Protocol of Pertuzumab study (produced by CinnaGen Co.) compared with Perjeta® (Pertuzumab, the reference drug, produced by Genentech)

Title

A Phase III, randomized, two armed, parallel, triple-blind, active controlled, equivalency clinical trial to determine the therapeutic efficacy and safety between Pertuzumab® (Manufactured by CinnaGen Co.) compared with active control group treated by Perjeta® (Pertuzumab, the reference drug, Manufactured by Genentech) in neoadjuvant treatment of HER 2 positive Breast Cancer patients

NCT Number: NCT04957212

Date: 18 January 2020

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This study will be conducted in accordance with Good Clinical Practices (GCPs), the Declaration of Helsinki, local, ethical, and legal requirements.

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Signature:

Date:

External Party Monitoring: CRO Trial

Signature:

Date

Sponsor Representative: Dr. Araz Sabzvari

Signature:

Date

Abbreviations

ANC	Absolute Neutrophil Count
ADR	Adverse Drug Reaction
AE	Adverse Event
ALT	Alanine Aminotransferase
ALP	Alkaline Phosphatase
AUC	Area Under the Curve
AST	Aspartate Aminotransferase
BUN	Blood Urea Nitrogen
CRF	Case Report Form
CISH	Chromogenic In Situ Hybridization
cCR	clinical Complete Response
cPR	clinical Partial Response
cPD	Clinical Progressive Disease
CRA	Clinical Research Associate
CRR	Clinical Response Rate
cSD	clinical Stable Disease
CTA	Clinical Trial Authorization
CBC	Complete Blood Count
CRO	Contract Research Organization
CIOMS	Council for International Organizations of Medical Sciences
Cr	Creatinine
ECOG	Eastern Cooperative Oncology Group
ECG	Electrocardiogram
ER	Estrogen Receptor
FISH	Fluorescence In Situ Hybridization
GFR	Glomerular Filtration Rate
GCP	Good Clinical Practices
hCG	human Chorionic Gonadotropin
HER2	Human Epidermal growth factor Receptor 2
IHC	Immunohistochemistry
IRB/IEC	Institutional Review Board/ Independent Ethics Committee
ICH	International Conference on Harmonization
IFDA	Iranian Food & Drug Administration
IRCT	Iranian Registry of Clinical Trial
LVEF	Left Ventricular Ejection Fraction
LVSD	Left Ventricular Systolic Dysfunction
MRI	Magnetic Resonance Imaging
MI	Myocardial Infarction
NYHA	New York Heart Association
NSAID	Nonsteroidal Anti-Inflammatory Drug
tpCR	Total Pathological Complete Response
bpCR	Breast Pathological Complete Response
Plt	Platelet
PI	Principal Investigator

PR	Progesterone Receptor
SAE	Serious Adverse Event
SAE/R	Serious Adverse Event/Reaction
SOP	Standard Operating Procedure
SUSARs	Suspected Unexpected Serious Adverse Reactions
ULN	Upper Limit of Normal

Administrative Information

Title

A Phase III, randomized, two armed, parallel, triple-blind, active controlled, equivalency clinical trial to determine the therapeutic efficacy and safety between Pertuzumab® (Manufactured by CinnaGen Co.) compared with active control group treated by Perjeta® (Pertuzumab, the reference drug, Manufactured by Genentech) in neoadjuvant treatment of HER 2 positive Breast Cancer patients

Study Registeration:

This protocol is registered according to the program in Iran Registration Clinical Trial (IRCT).

IRCT registration number: IRCT20150303021315N11

Roles and Responsibilities:

Sponsor

Name: Dr. Araz Sabzvari, representative from CinnaGen Co.

Roles:

- Preparing final draft of study protocol
- Providing electronic CRF
- Obtaining necessary licenses from foreign organizations for conducting the trial
- Providing standard operating procedure (SOP) for principal investigators
- Providing quality controlled medications in order to use during the study and transferring the medications to the centers of the study
- Providing financial aids for all anticipated activities in protocol by having agreement with principal investigator
- Providing any insurance or indemnity to cover the liability of the investigator and sponsor
- Providing protocol training and other necessary trainings for all the trial personnel
- Recruiting staff for monitoring the trial
- Responsibility of all trial costs for patients
- Monitoring the trial sites in order to check the progress of trial and reporting of protocol deviation
- Not disclosing protocol information and all the data without written permission of main principal investigator
- Providing financial aids for all anticipated activities in protocol by having agreement with principal investigator

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- Check the site's facilities for the performance and continuation of the clinical trials and take the necessary measures to provide them (to ensure that enough drugs are available to continue the study, to ensure that enough CRF and consent forms are provided and other requirements for the performance of the study).
- Persistent training of site personnel for protocol performance.
- Checking eCRFs and matching them with source data and filling out the query for noncompliance and reminding them to correct them.
- Provide reports of non-compliances and facilities required by the site and submit it to the company's managers.

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- Performing Statistical analysis and preparing the project report at the end of trial with coordination and supervision of main principal investigator
- Cooperation with the sponsor to perform monthly monitoring
- Cooperation with the sponsor for answering the methodology queries
- Regular visiting to all study sites

Principal Investigator

Name: Dr. Behrooz Najafi, Hematology and Oncology Specialist, Faculty member of Guilan University of Medical Sciences

Roles:

- Conducting the study according to the agreed protocol by IFDA and ethics committee
- Setting up a team for this purpose
- Organizing training session for their recruited team members wherever necessary
- Confirmation and supervision of suitable place for patient admission
- Confirmation and supervision of suitable place for drug storage in the trial
- Cooperation with monitors during the conduct of the study
- None disclosing protocol information and all the data without written permission of CinnaGen Co. (if needed to obtain informed consent from those persons to whom the products may be administered.)

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- Make the right decision in order to deal with the situations which affects balance of risk-benefit for participants in the study either individual or general. Regarding safety profile, this decision includes determining the causality between drug and adverse events
- Taking the right decision for continuing the study and excluding the participant who is not eligible for study anymore
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Introduction

Background and Rationals for the Study

Breast cancer

Breast cancer is a disease in which certain cells in the breast become abnormal and divide uncontrollably to form a tumor. Breast cancer is the second most commonly diagnosed cancer in women. About one in eight women in the United States will develop invasive breast cancer in her lifetime. Researchers estimate that more than 230,000 new cases of invasive breast cancer will be diagnosed in U.S. women in 2015 (1).

