This supplement contains the following items:

1. Original protocol (Page 2 – 44), final protocol (Page 45 –89), amendment list (Page 90– 94).

2. Original statistical analysis plan (Page 95–106), final statistical analysis plan (Page 107 – 118), amendment list (Page 119– 120).

A multicenter, phase III randomized study of metronomic capecitabine maintenance after standard treatment in patients with operable triple–negative breast cancer

(Protocol code: SYSUCC–EBC–CHEMO–001)

(Coding description: Sun Yat–sen University Cancer Center–Early Breast Cancer–Chemotherapy–001)

Version: 1.0

Principle Investigators

Zhong Yu YUAN, M.D. and Xi WANG, M.D.

Sun Yat–sen University Cancer Center

Date of approved version: April 05, 2010

Table of Contents

1. SYNOPSIS OF THE STUDY

1.1 Objectives

This study is designed to compare the efficacy (disease–free survival, DFS) and safety of metronomic capecitabine maintenance for one year with observation after standard local and systemic treatment in patients with operable triple negative breast cancer (TNBC).

1.2 Study Design

This study is to be a multi–center, phase III, randomized controlled trial. The study will include the following two treatment arms: 684 subjects will be randomized in a 1:1 fashion (342 in each arm) to receive either metronomic capecitabine maintenance (experimental arm) or observation (control arm) until objective disease recurrence, protocol violation, intolerable toxicity, death, or withdrawal of consent. Subjects will be stratified by lymph node status (positive or negative). Subjects discontinuing from the active treatment phase will enter the follow–up phase during which survival information will be collected.

1.3 Main Inclusion/ Exclusion Criteria

Main Inclusion Criteria:

1) Female, aged ≥ 18 years old and ≤ 75years.
2) Histologically confirmed invasive ductal carcinoma, no specific type (NOS).
3) Pathologic stage $T_{1c–3}N_{0–2}M_0$.
4) estrogen receptor (ER)–/progesterone receptor (PR)–negative and human epidermal growth factor receptor 2 (HER2) negative (ER– and PR–negative is defined by lower than 1% immunohistochemistry staining; HER2–negative is define by IHC score 0,1 or 2 with HER2–fluorescence in situ hybridization negative).
5) Have completed adequate surgery, neo–/adjuvant chemotherapy and radiation therapy (if indicated).
6) Available results for contralateral mammography, chest X–ray, abdominal ultrasonography, 99mTc–bone scanning (required for patients with stage ⅡB–ⅢA disease) within 3 months before randomization.
7) Adequate organ function including bone marrow, renal function, hepatic function, et al.
8) Compliance with the study protocol.
9) Have provided written and signed informed consent.

Main Exclusion Criteria:

1) Patients with T4, including inflammatory carcinomas.
2) Patients with N3.
3) Previously diagnosed with other malignancies (not including cured cervical carcinoma *in situ*, cutaneous squamous cell carcinoma, and cutaneous basal cell carcinoma).
4) History of invasive breast cancer.
5) Patients who are receiving or will receive other biological agents or immunotherapy.
6) Severe dysfunction of the heart, lung, liver, or kidney.
7) Patients with malabsorption syndrome diseases impairing GI function, resection of stomach or small intestine, or who are unable to swallow capecitabine tablets.
8) Patients who are pregnant or who are unwilling to use contraception during the study period.
9) Known intolerance to capecitabine or allergy to its excipients.

1.4 Investigational Drug and Administration

Capecitabine group (experimental arm): Capecitabine will be administered at a dose of 650 mg/m^2 orally twice daily (ie, total daily dose = 1300 mg/m^2) continuous for one year, starting within 2

weeks of randomization.

1.5 Study Endpoints

The primary efficacy parameter, DFS, will be analyzed in the full analysis set (FAS) population.

The secondary efficacy parameters, including overall survival (OS), disease–free survival (DDFS), and locoregional recurrence–free survival (LRFS), will be analyzed in the FAS populations.

Safety and tolerability will be assessed using reporting of adverse events (AEs), graded according to NCI–CTC (version 4.0).

2. BACKGROUND

Breast cancer comprises a group of diseases that show genetic heterogeneity and biological diversity [1, 2], which could be classified into five subtypes distinguished by their gene expression profiles [3, 4], including luminal A , luminal B, HER2+, normal breast, and basal–like [5]. The genotype of breast cancer is established using complicated gene analysis, which unsuitable for formalin–fixed specimens. Immunohistochemistry–based classification (using ER, PR, HER2, and KI–67) is more widely used in clinical practice [6–8], revealing a group of breast cancers characterized by

negative expression of ER, PR, and HER2, termed as “triple–negative” breast cancer (TNBC) [9]. Basal–like breast cancer and TNBC are differently defined, and might overlap with each other. The majority of basal–like breast cancers are triple–negative. Therefore, TNBC is used as an alternative histopathological definition of basal–like breast cancer in clinical practice, as well as in the inclusion criteria of most clinical trials.

TNBC comprises approximately 15%–25% of breast cancer in women [10–14], and is considered an independent clinicopathological subtype, with special clinical, pathological, and molecular genetic characteristics. In terms of clinical characteristics, TNBC is more common among young patients, with a high risk of early (within 2 years after surgery) recurrence, distant metastasis, and death [15–19]. TNBC has a shorter median survival after first recurrence than other types of breast cancer, with most deaths occurring within the first 5 years [10]. Visceral metastasis (especially in the lung and brain) is more frequent than bone metastasis, which might be one of the major contributors to the poor prognosis of TNBC. Pathologically, TNBC is associated with the presence of high histological grade, invasive ductal carcinoma, a high proliferation index, and high expression of p53 and EGFR

[20-24]. Molecularly, gene expression profiles of TNBC have revealed its high molecular homology [1, 4, 9].

For hormone receptor positive breast cancer, anti–estrogen therapies have significantly reduced recurrence and death [25]. For HER2+ breast cancers, anti–HER2 therapies (e.g., trastuzumab) have also significantly reduced recurrence [26]. Currently, there are few targeted therapies for TNBC, and chemotherapy is the only effective strategy to reduce recurrence, which is another reason for the poor prognosis of TNBC. Endocrine therapy for HR+ breast cancer and anti–HER2 therapy for HER2+ breast cancer are all long–term maintenance therapies after standard treatment [27]. Therefore, we propose that a long–term effective maintenance treatment might significantly improve the outcome in patients with early TNBC.

Most TNBC is more chemosensitive than HR+ breast cancer. Traditional regimens tend to achieve a better response in patients with TNBC; however, the duration of the response usually dose not last long. TNBC is still characterized with dismal DFS, PFS, and OS [14, 28, 29]. Therefore, the aggressive biological behavior and the lack of effective risk–reducing treatment have both contributed to the poor prognosis of TNBC.

Metronomic chemotherapy is a relatively novel regimen using continuous and low–dose chemotherapeutic agents with short or no intervals. Browder and Klement, et al. reported the anti–tumor activity of metronomic chemotherapy for the first time. The novel pattern of dosage has a different mechanism compared with conventional dosage regimens by exerting anti–angiogenesis effects [30, 31]. In addition, metronomic chemotherapy also produces antitumor effects by upregulating anti–tumor immune response in the host [32]. Metronomic chemotherapy had achieved good efficacy with low toxicity in advanced breast cancer [33–36]. Considering that angiogenesis and immune surveillance escape are major mechanisms of tumor metastasis, metronomic chemotherapy might be a potential therapeutic option for operable TNBC with high risk of distant metastasis.

Capecitabine is an effective agent with good tolerability and is convenient for breast cancer [37–41], which makes it an optimal choice for long–term metronomic use. The most common adverse events of capecitabine include hand–foot syndrome (HFS), diarrhea, and stomatitis, which are non–life threatening and can be managed using eduction without impairing efficacy [40]. Two recent phase III trials (FinXX and USO), which enrolled all subtypes of

breast cancer, have shown by subgroup analysis that the addition of capecitabine to standard treatment significantly reduced the risk of relapse for TNBC, especially the risk of distant metastases.

In summary, high rate of distant metastases and lack of effective treatment are the major reasons for the poor prognosis of TNBC. As a novel model of treatment, metronomic chemotherapy might be effective for TNBC by targeting angiogenesis and immune escape. The good efficacy and tolerability of capecitabine make it an optimal drug for metronomic chemotherapy. Clinical studies have also demonstrated a reduced risk of relapse in patients with TNBC receiving capecitabine in addition to standard treatment. This study aims to evaluate the efficacy and safety of capecitabine metronomic chemotherapy after standard treatment in patients with early TNBC.

3. OBJECTIVES

3.1 Primary Endpoint

To compare the DFS in patients who are randomized at enrollment to treatment with metronomic capecitabine maintenance (experimental arm) with in observation arm (control arm).

DFS is defined as time from randomization to the first of any of

the following events:

1) Relapse of invasive breast cancer in the ipsilateral chest wall and regional lymph nodes
2) Distant metastases (histologically confirmed or clinically diagnosed)
3) Breast cancer related, non–breast cancer related or unknown deaths
4) Contralateral invasive breast cancer.

3.2 Secondary Endpoints

To compare the overall survival (OS), distant disease–free survival (DDFS), locoregional recurrence–free survival (LRFS) and safety between the experimental arm and observation arm. In addition, exploratory analysis will include biomarkers that predict the efficacy and toxicity of capecitabine.

OS is defined as time from randomization to death caused by any reason.

DDFS is defined as time from randomization to the first occurrence of any of the following events: Distant metastases, death caused by any reason, and contralateral invasive breast cancer (NEJM 2005; 353:2747).

LRFS is defined as time from randomization to locoregional invasive recurrence or death.

Safety: The frequency and severity degree of AEs were judged based on NCI CTC V4.0.

4. STUDY DESIGN

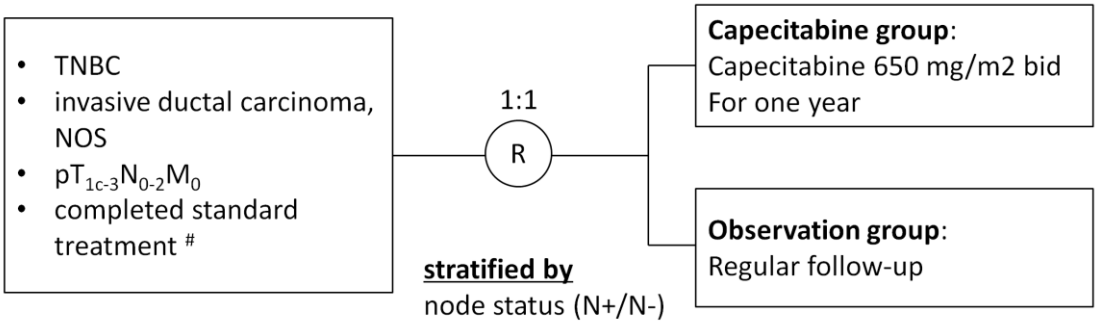
4.1 Summary of Design

This is a multi–center, phase III, randomized controlled study of metronomic capecitabine maintenance versus observation.

Approximately 684 subjects with TNBC will be randomized in a 1:1 fashion (342 in each arm) to receive treatment with either: Metronomic capecitabine maintenance (experimental arm); or observation (control arm) until objective disease recurrence, protocol violation, intolerable toxicity, death, or withdrawal of consent. Subjects will be stratified by lymph node status (positive or negative).

Subjects will participate in the study within 4 weeks after completion of standard curative treatment including surgery, neo-/adjuvant chemotherapy and radiotherapy. Patients in the two arms will be follow-up every 3 months using physical, laboratory, and radiological examinations according to the study protocol. This

study will be completed in approximately 72 months including 48 months for accrual and approximately 36 months of follow–up survival for the last subject enrolled. An overview of the study design is depicted below:



standard treatment including surgery, (neo)adjuvant chemotherapy and radiation therapy (if indicated)

Recommended chemotherapy regimens: According to the NCCN guidelines version 2010, recommended chemotherapy regimens and dosage are listed in Table 1. Dosages adjustment according to a patient's toleration will be allowed with no more than 25% reduction of standard dose. A minimum of four cycles of neo-/adjuvant chemotherapy should be delivered. For node–positive patients, chemotherapy regimens containing anthracyclines and taxanes are recommended.

Recommended indications for post–operative radiotherapy include: Involvement of ≥ four axillary nodes, primary tumor ≥ 5 cm in size, post breast conserving surgery, positive surgical margins,

involvement of internal mammary nodes (in selected cases), and involvement of 1–3 axillary nodes (in selected cases).

Table 1 Recommended chemotherapy regimen and dosage

Regimens(drugs)		Dose(mg/m^2)
CMF	cyclophosphamide/methotrexate/fluorouracil	500/ 40/ 600
AC	doxorubicin/cyclophosphamide	60/ 600
EC	epirubicin/cyclophosphamide	75–90/ 600
FAC	5–fluorouracil/doxorubicin/cyclophosphamide	500/ 50/ 500
FEC	5–fluorouracil/epirubicin/cyclophosphamide	500/ 75–90/ 500
TAC	docetaxel/doxorubicin/cyclophosphamide	75/ 50/ 500
TEC	docetaxel/epirubicin/cyclophosphamide	75/ 75/ 500
AC–P	doxorubicin/cyclophosphamide→paclitaxel weekly or every 3–weeks	60/ 600→80 (qw), 175 (q3w)
EC–P	epirubicin/cyclophosphamide→paclitaxel weekly or every 3–weeks	90/ 600→80 (qw), 175 (q3w)
AC–wP	doxorubicin/cyclophosphamide→paclitaxel (Dose–dense)	60/ 600→175 (q2w)
FEC–T	5–fluorouracil/epirubicin/cyclophosphamide→docetaxel, every 3 weeks	500/ 75–90/ 500→75
TC	docetaxel/cyclophosphamide	75/ 600

4.2 Randomization

On verification of the inclusion and exclusion criteria, eligible patients will be randomized using the method of stratified permuted blocks to receive metronomic capecitabine maintenance or observation in a 1:1 ratio. Patients will be stratified according to lymph node status (negative *vs.* positive). A computerized number generator in the SAS software (version 8.01) generate a randomization table, the results of which were placed in sequentially numbered opaque envelopes and remained concealed

until after enrollment.

