRBC infusion.
For any bleeding with grade 3 or 4 platelets (< 50,000), always grade Hemorrhage/bleeding with grade 3 or 4 thrombocytopenia. Also consider platelets, transfusion: pRBCS, and Transfusion: platelets in addition to grading severity by grading the site or type of bleeding.
If the site or type of Hemorrhage/bleeding is listed, also use the grading that incorporates the site of bleeding: CNS Hemorrhage/bleeding, Hematuria, Hematemesis, Hemoptysis, Hemorrhage/bleeding with surgery, Melena/lower GI bleeding, Petechiae/purpura (Hemorrhage/bleeding into skin), Rectal bleeding/hematochezia, Vaginal bleeding.
If the platelet count is ≥50,000 and the site or type of bleeding is listed, grade the specific site. If the site or type is not listed and the platelet count is ≥50,000, grade Hemorrhage/bleeding without grade 3 or 4 thrombocytopenia and specify the site or type in the OTHER category.

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Hemorrhage/bleeding with grade 3 or 4 thrombocytopenia	none	mild without transfusion		requiring transfusion	Catastrophic bleeding, requiring major non-elective intervention
Also consider Platelets, Hemoglobin, Transfusion: platelets, Transfusion: pRBCs, site or type of bleeding. If the site is not listed, grade as Hemorrhage-Other (Specify site, _____).					
Note: This adverse event must be graded for any bleeding with grade 3 or 4 thrombocytopenia.					
Hemorrhage/bleeding without grade 3 or 4 thrombocytopenia	none	mild without transfusion		requiring transfusion	Catastrophic bleeding requiring major non-elective intervention
Also consider Platelets, Hemoglobin, Transfusion: platelets, Transfusion:pRBCs, Hemorrhage – Other (Specify site, _____).					
Note: Bleeding in the absence of grade 3 or 4 thrombocytopenia is graded here only if the specific site or type of bleeding is not listed elsewhere in the HEMORRHAGE category. Also grade as Other in the HEMORRHAGE category.					
CNS hemorrhage/bleeding	none	-	-	bleeding noted on CT or other scan with no clinical consequences	hemorrhagic stroke or hemorrhagic vascular event (CVA) with neurologic signs and symptoms
Epistaxis	none	mild without transfusion	-	requiring transfusion	Catastrophic bleeding, requiring major non-elective intervention
Hematemesis	none	mild without transfusion	-	requiring transfusion	Catastrophic bleeding, requiring major non-elective intervention
Hematuria (in the absence of vaginal bleeding)	none	microscopic only	intermittent gross bleeding, no clots	persistent gross bleeding or clots; may require catheterization or instrumentation, or transfusion	open surgery or necrosis or deep bladder ulceration
Hemoptysis	none	mild without transfusion	-	requiring transfusion	Catastrophic bleeding, requiring major non-elective intervention
Hemorrhage/bleeding associated with surgery	none	mild without transfusion	-	requiring transfusion	Catastrophic bleeding, requiring major non-elective intervention
Note: Expected blood loss at the time of surgery is not graded as a toxicity.					
Melena/GI bleeding	none	mild without transfusion	-	requiring transfusion	Catastrophic bleeding, requiring major non-elective intervention
Petechiae/purpura (hemorrhage/bleeding into skin or mucosa)	none	rare petechiae of skin	petechiae or purpura in dependent areas of skin	generalized petechiae or purpura of skin or petechiae of any mucosal site	-