Some genes and the proteins can determine how a breast cancer behaves and how it might respond to a specific treatment. Human epidermal growth factor receptor 2 (HER2) is one of the such genes that can play a role in the development of breast cancer (2). The amplification of the HER2 gene leads to overexpression of the receptor, which is linked to the development of many types of human cancers including breast, ovarian and those of the gastrointestinal tract (3).

Human epidermal growth factor receptor 2 (HER2) is overexpressed in 25% to 30% of breast cancers, suggesting a role for overexpression in tumorigenesis. This overexpression is most commonly the result of gene amplification. Several lines of evidence support the role of HER2 overexpression in the pathogenesis and poor clinical outcome of human tumors. A number of these studies have shown that breast cancers that overexpress HER2 have a more aggressive course and higher relapse and mortality rates (4).

Knowledge of HER2 status is a prerequisite when considering a patient's eligibility for Herceptin (trastuzumab) therapy. Accurate assessment of HER2 status is essential to ensure that all patients who may benefit from Herceptin are correctly identified. There are several assays available to determine HER2 status: the most common in routine clinical practice are immunohistochemistry (IHC) and fluorescence in situ hybridization (FISH) (5).

Treatment

According to the studies, the most common and prevalent type of breast cancer would be breast cancers with positive esterogen/progestrone receptors. Endocrine treatment for breast cancers with positive non metastatic hormonal receptor includes tamoxifen and aromatase inhibitors (anastrazole, letrozole and exemestane). (6, 7)

Two target therapy method for breast cancer treatment include HER2 pathway and angiogenesis pathway. Studies shows that although prescription of Bevacizumab with angiogenesis inhibitor mechanism pathway can increase clinical complete response in patients with breast cancer, but still hasn't been approved the usefulness of the drug in increasing survival (8-10).

Several murine monoclonal antibodies against the extracellular domain of the HER2 protein were found to inhibit the proliferation of human cancer cells that overexpressed HER2, both in vitro and in vivo (11). In 1980, one monoclonal antibody was made against HER2 and named Trastuzumab. In 1998, trastuzumab (Herceptin®; Genentech Inc. South San Francisco, CA) as a humanized monoclonal antibody directed against the extracellular domain of HER2 was approved as adjuvant treatment for breast cancer and treatment of metastatic breast cancer (12).

In a study which has been done by Buzdar et al, the efficacy level of Trastuzumab was assessed along with chemotherapy regimen. Clinical complete response in Trastuzumab arm was 65.2% comparing to 26.3% in non-Trastuzumab chemotherapy regimen arm. The 3 years' survival rate was 100% in Trastuzumab arm comparing to 85.3% in non-Trastuzumab chemotherapy regimen arm (4).

In a meta-analysis study, which was done in 2012 and included 12000 patients, efficacy of Trastuzumab chemotherapy regimen was assessed by non-Trastuzumab chemotherapy regimen and confirmed prescription of Trastuzumab increases overall survival in 12 months. Although prescription of Trastuzumab less than 6 months, showed level of increment of overall survival (13).

Pertuzumab

Recently, new treatments with HER2 targeting have been developed and Pertuzumab as a monoclonal humanized antibody which connect to different epitope of HER2 external receptor (subgroup II) is one of them (14). Pertuzumab prevents HER2 dimerization with other HER receptors, especially HER3 (15).

Like Trastuzumab, Pertuzumab also induce antibody-dependent cellular cytotoxicity (ADCC) pathway. Since Pertuzumab and Trastuzumab attach to different epitopes of HER2 and have complementary effect, concurrent prescription of these two drugs causes more inhibition in HER2 signaling and anti tumor activity comparing to single use of them (16, 17). Efficacy of

Trastuzumab-Pertuzumab regimen in patients with HER2+ breast cancer patients has been shown in phase II trials (18-20).

In 2012, phase III clinical trial of Trastuzumab and Pertuzumab (CLEOPATRA) has been assessed safety and efficacy of Docetaxel and Trastuzumab plus Pertuzumab comparing to placebo as fist line treatment in HER2+ metastatic breast cancer patients. In this study, 808 patients in 204 centers were treated and the median of Progressive Free Survival (PFS) in Pertuzumab arm showed 6.1 months' increment compared to placebo arm. Furthermore, in this study, the ratio of treatment reponse in Pertuzumab arm showed 10.8% improvement compared to the placebo arm and reached to 80.2% (21).

Study on assessing safety and efficacy of neoadjuvant Pertuzumab and Trastuzumab in women with locally advanced breast cancer or HER2+ inflammation (NeoSphere study) caused accelerated approval in using this medication in neoadjuvant treatment. This phase II study conducted with 4 arms to assess effect of Docetaxel, Trastuzumab and Pertuzumab in different regimen. (19).

In 2013, another study with the aim of assessing the heart complications in neoadjuvant therapy combination of Trastuzumab and Pertuzumab for HER2+ breast cancer patients were done and checked the chemotherapy regimen with or without Antracycline. In this study, 225 patients were assessed and according to the results, heart complications did not show statistical significance difference between 3 treatment arm and the most treatment response (66.2%) was related to Docetaxel, Trastuzumab and Pertuzumab and carboplatin (22).

Pharmacological propertis

Pertuzumab is a recombinant humanised monoclonal antibody that specifically targets the extracellular dimerization domain (subdomain II) of the human epidermal growth factor receptor 2 protein (HER2), and thereby, blocks ligand-dependent heterodimerisation of HER2 with other HER family members, including EGFR, HER3 and HER4. As a result, pertuzumab inhibits ligand-initiated intracellular signalling through two major signal pathways, mitogen-activated protein (MAP) kinase and phosphoinositide 3-kinase (PI3K). Inhibition of these signalling pathways can result in cell growth arrest and apoptosis, respectively. In addition, pertuzumab mediates antibody-dependent cell-mediated cytotoxicity (ADCC).