Central randomization will be performed. When a suitable patient is to be enrolled into the study, the Investigator site will contact principal investigator (PI) site, and will be informed over the telephone at the time of individual patient enrollment what the treatment allocation is, and to which treatment arm the patient has been randomized. This is a multicenter study to be conducted at approximately 15 study sites.

4.3 Capecitabine Administration

4.3.1 Initiating Dose

The approved dose of capecitabine was 1250 mg/m^2 bid, days 1–14 every 21 days. However, the dose of capecitabine for metronomic chemotherapy is uncertain, particularly in the adjuvant setting. Several studies suggested that capecitabine at 650 mg/m^2 bid, continuously for one year in metastatic breast cancer had lower toxicity and was well tolerated [42–44]. The initiate dose of capecitabine was 650 mg/m^2 bid, continuously for one year.

Body surface area is calculated from height and body weight. Given that the height and weight of Chinese woman are 150–180 cm and 40–80 kg, respectively, their body surface area lie between

1.30 m^2 and 2.0 m^2. Combining the availability of capecitabine in China with the convenience of patients, the daily actual dose will be decided upon by using the **Table 2**.

Table 2 The daily dose of capecitabine

Body surface area (m^2)	Total Daily Dose (mg)	Morning dose (mg)	Evening dose (mg)
1.30–1.32	1690–1716	1000	500
1.33–1.71	1729–2223	1000	1000
1.72–2.0	2236–2600	1500	1000

4.3.2 Dose Adjustment

The most common AEs of capecitabine is HFS, and grading of HFS is listed in **Table 3**. Studies suggested that almost all AEs could improve after dose modification [45]. Dose adjustment of capecitabine in patients who experience HFS is listed in **Table 4**. Note, because of lower dose in patients with body surface areas 1.3–1.32 m^2, only one dose reduction of capecitabine is allowed, from 1500 mg to 1000mg (morning 500mg, evening 500mg). In addition, once a dose has been reduced for a subject, all subsequent doses should be administered at that dose, unless further dose reduction is required. Dose reescalation is not permitted. If dosage delay occurs because of AEs, whether to continue treatment should be determined by the investigator by

balancing the benefit and risk on an individual basis. Regardless of the cause of the delay, patients who discontinue dosage for more than 4 weeks should terminate treatment and withdraw from the trial.

Table 3 Grading of HFS Caused by Capecitabine

Grade	Manifestation
1	Numbness, tingling sensation, erythema of hands and/or feet that cause painless swelling or discomfort without affecting daily activities
2	Painful erythema or swelling of hands and/or feet that affect daily activities
3	Wet desquamation, ulceration, blistering, severe pain of hands and/or feet, and/or unable to work or perform daily activities

Table 4 Dose Adjustment of Capecitabine

Grade	Dose modification of capecitabine
1	Dose modifications are not recommended
2	–First appearance: Interrupt therapy until resolved to grade 0 or 1 and maintain the dose level for the next treatment at 100% –Second appearance: Interrupt therapy until resolved to grade 0 or 1 and maintain the dose level for the next treatment at 75% –Third appearance: Interrupt therapy until resolved to grade 0 or 1 and maintain the dose level for the next treatment at 50% –Fourth appearance: Discontinue therapy permanently
3	–First appearance: Interrupt therapy until resolved to grade 0 or 1 and begin the next cycle at 75% of the starting dose –Second appearance: Interrupt therapy until resolved to grade 0 or 1 and begin the next cycle at 50% of the starting dose –Third appearance: Discontinue therapy permanently
4	First appearance: Discontinue therapy permanently, or if the physician

	deems it to be in the patient's best interest to continue, interrupt until resolved to grade 0 or 1 and begin the next cycle at 50% of the starting dose

4.3.3 Concomitant and Prophylactic Medication

In addition to HFS, the other common toxicities of capecitabine are diarrhea and stomatitis. Symptom–relieving treatment can be given by investigators according to clinical need and should be recorded. Dose adjustment is not required for patients with mild to moderate hepatic impairment. Currently there are no data on the pharmacokinetics of capecitabine in patients with renal dysfunction (as evaluated by serum creatinine levels).

Mild myelosuppression related to capecitabine and the predominance of its active metabolic enzymes inside tumor cells mean that, hematologic toxicities of grade ≤ 2 can be managed according to clinical routine without discontinuation of capecitabine. For patients experiencing hematologic toxicities of grade ≥ 3, capecitabine should be interrupted until resolved to grade 0. Treatment should be terminated if dosage interruption occurs for more than 4 weeks. Treatment should be terminated if patients experience two episodes of grade ≥ 3 hematological toxicities consecutively, with any episode resulting in drug discontinuation for more than 2 weeks. Patient should be followed–up after termination

of therapy and toxicities and prognosis should be recorded.

5. SELECTION OF SUBJECTS

5.1 Enrollment

All patients meeting the inclusion criteria must be provided with detailed information about this study and written informed consent for participation must be obtained. The patients will then be randomly assigned into the observation arm or capecitabine arm using a random number table, and the assignment will be recorded on the case report form (CRF)by investigators. Analysis will be stratified by lymph node status (N0 or N+).

5.2 Inclusion Criteria

Patients must fulfill **ALL** of the following criteria to be eligible for study
enrollment and randomization.

1) Female, aged ≥ 18 years old and ≤ 75 years old.
2) Histologically confirmed invasive ductal carcinoma, no specific type (NOS).
3) Pathologic stage $T_{1c–3}N_{0–2}M_0$.
4) ER–, PR–, and HER2–negative (ER– and PR–negative is

defined by lower than 1% immunohistochemistry (IHC) staining; HER2 negative is define by an IHC score of 0, 1 or 2, with HER2–fluorescence *in situ* hybridization negative).

5) Have completed adequate surgery, neo-/adjuvant chemotherapy and radiation therapy (if indicated).
6) Available results of contralateral mammography, chest X–ray, abdominal ultrasonography, and ^{99m}Tc–bone scanning within 3 months before randomization.
7) Adequate organ function:
    a) Bone marrow: ANC ≥ 1.5 *10^9/L; platelet count ≥ 100*10^9/L; hemoglobin ≥ 10 g/dL
    b) Renal function: Serum creatinine ≤ 1.5×ULN by local laboratory
    c) Hepatic function: Total bilirubin ≤ 1.5×ULN; AST ≤ 1.5×ULN, ALT ≤ 1.5*ULN
8) Compliance with study protocol.
9) Providing written and signed informed consent.

5.3 Exclusion Criteria

Patients meeting **ANY** of the following criteria are not eligible for study enrollment and randomization.

1) Patients with T4, including inflammatory carcinomas.
2) Patients with N3.
3) Previously diagnosed other malignancies (not including cured cervical carcinoma in situ, cutaneous squamous cell carcinoma, and cutaneous basal cell carcinoma).
4) History of invasive breast cancer.
5) Patients who are receiving or will receive other biological agents or immunotherapy.
6) Severe dysfunction of the heart, lung, liver, or kidney.
7) Patients with malabsorption syndrome diseases impairing GI function, resection of stomach or small intestine, or unable to swallow capecitabine tablets.
8) Patient who are pregnant or who are unwilling to use contraception during the study period.
9) Known intolerance to capecitabine or allergy to its excipients.

5.4 Discontinuation Criteria

1) Recurrence of breast cancer.
2) Development for serious advent event develops.
3) Patients desire to withdraw from the study.
4) Patients are unable to receive treatment or follow–up

according to the study protocol.

5) Patients receive other anti–tumor treatment or other treatment that might affect the study results without the consent of the investigators.

6) Dosage discontinuation for more than 28 days.

6. STUDY PROTOCOL

6.1 Study Drug:

Capecitabine (Xeloda®, Roche, Basel, Switzerland), 500mg per tablet. The treatment schedule is described in section 4.3.

6.2 Assessment and Follow–up

The schedule of assessment during treatment and follow–up are showed in Appendix 1.

6.2.1 Baseline Assessment

Baseline assessment should complete within 1 week before enrollment).

- ✓ Screening form. Patients who meet all inclusion criteria and do not meet any exclusion criteria are eligible for this study. Investigators must complete a screening form at baseline.

- ✓ Medical history and clinical examination. Medical history, including risk factors for cardiac disease and their medical history of nervous system diseases must be collected before enrollment.
- ✓ Complete blood count, hepatic function (including AST, ALT, T–Bil, D–Bil, TP, and ALB), renal function (including BUN and Cr), serum electrolytes (including K^+ and $Ca2^+$), serum LDH, AKP, and blood glucose.
- ✓ Electrocardiogram and echocardiogram;
- ✓ Serum CEA and CA153;
- ✓ Imaging studies including chest X–ray, and abdominal ultrasonography. A bone ECT scan is recommended for patients with disease of stage ≥ IIb, unexplained bone pain, or elevated serum ALP
- ✓ 10 mL of peripheral blood will be collected for biomarker analysis.

6.2.2 Assessment during Treatment (repeated every 3 months)

Assessment during Treatment are to repeat every 3 monthsS.

- ✓ Physical examination and vital signs;
- ✓ Complete blood count, hepatic function, and renal function;

- ✓ Serum CEA and CA153;
- ✓ Electrocardiogram;
- ✓ Abdominal ultrasonography.

6.2.3 Assessment during Follow–up

Several randomized studies have shown that regular examination comprising bone scans, liver US, chest X–rays, and blood tests could not improve the survival and quality of life (QoL) of patients, compared with routine physical examination [46, 47]. Therefore, every 3 months during follow–up, physical examination and mammography are required for asymptomatic patients in both arms. However, this is the minimum requirement specified by the protocol, and investigators are allowed to perform additional evaluations according to the individual situation of the patients.

6.2.4 Follow–up

Follow–up of patients in both arms will be initiated after randomization and will be repeated every 3 months (± 28 days) during the first 2 years after randomization. Patients in the capecitabine arm are allowed to take medicine at home but must return to the study site every 3 months (± 28 days) for follow–up.

Follow–up will be repeated every 6 months (± 28 days) during the 3rd to 5th year after randomization, and then annually thereafter.

Diagnosis of relapse will be established on clinical manifestation, radiological findings, and/or histological evidence. If the diagnosis of relapse is based on clinical symptoms without laboratory or radiological evidence, other supporting evidence should be collected as much as possible. After a diagnosis of recurrence is established, the sites and date of relapse should be recorded.

Diagnosis of relapse could also be established if the treatment strategy is altered based on the hypothesis of relapse, even without adequate evidence.

Chest wall relapse: Defined as soft tissue recurrence in the area comprising the sternum as the middle line, the clavicle as the upper margin, the rib as the lower margin, and the posterior axillary line as lateral margin.

Regional relapse: Defined as relapse in the area of the supraclavicular fossa, subclavicular area, ipsilateral internal mammary area, and/or ipsilateral axillary lymph nodes. Tissue biopsy should be performed whenever possible.

Distant metastases: Cutaneous or subcutaneous metastasis should be supported by histological or cytological evidence. Bone

metastasis should be supported by imaging studies (e.g., X–ray or MR). Metastasis in the lung, liver, or brain should be supported by CT or MRI.

7. SAFETY ASSESSMENT

7.1 Adverse Events

7.1.1 Definition of Adverse Events

An AE is defined as any untoward medical occurrence during the period from randomization to the 28th day after the last dose or to the most recent follow–up, regardless of causal attribution with the study drug. An AE can be any of the following: A symptom, a sign, abnormal examination results, or a disease, which may occur at any time since the initiation of treatment.

An AE should be accurately recorded during the study, including its time, severity, duration, management, and prognosis.

7.1.2 Severity of Adverse Events

Severity of AEs is graded according to NCI CTCAE 4.0 (Appendix 2). Grades of AEs that are not listed in Appendix 2 are as follows:

- Mild: An effect on the daily function of subjects.
- Moderate: A mild effect on the daily function of subjects.

- Severe: A significant effect on the daily function of subjects.

7.1.3 Association between AEs and Study Treatment

The relationship between AEs and the study drug should be assessed by investigators according to the following criteria:

Definitely related: An AE that follows a reasonable temporal sequence from administration of the study intervention, follows a known or expected response pattern to the suspected intervention, and is confirmed by improvement on stopping and reappearance of the event on repeated exposure

Probably related: An AE that follows a reasonable temporal sequence from administration of the study intervention, follows a known or expected response pattern to the suspected intervention, but that could readily have been produced by the patient’s clinical conditions or other treatments.

Probably unrelated: An AE that does not follow a reasonable temporal sequence from administration of the study intervention, does not follow a known or expected response pattern to the suspected intervention, and could readily have been produced by the patient’s clinical conditions or other treatments.

Unrelated: An AE that does not follow a reasonable temporal

sequence from administration of the study intervention, but follows a known or expected response pattern to other treatments, and could readily have been produced by the patient's clinical conditions or other treatments. The AE can be relieved by improvement of the clinical conditions or stopping other treatments, and reappears after repeating other treatments.

Unable to determine: An AE that does not follow a reasonable temporal sequence from administration of the study intervention, but follows a known or expected response pattern to the study intervention, and could readily have been produced by other treatments.

7.2 Serious Adverse Events (SAEs)

7.2.1 Definition of SAEs

- Results in death.
- Is life–threatening.
- Requires or prolongs hospitalization.
- Causes persistent or significant disability or incapacity.
- Results in congenital anomalies or birth defects.

7.2.2 SAE Reporting

Any SAEs occurring during the study or follow–up should be reported to the PI and ethics committee by telephone within 24 hours regardless of their causal relationship with the study drug. The PI is responsible of reporting SAEs to the State Food and Drug Administration (SFDA) (also to the drug manufacturer within 24 hours if the SAE is considered to be related to the study drug).

8. STATISTICS

Additional details of the analysis will be provided in the statistical analysis plan.

8.1 Statistical Methods

The primary endpoint is DFS, defined as time from randomization to the first of breast cancer recurrence or death from any reason. The secondary endpoints include OS, DDFS, LRFS, and safety.

Efficacy analyses will be based on the FAS population, defined as all randomized patients excluding those who withdraw informed consent before protocol treatment, or who had no follow–up data after randomization. Safety analyses will be based on the safety analyses set (SAS) population, defined as all randomized patients who initiated the protocol treatment and who undergo safety assessment.

For the efficacy analysis, PFS, OS, DDFS, and LRFS will be analyzed using the Kaplan–Meier method and will be compared using the log–rank test. The hazard ratio and corresponding 95% confidence interval will be calculated using stratified Cox proportional hazard regression.