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Rectal bleeding/ hematochezia	none	mild without transfusion or medication	persistent, requiring medication (e.g., steroid suppositories) and/or break from radiation treatment	requiring transfusion	Catastrophic bleeding, requiring major non-elective intervention
Vaginal bleeding	none	spotting, requiring < 2 pads per day	requiring ≥ 2 pads per day, but not requiring transfusion	requiring transfusion	Catastrophic bleeding, requiring major non-elective intervention
Hemorrhage-Other (Specify site, _____)	none	mild without transfusion	-	requiring transfusion	Catastrophic bleeding, requiring major non-elective intervention
HEPATIC					
Alkaline phosphatase	WNL	> ULN - 2.5 x ULN	> 2.5 - 5.0 x ULN	> 5.0 - 20.0 x ULN	> 20.0 x ULN
Bilirubin	WNL	> ULN - 1.5 x ULN	> 1.5 - 3.0 x ULN	> 3.0 - 10.0 x ULN	> 10.0 x ULN
Bilirubin- graft versus host disease (GVHD)					
Note: The following criteria are used only for bilirubin associated with graft versus host disease.					
	normal	≥2 - <3 mg/100 ml	≥3 - <6 mg/100 ml	≥6 - <15 mg/100 ml	≥15 mg/100 ml
GGT (γ - Glutamyl transpeptidase)	WNL	> ULN - 2.5 x ULN	> 2.5 - 5.0 x ULN	> 5.0 - 20.0 x ULN	> 20.0 x ULN
Hepatic enlargement	absent	-	-	present	-
Note: Grade Hepatic enlargement only for treatment related adverse event including Veno-Occlusive Disease					
Hypoalbuminemia	WNL	<LLN - 3 g/dl	≥2 - <3 g/dl	<2 g/dl	-
Liver dysfunction/failure (clinical)	normal	-	-	asterixis	encephalopathy or coma
Portal vein flow	normal	-	decreased portal vein flow	reversal/retrograde portal vein flow	-
SGOT (AST) (serum glutamic oxaloacetic transaminase)	WNL	> ULN - 2.5 x ULN	> 2.5 - 5.0 x ULN	> 5.0 - 20.0 x ULN	> 20.0 x ULN
SGPT (ALT) (serum glutamic pyruvic transaminase)	WNL	> ULN - 2.5 x ULN	> 2.5 - 5.0 x ULN	> 5.0 - 20.0 x ULN	> 20.0 x ULN
Hepatic-Other (Specify, _____)	none	mild	Moderate	severe	life-threatening or disabling
INFECTION/FEBRILE NEUTROPENIA					
Catheter-related infection	none	mild, no active treatment	moderate, localized infection, requiring local or oral treatment	severe, systemic infection, requiring IV antibiotic or antifungal treatment or hospitalization	life-threatening sepsis (e.g., septic shock)

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Febrile neutropenia (fever of unknown origin without clinically or microbiologically documented infection)	none	-	-	Present	Life-threatening sepsis (e.g., septic shock)
(ANC < 1.0 x 10^9/L, fever ≥38.5°C)					
Also consider Neutrophils Note: Hypothermia instead of fever may be associated with neutropenia and is graded here.					
Infection (documented clinically or microbiologically) with grade 3 or 4 neutropenia	none	-	-	present	life-threatening sepsis (e.g., septic shock)
(ANC < 1.0 x 10^9/L)					
Note: Hypothermia instead of fever may be associated with neutropenia and is graded here. In the absence of documented infection with grade 3 or 4 neutropenia with fever is graded as Febrile neutropenia.					
Infection with unknown ANC	none	-	-	present	life-threatening sepsis (e.g., septic shock)
Note: This adverse event criterion is used in the rare case when ANC is unknown.					
Infection without neutropenia Also consider Neutrophils	none	mild, no active treatment	moderate, localized infection, requiring local or oral treatment	severe, systemic infection, requiring IV antibiotic or antifungal treatment, or hospitalization	life-threatening sepsis (e.g., septic shock)
Infection/Febrile Neutropenia-Other (Specify, _____)	none	mild	Moderate	severe	life-threatening or disabling
Wound-infectious is graded in the DERMATOLOGY/SKIN category.					
LYMPHATICS					
Lymphatics	normal	mild lymphedema	moderate lymphedema requiring compression; lymphocyst	severe lymphedema limiting function; lymphocyst requiring surgery	Severe lymphedema limiting function with ulceration
Lymphatics-Other (Specify, _____)	none	mild	Moderate	severe	life-threatening or disabling
METABOLIC/LABORATORY					
Acidosis (metabolic or respiratory)	normal	pH < normal, but ≥7.3	-	pH < 7.3	pH < 7.3 with life-threatening physiologic consequences
Alkalosis (metabolic or respiratory)	normal	pH > normal, but ≤7.5	-	pH > 7.5	pH > 7.5 with life-threatening physiologic consequences
Amylase Bicarbonate	WNL WNL	> ULN - 1.5 x ULN < LLN - 16 mEq/dl	> 1.5 - 2.0 x ULN 11 - 15 mEq/dl	> 2.0 - 5.0 x ULN 8 - 10 mEq/dl	>5.0 x ULN < 8 mEq/dl