While pertuzumab alone inhibited the proliferation of human tumour cells, the combination of pertuzumab and trastuzumab significantly augmented antitumour activity in HER2-overexpressing xenograft models (21).

Adverse events

>10%:

Central nervous system: Fatigue (26% to 36%), headache (11% to 21%), decreased left ventricular ejection fraction (8% to 16%), insomnia (8% to 13%), dizziness (3% to 13%)

Dermatologic: Alopecia (52% to 65%), skin rash (11% to 34%), pruritus (4% to 14%), palmar-plantar erythrodysesthesia (11%), xeroderma (9% to 11%)

Gastrointestinal: Diarrhea (46% to 67%), nausea (39% to 53%), vomiting (13% to 36%), decreased appetite (11% to 29%), mucositis (20% to 28%), constipation (23%), stomatitis (17% to 19%; grades 3/4: <1%), dysgeusia (13% to 18%)

Hematologic & oncologic: Neutropenia (47% to 53%; grades 3/4: 43% to 49%), anemia (3% to 23%; grades 3/4: 2% to 4%), leukopenia (9% to 16%; grades 3/4: 5% to 12%), febrile neutropenia (8% to 14%; grades 3/4: 9% to 13%)

Hypersensitivity: Hypersensitivity (1% to 11%)

Neuromuscular & skeletal: Asthenia (15% to 26%), myalgia (11% to 22%), arthralgia (10% to 12%)

Respiratory: Upper respiratory tract infection (4% to 17%), epistaxis (11%)

Miscellaneous: Fever (9% to 19%), infusion reactions (13%)

1% to 10%:

Cardiovascular: Left ventricular dysfunction (3% to 4%), peripheral edema (3% to 4%)

Central nervous system: Peripheral sensory neuropathy (8%; grades 3/4: <1%), peripheral neuropathy (1%)

Dermatologic: Nail disease (7%), paronychia (1% to 7%)

Gastrointestinal: Dyspepsia (8%)

Hematologic & oncologic: Thrombocytopenia (1%)

Hepatic: Increased serum alanine aminotransferase (3%)

Ophthalmic: Increased lacrimation (4% to 5%)

Respiratory: Dyspnea (8%), nasopharyngitis (7%), oropharyngeal pain (7%), cough (5%)

<1%, postmarketing, and/or case reports: Left systolic heart failure, pleural effusion, sepsis, tumor lysis syndrome

Contraindications:

Any known sensitivity to Pertuzumab or each of compounds (23)

Rationale for the Study

Different studies showed enhanced anti tumor activity of Pertuzumab along with trastuzumab in breast cancer patients. Due to high price of this antibody and due to not availability of this drug in Iran's drug list, the CinnaGen Co.decided to produce this drug. This study has been designed as a neoadjuvant treatment in patients with HER2+ breast cancer and patients randomized in 2 gropus receiving Pertuzumab (manufactured by CinnaGen Co.) and Perjeta (Refrence drug, manufactured by Genentech). Patients in this study will receive Docetaxel, Trastuzumab and Pertuzumab and carboplatin regimen every 3 weeks for 6 periods in the form of IV.

The most important objectives of this study would be efficacy and safety comparison of Pertuzumab (manufactured by CinnaGen Co.) and Perjeta manufactured by Genentech in neoadjuvant setting for HER2+ breast cancer patients.

Objectives

Pertuzumab (manufactured by CinnaGen Co.) and Perjeta (manufactured by Genentech) are equivalence in neoadjuvant setting treatment of patients with HER2+ breast cancer.

Primary Objectives

Primary objective of this study is to evaluate equivalence of Pertuzumab (Manufactured by CinnaGen Co.) compared to the Perjeta® (Reference drug, Genentech), when added to Carboplatin+Docetaxel+Trastuzumab regimen, based on the pathological complete response (pCR) in neoadjuvant treatment setting in HER2+ breast cancer patients

Secondary Objetcives

Clinical Response Rate

Total Pathological complete response (tpCR) in breast and axillary lymph nodes

Breast conserving surgery rate

Safety

Immunogenicity

Study Design

Procedures

Phase III clinical trial, randomized, 2 arms, triple blind, parallel, with active control in order to assess equivalency with 1: 1 ratio

Study Sites

This study will be done from Jan 2018 till Oct 2019 on patients in below centers. All the sites will use the same protocol and procedures. The methods are standardized as much as possible in order to reduce differences and all the staff will be trained accordingly. The name and city of study cenetrs is as below:

1- Razi Hospital-Guilan
2- Golsar Hospital-Guilan
3- Mehrad Hospital-Tehran
4- Imam Khomeini Hospital-Tehran
5- Sina Hospital-Tehran
6- Firouzgar Hospital-Tehran
7- Masoud Clinic-Tehran
8- Dr. Safa Najar Najafi clinic-Tehran
9- Imam Reza Hospital-Mashad
10- Dr. Mehrdad Payandeh Clinic-Kermanshah
11- Dr. Mortazavizadeh Clininc-Yazd
12- Namazi Hospital-Shiraz
13- Shafa Hospital-Ahwaz
14- Shahid Bahonar Hospital-Kerman
15- Jahad Daneshgahi center-Tehran
16- Rasool Akram Hospital-Tehran
17- Taleghani Hospital-Tehran
18- Faghihi Hospital-Tehran
19- Shams Hospital-Tabriz
20- Seyedolshohada Hospital-Isfahan
21- Bou Ali Hospital-Tehran
22- Masih Daneshvari Hospital-Tehran
23- Sheikh Mofid clinic-Isfahan
24- Ghaem Hospital-Mashhad
25- Sanabd Clinic-Mashhad
26- Arvand Hospital-Ahwaz
27- Saba Clinic-Isfahan
28- Naft Hospital-Tehran

Study Population Criteria

Inclusion Critera:

- Female patients aged 18 - 70
- Operable (T2-3, N0-1, M0), locally advanced (T2-3, N2 or N3, M0; T4a-c, any N, M0), or inflammatory (T4d, any N, M0) breast cancer
- Primary tumor diameter should be more than 2 centimeters
- Positive HER2 status approved by immunohistochemistry (IHC 3+ or IHC 2+ verified by fluorescence in situ hybridization (FISH) or chromogenic in situ hybridization (CISH))
- Eastern Cooperative Oncology Group performance status (ECOG PS) of 0 or 1
- LVEF ≥55% at baseline assessed by echocardiography
- Able and willing to sign an informed consent