AEs and SAEs will be summarized by arm. The incidence of grade 3 HFS will be compared between the two arms using Fisher's exact test.

For continuous variables, the distribution, mean, median, standard deviation, and interquartile rang (IQR) will be calculated and compared using a *t*–test or non–parametric test. For categorical variables, the number and percentage will be presented in contingency table data and compared using the chi–squared test or Fisher's exact test.

All statistical tests are two–sided with a *P* value of < 0.05 being considered statistically significant.

8.2 Sample Size

The assumptions for sample size calculations are as follows: 5–year DFS is 68% in the control arm [10, 13, 28], and 80% in the experimental arm. The estimated periods of enrollment and

follow–up will be 48 and 36 months, respectively. The design is based on a 2–sided log–rank test with alpha = 0.05, power = 90%, and an interim analysis when the last one patient has completed 12 months of follow–up. The dropout rate is assumed to be 20%. Approximately 684 patients (342 patients in each arm) will be enrolled.

9. ETHICS

9.1 Informed Consent

Before enrollment, study physicians are responsible for a complete and comprehensive presentation to patients of the study purpose, the properties of the drug, its possible side effects and potential risks. Patients should be informed of their rights, the risk, and the benefit. It should be emphasized that they can withdraw from the trial at any stage without affecting their subsequent treatment. Subjects should be promptly informed of any updates of the study, and a renewed informed consent to continue in the study should be obtained. Patients should sign the informed consent in duplicate with their name and date. One copy is given to the patient and the other is kept in the study archives.

9.2 Ethic Policies and Regulations

The investigator will ensure that this study is conducted in full conformance with the principles of the “Declaration of Helsinki” as well as “Guideline for Good Clinical Practice (GCP)” and relevant laws and regulations of the SFDA, whichever affords the greater protection to the individual.

The study will be initiated only after the protocol is approved by the ethics committee of the Sun Yat–sen University Cancer Center. Any changes to the protocol during the study should be reported to the ethics committee and filed.

9.3 Protocol Modifications

All protocol modifications must be submitted to the Independent Ethics Committee (IEC). Approval must be awaited before any changes can be implemented, except for changes necessary to eliminate an immediate hazard to the trial patients, or when the change involve only logistical or administrative aspects of the trial.

10. QUALITY ASSURANCE

To ensure accordance with study protocols, physicians are asked to strictly follow the requirements of GCP throughout the trial, to

achieve standard procedures, accurate data, and reliable conclusions. Specific requirements are as follows:

- ✓ Obtain informed consent that is signed by each subject or their agents.
- ✓ Complete the case report form (CRF) as required.
- ✓ Follow–up on schedule.
- ✓ Keep complete records of laboratory examinations, clinical records, and the original medical documents of the subjects.

11. DATA PROCESSING AND STORAGE

11.1 Case Report Form (CRF)

The CRF will be completed by investigators in a timely manner to ensure the accuracy and timeliness of the content. Generally, the CRF should not be altered. If there are any errors to be corrected, the original record should be crossed out with a horizontal line, and the modified text should be signed and dated. The completed CRFs are reviewed by the quality control officer for data input. No further modification of CRFs is allowed once the database is locked.

11.2 Database Establishment

Statisticians will have questions in the CRFs checked with investigators, who should reply and return the CRFs promptly.

Statisticians should establish the database in a timely manner, and the data will be locked by investigators, statisticians, and research assistants after the database has been reviewed. To ensure data security, an non-permitted person cannot modify the data, and the data must be backed up.

11.3 Data Storage

Investigators should keep the data intact. According to the principle of GCP in China, research data should be stored for at least five years.

12. REFERENCES

1. Perou CM, Sorlie T, Eisen MB, et al.: Molecular portraits of human breast tumours. Nature 2000, 406: 747–52.
2. Sorlie T. Molecular portraits of breast cancer: tumour subtypes as distinct disease entities. Eur J Cancer 2004; 406: 2667–75.
3. Sorlie T, Perou CM, Tibshirani R, et al.: Gene expression patterns of breast carcinomas distinguish tumor subclasses with clinical implications. Proc Natl Acad Sci USA 2001, 98: 10869–74.
4. Sotiriou C, Neo SY, McShane LM, et al. Breast cancer classification and prognosis based on gene expression profiles from a population–based study. PNAS, 2003; 100(18): 10393–8.
5. Weigelt B, Glas AM, Wessels LF, et al. Gene expression profiles of primary breast tumors maintained in distant metastases. Proc Natl Acad Sci USA. 2003; 100(26): 15901–5.
6. Abd El–Rehim DM, Pinder SE, Paish CE, et al. Expression of luminal and basal cytokeratins in human breast carcinoma. J Pathol. 2004; 203(2): 661–71.
7. Jacquemier J, Ginestier C, Rougemont J, et al. Protein expression profiling identifies subclasses of breast cancer and predicts prognosis. Cancer Res. 2005; 65(3): 767–79.
8. Nielsen TO, Hsu FD, Jensen K, et al. Immunohistochemical and clinical characterization of the basal–like subtype of invasive breast carcinoma. Clin Cancer Res. 2004; 10: 5367–74.
9. Sørlie T, Tibshirani R, Parker J, et al. Repeated observation of breast tumor subtypes in independent gene expression data sets. PNAS, 2003; 100(14): 8418–23.
10. Dent R, Trudeau M, Pritchard KI et al. Triple–negative breast cancer: clinical features and patterns of recurrence. Clin. Cancer Res. 2007; 13; 4429–34.
11. Tischkowitz M, Brunet JS, Begin LR et al. Use of immunohistochemical markers can refine prognosis in triple negative breast cancer. BMC Cancer 2007; 7; 134.

12. Bauer KR, Brown M, Cress RD, et al. Descriptive analysis of estrogen receptor (ER)–negative, progesterone receptor (PR)–negative, and HER2–negative invasive breast cancer, the so–called triple–negative phenotype: a population–based study from the California cancer Registry. Cancer 2007; 109; 1721–8.
13. Haffty BG, Yang Q, Reiss M, et al. Locoregional Relapse and Distant Metastasis in Conservatively Managed Triple Negative Early-Stage Breast Cancer. J Clin Oncol 2006;24(36):5652-7.
14. Carey LA, Dees EC, Sawyer L, et al: The triple negative paradox: Primary tumor chemosensitivity of breast cancer subtypes. Clin Cancer Res, 2007; 13:2329–34.
15. Abd El–Rehim DM, Ball G, Pinder SE, et al. High–throughput protein expression analysis using tissue microarray technology of a large well–characterised series identifies biologically distinct classes of breast cancer confirming recent cDNA expression analyses. Int J Cancer 2005; 116: 340–50.
16. van de Rijn M, Perou CM, Tibshirani R, et al. Expression of cytokeratins 17 and 5 identifi es a group of breast carcinomas with poor clinical outcome. Am J Pathol 2002; 161:1991–96.
17. Banerjee S, Reis–Filho JS, Ashley S, et al. Basal–like breast carcinomas: clinical outcome and response to chemotherapy. J Clin Pathol 2006; 59: 729–35.
18. Rodriguez–Pinilla SM, Sarrio D, Honrado E, et al. Prognostic significance of basal–like phenotype and fascin expression in node negative invasive breast carcinomas. Clin Cancer Res 2006; 12: 1533–39.
19. Tsuda H, Takarabe T, Hasegawa F, et al. Large, central cellular zones indicating myoepithelial tumor differentiation in high–grade invasive ductal carcinomas as markers of predisposition to lung and brain metastases. Am J Surg Pathol 2000; 24: 197–202.
20. Korsching E, Packeisen J, Agelopoulos K, et al. Cytogenetic alterations and cytokeratin expression patterns in breast cancer: integrating a new model of breast differentiation into cytogenetic pathways of breast carcinogenesis. Lab Invest 2002; 82: 1525–33.

21. Fulford LG, Easton DF, Reis–Filho JS, et al. Specific morphological features predictive for the basal phenotype in grade 3 invasive ductal carcinoma of breast. Histopathology 2006; 49; 22–34.
22. Livasy CA, Karaca G, Nanda R et al. Phenotypic evaluation of the basal–like subtype of invasive breast carcinoma. Mod. Pathol. 2006; 19; 264–71.
23. Jacquemier J, Padovani L, Rabayrol L et al. Typical medullary breast carcinomas have a basal／myoepithelial phenotype. J. Pathol. 2005; 207; 260–8.
24. Rakha EA, El–Sayed ME, Green AR et al. Prognostic markers in triple–negative breast cancer. Cancer 2007; 109; 25–32.
25. Early Breast Cancer Trialists' Collaborative Group (EBCTCG). Effects of chemotherapy and hormonal therapy for early breast cancer on recurrence and 15–year survival: an overview of the randomised trials. Lancet. 2005; 365(9472): 1687–717.
26. Dahabreh IJ, Linardou H, Siannis F, Fountzilas G, Murray S. Trastuzumab in the adjuvant treatment of early–stage breast cancer: a systematic review and meta–analysis of randomized controlled trials. Oncologist. 2008;13(6):620–30.
27. Cole BF, Gelber RD, Gelber S, Coates AS, Goldhirsch A. Polychemotherapy for early breast cancer: an overview of the randomised clinical trials with quality-adjusted survival analysis. Lancet 2001; 358(9278): 277-86.
28. Liedtke C, Mazouni C, Hess KR, et al. Response to neoadjuvant therapy and long–term survival in patients with triple–negative breast cancer. J Clin Oncol. 2008; 26(8): 1275–81.
29. Rouzier R, Perou CM, Symmans WF, et al. Breast Cancer Molecular Subtypes Respond Differently to Preoperative Chemotherapy. Clin Cancer Res, 2005; 11(16): 5678–5685.
30. Browder T, Butterfield CE, Kräling BM, et al. Antiangiogenic scheduling of chemotherapy improves efficacy against experimental drug–resistant cancer. Cancer Res 2000; 60: 1878–86.
31. Klement G, Baruchel S, Rak J, et al. Continuous low–dose therapy with

824 vinblastine and vegf receptor–2 antibody induces sustained tumor
825 regression without overt toxicity. J Clin Invest 2000; 105: R15–24
826 32. Ghiringhelli F, Menard C, Puig PE, et al. Metronomic cyclophosphamide
827 regimen selectively depletes CD4+ CD25+ regulatory T cells and restores
828 T and NK effector functions in end stage cancer patient. Cancer Immunol
829 Immunother 2006; 56: 641–8.
830 33. Colleoni M, Rocca A, Sandri MT, et al. Low–dose oral methotrexate and
831 cyclophosphamide in metastatic breast cancer: antitumor activity and
832 correlation with vascular endothelial growth factor levels. Ann Oncol 2002;
833 13: 73–80.
834 34. Colleoni M, Orlando L, Sanna G, et al. Metronomic low–dose oral
835 cyclophosphamide and methotrexate plus or minus thalidomide in
836 metastatic breast cancer: antitumor activity and biological effects. Ann
837 Oncol 2006; 17: 232–8.
838 35. Orlando L, Cardillo A, Rocca A, et al. Prolonged clinical benefit with
839 metronomic chemotherapy in patients with metastatic breast cancer.
840 Anticancer Drugs 2006; 17: 961–7.
841 36. Emmenegger U, Man S, Shaked Y, et al. A comparative analysis of
842 low–dose metronomic cyclophosphamide reveals absent or low–grade
843 toxicity on tissues highly sensitive to the toxic effects of maximum tolerated
844 dose regimens. Cancer Res 2004; 64: 3994–4000.
845 37. Blum JL, Dieras V, Lo Russo PM et al. Multicenter, phase II study of
846 capecitabine in taxane–pretreated metastatic breast carcinoma patients.
847 Cancer 2001; 92: 1759–68.
848 38. Reichardt P, Von Minckwitz G, Thuss–Patience PC et al. Multicenter phase
849 II study of oral capecitabine (Xeloda) in patients with metastatic breast
850 cancer relapsing after treatment with a taxane–containing therapy. Ann
851 Oncol 2003; 14: 1227–33.
852 39. O’Shaughnessy J, Miles D, Vukelja S et al. Superior survival with
853 capecitabine plus docetaxel combination therapy in
854 anthracyclinepretreated patients with advanced breast cancer: Phase III
855 trial results. J Clin Oncol 2002; 20: 2812–23.
856 40. Leonard R, O'Shaughnessy J, Vukelja S, et al. Detailed analysis of a

randomized Phase III trial: can the tolerability of capecitabine plus docetaxel be improved without compromising its survival advantage? Ann Oncol. 2006; 17(9): 1379–85.

41. Blum JL, Jones SE, Buzdar AU, et al. Multicenter phase II study of capecitabine in paclitaxel– refractory metastatic breast cancer. J Clin Oncol. 1999; 17(2): 485–93
42. Martin M, Calvo L, Martinez N, et al. Standard Versus Continuous Administration of Capecitabine in Metastatic Breast Cancer (GEICAM/2009-05): A Randomized, Noninferiority Phase II Trial With a Pharmacogenetic Analysis. Oncologist 2015; 20(2): 111-2.
43. Stockler MR, Harvey VJ, Francis PA, et al. Capecitabine versus classical cyclophosphamide, methotrexate, and fluorouracil as first-line chemotherapy for advanced breast cancer. J Clin Oncol 2011; 29: 4498-504.
44. Blum JL, Jones SE, Buzdar AU. Blum J, Jones S, Buzdar A. Capecitabine (Xeloda) in 162 patients with paclitaxel-pretreated mbc: updated results and analysis of dose modification. Eur J Cancer 2001; 37(6): S190-S.
45. Rosselli Del Turco M, Palli D, Cariddi A, et al. Intensive diagnostic follow–up aftertreatment of primary breast cancer. A randomized tial. National Research Council Project on Breast Cancer follow–up. JAMA 1994; 271:1593–7
46. Rosselli Del Turco M, Palli D, Cariddi A, Ciatto S, Pacini P, Distante V. Intensive diagnostic follow-up after treatment of primary breast cancer. A randomized trial. National Research Council Project on Breast Cancer follow-up. Jama 1994; 271(20): 1593-7.
47. Impact of follow–up testing on survival and health–related quality of life in breast cancer patients. A multicenter randomized controlled trial. The GIVIO Investigator. JAMA 1994; 271(20):1587–92.