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
CPK (creatine phosphokinase)	WNL	> ULN - 2.5 x ULN	> 2.5 - 5 x ULN	> 5 - 10 x ULN	> 10 x ULN
Hypercalcemia	WNL	> ULN - 11.5 mg/dl > ULN - 2.9 mmol/L	>11.5 - 12.5 mg/dl > 2.9 - 3.1 mmol/L	>12.5 - 13.5 mg/dl > 3.1 - 3.4 mmol/L	> 13.5 mg/dl > 3.4 mmol/L
Hypercholesterolemia	WNL	> ULN - 300 mg/dl > ULN - 7.75 mmol/L	> 300 - 400 mg/dl > 7.75 - 10.34 mmol/L	> 400 - 500 mg/dl >10.34 - 12.92 mmol/L	> 500 mg/dl > 12.92 mmol/L
Hyperglycemia	WNL	> ULN - 160 mg/dl > ULN - 8.9 mmol/L	> 160 - 250 mg/dl > 8.9 - 13.9 mmol/L	> 250 - 500 mg/dl > 13.9 - 27.8 mmol/L	> 500 mg/dl > 27.8 mmol/L or ketoacidosis
Hyperkalemia	WNL	> ULN - 5.5 mmol/L	> 5.5 - 6.0 mmol/L	> 6.0 - 7.0 mmol/L	> 7.0 mmol/L
Hypermagnesemia	WNL	> ULN - 3.0 mg/dl > ULN - 1.23 mmol/L	-	> 3.0 - 8.0 mg/dl > 1.23 - 3.30 mmol/L	> 8.0 mg/dl > 3.30 mmol/L
Hypernatremia	WNL	> ULN - 150 mmol/L	>150 - 155 mmol/L	>155 - 160 mmol/L	>160 mmol/L
Hypertriglyceridemia	WNL	> ULN - 2.5 x ULN	> 2.5 - 5.0 x ULN	> 5.0 - 10 x ULN	> 10 x ULN
Hyperuricemia	WNL	> ULN - ≤ 10 mg/dl ≤ 0.59 mmol/L without physiologic consequences	-	> ULN - ≤ 10 mg/dl ≤ 0.59 mmol/L with physiologic consequences	> 10 mg/dl > 0.59 mmol/L
Also consider Tumor lysis syndrome, Renal failure, Creatinine,Hyperkalemia.					
Hypocalcemia	WNL	<LLN - 8.0 mg/dl <LLN - 2.0 mmol/L	7.0 - < 8.0 mg/dl 1.75 - < 2.0 mmol/L	6.0 - < 7.0 mg/dl 1.5 - < 1.75 mmol/L	<6.0 mg/dl < 1.5 mmol/L
Hypoglycemia	WNL	<LLN - 55 mg/dl <LLN - 3.0 mmol/L	40 - < 55 mg/dl 2.2 - < 3.0 mmol/L	30 - < 40 mg/dl 1.7 - < 2.2 mmol/L	< 30 mg/dl < 1.7 mmol/L
Hypokalemia	WNL	<LLN - 3.0 mmol/L	-	2.5 - <3.0 mmol/L	<2.5 mmol/L
Hypomagnesemia	WNL	<LLN - 1.2 mg/dl <LLN - 0.5 mmol/L	0.9 - <1.2 mg/dl 0.4 - < 0.5 mmol/L	0.7 - < 0.9 mg/dl 0.3 - < 0.4 mmol/L	< 0.7 mg/dl < 0.3 mmol/L
Hyponatremia	WNL	<LLN - 130 mmol/L	-	120 - <130 mmol/L	<120 mmol/L
Hypophosphatemia	WNL	<LLN -2.5 mg/dl <LLN - 0.8 mmol/L	≥2.0 - <2.5 mg/dl ≥0.6 - <0.8 mmol/L	≥1.0 - <2.0 mg/dl ≥0.3 - <0.6 mmol/L	< 1.0 mg/dl <0.3 mmol/L
Hypothyroidism is graded in the ENDOCRINE category.					
Lipase	WNL	> ULN - 1.5 x ULN	> 1.5 - 2.0 x ULN	> 2.0 - 5.0 x ULN	> 5.0 x ULN
Metabolic/Laboratory-Other (Specify, _____)	none	mild	Moderate	severe	life-threatening or disabling
MUSCULOSKELETAL					
Arthralgia is graded in the PAIN category.					
Arthritis	none	mild pain with inflammation, erythema or joint swelling but not interfering with function	moderate pain with inflammation, erythema, or joint swelling interfering with function, but not interfering with activities of daily living	severe pain with inflammation, erythema, or joint swelling and interfering with activities of daily living	disabling