Exclusion Criteria:

- Metastatic (stage IV) or bilateral breast cancer
- Previous systemic or local anticancer therapy (chemotherapy or radiotherapy) for any type of cancer
- Any other malignancy except for carcinoma in situ of the cervix, basal cell carcinoma, or squamous cell carcinoma of the skin
- Use of another research drug in the four weeks before the start of the study
- Major surgery four weeks before the start of the study
- Uncontrolled hypertension (systolic blood pressure more than 150 mmHg or/and diastolic blood pressure more than 100 mmHg), unstable angina, Congestive heart failure with any class based on the NYHA classification, serious arrhythmia which needs treatment, or history of MI 6 months prior to enrollment
- Bone marrow malfunction:
  - ANC < 1500/µL
  - Plt < 100,000/µL
  - Hb < 9 g/dL
- Liver malfunction:
  - ALT/AST>1.5, ALP>2.5 ULN
  - Total serum Blirubin>1.25 ULN
- Kidney malfunction:

- ❖ Serum Creatinine >1.5 ULN

- Shortness of breath during rest or any other disease that requires continuous oxygen therapy
- Any severe uncontrolled systemic disease (cardiovascular, pulmonary, metabolic, etc.)
- Chronic treatment with corticosteroids with a daily dose of ≥ 10 mg oral prednisolone or equivalent of other types (other than inhaled corticosteroid drugs)
- Patients with HIV, HBV, and HCV infections
- Hypersensitivity to any of the studied drugs or excipients
- Pregnancy, lactating or fertile women who do not want to use contraceptive methods (contraceptives should be taken in to consideration up to six months after the last dose of the drug)
- Unwillingness or inability to fulfill the requirements of the protocol, including any kind of condition (physical, mental or social) that affects one's ability to fulfill the requirements of the protocol

Interventions

All of the participants in the study, will receive the below regimens. In order to increase patient' compliance and safety, study drug wil be injected by a trained nurse.

In the test drug arm (group 1), Pertuzumab (manufactured by CinnaGen Co.) and in the control arm Perjeta® (Reference drug) will be administered.

All of the patients will receive Trastuzumab, Pertuzumab, Carboplatin and Docetaxel as follow:

Duration of each cycle: 21 days			**Number of cycles: 6 cycles**
Drugs	**Dose and route of administration**	**Adminstration**	**Days**
Trastuzumab	8 mg/kg IV loading dose (at cycle 1), followed by 6 mg/kg at subsequent cycles Change in dosage during the treatment is not allowed.	IV infusion in 250ml NS over 90 minutes for first dose IV infusion in 250 mL NS over 30 min on all subsequent doses if no adverse reactions. DO NOT mix with D5W, and DO NOT infuse as an IV push or bolus.	Day 1
Pertuzumab	Fixed dose of 840 mg IV infusion in first cycle, followed by fixed IV infusion of 420 mg in next cycles Changing in dosage during the treatment is not allowed.	Dilute into a 250 mL normal saline. Dose in first cycle in over 60 min and in second till 6th cycle during 30- 60 min, IV infusion DO NOT mix with D5W, and DO NOT infuse as an IV push or bolus.	Day 1
Carboplatin	AUC* = 6 mg/mL x min IV infusion	Dilute into a 250 mL 5% Dextrose and administered as a 30-minute IV infusion. Infusion of carboplatin should start 30-60 min after completion of Pertuzumb administration.	Day 1
Docetaxel	75mg/m^2, IV infusion, Increasing dosage of Docetaxel is not allowed.	Dilute into a 250 mL NS till the final concentration of 0.3-0.74 mg/mL and then administered as a 60-minute IV infusion.	Day 1

In case of variation more than 10% in weight, dose should be calculated for all drugs again.

AUC* or area under curve with regards to renal activity and by using Clavert formula can be converted to required dose in milligram.

According to Clavert formula, final dose (milligram) would be multiplication of target AUC in GFR+25. In case of using creatinine, final limit of GFR in this calculation would be 125 ml/min.

Carboplatin Dose (mg) = Target AUC * (GFR + 25)

Pre treatment Consideration

Prevention of nausea

Recommened to be premedicated with a serotonin antagonist (8-16 mg Ondansetron with 1-2 mg Granisetron), Neurokinin inhibitor (125 mg Aperpitant) and Dexametasone (8mg) in the day of chemotherapy.

Diarrhea

It is recommened to use Loperamide (4mg and then 2 mg every 4 hours up to 16 mg daily) till 12 hours after cessation of diarrhea.

Prevention of infusion related recations

Dexamethasone 8 mg PO bid for 3 days, starting one day prior to each Docetaxel administration. Pre treatment usually is not used for Pertuzumab and carboplatin, however in the case of flu-like side effects within 24 hours after Trastuzumab administration; it is recommened to use Acetaminophen or NSAID.

Dose Adjust in Hepatic or Renal impairment

Docetaxel should not be administered to patients with bilirubin more than ULN or with transaminase more than 1.5 times ULN and alkaline phosphatase more than 2.5 times ULN. Each dose of Carboplatin should be caculated according to kidney activity and based on the Calvert formula.

Cardiac issue

Cardiotoxicity has been observed by Trastuzumab and Pertuzumb, So LVEF should be measured before the treatment.

Prevention of Thrombocytopenia

It is necessary to administer amyloid growth factor 24-72 hours after each chemotherapy cycle.

Dose adjustment:

Myelotoxicity:

In the case of ANC>1000 cell and Platelet>100000, complete dose of chemotherapy regimen drugs should be administered. In case, count of cell or platelet are less than specified amount, chemotheraoy should be postponed for 1 week.

ANC<1000 and two times of one week delay	Dose adjustment of Docetaxel by 75% dose
More than 1 week delay due to thrombocytopenia and Platelet <50000	Dose adjustment of Docetaxel by 75% dose

In case of Febrile Neutropenia (ANC<500, Fever>38.3 required treatment by IV antibiotic with or without hospitalization), dose of chemotherapy drugs should be adjusted.