13. APPENDIX 1

Schedule of the Study

		Baseline (Within 7 days before enrollment)	Treatment	End of treatment	Follow–up (d)
Informed consent		√			
Screening form		√			
Blood samples		√	√	√	as clinically indicated
Medical records and examinations					
Observation items	Medical history(a)	√			
	Physical examination	√	√	√	√
	Vital signs	√	√	√	√
	ECOG score	√	√	√	√
	Complete blood count	√	√	√	as clinically indicated
	Blood chemistry test(b)	√	√	√	as clinically indicated
	Coagulation function (4 items)	√	√	√	as clinically indicated
	CEA/CA153	√	√	√	as clinically indicated
	Electrocardiogram	√	√	√	as clinically indicated
	Echocardiogram	√		√	as clinically indicated
	Imaging examination(c)	√		√	as clinically indicated
	Adverse events		√	√	√
	Concomitant medication		√	√	√
	Assessment of recurrence and metastasis		√	√	√

NOTES:

a) Medical history: Risk factors of heart disease and history of nervous system disease should be recorded;

b) Blood chemistry tests: Hepatic function (AST, ALT, T–Bil, D–Bil, TP, and ALB), renal function (BUN and Cr), serum electrolytes (K^+ and $Ca2^+$), serum LDH and AKP, and serum glucose.

c) Imaging examination: Including chest X–ray and abdominal ultrasonography. Bone ECT scan is recommended for patients with disease of stage ≥ IIB, unexplained bone pain, or elevated serum ALP; Mammography is repeated annually;

d) Follow–up is repeated every 3 months (± 28 days) during the first 2 years after randomization, every 6 months (± 28 days) during the 3rd to 5th year after randomization, and then annually thereafter.

APPENDIX 2 Common Terminology Criteria for Adverse Events v4.0

- ✓ The CTCAE v4.0 manual can be found at the following URL: http://ctep.cancer.gov/forms/CTCAEv4.pdf.

A multicenter, phase III randomized study of metronomic capecitabine maintenance after standard treatment in patients with operable triple–negative breast cancer

(Protocol code: SYSUCC–EBC–CHEMO–001)

(Coding description: Sun Yat–sen University Cancer Center–Early Breast Cancer–Chemotherapy–001)

Version: 3.0

Principle Investigators

Zhong Yu YUAN, M.D. and Xi WANG, M.D.

Sun Yat–sen University Cancer Center

Date of approved version: January 19, 2017

Table of Contents

1. SYNOPSIS OF THE STUDY

1.1 Objectives

This study is designed to compare the efficacy (disease–free survival, DFS) and safety of metronomic capecitabine maintenance for one year with observation after standard local and systemic treatment in patients with operable triple negative breast cancer (TNBC).

1.2 Study design

This study is to be a multi–center, phase III, randomized controlled trial. The study will include the following two treatment arms: 424 subjects will be randomized in a 1:1 fashion (212 in each arm) to receive treatment with either: Metronomic capecitabine maintenance (experimental arm); or observation (control arm) until objective disease recurrence, protocol violation, intolerable toxicity, death, or withdrawal of consent. Subjects will be stratified by lymph node status (positive or negative). Subjects discontinuing from the active treatment phase will enter the follow–up phase during which survival information will be collected.

1.3 Main Inclusion/Exclusion Criteria

Main Inclusion Criteria:

1) Female, aged ≥ 18 years old and ≤ 70years.
2) Histologically confirmed invasive ductal carcinoma, no specific type (NOS).
3) Stage Ib–IIIc disease (N3 disease with involvement of the supraclavicular or internal mammary lymph nodes will be excluded).
4) estrogen receptor (ER)–/progesterone receptor (PR)–negative and human epidermal growth factor receptor 2 (HER2) negative (ER– and PR–negative is defined by lower than 1% immunohistochemistry staining; HER2–negative is define by IHC score 0,1 or 2 with HER2–fluorescence in situ hybridization negative).
5) Have completed adequate surgery, neo-/adjuvant chemotherapy and radiation therapy (if indicated).
6) Available results for contralateral mammography, chest X–ray, abdominal ultrasonography, 99mTc–bone scanning (required for patients with stage IIb–IIIc disease) within 3 months before randomization.
7) Adequate organ function including bone marrow, renal function, hepatic function.

8) Compliance with the study protocol.

9) Have provided written and signed informed consent.

Main Exclusion Criteria:

1) Inflammatory or bilateral breast cancer.

2) Previously diagnosed with other malignancies (not including cured cervical carcinoma *in situ*, cutaneous squamous cell carcinoma, and cutaneous basal cell carcinoma).

3) History of invasive breast cancer.

4) Patients who are receiving or will receive other biological agents or immunotherapy.

5) Severe dysfunction of the heart, lung, liver, or kidney.

6) Patients with malabsorption syndrome diseases impairing GI function, resection of stomach or small intestine, or who are unable to swallow capecitabine tablets.

7) Patients who are pregnant or who are unwilling to use contraception during the study period.

8) Known intolerance to capecitabine or allergy to its excipients.

1.4 Investigational Drug and Administration

Capecitabine group (experimental arm): Capecitabine will be administered at a dose of 650 mg/m^2 orally twice daily (total daily

dose = 1300 mg/m^2) continuously for one year, starting within 2 weeks from randomization.

1.5 Study Endpoints

The primary efficacy parameter, DFS, will be analyzed in the full analysis set (FAS) population.

The secondary efficacy parameters, including overall survival (OS), disease–free survival (DDFS), and locoregional recurrence–free survival (LRFS), will be analyzed in FAS population.

Safety and tolerability will be assessed using reporting of adverse events (AEs), graded according to NCI–CTC (version 4.0).

2. BACKGROUND

Breast cancer comprises a group of diseases that show genetic heterogeneity and biological diversity [1, 2], which could be classified into five subtypes distinguished by their gene expression profiles [3, 4], including luminal A , luminal B, HER2+, normal breast, and basal–like [5]. The genotype of breast cancer is established using complicated gene analysis, which unsuitable for formalin–fixed specimens. Immunohistochemistry–based classification (using ER, PR, HER2, and KI–67) is more widely used in clinical

practice [6–8], revealing a group of breast cancers characterized by negative expression of ER, PR, and HER2, termed as “triple–negative” breast cancer (TNBC) [9]. Basal–like breast cancer and TNBC are differently defined, and might overlap with each other. The majority of basal–like breast cancers are triple–negative. Therefore, TNBC is used as an alternative histopathological definition of basal–like breast cancer in clinical practice, as well as in the inclusion criteria of most clinical trials.

TNBC comprises approximately 15%–25% of breast cancer in women [10–14], and is considered an independent clinicopathological subtype, with special clinical, pathological, and molecular genetic characteristics. In terms of clinical characteristics, TNBC is more common among young patients, with a high risk of early (within 2 years after surgery) recurrence, distant metastasis, and death [15–19]. TNBC has a shorter median survival after first recurrence than other types of breast cancer, with most deaths occurring within the first 5 years [10]. Visceral metastasis (especially in the lung and brain) is more frequent than bone metastasis, which might be one of the major contributors to the poor prognosis of TNBC. Pathologically, TNBC is associated with the presence of high histological grade, invasive ductal carcinoma,

a high proliferation index, and high expression of p53 and EGFR [20-24]. Molecularly, gene expression profiles of TNBC have revealed its high molecular homology [1, 4, 9].

For hormone receptor positive breast cancer, anti–estrogen therapies have significantly reduced recurrence and death [25]. For HER2+ breast cancers, anti–HER2 therapies (e.g., trastuzumab) have also significantly reduced recurrence [26]. Currently, there are few targeted therapies for TNBC, and chemotherapy is the only effective strategy to reduce recurrence, which is another reason for the poor prognosis of TNBC. Endocrine therapy for HR+ breast cancer and anti–HER2 therapy for HER2+ breast cancer are all long–term maintenance therapies after standard treatment [27]. Therefore, we propose that a long–term effective maintenance treatment might significantly improve the outcome in patients with early TNBC.

Most TNBC is more chemosensitive than HR+ breast cancer. Traditional regimens tend to achieve a better response in patients with TNBC; however, the duration of the response usually dose not last long. TNBC is still characterized with dismal DFS, PFS, and OS [14, 28, 29]. Therefore, the aggressive biological behavior and the lack of effective risk–reducing treatment have both contributed to

the poor prognosis of TNBC.

Metronomic chemotherapy is a relatively novel regimen using continuous and low–dose chemotherapeutic agents with short or no intervals. Browder and Klement, et al. reported the anti–tumor activity of metronomic chemotherapy for the first time. The novel pattern of dosage has a different mechanism compared with conventional dosage regimens by exerting anti–angiogenesis effects [30, 31]. In addition, metronomic chemotherapy also produces antitumor effects by upregulating anti–tumor immune response in the host [32]. Metronomic chemotherapy had achieved good efficacy with low toxicity in advanced breast cancer [33–36]. Considering that angiogenesis and immune surveillance escape are major mechanisms of tumor metastasis, metronomic chemotherapy might be a potential therapeutic option for operable TNBC with high risk of distant metastasis.

Capecitabine is an effective agent with good tolerability and is convenient for breast cancer [37–41], which makes it an optimal choice for long–term metronomic use. The most common adverse events of capecitabine include hand–foot syndrome (HFS), diarrhea, and stomatitis, which are non–life threatening and can be managed using eduction without impairing efficacy [40]. Two recent

phase III trials (FinXX and USO), which enrolled all subtypes of breast cancer, have shown by subgroup analysis that the addition of capecitabine to standard treatment significantly reduced the risk of relapse for TNBC, especially the risk of distant metastases.

In summary, high rate of distant metastases and lack of effective treatment are the major reasons for the poor prognosis of TNBC. As a novel model of treatment, metronomic chemotherapy might be effective for TNBC by targeting angiogenesis and immune escape. The good efficacy and tolerability of capecitabine make it an optimal drug for metronomic chemotherapy. Clinical studies have also demonstrated a reduced risk of relapse in patients with TNBC receiving capecitabine in addition to standard treatment. This study aims to evaluate the efficacy and safety of capecitabine metronomic chemotherapy after standard treatment in patients with early TNBC.

3. OBJECTIVES

3.1 Primary Endpoint

To compare the DFS in patients who are randomized at enrollment to treatment with metronomic capecitabine maintenance (experimental arm) with in observation arm (control arm).

DFS is defined as time from randomization to the first of any of the following events:

5) Relapse of invasive breast cancer in the ipsilateral chest wall and regional lymph nodes
6) Distant metastases (histologically confirmed or clinically diagnosed)
7) Breast cancer related, non–breast cancer related or unknown deaths
8) Contralateral invasive breast cancer

3.2 Secondary Endpoints

To compare the overall survival (OS), distant disease–free survival (DDFS), locoregional recurrence–free survival (LRFS) and safety between the experimental arm and observation arm. In addition, exploratory analysis will include biomarkers that predict the efficacy and toxicity of capecitabine.

OS is defined as time from randomization to death caused by any reason.

DDFS is defined as time from randomization to the first occurrence of any of the following events: Distant metastases, death caused by any reason, and contralateral invasive breast

cancer (NEJM 2005; 353:2747).

LRFS is defined as time from randomization to locoregional invasive recurrence or death.

Safety: The frequency and severity degree of AEs were judged based on NCI CTC V4.0.

4. STUDY DESIGN

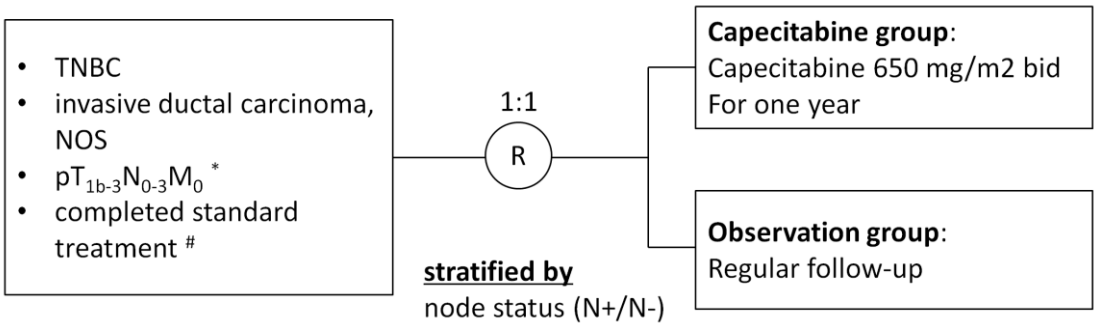
4.1 Summary of Design

This is a multi–center, phase III, randomized controlled study of metronomic capecitabine maintenance versus observation.

Approximately 424 subjects with TNBC will be randomized in a 1:1 fashion (212 in each arm) to receive treatment with either: Metronomic capecitabine maintenance (experimental arm); or observation (control arm) until objective disease recurrence, protocol violation, intolerable toxicity, death, or withdrawal of consent. Subjects will be stratified by lymph node status (positive or negative).

Subjects will participate in the study within 4 weeks after completion of standard curative treatment including surgery, neo-/adjuvant chemotherapy and radiotherapy. Patients in the two arms will be followed-up every 3 months using physical, laboratory,

and radiological examinations according to the study protocol. This study will be completed in approximately 108 months including 72 months for accrual and approximately 36 months of follow–up survival for the last subject enrolled. An overview of the study design is depicted below:



*N3: Not including internal mammary or supraclavicular nodes involvement
standard treatment including surgery, (neo)adjuvant chemotherapy and radiation therapy (if indicated)

Recommended chemotherapy regimens: According to the NCCN guidelines version 2010, recommended chemotherapy regimens and dosages are listed in Table 1. Dosage adjustment according to a patient's toleration will be allowed with no more than 25% reduction of the standard dose. A minimum of four cycles of neo-/adjuvant chemotherapy should be delivered. For node–positive patients, chemotherapy regimens containing anthracyclines and taxanes are recommended.

Recommended indications for post–operative radiotherapy

include: Involvement of ≥ four axillary nodes, primary tumor≥ 5 cm in size, post breast conserving surgery, positive surgical margins, involvement of internal mammary nodes (in selected cases), and involvement of 1–3 axillary nodes (in selected cases).