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Muscle weakness (not due to neuropathy)	normal	asymptomatic with weakness on physical exam	symptomatic and interfering with function, but not interfering with activities of daily living	symptomatic and interfering with activities of daily living	bedridden or disabling
Myalgia [tenderness or pain in muscles] is graded in the PAIN category.					
Myositis (inflammation/damage of muscle)	none	mild pain, not interfering with function	pain interfering with function, but not interfering with activities of daily living	pain interfering with function and interfering with activities of daily living	bedridden or disabling
Also consider CPK.					
Note: Myositis implies muscle damage (i.e., elevated CPK).					
Osteonecrosis (avascular necrosis)	none	asymptomatic and detected by imaging only	symptomatic and interfering with function, but not interfering with activities of daily living	symptomatic and interfering with activities of daily living	symptomatic; or disabling
Musculoskeletal-Other (Specify, _____)	none	mild	Moderate	severe	life-threatening or disabling
NEUROLOGY					
Aphasia, receptive and/or expressive, is graded under Speech impairment in the NEUROLOGY category.					
Arachnoiditis/meningismus/ radiculitis	absent	mild pain not interfering with function	moderate pain interfering with function, but not interfering with activities of daily living	severe pain interfering with activities of daily living	unable to function or perform activities of daily living; bedridden; paraplegia
Also consider Headache, Vomiting, Fever.					
Ataxia (incoordination)	normal	asymptomatic but abnormal on physical exam, and not interfering with function	mild symptoms interfering with function, but not interfering with activities of daily living	moderate symptoms interfering with activities of daily living	bedridden or disabling
CNS cerebrovascular ischemia	none	-	-	transient ischemic event or attack (TIA)	permanent event (e.g., cerebral vascular accident)
CNS hemorrhage/bleeding is graded in the HEMORRHAGE category.					
Cognitive disturbance/ learning problems	*none*	*cognitive disability; not interfering with work/school performance; preservation of intelligence*	*cognitive disability; interfering with work/school performance; decline of 1 SD (Standard Deviation) or loss of developmental milestones*	*cognitive disability; resulting in significant impairment of work/school performance; cognitive decline > 2 SD*	*inability to work/frank mental retardation*

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Confusion	normal	confusion or disorientation or attention deficit of brief duration; resolves spontaneously with no sequelae	confusion or disorientation or attention deficit interfering with function, but not interfering with activities of daily living	confusion or delirium interfering with activities of daily living	harmful to others or self; requiring hospitalization
Cranial neuropathy is graded in the NEUROLOGY category as Neuropathy-cranial.					
Delusions	normal	-	-	present	toxic psychosis
Depressed level of consciousness	normal	somnolence or sedation not interfering with function	somnolence or sedation interfering with function, but not interfering with activities of daily living	obtundation or stupor; difficult to arouse; interfering with activities of daily living	coma
Note: Syncope (fainting) is graded in the NEUROLOGY category.					
Dizziness/lightheadedness	none	not interfering with function	interfering with function, but not interfering with activities of daily living	interfering with activities of daily living	bedridden or disabling
Dysphasia, receptive and/or expressive, is graded under Speech impairment in the NEUROLOGY category.					
Extrapyramidal/ involuntary movement/ restlessness	none	mild involuntary movements not interfering with function	moderate involuntary movements interfering with function, but not interfering with activities of daily living	severe involuntary movements or torticollis interfering with activities of daily living	bedridden or disabling
Hallucinations	normal	-	-	present	toxic psychosis
Headache is graded in the PAIN category.					
Insomnia	normal	occasional difficulty sleeping not interfering with function	difficulty sleeping interfering with function, but not interfering with activities of daily living	frequent difficulty sleeping, interfering with activities of daily living	-
Note: This toxicity is graded when insomnia is related to treatment. If pain or other symptoms interfere with sleep do NOT grade as insomnia.					
Irritability *(children <3 years of age)*	*normal*	*mild; easily consolable*	*moderate; requiring increased attention*	*severe; inconsolable*	-
Leukoencephalopathy associated radiological findings	*none*	*mild increase in SAS (subarachnoid space) and/or mild ventriculomegaly; and/or small (+/- multiple) focal T2 hyperintensities, involving periventricular white matter or < 1/3 of susceptible areas of cerebrum*	*moderate increase in SAS; and/or moderate ventriculomegaly; and/or focal T2 hyperintensities extending into centrum ovale; or involving 1/3 to 2/3 of susceptible areas of cerebrum*	*severe increase in SAS; severe ventriculomegaly; near total white matter T2 hyperintensities or diffuse low attenuation (CT); focal white matter necrosis (cystic)*	*severe increase in SAS; severe ventriculomegaly; diffuse low attenuation with calcification (CT); diffuse white matter necrosis (MRI)*