In case of febrile neutropenia	Antibiotic treatment according to NCCN and ESMO guideline
Episode 1	Docetaxel dose reduced by 75% and Carboplatin dose reduce to 5 AUC
Episode 2	Docetaxel dose reduced by 60% and Carboplatin dose reduce to 4 AUC

Hepatic toxicity:

Docetaxel dose can be reduced to 60 mg/m^2 in patients with significant change of liver factors (increase of Transaminase>1.5 ULN and increase of Alkaline phosphatase>2.5 ULN) and it cannot be level up to 75 mg/m^2 in next cycles. In case of Transaminase increase to more than 3.5 ULN and increase of alkaline phosphatase to more than 6 ULN, it is not recommended to use Docetaxel.

Docetaxel dose	ALP	AST +/or ALT
100%	< 2.5 ULN	< 1.5 ULN and

75%	2.5-6 ULN	1.5-3.5 ULN and
Not recommended	6-10 ULN	> 3.5 ULN and

Cardiotoxicity:

In case of left ventricular ejection fraction (LVEF) reduced to less than 45% or decrease of 10% or more than from baseline amount to 45-49%, administration of Trastuzumab or Pertuzumab should be stopped for at least one cycle until recovering to normal condition (**Appendix 4**).

Infusion-related reactions:

In case of infusion realted reactions, the rate of infusion should be decreased or discontinued in order to have supportive therapy. In case of severe adverse events or anaphylactic shock, the administration of drug should be discontinued.

Dermatological, mucosal and neurotoxicity:

In case of severe or cumulative cutaneous reactions, grade 3 or 4 stomatitis or moderate neurosensory signs and/or symptoms, Reduce dose to 60 mg/m^2, if reactions persist at 60 mg/m^2, therapy should be discontinued.

Endpoints

Endpoints will be evaluated during 20 weeks after study start.

Primary Endpoints

- Pathologic Complete Response (pCR):

pCR Defined as the absence of invasive neoplastic cells at microscopic examination of the primary tumor at surgery (ypT0/is)

Secondary Endpoints

- Clinical Response Rate (CRR) according to RECIST 1.1 (Appendix 3):

Sum of patients who had clinical complete response (cCR) or clinical partial response (cPR) before surgery.

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

- Total Pathological complete response (tpCR) in breast and axillary lymph nodes:
  Defined as the absence of invasive neoplastic cells at microscopic examination of the primary tumor at surgery in breast and axillary lymph nodes, without considering in situ involvement (ypT0/is ypN0)
- Conserving surgery rate in patients who are mastectomoy candidate (T2-3)
- Adverse Drug Reaction (Safety)

Safety Endpoints

Adverse events (AEs) and Adverse drug reactions (ADRs) including:

- Incidence of Left Ventricular Systolic Dysfunctionn (LVSD)
- 10% or more reduction of LVEF compared to the baseline limit to less than 50%
- Other adverse drug reactions
- Abnormal lab test
- Vital sign assessment
- Immunogenicity

End of Study Policy

This decision is under authorization of DSMB committee and should be confirmed by IFDA and ethical committee of Tehran Univeristy of Medical Sciences and Guilan Univeristy of Medical Sciences.

Timeline of the Study

	Duration of study								
	Screening	Interventions							End of study
Visits	Visit 0	Visit 1	Visit 2	Visit 3	Visit 4	Visit 5	Visit 6	Visit 7	Surgery
Weeks	Day -1 to -14	Day 0	Week 3	Week 6	Week 9	Week 12	Week 15	Week 18	Week 18-20
Eligibility evaluation	X								
Informed consent form	X								
Medical history	X								
Physical examination and vital sign	X	X	X	X	X	X	X	X	
ECG	X								
Laboratory	X	X	X	X	X	X	X	X	
Pregnancy (β-hCG)	X			X			X		
Echocardiography	X			X		X		X	
Metastasis	X								
HER2 و ER/PR	X								
MRI*)Tumor assessment)	X							X	
Concomitant drug record	X	X	X	X	X	X	X	X	
Adverse events	X	X	X	X	X	X	X	X	
Randomization		X							
Drug intervention		X	X	X	X	X	X		
Heart evaluation questionnaire **		X	X	X	X	X	X		
Immunogenicity		X	X	X	X	X	X	X	
Surgery and pathological sampling									X

*In case of patient early drop out due to any reason before completion of the treatment period, MRI should be done.

** Refer to Appendix 6

Description of Study Visits

Visit 0: Screening, eligibility criteria evaluation (Day -1 to -14)

1. Eligibility criteria evaluations
2. Explaining protocol, preparing informed consent form (ICF), asking for reading and understanding and then asking for signing of ICF
3. Medical history including underlying disease and recent surgery
4. Pysical examination, vital signs, height, weight
5. Electrocardiogram test
6. Laboratory test including: CBC diff, BUN, Cr, ALT, AST, Bilirubin, Na, K, Ca, P, Mg, Cl, albumin, glucose, LDH ،INR ،PTT/aPTT, urine analysis, HBV, HCV, HIV tests.
7. ECOG evaluations
8. Pregnancy test (β-hCG)
9. Echocardiography for LVEF
10. Assessing of non-metastatic in suspicious patients by bone scan،CT scan of thorax, abdomen, pelvic and if needed brain MRI.
11. Assessment of HER2 and ER/PR receptors
12. Assessment of basis tumor specifications. In this evaluation, size of tumor, grade of tumour, inflammation, locally advanced, capability of surgery is determined by surgeon to be conservative. MRI should be done in this visit under physician decision.
13. Record concomitant drugs and observed adverse events
14. In screeing visit, the first step is determining ER/PR, MRI, and type of breast cancer

Visit 1 (Day 0, patient allocation)

When the investigator is ensured about the eligibility, patients allocated to one of the below groups:

Group 1: Pertuzumab (manufactured by CinnaGen Co.)

Drug: Pertuzumab vial (840 mg dose)(Initial dose), IV infusion every 3 weeks for 6 cycles.