Table 1 Recommended chemotherapy regimen and dosage

Regimens(drugs)		Dose(mg/m^2)
CMF	cyclophosphamide/methotrexate/fluorouracil	500/ 40/ 600
AC	doxorubicin/cyclophosphamide	60/ 600
EC	epirubicin/cyclophosphamide	75–90/ 600
FAC	5–fluorouracil/doxorubicin/cyclophosphamide	500/ 50/ 500
FEC	5–fluorouracil/epirubicin/cyclophosphamide	500/ 75–90/ 500
TAC	docetaxel/doxorubicin/cyclophosphamide	75/ 50/ 500
TEC	docetaxel/epirubicin/cyclophosphamide	75/ 75/ 500
AC–P	doxorubicin/cyclophosphamide→weekly or every–3–week paclitaxel	60/ 600→80 (qw), 175 (q3w)
EC–P	epirubicin/cyclophosphamide→weekly or every–3–week paclitaxel	90/ 600→80 (qw), 175 (q3w)
AC–wP	doxorubicin/cyclophosphamide→paclitaxel (Dose–dense)	60/ 600→175 (q2w)
FEC–T	5–fluorouracil/epirubicin/cyclophosphamide→docetaxel, every 3 weeks	500/ 75–90/ 500→75
TC	docetaxel/cyclophosphamide	75/ 600

4.2 Randomization

On verification of inclusion and exclusion criteria, eligible patients will be randomized using the method of stratified permuted blocks to receive metronomic capecitabine maintenance or observation in a 1:1 ratio. Patients will be stratified according to lymph node status

(negative *vs.* positvie). A computerized number generator in the SAS Software (version 8.01) will generate a randomization table, the results of which were placed in sequentially numbered opaque envelopes and remained concealed until after enrollment.

Central randomization will be performed. When a suitable patient is to be enrolled into the study, the Investigator site will contact principal investigator (PI) site, and will be informed over the telephone system at the time of individual patient enrollment what the treatment allocation is, and to which treatment arm the patient has been randomized. This is a multicenter study to be conducted at approximately 15 study sites.

4.3 Capecitabine Administration

4.3.1 Initiating Dose

The approved dose of capecitabine was 1250 mg/m^2 bid, days 1–14 every 21 days. However, the dose of capecitabine for metronomic chemotherapy is uncertain, particularly in the adjuvant setting. Some small sample studies suggested that capecitabine at 650 mg/m^2 bid, continuously for one year in metastatic breast cancer had lower toxicity and was well tolerated [42–44]. The initiate dose of capecitabine was 650 mg/m^2 bid, continuously for

one year.

Body surface area is calculated from height and body weight. Given that the height and weight of Chinese woman are 150–180 cm and 40–80 kg, respectively, their body surface area lie between 1.30 m^2 and 2.0 m^2. Combining the availability of capecitabine in China with the convenience of patients, the daily actual dose will be decided upon by using the **Table 2**.

Table 2 The daily dose of capecitabine

Body surface area (m^2)	Total Daily Dose (mg)	Morning dose (mg)	Evening dose (mg)
1.30–1.32	1690–1716	1000	500
1.33–1.71	1729–2223	1000	1000
1.72–2.0	2236–2600	1500	1000

4.3.2 Dose Adjustment

The most common AEs of capecitabine is HFS, and grading of HFS is listed in **Table 3**. Studies suggested that almost all AEs could improve after dose modification [45]. Dose adjustment of capecitabine in patients who experience HFS is listed in **Table 4**. Note, because of lower dose in patients with body surface areas 1.3–1.32 m^2, only one dose reduction of capecitabine is allowed, from 1500 mg to 1000mg (morning 500mg, evening 500mg). In addition, once a dose has been reduced for a subject, all

subsequent doses should be administered at that dose, unless further dose reduction is required. Dose reescalation is not permitted. If dosage delay occurs because of AEs, whether to continue treatment should be determined by the investigator by balancing the benefit and risk on an individual basis. Regardless of the cause of the delay, patients who discontinue dosage for more than 4 weeks should terminate treatment and withdraw from the trial.

Table 3 Grading of HFS Caused by Capecitabine

Grade	Manifestation
1	Numbness, tingling sensation, erythema of hands and/or feet that cause painless swelling or discomfort without affecting daily activities
2	Painful erythema or swelling of hands and/or feet that affect daily activities
3	Wet desquamation, ulceration, blistering, severe pain of hands and/or feet, and/or unable to work or perform daily activities

Table 4 Dose Adjustment of Capecitabine

Grade	Dose modification of capecitabine
1	Dose modifications are not recommended
2	–First appearance: Interrupt therapy until resolved to grade 0 or 1 and maintain the dose level for the next treatment at 100% –Second appearance: Interrupt therapy until resolved to grade 0 or 1 and maintain the dose level for the next treatment at 75% –Third appearance: Interrupt therapy until resolved to grade 0 or 1 and maintain the dose level for the next treatment at 50% –Fourth appearance: Discontinue therapy permanently

3	–First appearance: Interrupt therapy until resolved to grade 0 or 1 and begin the next cycle at 75% of the starting dose –Second appearance: Interrupt therapy until resolved to grade 0 or 1 and begin the next cycle at 50% of the starting dose –Third appearance: Discontinue therapy permanently
4	First appearance: Discontinue therapy permanently, or if the physician deems it to be in the patient's best interest to continue, interrupt until resolved to grade 0 or 1 and begin the next cycle at 50% of the starting dose

4.3.3 Concomitant and Prophylactic Medication

In addition to HFS, the other common toxicities of capecitabine are diarrhea and stomatitis. Symptom–relieving treatment can be given by investigators according to clinical need and should be recorded. Dose adjustment is not required for patients with mild to moderate hepatic impairment. Currently there are no data on the pharmacokinetics of capecitabine in patients with renal dysfunction (as evaluated by serum creatinine levels).

Mild myelosuppression related to capecitabine and the predominance of its active metabolic enzymes inside tumor cells mean that, hematologic toxicities of grade ≤ 2 can be managed according to clinical routine without discontinuation of capecitabine. For patients experiencing hematologic toxicities of grade ≥ 3, capecitabine should be interrupted until resolved to grade 0. Treatment should be terminated if dosage interruption occurs for

more than 4 weeks. Treatment should be terminated if patients experience two episodes of grade ≥ 3 hematological toxicities consecutively, with any episode resulting in drug discontinuation for more than 2 weeks. Patient should be followed–up after termination of therapy and toxicities and prognosis should be recorded.

5. SELECTION OF SUBJECTS

5.1 Enrollment

All patients meeting the inclusion criteria must be provided with detailed information about this study and written informed consent for participation must be obtained. The patients will then be randomly assigned into the observation arm or capecitabine arm using a random number table, and the assignment will be recorded on the case report form (CRF) by investigators. Analysis will be stratified by lymph node status (N0 or N+).

5.2 Inclusion Criteria

Patients must fulfill **ALL** of the following criteria to be eligible for study enrollment and randomization.

1) Female, aged ≥ 18 years old and ≤ 70years old.
2) Histologically confirmed invasive ductal carcinoma, no specific

type (NOS).

3) Stage Ib–IIIc (N3 not including internal mammary or supraclavicular nodes involvement).
4) ER–, PR–, and HER2–negative (ER– and PR–negative is defined by lower than 1% immunohistochemistry (IHC) staining; HER2 negative is define by an IHC score of 0,1 or 2 with HER2–fluorescence *in situ* hybridization negative).
5) Have completed adequate surgery, neo-/adjuvant chemotherapy and radiation therapy (if indicated).
6) Available results of contralateral mammography, chest X–ray, abdominal ultrasonography, and ^{99m}Tc–bone scanning within 3 months before randomization.
7) Adequate organ function:
    d) Bone marrow: ANC ≥ 1.5 *10^9/L; platelet count ≥ 100*10^9/L; hemoglobin ≥ 10 g/dL
    e) Renal function: serum creatinine ≤ 1.5×ULN by local laboratory
    f) Hepatic function: total bilirubin ≤ 1.5×ULN; AST ≤ 1.5×ULN, ALT ≤ 1.5*ULN
8) Compliance with study protocol.
9) Providing written informed and signed consent.

5.3 Exclusion Criteria

Patients meeting **ANY** of the following criteria are not eligible for study enrollment and randomization.

1) Inflammatory or bilateral breast cancer.
2) Other previously diagnosed other malignancies (not including cured cervical carcinoma *in situ*, cutaneous squamous cell carcinoma, and cutaneous basal cell carcinoma).
3) History of invasive breast cancer.
4) Patients who are receiving or will receive other biological agents or immunotherapy.
5) Severe dysfunction of the heart, lung, liver, or kidney.
6) Patients with malabsorption syndrome diseases impairing GI function, resection of stomach or small intestine, or unable to swallow capecitabine tablets.
7) Patient who are pregnant or who are unwilling to use contraception during the study period.
8) Known intolerance to capecitabine or allergy to its excipients.

5.4 Discontinuation Criteria

1) Recurrence of breast cancer.

2) Development of serious AEs.

3) Patients desire to withdraw from the study.

4) Patients are unable to receive treatment or follow–up according to the study protocol.

5) Patients receive other anti–tumor treatment or other treatment that mighty affect the study results without the consent of the investigators.

6) Dosage discontinuation for more than 28 days.

6. STUDY PROTOCOL

6.1 Study Drug:

Capecitabine (Xeloda®, Roche, Basel, Switzerland), 500mg per tablet. The treatment schedule is described in section 4.3

6.2 Assessment and Follow–up

The schedule of assessment during treatment and follow–up are shown in Appendix 1.

6.2.1 Baseline Assessment

Baseline assessment should complete within 1 week before enrollment.

- ✓ Screening form. Patients who meet all inclusion criteria and do not meet any exclusion criteria are eligible for this study. Investigators must complete a screening form at baseline.
- ✓ Medical history and clinical examination. Medical history, including risk factors for cardiac disease and their medical history of nervous system diseases must be collected before enrollment.
- ✓ Complete blood count, hepatic function (including AST, ALT, T–Bil, D–Bil, TP, and ALB), renal function (including BUN and Cr), serum electrolytes (including K^+ and Ca^{2+}), serum LDH, AKP, and blood glucose.
- ✓ Electrocardiogram and echocardiogram;
- ✓ Serum CEA and CA153;
- ✓ Imaging studiesy including chest X–ray, and abdominal ultrasonography. A Bone ECT scan is recommended for patients with disease of stage ≥ IIB, unexplained bone pain, or elevated serum ALP
- ✓ 10 mL of peripheral blood was collected for biomarker analysis.

6.2.2 Assessment during Treatment

Assessment during treatment are to repeat every 3 months.

- ✓ Physical examination and vital signs;
- ✓ Complete blood count, hepatic function, and renal function;
- ✓ Serum CEA and CA153;
- ✓ Electrocardiogram;
- ✓ Abdominal ultrasonography.

6.2.3 Assessment during Follow–up

Several randomized studies have shown that regular examination comprising bone scans, liver US, chest X–rays, and blood tests could not improve the survival and quality of life (QoL) of patients, compared with routine physical examination [46, 47]. Therefore, every 3 months during follow–up, physical examination and mammography are required for asymptomatic patients in both arms. However, this is the minimum requirement specified by the protocol, and investigators are allowed to perform additional evaluations according to the individual situation of the patients.

6.2.4 Follow–up

Follow–up of patients in both arms will be initiated after randomization and will be repeated every 3 months (± 28 days)

during the first 2 years after randomization. Patients in the capecitabine arm are allowed to take medicine at home but must return to the study site every 3 months (± 28 days) for follow–up. Follow–up will be repeated every 6 months (± 28 days) during the 3rd to 5th year after randomization, and then annually thereafter.

Diagnosis of relapse will be established on clinical manifestation, radiological findings, and/or histological evidence. If the diagnosis of relapse is based on clinical symptoms without laboratory or radiological evidence, other supporting evidence should be collected as much as possible. After a diagnosis of recurrence is established, the sites and date of relapse should be recorded.

Diagnosis of relapse could also be established if the treatment strategy is altered based on the hypothesis of relapse, even without adequate evidence.

Chest wall relapse: Defined as soft tissue recurrence in the area comprising the sternum as the middle line, the clavicle as the upper margin, the rib as the lower margin, and the posterior axillary line as lateral margin.

Regional relapse: Defined as relapse in the area of the supraclavicular fossa, subclavicular area, ipsilateral internal mammary area, and/or ipsilateral axillary lymph nodes. Tissue

biopsy should be performed whenever possible.

Distant metastases: Cutaneous or subcutaneous metastasis should be supported by histological or cytological evidence. Bone metastasis should be supported by imaging studies (e.g., X–ray or MR). Metastasis in the lung, liver, or brain should be supported by CT or MRI.

7. SAFETY ASSESSMENT

7.1 Adverse Events

7.1.1 Definition of Adverse Events

An AE is defined as any untoward medical occurrence during the period from randomization to the 28th day after the last dose or to the most recent follow–up, regardless of causal attribution with the study drug. An AE can be any of the following: A symptom, a sign, abnormal examination results, or a disease, which may occur at any time since the initiation of treatment.

An AE should be accurately recorded during the study, including its time, severity, duration, management, and prognosis.

7.1.2 Severity of AEs

Severity of AEs is graded according to NCI CTCAE 4.0 (Appendix

2). Grades of AEs that are not listed in Appendix 2 are as follows:

- Mild: An effect on the daily function of subjects.
- Moderate: A mild effect on the daily function of subjects.
- Severe: A significant effect on the daily function of subjects.

7.1.3 Association between AEs and Study Treatment

The relationship between AEs and the study drug should be assessed by investigators according to the following criteria:

Definitely related: An AE that follows a reasonable temporal sequence from administration of the study intervention, follows a known or expected response pattern to the suspected intervention, and is confirmed by improvement on stopping and reappearance of the event on repeated exposure

Probably related: An AE that follows a reasonable temporal sequence from administration of the study intervention, follows a known or expected response pattern to the suspected intervention, but that could readily have been produced by the patient’s clinical conditions or other treatments.

Probably unrelated: An AE that does not follow a reasonable temporal sequence from administration of the study intervention, does not follow a known or expected response pattern to the

suspected intervention, and could readily have been produced by the patient's clinical conditions or other treatments.