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Memory loss	normal	memory loss not interfering with function	memory loss interfering with function, but not interfering with activities of daily living	memory loss interfering with activities of daily living	amnesia
Mood alteration-anxiety agitation	normal	mild mood alteration not interfering with function	moderate mood alteration interfering with function, but not interfering with activities of daily living	severe mood alteration interfering with activities of daily living	suicidal ideation or danger to self
Mood alteration-depression	normal	mild mood alteration not interfering with function	moderate mood alteration interfering with function, but not interfering with activities of daily living	severe mood alteration interfering with activities of daily living	suicidal ideation or danger to self
Mood alteration-euphoria	normal	mild mood alteration not interfering with function	moderate mood alteration interfering with function, but not interfering with activities of daily living	severe mood alteration interfering with activities of daily living	danger to self
Neuropathic pain is graded in the PAIN category.					
Neuropathy- cranial	absent	-	present, not interfering with activities of daily living	present, interfering with activities of daily living	life-threatening, disabling
Neuropathy- motor	normal	subjective weakness but no objective findings	mild objective weakness interfering with function, but not interfering with activities of daily living	objective weakness interfering with activities of daily living	paralysis
Neuropathy-sensory	normal	loss of deep tendon reflexes or paresthesia (including tingling) but not interfering with function	objective sensory loss or paresthesia (including tingling), interfering with function, but not interfering with activities of daily living	sensory loss or paresthesia interfering with activities of daily living	permanent sensory loss that interferes with function
Nystagmus	absent	present	-	-	-
Also consider Vision-double vision.					
Personality/behavioral	normal	change, but not disruptive to patient or family	disruptive to patient or family	disruptive to patient and family; requiring mental health intervention	harmful to others or self; requiring hospitalization
Pyramidal tract dysfunction (e.g., ↑ tone, hyperreflexia, positive Babinski, ↓ fine motor coordination)	normal	asymptomatic with abnormality on physical examination	symptomatic or interfering with function but not interfering with activities of daily living	interfering with activities of daily living	bedridden or disabling; paralysis

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Seizure(s)	none	-	seizure(s) self-limited and consciousness is preserved	seizure(s) in which consciousness is altered	seizures of any type which are prolonged, repetitive, or difficult to control (e.g., status epilepticus, intractable epilepsy)
Speech impairment (e.g., dysphasia or aphasia)	normal	-	awareness of receptive or expressive dysphasia, not impairing ability to communicate	receptive or expressive dysphasia, impairing ability to communicate	inability to communicate
Syncope (fainting)	absent	-	-	present	-
Also consider CARDIOVASCULAR (ARRHYTHMIA), Vasovagal episode, CNS cerebrovascular ischemia.					
Tremor	none	mild and brief or intermittent but not interfering with function	moderate tremor interfering with function, but not interfering with activities of daily living	severe tremor interfering with activities of daily living	-
Vertigo	none	not interfering with function	interfering with function, but not interfering with activities of daily living	interfering with activities of daily living	bedridden or disabling
Neurology-Other (Specify, _____)	none	mild	Moderate	severe	life-threatening or disabling
OCULAR/VISUAL					
Cataract	none	asymptomatic	symptomatic, partial visual loss	symptomatic, visual loss requiring treatment or interfering with function	-
Conjunctivitis	none	abnormal ophthalmologic changes, but asymptomatic or symptomatic without visual impairment (i.e., pain and irritation)	symptomatic and interfering with function, but not interfering with activities of daily living	symptomatic and interfering with activities of daily living	-
Dry eye	normal	mild, not requiring treatment	moderate or requiring artificial tears	-	-
Glaucoma	none	increase in intraocular pressure but no visual loss	increase in intraocular pressure with retinal changes	visual impairment	unilateral or bilateral loss of vision (blindness)
Keratitis (corneal inflammation/ corneal ulceration)	none	abnormal ophthalmologic changes but asymptomatic or symptomatic without visual impairment (i.e., pain and irritation)	symptomatic and interfering with function, but not interfering with activities of daily living	symptomatic and interfering with activities of daily living	unilateral or bilateral loss of vision (blindness)

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Tearing (watery eyes)	none	mild: not interfering with function	moderate: interfering with function, but not interfering with activities of daily living	interfering with activities of daily living	-
Vision- blurred vision	normal	-	symptomatic and interfering with function, but not interfering with activities of daily living	symptomatic and interfering with activities of daily living	-
Vision- double vision (diplopia)	normal	-	symptomatic and interfering with function, but not interfering with activities of daily living	symptomatic and interfering with activities of daily living	-
Vision- flashing lights/floaters	normal	mild, not interfering with function	symptomatic and interfering with function, but not interfering with activities of daily living	symptomatic and interfering with activities of daily living	-
Vision- night blindness (nyctalopia)	normal	abnormal electro-retinography but asymptomatic	symptomatic and interfering with function, but not interfering with activities of daily living	symptomatic and interfering with activities of daily living	-
Vision- photophobia	normal	-	symptomatic and interfering with function, but not interfering with activities of daily living	symptomatic and interfering with activities of daily living	-
Ocular/Visual-Other (Specify, _____)	normal	mild	Moderate	severe	unilateral or bilateral loss of vision (blindness)
PAIN					
Abdominal pain or cramping	none	mild pain not interfering with function	moderate pain: pain or analgesics interfering with function, but not interfering with activities of daily living	severe pain: pain or analgesics severely interfering with activities of daily living	disabling
Arthralgia (joint pain)	none	mild pain not interfering with function	moderate pain: pain or analgesics interfering with function, but not interfering with activities of daily living	severe pain: pain or analgesics severely interfering with activities of daily living	disabling
Arthritis (joint pain with clinical signs of inflammation) is graded in the MUSCULOSKELETAL category.					