Group 2: Perjeta® (Manufacutured by Genentech)

Drug: Pertuzumab vial (840 mg dose)(Initial dose), IV infusion every 3 weeks for 6 cycles

After randomization, each patient will receive a randomization code. Patient will receive the treatment based on her group and will received these interventions:

1. Laboratory tests (CBC diff, BUN, Cr, ALT, AST, Bilirubin, Na, K, Ca, Mg, P, Cl, albumin, glucose, LDH) before administration of chemotherapy regimen in order to diagnose any hematologic changes and patient ability for receiving next cycle will be evaluated. Moreover sampling for immunogenicity will be done.
2. Pysical examination, vital signs, height, weight
3. MRI by decision of investigator (in case of early withdrawal) can be done in this visit
4. Chemotherapy regimen drugs (Trastuzumab, Pertuzumab, Carboplatin, Docetaxel)
5. Heart evaluation questionnaire
6. Concomitant drugs, adverse events record
7. AE of previous visit will be checked and results will be recorded, if applicable.
8. Determination of lab visit time, one day prior to next chemotherapy cycle
9. Information about the next visit time
10. Record of information in CRF

Visit 2 (week 3)

1. Laboratory tests (CBC diff, BUN, Cr, ALT, AST, Bilirubin, Na, K, Ca, Mg, P, Cl, albumin, glucose, LDH) before administration of chemotherapy regimen in order to diagnose any hematologic changes and patient ability for receiving next cycle will be evaluated. Moreover sampling for immunogenicity will be done.
2. Pysical examination, vital signs, height, weight
3. MRI by decision of investigator (in case of early withdrawal) can be done in this visit
4. Chemotherapy regimen drugs (Trastuzumab, Pertuzumab, Carboplatin, Docetaxel)
5. Heart evaluation questionnaire
6. Concomitant drugs, adverse events record
7. AE of previous visit will be checked and results will be recorded, if applicable.
8. Determination of lab visit time, one day prior to next chemotherapy cycle
9. Information about the next visit time
10. Record of information in CRF

Visit 3 (week 6)

1. Laboratory tests (CBC diff, BUN, Cr, ALT, AST, Bilirubin, Na, K, Ca, Mg, P, Cl, albumin, glucose, LDH) before administration of chemotherapy regimen in order to diagnose any hematologic changes and patient ability for receiving next cycle will be evaluated. Moreover sampling for immunogenicity will be done.
2. Pregnancy test (β-hCG)
3. Pysical examination, vital signs, height, weight
4. MRI by decision of investigator (in case of early withdrawal) can be done in this visit
5. Echocardiography for LVEF evaluation
6. Chemotherapy regimen drugs (Trastuzumab, Pertuzumab, Carboplatin, Docetaxel)
7. Heart evaluation questionnaire
8. Concomitant drugs, adverse events record
9. AE of previous visit will be checked and results will be recorded, if applicable.
10. Determination of lab visit time,one day prior to next chemotherapy cycle
11. Information about the next visit time
12. Record of information in CRF

Visit 4 (week 9)

1. Laboratory tests (CBC diff, BUN, Cr, ALT, AST, Bilirubin, Na, K, Ca, Mg, P, Cl, albumin, glucose, LDH) before administration of chemotherapy regimen in order to diagnose any hematologic changes and patient ability for receiving next cycle will be evaluated. Moreover sampling for immunogenicity will be done.
2. Pysical examination, vital signs, height, weight
3. MRI by decision of investigator (in case of early withdrawal) can be done in this visit
4. Chemotherapy regimen drugs (Trastuzumab, Pertuzumab, Carboplatin, Docetaxel)
5. Heart evaluation questionnaire
6. Concomitant drugs, adverse events record
7. AE of previous visit will be checked and results will be recorded, if applicable.
8. Determination of lab visit time, one day prior to next chemotherapy cycle
9. Information about the next visit time
10. Record of information in CRF

Visit 5 (week 12)

1. Laboratory tests (CBC diff, BUN, Cr, ALT, AST, Bilirubin, Na, K, Ca, Mg, P, Cl, albumin, glucose, LDH) before administration of chemotherapy regimen in order to diagnose any hematologic changes and patient ability for receiving next cycle will be evaluated. Moreover sampling for immunogenicity will be done.
2. Pysical examination, vital signs, height, weight
3. MRI by decision of investigator (in case of early withdrawal) can be done in this visit
4. Echocardiography for LVEF evaluation
5. Chemotherapy regimen drugs (Trastuzumab, Pertuzumab, Carboplatin, Docetaxel)
6. Heart evaluation questionnaire
7. Concomitant drugs, adverse events record
8. AE of previous visit will be checked and results will be recorded, if applicable.
9. Determination of lab visit time, one day prior to next chemotherapy cycle
10. Information about the next visit time
11. Record of information in CRF

Visit 6 (week 15)

1. Laboratory tests (CBC diff, BUN, Cr, ALT, AST, Bilirubin, Na, K, Ca, Mg, P, Cl, albumin, glucose, LDH) before administration of chemotherapy regimen in order to diagnose any hematologic changes and patient ability for receiving next cycle will be evaluated. Moreover sampling for immunogenicity will be done.
2. Pregnancy test (β-hCG)
3. Pysical examination, vital signs, height, weight
4. MRI by decision of investigator (in case of early withdrawal) can be done in this visit
5. Chemotherapy regimen drugs (Trastuzumab, Pertuzumab, Carboplatin, Docetaxel)
6. Heart evaluation questionnaire
7. Concomitant drugs, adverse events record
8. AE of previous visit will be checked and results will be recorded, if applicable.
9. Determination of lab visit time, one day prior to next chemotherapy cycle
10. Information about the next visit time
11. Record of information in CRF

Visit 7 (week 18)

1. Laboratory tests (CBC diff, BUN, Cr, ALT, AST, Bilirubin, Na, K, Ca, Mg, P, Cl, albumin, glucose, LDH) before administration of chemotherapy regimen in order to diagnose any hematologic changes and patient ability for receiving next cycle will be evaluated. Moreover sampling for immunogenicity will be done.
2. Pysical examination, vital signs, height, weight
3. MRI by decision of investigator (in case of early withdrawal) can be done in this visit
4. Echocardiography for LVEF evaluation
5. Concomitant drugs, adverse events record
6. Follow up of AEs and record
7. Record of information in CRF

Surgery visit (week 18-20)

Surgery and pathological sampling should be done 2 weeks later than visit 7 (week 18-20)

Sample Size

Using an equivalence margin of 0.2, a sample size of 96 subjects in each one of both Pertuzumab (CinnaGen Co.) and Perjeta® (Genentech Co.) groups achieves 80% power to detect bioequivalence for the difference between groups. The actual difference is considered zero. The PCR efficacy for Perjeta® group is 0.66 (22, 24, 25). Two-way independent T-test with 5% significance level will be used to analyze the results. Considering 10% dropout, final sample size is considered 107 in each group.