Unrelated: An AE that does not follow a reasonable temporal sequence from administration of the study intervention, but follows a known or expected response pattern to other treatments, and could readily have been produced by the patient's clinical conditions or other treatments. The AE can be relieved by improvement of the clinical conditions or stopping other treatments, and reappears after repeating other treatments.

Unable to determine: An AE that does not follow a reasonable temporal sequence from administration of the study intervention, but follows a known or expected response pattern to the study intervention, and could readily have been produced by other treatments.

7.2 Serious Adverse Events (SAEs)

7.2.1 Definition of SAEs

- Results in death.
- Is life–threatening.
- Requires or prolongs hospitalization.
- Causes persistent or significant disability or incapacity.

- Results in congenital anomalies or birth defects.

7.2.2 SAEs Reporting

Any SAEs occurring during the study or follow–up should be reported to the PI and ethics committee by telephone within 24 hours regardless of their causal relationship with the study drug. The PI is responsible of reporting SAEs to the State Food and Drug Administration (SFDA) (also to the drug manufacturer within 24 hours if the SAE is considered to be related to the study drug).

8. STATISTICS

Additional details of the analysis will be provided in the statistical analysis plan.

8.1 Statistical Methods

The primary endpoint is DFS, defined as time from randomization to the first of breast cancer recurrence or death from any reason. The secondary endpoints include OS, DDFS, LRFS, and safety.

Efficacy analyses will be based on the FAS population, defined as all randomized patients excluding those who withdraw informed consent before protocol treatment, or who had no follow–up data after randomization. Safety analyses will be based on the safety

analyses set (SAS) population, defined as all randomized patients who initiated the protocol treatment and who undergo safety assessment.

For the efficacy analysis, PFS, OS, DDFS, and LRFS will be analyzed using the Kaplan–Meier method and will be compared using the log–rank test. The hazard ratio and corresponding 95% confidence interval will be calculated using stratified Cox proportional hazard regression.

AEs and SAEs will be summarized by arm. The incidence of grade 3 HFS will be compared between the two arms using Fisher’s exact test.

For continuous variables, the distribution, mean, median, standard deviation, and interquartile rang (IQR) will be calculated and compared using a *t*–test or non–parametric test. For categorical variables, the number and percentage will be presented in contingency table data and compared using the chi–squared test or Fisher's exact test.

All statistical tests are two–sided with a *P* value of < 0.05 being considered statistically significant.

8.2 Sample Size

The assumptions for sample size calculations as the follows: 5–year DFS is 68% in the control arm [10, 13, 28], and 80% in the experimental arm. The estimated period of enrollment and follow–up will be 72 and 36 months, respectively. The design is based on a 2–sided log–rank test with alpha = 0.05, power = 80%. The dropout rate is assumed to be 9%. Approximately 424 patients (212 patients in each arm) will be enrolled.

9. Ethics

9.1 Informed Consent

Before enrollment, study physicians are responsible for a complete and comprehensive presentation to patients of the study purpose, the properties of the drug, its possible side effects and potential risks. Patients should be informed of their rights, risk, and benefit. It should be emphasized that they can withdraw from the trial at any stage of the trial without affecting their subsequent treatment. Subjects should be promptly informed of any updates of the study, and a renewed informed consent to continue in the study should be obtained. Patients should sign the informed consent in duplicate with their name and date. The two copies are given to the patient and kept in study archives, respectively.

9.2 Ethic Policies and Regulations

The investigator will ensure that this study is conducted in full conformance with the principles of the “Declaration of Helsinki” as well as “Guideline for Good Clinical Practice (GCP)” and relevant laws and regulations of the SFDA, whichever affords the greater protection to the individual.

The study will be initiated only after the protocol is approved by the ethics committee of the Sun Yat–sen University Cancer Center. Any changes to the protocol during the study should be reported to the ethics committee and filed.

9.3 Protocol Modifications

All protocol modifications must be submitted to the Independent Ethics Committee (IEC). Approval must be awaited before any changes can be implemented, except for changes necessary to eliminate an immediate hazard to the trial patients, or when the change involve only logistical or administrative aspects of the trial.

10. QUALITY ASSURANCE

To ensure accordance with study protocols, physicians are asked to

strictly follow the requirements of GCP throughout the trial, to achieve standard procedures, accurate data, and reliable conclusions. Specific requirements are as follows:

- ✓ Obtain informed consent that is signed by each subject or their agents.
- ✓ Complete the case report form (CRF) as required.
- ✓ Follow–up on schedule.
- ✓ Keep complete records of laboratory examinations, clinical records, and the original medical documents of the subjects.

11. DATA PROCESSING AND STORAGE

11.1 Case Report Form (CRF)

The CRF will be completed by investigators in a timely manner to ensure the accuracy and timeliness of the content. Generally, the CRF should not be altered. If there are any errors to be corrected, the original record should be crossed out with a horizontal line, and the modified text should be signed and dated. The completed CRFs are reviewed by the quality control officer for data input. No further modification of CRFs is allowed once the database is locked.

11.2 Database Establishment

Statisticians will have questions in the CRFs checked with investigators, who should reply and return the CRFs promptly. Statisticians should establish the database in a timely manner, and the data will be locked by investigators, statisticians, and research assistants after the database has been reviewed. To ensure data security, an non-permitted person cannot modify the data, and the data must be backed up.

11.3 Data Storage

Investigators should keep the data intact. According to the principle of GCP in China, research data should be stored for at least five years.

12. REFERENCES

1. Perou CM, Sorlie T, Eisen MB, et al.: Molecular portraits of human breast tumours. Nature 2000, 406: 747–752.
2. Sorlie T. Molecular portraits of breast cancer: tumour subtypes as distinct disease entities. Eur J Cancer 2004; 406: 2667–75.
3. Sorlie T, Perou CM, Tibshirani R, et al.: Gene expression patterns of breast carcinomas distinguish tumor subclasses with clinical implications. Proc Natl Acad Sci USA 2001, 98: 10869–10874.
4. Sotiriou C, Neo SY, McShane LM, et al. Breast cancer classification and prognosis based on gene expression profiles from a population–based study. PNAS, 2003; 100(18): 10393–10398
5. Weigelt B, Glas AM, Wessels LF, et al. Gene expression profiles of primary breast tumors maintained in distant metastases. Proc Natl Acad Sci USA. 2003; 100(26): 15901–5.
6. Abd El–Rehim DM, Pinder SE, Paish CE, et al. Expression of luminal and basal cytokeratins in human breast carcinoma. J Pathol. 2004; 203(2): 661–71.
7. Jacquemier J, Ginestier C, Rougemont J, et al. Protein expression profiling identifies subclasses of breast cancer and predicts prognosis. Cancer Res. 2005; 65(3): 767–79.
8. Nielsen TO, Hsu FD, Jensen K, et al. Immunohistochemical and clinical characterization of the basal–like subtype of invasive breast carcinoma. Clin Cancer Res. 2004; 10: 5367–5374.
9. Sørlie T, Tibshirani R, Parker J, et al. Repeated observation of breast tumor subtypes in independent gene expression data

sets. PNAS, 2003; 100(14): 8418–23.
10. Dent R, Trudeau M, Pritchard KI et al. Triple–negative breast cancer: clinical features and patterns of recurrence. Clin. Cancer Res. 2007; 13; 4429–4434.
11. Tischkowitz M, Brunet JS, Begin LR et al. Use of immunohistochemical markers can refine prognosis in triple negative breast cancer. BMC Cancer 2007; 7; 134.
12. Bauer KR, Brown M, Cress RD, et al. Descriptive analysis of estrogen receptor (ER)–negative, progesterone receptor (PR)–negative, and HER2–negative invasive breast cancer, the so–called triple–negative phenotype: a population–based study from the California cancer Registry. Cancer 2007; 109; 1721–1728.
13. Haffty BG, Yang Q, Reiss M, et al. Locoregional Relapse and Distant Metastasis in Conservatively Managed Triple Negative Early-Stage Breast Cancer. J Clin Oncol 2006;24(36):5652-7.
14. Carey LA, Dees EC, Sawyer L, et al: The triple negative paradox: Primary tumor chemosensitivity of breast cancer subtypes. Clin Cancer Res, 2007; 13:2329–2334.
15. Abd El–Rehim DM, Ball G, Pinder SE, et al. High–throughput protein expression analysis using tissue microarray technology of a large well–characterised series identifies biologically distinct classes of breast cancer confirming recent cDNA expression analyses. Int J Cancer 2005; 116: 340–50.
16. van de Rijn M, Perou CM, Tibshirani R, et al. Expression of cytokeratins 17 and 5 identifi es a group of breast carcinomas with poor clinical outcome. Am J Pathol 2002; 161:1991–96.
17. Banerjee S, Reis–Filho JS, Ashley S, et al. Basal–like breast

carcinomas: clinical outcome and response to chemotherapy. J Clin Pathol 2006; 59: 729–35.

18. Rodriguez–Pinilla SM, Sarrio D, Honrado E, et al. Prognostic significance of basal–like phenotype and fascin expression in node negative invasive breast carcinomas. Clin Cancer Res 2006; 12: 1533–39.
19. Tsuda H, Takarabe T, Hasegawa F, et al. Large, central cellular zones indicating myoepithelial tumor differentiation in high–grade invasive ductal carcinomas as markers of predisposition to lung and brain metastases. Am J Surg Pathol 2000; 24: 197–202.
20. Korsching E, Packeisen J, Agelopoulos K, et al. Cytogenetic alterations and cytokeratin expression patterns in breast cancer: integrating a new model of breast differentiation into cytogenetic pathways of breast carcinogenesis. Lab Invest 2002; 82: 1525–33.
21. Fulford LG, Easton DF, Reis–Filho JS, et al. Specific morphological features predictive for the basal phenotype in grade 3 invasive ductal carcinoma of breast. Histopathology 2006; 49; 22–34.
22. Livasy CA, Karaca G, Nanda R et al. Phenotypic evaluation of the basal–like subtype of invasive breast carcinoma. Mod. Pathol. 2006; 19; 264–271.
23. Jacquemier J, Padovani L, Rabayrol L et al. Typical medullary breast carcinomas have a basal／myoepithelial phenotype. J. Pathol. 2005; 207; 260–268.
24. Rakha EA, El–Sayed ME, Green AR et al. Prognostic markers in triple–negative breast cancer. Cancer 2007; 109; 25–32.

25. Early Breast Cancer Trialists' Collaborative Group (EBCTCG). Effects of chemotherapy and hormonal therapy for early breast cancer on recurrence and 15–year survival: an overview of the randomised trials. Lancet. 2005; 365(9472): 1687–717.
26. Dahabreh IJ, Linardou H, Siannis F, Fountzilas G, Murray S. Trastuzumab in the adjuvant treatment of early–stage breast cancer: a systematic review and meta–analysis of randomized controlled trials. Oncologist. 2008;13(6):620–30.
27. Cole BF, Gelber RD, Gelber S, Coates AS, Goldhirsch A. Polychemotherapy for early breast cancer: an overview of the randomised clinical trials with quality-adjusted survival analysis. Lancet 2001; 358(9278): 277-86.
28. Liedtke C, Mazouni C, Hess KR, et al. Response to neoadjuvant therapy and long–term survival in patients with triple–negative breast cancer. J Clin Oncol. 2008; 26(8): 1275–81.
29. Rouzier R, Perou CM, Symmans WF, et al. Breast Cancer Molecular Subtypes Respond Differently to Preoperative Chemotherapy. Clin Cancer Res, 2005; 11(16): 5678–5685.
30. Browder T, Butterfield CE, Kräling BM, et al. Antiangiogenic scheduling of chemotherapy improves efficacy against experimental drug–resistant cancer. Cancer Res 2000; 60: 1878–86.
31. Klement G, Baruchel S, Rak J, et al. Continuous low–dose therapy with vinblastine and vegf receptor–2 antibody induces sustained tumor regression without overt toxicity. J Clin Invest 2000; 105: R15–24
32. Ghiringhelli F, Menard C, Puig PE, et al. Metronomic

cyclophosphamide regimen selectively depletes CD4+ CD25+ regulatory T cells and restores T and NK effector functions in end stage cancer patient. Cancer Immunol Immunother 2006; 56: 641–8.

33. Colleoni M, Rocca A, Sandri MT, et al. Low–dose oral methotrexate and cyclophosphamide in metastatic breast cancer: antitumor activity and correlation with vascular endothelial growth factor levels. Ann Oncol 2002; 13: 73–80.
34. Colleoni M, Orlando L, Sanna G, et al. Metronomic low–dose oral cyclophosphamide and methotrexate plus or minus thalidomide in metastatic breast cancer: antitumor activity and biological effects. Ann Oncol 2006; 17: 232–8.
35. Orlando L, Cardillo A, Rocca A, et al. Prolonged clinical benefit with metronomic chemotherapy in patients with metastatic breast cancer. Anticancer Drugs 2006; 17: 961–7.
36. Emmenegger U, Man S, Shaked Y, et al. A comparative analysis of low–dose metronomic cyclophosphamide reveals absent or low–grade toxicity on tissues highly sensitive to the toxic effects of maximum tolerated dose regimens. Cancer Res 2004; 64: 3994–4000.
37. Blum JL, Dieras V, Lo Russo PM et al. Multicenter, phase II study of capecitabine in taxane–pretreated metastatic breast carcinoma patients. Cancer 2001; 92: 1759–68.
38. Reichardt P, Von Minckwitz G, Thuss–Patience PC et al. Multicenter phase II study of oral capecitabine (Xeloda) in patients with metastatic breast cancer relapsing after treatment with a taxane–containing therapy. Ann Oncol 2003; 14: 1227–33.