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Bone pain	none	mild pain not interfering with function	moderate pain: pain or analgesics interfering with function, but not interfering with activities of daily living	severe pain: pain or analgesics severely interfering with activities of daily living	disabling
Chest pain (non-cardiac and non-pleuritic)	none	mild pain not interfering with function	moderate pain: pain or analgesics interfering with function, but not interfering with activities of daily living	severe pain: pain or analgesics severely interfering with activities of daily living	disabling
Dysmenorrhea	none	mild pain not interfering with function	moderate pain: pain or analgesics interfering with function, but not interfering with activities of daily living	severe pain: pain or analgesics severely interfering with activities of daily living	disabling
Dyspareunia	none	mild pain not interfering with function	moderate pain interfering with sexual activity	severe pain preventing sexual activity	-
Dysuria is graded in the RENAL/GENITOURINARY category.					
Earache (otalgia)	none	mild pain not interfering with function	moderate pain: pain or analgesics interfering with function, but not interfering with activities of daily living	severe pain: pain or analgesics severely interfering with activities of daily living	disabling
Headache	none	mild pain not interfering with function	moderate pain: pain or analgesics interfering with function, but not interfering with activities of daily living	severe pain: pain or analgesics severely interfering with activities of daily living	disabling
Hepatic pain	none	mild pain not interfering with function	moderate pain: pain or analgesics interfering with function, but not interfering with activities of daily living	severe pain: pain or analgesics severely interfering with activities of daily living	disabling
Myalgia (muscle pain)	none	mild pain not interfering with function	moderate pain: pain or analgesics interfering with function, but not interfering with activities of daily living	severe pain: pain or analgesics severely interfering with activities of daily living	disabling
Neuropathic pain (e.g., jaw pain, neurologic pain, phantom limb pain, post-infectious neuralgia, or painful neuropathies)	none	mild pain not interfering with function	moderate pain: pain or analgesics interfering with function, but not interfering with activities of daily living	severe pain: pain or analgesics severely interfering with activities of daily living	disabling

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Pain due to radiation	none	mild pain not interfering with function	moderate pain: pain or analgesics interfering with function, but not interfering with activities of daily living	severe pain: pain or analgesics severely interfering with activities of daily living	disabling
Pelvic pain	none	mild pain not interfering with function	moderate pain: pain or analgesics interfering with function, but not interfering with activities of daily living	severe pain: pain or analgesics severely interfering with activities of daily living	disabling
Pleuritic pain	none	mild pain not interfering with function	moderate pain: pain or analgesics interfering with function, but not interfering with activities of daily living	severe pain: pain or analgesics severely interfering with activities of daily living	disabling
Rectal or perirectal pain (proctalgia)	none	mild pain not interfering with function	moderate pain: pain or analgesics interfering with function, but not interfering with activities of daily living	severe pain: pain or analgesics severely interfering with activities of daily living	disabling
Tumor pain (onset or exacerbation of tumor pain due to treatment)	none	mild pain not interfering with function	moderate pain: pain or analgesics interfering with function, but not interfering with activities of daily living	severe pain: pain or analgesics severely interfering with activities of daily living	disabling
Tumor flair is graded in the SYNDROME category.					
Pain-Other (Specify, _____)	none	mild	Moderate	severe	disabling
PULMONARY					
Adult Respiratory Distress Syndrome (ARDS)	absent	-	-	-	present
Apnea	none	-	-	present	requiring intubation
Carbon monoxide diffusion capacity (DL_{CO})	≥ 90% of pretreatment or normal value	≥75 - <90% of pretreatment or normal value	≥50 - <75% of pretreatment or normal value	≥25 - <50% of pretreatment or normal value	< 25% of pretreatment or normal value
Cough	absent	mild, relieved by non-prescription medication	requiring narcotic antitussive	severe cough or coughing spasms, poorly controlled or unresponsive to treatment	-
Dyspnea (shortness of breath)	normal	-	dyspnea on exertion	dyspnea at normal level of activity	dyspnea at rest or requiring ventilator support
FEV_1	≥ 90% of pretreatment or normal value	≥75 - <90% of pretreatment or normal value	≥50 - <75% of pretreatment or normal value	≥25 - <50% of pretreatment or normal value	< 25% of pretreatment or normal value
Hiccoughs (hiccups, singultus)	none	mild, not requiring treatment	moderate, requiring treatment	severe, prolonged, and refractory to treatment	-