Recruitment

This study has two arms and 214 patients will be enrolled (107 patients in each arm)

1) Group 1: Patients who receive Pertuzumab (Manufactured by CinnaGen Co.) every 3 weeks with Trastuzumab, Carboplatin, Docetaxel

2) Group 2: Patients who receive Perjeta® (Manufactured by Genentech) every 3 weeks with Trastuzumab, Carboplatin, Docetaxel

Randomization

Patients will be dynamically randomized to different groups during the study using Minimization method (with a 90% random component). The two main variables on which the balance is performed will be as follows:

1- ER/PR: ER/PR+, ER/PR-

2- Type of breast cancer: operable, locally advanced, inflammatory

At the beginning of the study, each patient will receive a unique code in order to be recognized during the study. The assigned code consists of 4 letters (corresponding to the first two letters of the first name and the first two letters of the surname), three digits (center code), the first three letters of the generic drug name (which is PER-) and 3 digits (corresponding to randomization code). For example: ABCD001PER-001.

Each Pertuzumab package which will be used for a patient's treatment during the study and will be recognized by a unique 3-digit code similar to the randomization code (this number includes two English letters and a number which will be unique for each subjetcs).

Blinding

Both Pertuzumab drugs in this study are not identifiable by patients and treatment group. Since the administration route of this drug is IV infusion, patients' blinding in medication groups would be possible. Furthermore, persons who are doing the data analysis are not aware of patients' grouping.

Patients' groups and type of their medications will not disclose to investigators and will be in the stamped pocket and will be provided to investigator of each site. Persons who are doing analysis are not aware of grouping.

Data Collection Methods

Data collection will be electronic-based and the data will be recorded in eCRF. Adequate attention will be given to collect accurate and valid data. Investigators are responsible for completing the eCRF in study centers. The sites will be equipped with personal computers or tablets. Only the investigator or the person assigned by the investigator will have access to the database for data entry.

Data Management

The principal investigator, investigators, and other personnel assigned by the investigators will be responsible for eCRF data entry. Each investigator will be given a specific username and password. The PI and investigators must not disclose the data obtained from the study. All of the Investigators are responsible for keeping the study data safe. Sending and receiving the patients' information must be done considering safety and security procedures. Trial CRO is responsible for planning a monitoring which will be conducted by qualified personnel in order to check the eCRF for discrepancies with the patients' source documents. The monitors or auditors are not able to change eCRF data. However, they can make queries for blatant mistakes in data entry and the investigator is responsible for rectifying such mistakes or answering to the queries. The history of modification in eCRF data will be recorded meticulously and can be observed by the monitors or auditors.

Data Monitoring

The objectives of the data quality control are:

- To ensure the existence of the patients and the respect of ethics (including signed patient informed consent)

- To detect the issues (including systematic errors) as early as possible for appropriate measures to be taken
- To ensure the validity of the data

To meet these objectives, quality control should be applied via the following activities:
The schedule of site quality control evaluation, performed by the monitoring team must be explained to the investigators at the time of site initiation visit and agreed upon. QC will be performed in study sites by the monitoring team who are in charge of planning for action plans to improve study site quality.
QC of the study site will be carried out during two main monitoring visits (30% and 70% of study completion) and several periodic monitoring visits throughout the study.
In every monitoring visit the following items will be evaluated according to a pre-prepared check list and the quality control report will be completed.

- eCRF forms will be assessed in terms of completeness, the quality of data entry and accordance with source data.
- Informed consent forms signed by the patient and the physician for all the patients who have gone through the screening visit
- Evaluation of key variables regarding the wrong and missing data

Statistical Analysis

Statistical analysis of data will be conducted by the study statistical consultant. Additionally, the available data will be evaluated separately by the Trial CRO representative and the results will be compared. The final analysis will be assessed by the Steering Committee prior to submission to the Food and Drug Administration.

It should also be noted that in order to prevent any bias in the analysis, all the results will be reviewed in joint meetings with Trial CRO on behalf of Dr. Hamed Hosseini and approved by the Trial CRO team.

Statistical Methods

The primary endpoints will be analysed for Per-protocol population. Moreover, primary endpoints and secondary endpoints in intention-to-treat/FAS population will be assessed and for comparison of primary endpoints, sensisitivity anlysis will be applied.

All adverse events analysis was conducted based on the Saftey population

1. Per-protocol population: All randomized patients who treated with trial drugs and does not have a major violation with protocol.

2. **Intention- to – treat population:** All randomized patients who were allocated to treatment groups and received at least one dose of study drug. The patients were analyzed based on their treatment group which was randomized.

3. **Safety population:** All patients who randomly received at least one dose of the drugs. The patients were analyzed based on the randomized received drugs.

Missing Data

Graphical methods (standard tables and figures) will be used to recognize potential outliers. Then, the analyses will be conducted including/excluding outliers. Finally, the results of the two model will be compared considering the effect of outliers.

Both pre-protocol and FAS/ITT populations (with and without imputation) will be reported for the primary endpoint analysis. In case there is no final answer, the worst-case scenario “NO PCR” will be applied. Then, the results will be comaprd and if there was any significant differences, the reason will be evaluated.

Covariate Adjustment

Since the randomization method in this study is Minimization, the results of patients who will be randomly assigned to one of the treatment groups at the beginning of the study will be compared with that of other patients who will assigned to their treatment group using Minimization method during the study.