39. O’Shaughnessy J, Miles D, Vukelja S et al. Superior survival with capecitabine plus docetaxel combination therapy in anthracyclinepretreated patients with advanced breast cancer: Phase III trial results. J Clin Oncol 2002; 20: 2812–23.
40. Leonard R, O'Shaughnessy J, Vukelja S, et al. Detailed analysis of a randomized Phase III trial: can the tolerability of capecitabine plus docetaxel be improved without compromising its survival advantage? Ann Oncol. 2006; 17(9): 1379–85.
41. Blum JL, Jones SE, Buzdar AU, et al. Multicenter phase II study of capecitabine in paclitaxel– refractory metastatic breast cancer. J Clin Oncol. 1999; 17(2): 485–93.
42. Stockler MR, Harvey VJ, Francis PA, et al. Capecitabine versus classical cyclophosphamide, methotrexate, and fluorouracil as first-line chemotherapy for advanced breast cancer. J Clin Oncol 2011; 29: 4498-504.
43. Martin M, Calvo L, Martinez N, et al. Standard Versus Continuous Administration of Capecitabine in Metastatic Breast Cancer (GEICAM/2009-05): A Randomized, Noninferiority Phase II Trial With a Pharmacogenetic Analysis. Oncologist 2015; 20(2): 111-2.
44. Blum JL, Jones SE, Buzdar AU. Blum J, Jones S, Buzdar A. Capecitabine (Xeloda) in 162 patients with paclitaxel-pretreated mbc: updated results and analysis of dose modification. Eur J Cancer 2001; 37(6): S190-S.
45. Rosselli Del Turco M, Palli D, Cariddi A, et al. Intensive diagnostic follow–up aftertreatment of primary breast cancer. A randomized tial. National Research Council Project on Breast

Cancer follow–up. JAMA 1994; 271:1593–7

46. Rosselli Del Turco M, Palli D, Cariddi A, Ciatto S, Pacini P, Distante V. Intensive diagnostic follow-up after treatment of primary breast cancer. A randomized trial. National Research Council Project on Breast Cancer follow-up. Jama 1994; 271(20): 1593-7

47. Impact of follow–up testing on survival and health–related quality of life in breast cancer patients. A multicenter randomized controlled trial. The GIVIO Investigator. JAMA 1994; 271:1587–92.

13. APPENDIX 1

Schedule of the study

		Baseline (within 7 days before enrollment)	Treatment	End of treatment	Follow–up (d)
Informed consent		√			
Screening form		√			
Blood samples		√	√	√	as clinically indicated
Medical records and examinations					
Observation items	Medical history(a)	√			
	Physical examination	√	√	√	√
	Vital signs	√	√	√	√
	ECOG score	√	√	√	√
	Complete blood count	√	√	√	as clinically indicated
	Blood chemistry test(b)	√	√	√	as clinically indicated
	Coagulation function (4 items)	√	√	√	as clinically indicated
	CEA/CA153	√	√	√	as clinically indicated
	Electrocardiogram	√	√	√	as clinically indicated

	Echocardiogram	√		√	as clinically indicated
	Imaging examination(c)	√		√	as clinically indicated
	Adverse events		√	√	√
	Concomitant medication		√	√	√
	Assessment of recurrence and metastasis		√	√	√

NOTES:

a) Medical history: Risk factors of heart disease and history of nervous system disease should be recorded;

b) Blood chemistry tests: Hepatic function (AST, ALT, T–Bil, D–Bil, TP, and ALB), renal function (BUN and Cr), serum electrolytes (K^+ and Ca^{2+}), serum LDH and AKP, and serum glucose.

c) Imaging examination: Including chest X–ray and abdominal ultrasonography. Bone ECT scan is recommended for patients with disease of stage ≥ IIB, unexplained bone pain, or elevated serum ALP; Mammography is repeated annually;

d) Follow–up is repeated every 3 months (± 28 days) during the first 2 years after randomization, every 6 months (± 28 days) during the 3rd to 5th year after randomization, and then annually thereafter.

APPENDIX 2 Common Terminology Criteria for Adverse Events v4.0

✓ The CTCAE v4.0 manual can be found at the following URL: http://ctep.cancer.gov/forms/CTCAEv4.pdf.

`

SYSUCC–001 Study Protocol Amendment List

page	item	Before amendment (Protocol Ver.**2.0** November 30, 2012)	After amendment (Protocol Ver.**3.0** January 9, 2017)	Reasons
cover		Ver.2.0 approved date: November 30, 2012	Ver.3.0 approved date: January 9, 2017	–
P76	8.2 Sample size	An interim analysis when the last one patient has completed 18 months of follow–up.	(None)	The number of DFS events was much lower than expected after the last one patient has completed 12 months of follow–up

1834

page	item	Before amendment (Protocol Ver.**1.0** April 5, 2010)	After amendment (Protocol Ver.**2.0** October 20, 2012)	Reasons
Cover	Cover	Ver.1.0 approved date: April 5, 2010	Ver.2.0 approved date: October 20, 2012	–
P5	1.1 Study Design	684 subjects will be randomized in a 1:1 fashion (342 in each arm)	424 subjects will be randomized in a 1:1 fashion (212 in each arm)	Considering the influence of duration of enrollment and follow-up on sample size, also too high drop-out rate
P6	1.3 Main Inclusion/ Exclusion Criteria□	1) Female, aged >= 18 years old and <= 75 years.	1) Female, aged >= 18 years old and <= 70 years.	Fewer patients and poorer compliance
P6	1.3 Main Inclusion/ Exclusion Criteria	3) Pathologic stage $T_{1c-3}N_{0-2}M_0$	3) Pathologic stage $T_{1b-3}N_{0-3}M_0$	Findings from retrospective studies showed the number of positive lymph nodes could not be used for predicting the survival rate. Adjuvant chemotherapy was recommended to

				patients with T_{1b} disease by NCCN guideline
P6	1.3 Main Inclusion/ Exclusion Criteria	1) Patients with T4, including inflammatory carcinomas.	1) Patients with bilateral breast cancer, inflammatory carcinomas.	according to the American Joint Committee on Cancer 2010 staging system
P7	1.3 Main Inclusion/ Exclusion Criteria	2) Patients with N3.	2) Patients with positive supraclavicular or internal mammary lymph node.	Treatment for positive supraclavicular or internal mammary lymph node remains controversial
P14	4.1 Summary of Design	Approximately 684 subjects with TNBC will be randomized in a 1:1 fashion (342 in each arm) to receive treatment	Approximately 424 subjects with TNBC will be randomized in a 1:1 fashion (212 in each arm) to receive treatment	Considering the influence of duration of enrollment and follow-up on sample size, also too high drop-out rate
P14	4.1 Summary of Design	This study will be completed in approximately 84 months including 48 months for accrual and approximately 36 months follow–up	This study will be completed in approximately 96 months including 60 months for accrual and approximately 36 months follow–up survival for the last subject	Slower enrollment than expected

		survival for the last subject enrolled.	enrolled.	
P21	5.2 Inclusion Criteria	1) Female, aged >= 18 years old and <= 75 years.	1) Female, aged >= 18 years old and <= 70 years.	See above
P21	5.2 Inclusion Criteria	3) Pathologic stage $T_{1c–3}N_{0–2}M_0$	3) Pathologic stage $T_{1b–3}N_{0–3}M_0$	See above
P22	5.3 Exclusion Criteria	1) Patients with T4, including inflammatory carcinomas.	1) Patients with bilateral breast cancer, inflammatory carcinomas.	See above
P22	5.3 Exclusion Criteria	2) Patients with N3.	2) Patients with positive supraclavicular or internal mammary lymph node.	See above
P32	8.2 Sample Size	The estimated period of enrollment and follow–up will be 48 and 36 months, respectively. The design is based on 2–sided log–rank test with alpha=0.05, power=90%, and an	The estimated period of enrollment and follow–up will be 60 and 36 months, respectively. The design is based on 2–sided log–rank test with alpha=0.05, power=80%, and an interim	See above

		interim analysis when the last one patient has completed 12 months of follow–up. The dropout rate is assumed to be 20%. Approximately 684 patients (342 patients in each arm) will be enrolled.	analysis when the last one patient has completed 18 months of follow–up. The dropout rate is assumed to be 9%. Approximately 424 patients (212 patients in each arm) will be enrolled.	

STATISTICAL ANALYSIS PLAN

TITLE:	**A MULTICENTER, PHASE III RANDOMIZED STUDY OF METRONOMIC CAPECITABINE MAINTENANCE AFTER STANDARD TREATMENT IN PATIENTS WITH OPERABLE TRIPLE–NEGATIVE BREAST CANCER**
PROTOCOL NUMBER:	**SYSUCC–EBC–CHEMO–001**
STUDY DRUG:	**Capecitabine**
PLAN PREPARED BY:	**Ying Guo and Ji–Bin Li**
PLAN VERSION:	**1.0**
APPROVAL DATE:	**April 5, 2010**

CONTENTS

1. BACKGROUND

The SYSUCC–001 trial is a multicenter, phase III, randomized controlled study to compare the efficacy and safety of metronomic capecitabine maintenance for one year with observation after standard local and systemic treatment in patients with operable triple negative breast cancer (TNBC).

The primary objective of this study is to evaluate whether the addition of metronomic capecitabine maintenance to standard alone treatment improves disease–free survival (DFS), compared with standard treatment. Secondary objectives include determining whether the addition of metronomic capecitabine maintenance to standard treatment could improve overall survival (OS), distant disease–free survival (DDFS), locoregional recurrence–free survival (LRFS) and safety.

The purpose of this Statistical Analysis Plan (SAP) is to provide the details of the proposed analyses of the data collected during this tial.

2. STUDY DESIGN

The SYSUCC–001 trial is a multicenter, phase III, randomized controlled study. A total of 684 patients will be enrolled from

approximately 18 sites in China. Eligible patients will be randomized between the two study arms in a 1:1 ratio.

The sample size of the study is primarily driven by the analysis of DFS. To detect a hazard ratio (HR) of 0.58 in DFS (an estimated improvement of 12% in the 5–year DFS from 68% in the control arm to 80% in the capecitabine maintenance arm), approximately 148 DFS events will be required to achieve a statistical power of 90% at a 2–sided significance level of 5%. The estimated periods of enrollment and follow–up will be 48 and 36 months, respectively. After considering a dropout rate of 20%, approximately 684 patients (342 patients in each group) will be enrolled in the study [1-5].

Interim Analyses: One interim analysis of DFS is planned on the basis of the results of the regular follow–up at 12 months after the completion of enrolment. This interim analysis consists of a comparison of the primary endpoint, the DFS, between groups; if significant differences are found, a secondary endpoint, OS, will be likewise compared between the groups. To maintain the primary errors in the whole study at a level of 5% (with two sided), the multiplicity in the primary endpoint analysis was adjusted using the

Lan–DeMets alpha spending function with an O'Brien−Fleming boundary method. The p–value will be 0.003 for the interim DFS analyses, and 0.047 for final DFS analysis [1-5].

3. RANDOMIZATION

After verification of the inclusion and exclusion criteria, eligible patients will be randomized using the method of stratified permuted blocks to receive metronomic capecitabine maintenance treatment or observation. Patients will be stratified according to lymph node status (negative *vs.* positive). A computerized number generator using Software SAS (version 8.01) generated a randomization table, the results of which were placed in sequentially numbered opaque envelopes and remained concealed until after enrollment.

4. STATISTICAL METHODS

4.1 Analysis populations

4.1.1 Full Analysis Set (FAS) Population

The Full Analysis Set (FAS) is defined as all randomized patients excluding those who withdraw informed consent before protocol treatment, or who had no follow–up data after randomization. The primary analysis population for all efficacy endpoints will be the

FAS population.

4.1.2 Per–Protocol Set (PPS) Population

The Per Protocol Set (PPS) is defined as all randomized patients who have completed the study without major protocol violations, such as patients who discontinue the study across the protocol treatment for reasons determined to be unrelated to breast cancer treatment, and patients who refuse any follow–up or visit, not including breast cancer recurrence or death.

4.1.3 Safety Population

The Safety Analyses Set (SAS) is defined as all randomized patients who initiate the protocol treatment.

4.2 Efficacy Analysis

The following sections outline the planned analysis of the primary and secondary efficacy endpoints of this study. All efficacy analysis will be performed based on the FAS population.

4.2.1 Analysis of the Primary Endpoint

The primary endpoint is DFS, defined as the time from

randomization to the first occurrence of the following events:

1) Relapse of breast cancer in the ipsilateral chest wall and regional lymph nodes
2) Distant metastases (histologically confirmed or clinically diagnosed)
3) Breast cancer related, non–breast cancer–related or unknown deaths
4) Contralateral breast cancer

Patients who have not had an event at the time of data analysis will be censored at the last date they were known to be alive and event–free.

The null hypothesis for the primary endpoint is that the survival distributions of DFS in the two treatment groups are the same. The alternative hypothesis is that the survival distributions of DFS in the treatment and the control arm are different:

$\mathbf{H}_0$: $S_{<capecitabine>} = S_{<observation>}$ **vs.** $\mathbf{H_1}$: $S_{<capecitabine>} \neq S_{<observation>}$

We will estimate survival curves in each treatment arm using the Kaplan–Meier estimator and the hazard ratio with 95%CI between treatment arms based on the proportional hazards model, with assumptions of proportional hazards confirmed based on the Schoenfeld residuals for the final dataset. We will use a two–sided

log–rank test at the final analysis (at a significance level of 0.047). Stratified analyses of the lymph node status will also be conducted.

4.2.2 Analysis of the Secondary Endpoints

The Secondary Endpoints are defined as follows:

Overall Survival (OS) is defined as the time from randomization to death caused by any reason. Patients who are alive (including lost to follow–up) at the time of the analysis will be censored at the date when they were last known to be alive.

Distant disease–free survival (DDFS) is defined as the time from randomization to the first occurrence of any of the following events: Distant metastases, death caused by any reason, and contralateral invasive breast cancer. Patients who have not had a distant recurrence event at the time of data analysis will be censored at the date when they were last known to be alive.

Locoregional recurrence–free survival (LRFS) is defined as the time from randomization to the first occurrence of any of the following events: ipsilateral breast or chest wall, regional lymph node, and death caused by any reason.

The primary analyses for all secondary endpoints will be performed at the time of the primary analysis of the primary endpoint DFS. The

estimated Kaplan–Meier curves and the hazard ratio with 95% CI will be calculated based on the proportional hazards model and the endpoints will be compared using a two–sided log–rank test (at a significance level of 0.05).

4.2.3 Subgroup Analysis

At the time of the primary analysis, exploratory analyses will be performed for DFS to determine whether the magnitude of the effectiveness of the addition of capecitabine maintenance might differ according to patient sub–populations.