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Hypoxia	normal	-	decreased O_2 saturation with exercise	decreased O_2 saturation at rest, requiring supplemental oxygen	decreased O_2 saturation, requiring pressure support (CPAP) or assisted ventilation
Pleural effusion (non-malignant)	none	asymptomatic and not requiring treatment	symptomatic, requiring diuretics	symptomatic, requiring O_2 or therapeutic thoracentesis	life-threatening (e.g., requiring intubation)
Pleuritic pain is graded in the PAIN category.					
Pneumonitis/pulmonary infiltrates	none	radiographic changes but asymptomatic or symptoms not requiring steroids	radiographic changes and requiring steroids or diuretics	radiographic changes and requiring oxygen	radiographic changes and requiring assisted ventilation
Pneumothorax	none	no intervention required	chest tube required	sclerosis or surgery required	life-threatening
Pulmonary embolism is graded as Thrombosis/embolism in the CARDIOVASCULAR (GENERAL) category.					
Pulmonary fibrosis	none	radiographic changes, but asymptomatic or symptoms not requiring steroids	requiring steroids or diuretics	requiring oxygen	requiring assisted ventilation
Note: Radiation-related pulmonary fibrosis is graded in the RTOG/EORTC Late Radiation Morbidity Scoring Scheme- Lung. (See Appendix IV)					
Voice changes/stridor/larynx (e.g., hoarseness, loss of voice, laryngitis)	normal	mild or intermittent hoarseness	persistent hoarseness, but able to vocalize; may have mild to moderate edema	whispered speech, not able to vocalize; may have marked edema	marked dyspnea/stridor requiring tracheostomy or intubation
Note: Cough from radiation is graded as cough in the PULMONARY category.					
Radiation-related hemoptysis from larynx/pharynx is graded as Grade 4 Mucositis due to radiation in the GASTROINTESTINAL category. Radiation-related hemoptysis from the thoracic cavity is graded as Grade 4 Hemoptysis in the HEMORRHAGE category.					
Pulmonary-Other (Specify, _____)	none	mild	moderate	severe	life-threatening or disabling
RENAL/GENITOURINARY					
Bladder spasms	absent	mild symptoms, not requiring intervention	symptoms requiring antispasmotic	severe symptoms requiring narcotic	-
Creatinine	WNL	> ULN - 1.5 x ULN	> 1.5 - 3.0 x ULN	> 3.0 - 6.0 x ULN	> 6.0 x ULN
Note: Adjust to age-appropriate levels for pediatric patients.					
Dysuria (painful urination)	none	mild symptoms requiring no intervention	symptoms relieved with therapy	symptoms not relieved despite therapy	-
Fistula or GU fistula (e.g., vaginal, vesicovaginal)	none	-	-	requiring intervention	requiring surgery
Hemoglobinuria	-	present	-	-	-
Hematuria (in the absence of vaginal bleeding) is graded in the HEMORRHAGE category.					
Incontinence	none	with coughing, sneezing, etc.	spontaneous, some control	no control (in the absence of fistula)	-
Operative injury to bladder and/or ureter	none	-	injury of bladder with primary repair	sepsis, fistula, or obstruction requiring secondary surgery; loss of one kidney; injury requiring anastomosis or re-implantation	septic obstruction of both kidneys or vesicovaginal fistula requiring diversion

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Proteinuria	normal or < 0.15 g/24 hours	1+ or 0.15 - 1.0 g/24 hours	2+ to 3+ or 1.0 - 3.5 g/24 hours	4+ or > 3.5 g/24 hours	nephrotic syndrome
Note: If there is an inconsistency between absolute value and dip stick reading, use the absolute value for grading.					
Renal failure	none	-	-	requiring dialysis, but reversible	requiring dialysis and irreversible
Ureteral obstruction	none	unilateral, not requiring surgery	-	bilateral, not requiring surgery	stent, nephrostomy tube, or surgery
Urinary electrolyte wasting (e.g., Fanconi's syndrome, renal tubular acidosis)	none	asymptomatic, not requiring treatment	mild, reversible and manageable with oral replacement	reversible but requiring IV replacement	irreversible, requiring continued replacement
Also consider Acidosis, Bicarbonate, Hypocalcemia, Hypophosphatemia.					
Urinary frequency/urgency	normal	increase in frequency or nocturia up to 2 x normal	increase > 2 x normal but < hourly	hourly or more with urgency, or requiring catheter	-
Urinary retention	normal	hesitancy or dribbling, but no significant residual urine; retention occurring during the immediate postoperative period	hesitancy requiring medication or occasional in/out catheterization (<4 x per week), or operative bladder atony requiring indwelling catheter beyond immediate postoperative period but for < 6 weeks	requiring frequent in/out catheterization (≥ 4 x per week) or urological intervention (e.g., TURP, suprapubic tube, urethrotomy)	bladder rupture
Urine color change (not related to other dietary or physiologic cause e.g., bilirubin, concentrated urine, hematuria)	normal	asymptomatic, change in urine color	-	-	-
Vaginal bleeding is graded in the HEMORRHAGE category.					
Vaginitis (not due to infection)	none	mild, not requiring treatment	moderate, relieved with treatment	severe, not relieved with treatment, or ulceration not requiring surgery	ulceration requiring surgery
Renal/Genitourinary-Other (Specify, _____)	none	mild	moderate	severe	life-threatening or disabling
SECONDARY MALIGNANCY					
Secondary Malignancy-Other (Specify type, _____) excludes metastastic tumors	none	-	-	-	present
SEXUAL/REPRODUCTIVE FUNCTION					
Dyspareunia is graded in the PAIN category.					
Dysmenorrhea is graded in the PAIN category.					
Erectile impotence	normal	mild (erections impaired but satisfactory)	moderate (erections impaired, unsatisfactory for intercourse)	no erections	-
Female sterility	normal	-	-	sterile	-
Femininization of male is graded in the ENDOCRINE category.					

	Grade				
Adverse Event	**0**	**1**	**2**	**3**	**4**
Irregular menses (change from baseline)	normal	occasionally irregular or lengthened interval, but continuing menstrual cycles	very irregular, but continuing menstrual cycles	persistent amenorrhea	-
Libido	normal	decrease in interest	severe loss of interest	-	-
Male infertility	-	-	Oligospermia (low sperm count)	Azoospermia (no sperm)	-
Masculinization of female is graded in the ENDOCRINE category.					
Vaginal dryness	normal	mild	requiring treatment and/or interfering with sexual function, dyspareunia	-	-
Sexual/Reproductive Function-Other (Specify, _____)	none	mild	moderate	severe	disabling
SYNDROMES (not included in previous categories)					
Acute vascular leak syndrome is graded in the CARDIOVASCULAR (GENERAL) category.					
ARDS (Adult Respiratory Distress Syndrome) is graded in the PULMONARY category.					
Autoimmune reactions are graded in the ALLERGY/IMMUNOLOGY category.					
DIC (disseminated intravascular coagulation) is graded in the COAGULATION category.					
Fanconi's syndrome is graded as Urinary electrolyte wasting in the RENAL/GENITOURINARY category.					
Renal tubular acidosis is graded as Urinary electrolyte wasting in the RENAL/GENITOURINARY category.					
Stevens-Johnson syndrome (erythema multiforme) is graded in the DERMATOLOGY/SKIN category.					
SIADH (syndrome of inappropriate antidiuretic hormone) is graded in the ENDOCRINE category.					
Thrombotic microangiopathy (e.g., thromboitic thrombocytopenic purpura/TTP or hemolytic uremic syndrom/HUS) is graded in the COAGULATION category.					
Tumor flare	none	mild pain not interfering with function	moderate pain; pain or analgesics interfering with function, but not interfering with activities of daily living	severe pain; pain or analgesics interfering with function and interfering with activities of daily living	Disabling
Also consider Hypercalcemia.					
Note: Tumor flare is characterized by a constellation of symptoms and signs in direct relation to initiation of therapy (e.g., anti-estrogens/androgens or additional hormones). The symptoms/signs include tumor pain, inflammation of visible tumor, hypercalcemia, diffuse bone pain, and other electrolyte disturbances.					
Tumor lysis syndrome	absent	-	-	present	-
Also consider Hyperkalemia, Creatinine.					
Urinary electrolyte wasting (e.g., Fanconi's syndrome, renal tubular acidosis) is graded in the RENAL/GENITOURINARY category.					
Syndromes-Other (Specify, _____)	none	mild	moderate	severe	life-threatening or disabling

APPENDIX 13 - HEMATOLOGY NORMAL LABORATORY VALUES

BCIRG 006 (TAX GMA 302)
HEMATOLOGY NORMAL LABORATORY VALUES

Test	Lower limit	Upper limit	Units
Hemoglobin	12.00 (F)	15.60 (F)	g/dL
Platelets	150.00	400.00	10^9/L
White Blood Cells	4.00	10.00	10^9/L
Neutrophils	2.00	6.00	10^9/L
Lymphocytes	2.00	4.00	10^9/L
Monocytes	0.20	0.95	10^9/L
Eosinophils	0.04	0.60	10^9/L
Basophils	0.01	0.05	10^9/L
Atypical lymphocytes	< 0		

The table above is to inform you of the standard normal ranges which will be used to analyse hematological parameters for the study. The standard values have been taken from NCI Common Toxicity Criteria (Version 2.0) and Clinical Decisions for Lab Tests by B.E. Statland, published by Medical Economics Books, 1987 when not defined in the NCI Common Toxicity Criteria.