Variables to be considered for defining subgroups of interest include the node status as well as other disease– or patient–related prognostic or predictive factors. We will conduct the subgroup analysis by estimating the hazard ratio with 95% CI and the test interaction, if applicable, among subgroups with two–sided p–values) for the following items:

- ✓ Age (≤40 / >40 and median and range)
- ✓ Tumor size at diagnosis (T1 /T2/T3)
- ✓ Histological grade (I/II/III)
- ✓ Nodal stage (N0 / N+)
- ✓ Stage (I / II / III)

- ✓ KI–67 (≤14% / >14%)
- ✓ Lymphovascular invasion (positive / negative)
- ✓ Neo-/adjuvant regimens (anthracycline–based/ taxane–based/ anthracycline– and taxane–based)

The above background variables will be compared using statistical test (at a two–sided significance level of 0.05).

4.2.4 Exploratory Analysis

In the capecitabine arm, a tabulation of those patients who have completed the protocol as planned against those who did not complete the protocol as planned will be performed. Completion/ reduction/stop numbers and proportion of capecitabine are calculated at every 3–month visit, to show relative dose intensity (RDI) of capecitabine, which is defined as the actual cumulative dose compared to planned total dose.

To determine the relationship between the RDI of capecitabine and DFS, estimated Kaplan–Meier curves and the hazard ratio with 95% CI will be calculated based on the proportional hazards model and the comparison will be tested using a two–sided log–rank test (at a significance level of 0.05).

4.3 Safety Analyses

Safety data will be summarized based on the Safety Population. Verbatim descriptions of treatment–emergent adverse events (AEs) will be mapped to MedDRA thesaurus terms and graded according to NCI–CTCAE version 4.0. All AEs, including serious adverse events (SAEs), will be summarized by treatment arm and NCI–CTCAE grade. Comparisons between treatment groups will use the chi squared test (grade0–2/grade3–4) with a two–sided p–value (at a significance level of 0.05). The variables to be tested are:

- ✓ White blood cell count
- ✓ Neutrophil count
- ✓ Platelet count
- ✓ Hemoglobin
- ✓ AST
- ✓ ALT
- ✓ Total Bilirubin
- ✓ Creatinine
- ✓ Appetite loss
- ✓ Abdominal pain / Diarrhea
- ✓ Nausea

- ✓ Vomiting
- ✓ Stomatitis
- ✓ Fatigue
- ✓ Hand–foot syndrome (HFS)

5. REFERENCES

1. Chow, S.C.; Shao, J.; Wang, H. 2003. Sample Size Calculations in Clinical Research. Marcel Dekker. New York.
2. Lan, K.K.G. and DeMets, D.L. 1983. 'Discrete sequential boundaries for clinical trials.' Biometrika, 70, pages 659-663.
3. O'Brien, P.C. and Fleming, T.R. 1979. 'A multiple testing procedure for clinical trials.' Biometrics, 35, pages 549-556.
4. Pocock, S.J. 1977. 'Group sequential methods in the design and analysis of clinical trials.' Biometrika, 64, pages 191-199.
5. Reboussin, D.M., DeMets, D.L., Kim, K, and Lan, K.K.G. 1992. 'Programs for computing group sequential boundaries using the Lan-DeMets Method.' Technical Report 60, Department of Biostatistics, University of Wisconsin-Madison.

STATISTICAL ANALYSIS PLAN

TITLE:	**A MULTICENTER, PHASE III RANDOMIZED STUDY OF METRONOMIC CAPECITABINE MAINTENANCE AFTER STANDARD TREATMENT IN PATIENTS WITH OPERABLE TRIPLE–NEGATIVE BREAST CANCER**
PROTOCOL NUMBER:	**SYSUCC–EBC–CHEMO–001**
STUDY DRUG:	**Capecitabine**
PLAN PREPARED BY:	**Ying Guo and Ji–Bin Li**
PLAN VERSION:	**3.0**
APPROVAL DATE:	**January 19, 2017**

CONTENTS

1. BACKGROUND

The SYSUCC–001 trial is a multicenter, phase III, randomized controlled study to compare the efficacy and safety of metronomic capecitabine maintenance for one year with observation after standard local and systemic treatment in patients with operable triple negative breast cancer (TNBC).

The primary objective of this study is to evaluate whether the addition of metronomic capecitabine maintenance to standard treatment improves disease–free survival (DFS), compared with standard treatment alone. Secondary objectives include determining whether the addition of metronomic capecitabine maintenance to standard treatment could improve overall survival (OS), distant disease–free survival (DDFS), locoregional recurrence–free survival (LRFS) and safety.

The purpose of this Statistical Analysis Plan (SAP) is to provide the details of the proposed analyses of the data collected during this trial.

2. STUDY DESIGN

The SYSUCC–001 trial is a multicenter, phase III, randomized controlled study. A total of 424 patients will be enrolled from

approximately 13 sites in China. Eligible patients will be randomized between the two study arms in a 1:1 ratio.

The sample size of the study is primarily driven by the analysis of DFS. To detect a hazard ratio (HR) of 0.58 in DFS (an estimated improvement of 12% in the 5–year DFS from 68.0% in the control arm to 80.0% in the capecitabine maintenance arm), approximately 109 DFS events will be required to achieve a statistical power of 80% at a 2–sided significance level of 5%. The estimated periods of enrollment and follow–up will be 60 and 36 months, respectively. After considering a dropout rate of 9%, approximately 424 patients (212 patients in each group) will be enrolled in the study [1, 2].

Interim Analyses: One interim analysis of DFS is planned on the basis of the results of the regular follow–up at 18 months after the completion of enrolment. So far, however, the number of events is too much lower than expected. This interim analysis is cancelled and approved by SYSUCC Ethics Committee. The p–value for final DFS analysis will be 0.047 yet.

3. RANDOMIZATION

After verification of the inclusion and exclusion criteria, eligible

patients will be randomized using the method of stratified permuted blocks to receive metronomic capecitabine maintenance treatment or observation. Patients will be stratified according to lymph node status (negative *vs.* positive). A computerized number generator using Software SAS (version 8.01) generated a randomization table, the result of which were placed in sequentially numbered opaque envelopes and remained concealed until after enrollment.

4. STATISTICAL METHODS

4.1 Analysis populations

4.1.1 Full Analysis Set (FAS) Population

The Full Analysis Set (FAS) is defined as all randomized patients excluding those who withdraw informed consent before protocol treatment, or who had no follow–up data after randomization. The primary analysis population for all efficacy endpoints will be the FAS population.

4.1.2 Per–Protocol Set (PPS) Population

The Per Protocol Set (PPS) is defined as all randomized patients who have completed the study without major protocol violations, such as patients who discontinue the study across the protocol

treatment for reasons determined to be unrelated to breast cancer treatment, and patients who refuse any follow–up or visit, not including breast cancer recurrence or death.

4.1.3 Safety Population

The Safety Analyses Set (SAS) is defined as all randomized patients who initiate the protocol treatment.

4.2 Efficacy Analysis

The following sections outline the planned analysis of the primary and secondary efficacy endpoints of this study. All efficacy analysis will be performed based on the FAS population.

4.2.1 Analysis of the Primary Endpoint

The primary endpoint is DFS, defined as the time from randomization to the first occurrence of the following events:

5) Relapse of breast cancer in the ipsilateral chest wall and regional lymph nodes
6) Distant metastases (histologically confirmed or clinically diagnosed)
7) Breast cancer related, non–breast cancer related or unknown

deaths

8) Contralateral breast cancer

Patients who have not had an event at the time of data analysis will be censored at the last date they were known to be alive and event–free.

The null hypothesis for the primary endpoint is that the survival distributions of DFS in the two treatment groups are the same. The alternative hypothesis is that the survival distributions of DFS in the treatment and the control arm are different:

$\mathbf{H_0}$: $S_{<capecitabine>} = S_{<observation>}$ **vs.** $\mathbf{H_1}$: $S_{<capecitabine>} \neq S_{<observation>}$

We will estimate survival curves in each treatment arm using Kaplan–Meier estimator and the hazard ratio with 95%CI between treatment arms based on the proportional hazards model, with assumptions of proportional hazards confirmed based on the Schoenfeld residuals in the final dataset. We will use a two–sided log–rank test for the final analysis (at a significance level of 0.047). Stratified analyses of the lymph node status will also be conducted.

4.2.2 Analysis of the Secondary Endpoints

The Secondary Endpoints are defined as follows:

Overall Survival (OS) is defined as the time from randomization to death caused by any reason. Patients who are alive (including lost to follow–up) at the time of the analysis will be censored at the date when they were last known to be alive.

Distant disease–free survival (DDFS) is defined as the time from randomization to the first occurrence of any of the following events: distant metastases, death caused by any reason, and contralateral invasive breast cancer. Patients who have not had a distant recurrence event at the time of data analysis will be censored at the date when they were last known to be alive.

Locoregional recurrence–free survival (LRFS) is defined as the time from randomization to the first occurrence of any of the following events: ipsilateral breast or chest wall, regional lymph node, and death caused by any reason.

The primary analyses for all secondary endpoints will be performed at the time of the primary analysis of the primary endpoint (DFS). The estimated Kaplan–Meier curves, and the hazard ratio with 95% CI, will be calculated based on the proportional hazards model, and the endpoints will be compared using a two–sided log–rank test (at a significance level of 0.05).

4.2.3 Subgroup Analysis

At the time of the primary analysis, exploratory analyses will be performed for DFS to determine whether the magnitude of the effectiveness of the addition of capecitabine maintenance might differ according to patient sub–populations.

Variables to be considered for defining subgroups of interest include the node status as well as other disease– or patient–related prognostic or predictive factors. We will conduct the subgroup analysis by estimating the hazard ratio with 95%CI and test the interaction, if applicable, among subgroups with two–sided p–value for the following items:

- ✓ Age (≤40 / >40 and median and range)
- ✓ Tumor size at diagnosis (T1 / >=T2)
- ✓ Histological grade (I+II/III)
- ✓ Nodal stage (N0 / N+)
- ✓ Stage (I / II / III)
- ✓ Ki–67 (< 30% / ≥ 30%)
- ✓ Lymphovascular invasion (positive / negative)
- ✓ Neo-/adjuvant regimens (anthracycline– or taxane–based/ anthracycline– and taxane–based)

The above background variables will be compared using statistical

test (two–sided significance level is 0.05).

4.2.4 Exploratory Analysis

In the capecitabine arm, a tabulation of those patients who have completed the protocol as planned against those who did not complete the protocol as planned will be perform. Completion / reduction / stop numbers and proportion of capecitabine are calculated at every 3–months visit to show the relative dose intensity (RDI) of capecitabine, which is defined as the actual cumulative dose compared to planned total dose.

To determine the relationship between the RDI of capecitabine and DFS, estimated Kaplan–Meier curves and the hazard ratio with 95% CI will be calculated based on the proportional hazards model and the comparison will be tested using a two–sided log–rank test (at a significance level of 0.05).

4.3 Safety Analyses

Safety data will be summarized based on the Safety Population.

Verbatim descriptions of treatment–emergent adverse events (AEs) will be mapped to MedDRA thesaurus terms and graded according to NCI–CTCAE version 4.0. All AEs, including serious adverse

events (SAEs), will be summarized by treatment arm and NCI–CTCAE grade. Comparisons between treatment groups will use the chi squared test (grade0–2/grade3–5(4)) with a two–sided p–value (at a significance level of 0.05). The variables to be tested are:

- ✓ White blood cell count
- ✓ Neutrophil count
- ✓ Platelet count
- ✓ Hemoglobin
- ✓ AST
- ✓ ALT
- ✓ Total Bilirubin
- ✓ Creatinine
- ✓ Appetite loss
- ✓ Abdominal pain / Diarrhea
- ✓ Nausea
- ✓ Vomiting
- ✓ Stomatitis
- ✓ Fatigue
- ✓ Hand–foot syndrome (HFS)

5. REFERENCES

1. Lakatos, Edward. 1988. 'Sample Sizes Based on the Log-Rank Statistic in Complex Clinical Trials', Biometrics, Volume 44, March, pages 229-241.

2. Lakatos, Edward. 2002. 'Designing Complex Group Sequential Survival Trials', Statistics in Medicine, Volume 21, pages 1969-1989.

SYSUCC–001 Statistical Analysis Plan Amendment List

page	item	before amendment (ver. 2.0)	after amendment (ver.3.0)
Cover	Cover	Version 2.0 approved date: November 30, 2012	Version 3.0 approved date: January 19, 2017
Page 107	2. STUDY DESIGN	(None)	So far, however, the number of events is too much lower than expected. This interim analysis is cancelled and approved by SYSUCC Ethics Committee. The p–value for final DFS analysis will be 0.047 yet.
Page 112	4.2.3 Subgroup Analysis	Tumor size at diagnosis (T1 /T2/T3)	Tumor size at diagnosis (T1 / >=T2)
Page 112	4.2.3 Subgroup Analysis	Histological grade (Ⅰ / Ⅱ /Ⅲ)	Histological grade (Ⅰ + Ⅱ /Ⅲ)
Page 112	4.2.3 Subgroup Analysis	KI–67 (≤14% / >14%)	KI–67 (<30% / ≥30%)
Page 112	4.2.3 Subgroup Analysis	Neo-/adjuvant regimens (anthracycline–based/ taxane–based/ anthracycline– and taxane–based)	Neo-/adjuvant regimens (anthracycline– or taxane–based/ anthracycline– and taxane–based)

page	item	before amendment (ver. 1.0)	after amendment (ver.2.0)
Cover	Cover	Version 1.0 approved date: April 5, 2010	Version 2.0 approved date: November 30, 2012
Page 96	2. STUDY DESIGN	A total of 684 patients will be enrolled from approximately 18 sites in China.	A total of 424 patients will be enrolled from approximately 13 sites in China.
Page 96	2. STUDY DESIGN	Approximately 148 DFS events will be required to achieve 90% power at a 2–sided significance level of 5%. The estimated period of enrollment and follow–up will be 48 and 36 months, respectively. After considering 20% dropout rate	Approximately 109 DFS events will be required to achieve 80% power at a 2–sided significance level of 5%. The estimated period of enrollment and follow–up will be 60 and 36 months, respectively. After considering 9% dropout rate
Page 96	2. STUDY DESIGN	One interim analysis of DFS is planned on the basis of the results of the regular follow–up study 12 months after the completion of enrolment.	One interim analysis of DFS is planned on the basis of the results of the regular follow–up study 18 months after the completion of enrolment.