Patient Characteristics and Baseline Comparisons

Descriptive statistics of thew data will be presented in the study. For categorical variables, number and percentage of patients in each category, and for continuous variables, mean, standard deviation, and quartiles will be calculated.

Primary Endpoint Analysis

Primary endpoint (pCR) analysis, will be conducted using a two-sided 95% confidence interval with equivalence margin of 0.2. Equivalence of Pertuzumab (CinnaGen Co.) and Perjeta® will be concluded if the resulted 95% confidence interval for the difference will be within the predetermined range (-0.2, 0.2)

Secondary Endpoint Analysis

The clinical response rate and tpCR in both treatment groups will be calculated and compared using the ratio test.

The conserving surgery rate in both treatment groups will be calculated and their comparison will be done using the ratio test.

Safety Endpoint Analysis

All adverse events data will be descriptively analyzed.

Adverse events: The number and percentage of adverse events caused by treatment will be classified by organ system class and preferred term will be applied. Laboratory data of hematology and biochemistry will be graphically displayed to show the trend of changes between treatment groups.

Frequency of abnormal parameters will be reported in each visit. Furthermore, clinical significant items will be determined in the study. Change in vital sign parameters will be determined in each visit and change from Normal to Abnormal for physical evaluation will be evaluated.

Immunogenicity Analysis

Samples for immunogenicity assessment (antidrug antibody [ADA] and neutralizing antibody [nAb]) will be collected prior Pertuzumab administration in all visits. All patients in each treatment arm will be assessed for ADA formation to Pertuzumab. In Institute Pasteur, with validated kit, ADA formation will be detected and characterized. These tests will be done in 3 following steps:

- A screening assay to detect anti-Pertuzumab antibodies;
- A confirmatory assay for the samples that were positive in previous step
- A titration assay to determine antibody titers

Monitoring

The purpose of monitoring is to assuring conducting the trial according to protocol and GCP standards.

Monitor should prepare a written report for each visit to sponsor and PI. This report should include monitor's name, date of visit, center name, summary of visit, important findings, deviations, final conclusion, and CAPA.

Site Initiation Visit (SIV)

SIV will be performed to ensure that the facility and medications required for the trial are available in the study site and the investigators and staff involved in the study are aware of study objectives and GCP principles.

In the SIV, the role of the monitor, CRA, project manager, and the auditor will be defined clearly. After the introduction of the roles and responsibilities, staff will be taught the protocol details including timelines, sample size, eligibility criteria, protocol conformity, deviation, and reporting. The training should be documented. The eCRF details, ICD process, source documentation, randomization and serious adverse event reporting will be discussed and documented as well.

Site Monitoring Visit

Site monitoring visit will be performed to ensure the conduct of the study according to approved protocol and GCP principles.

Before the trial starts, monitoring visits will be planned for each study site and will be confirmed by the PI and other investigators through email or letter. The monitoring visits during the study will be scheduled in order to visit when the planned recruitment of patients will be in its 30% and 70% progress. Sponsor is responsible for the 30% monitoring visit and "Trial" CRO is in charge of the 70% visit. In each visit, study elements will be monitored including source data, consent forms, trial medicinal products accountability, adverse events, protocol compliance, team qualification, and training. The monitoring report will be prepared and reported to both PI and sponsor preferably in an arranged meeting.

Study Close-Out

Study close-out visit will be performed to ensure the proper documentation of the data and return of the medicinal product and the equipments related to the trial.

After the last visit of the last participant, site close-out will be scheduled in a meeting and in the presence of the investigator. The site close-out visit will start with a brief meeting with the investigator and a decision will be made regarding the disposal of the remaining investigational medicinal product. The study documents will be archived at the trial site. A copy all safety reports will remain with the investigator and one copy will be rendered to the sponsor. There will be a brief closure meeting with the investigator at the site and the site close-out report will be prepared after the visit.

Adverse Events

Adverse Event will be considered any medical event presented by the subjects involved in the study, which do not necessarily have a causal relation with the treatment in study. It will be reported as an adverse effect any symptom, sign (including any abnormal laboratory determination) or temporary disease associated to the use if the drug in study, whether they are or not etiologically related to it. The medical conditions present before the beginning of the study will only be considered as adverse events if they worsen during the study and cannot be attributable to the natural evolution of the disease.

Adverse drug reaction (ADR)—The World Health Organization defines an ADR as "any response to a drug which is noxious and unintended, and which occurs at doses normally used in man for prophylaxis, diagnosis, or therapy of disease, or for the modification of physiological function."

AEs will be considered a Serious Adverse Event (SAE) if the event:

- Results in death;
- Implies a death risk;
- Requires hospitalization;
- Extends prior hospitalization;
- Results in persistent or significant incapacity;
- Produces a congenital anomaly or malformation, or
- Requires medical or surgical intervention to avoid a permanent damage.

AEs Classification Based on Severity:

AEs will be classified according to its severity in relation to the guidelines established in the Common Terminology Criteria for Adverse Events (CTCAE, version 5.0, published on November 27, 2017). Grade refers to the severity of the AE. The CTCAE displays Grades 1 through 5 with unique clinical descriptions of severity for each AE based on this general guideline:

Grade 1: Mild; asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated.

Grade 2: Moderate; minimal, local or noninvasive intervention indicated; limiting ageappropriate instrumental ADL*.

Grade 3: Severe or medically significant but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care ADL**.

Grade 4: Life-threatening consequences; urgent intervention indicated.

Grade 5: Death related to AE.

* A Semi-colon indicates 'or' within the description of the grade.

* Activities of Daily Living (ADL)

*Instrumental ADL refer to preparing meals, shopping for groceries or clothes, using the telephone, managing money, etc.

**Self-care ADL refer to bathing, dressing and undressing, feeding self, using the toilet, taking medications, and not bedridden.

AE classification based on its relation with the treatment (Causality assessment):

In order to establish the relation between the AE or the DAR and the treatment in study, the following definitions will be considered:

- **Certain**: a clinical event including alterations in the laboratory tests manifesting with a reasonable temporal sequence related to the administration of the drug and cannot be explained by the current disease, or by other drugs or substances. The response to the drug suppression (released; dechallenge) should be clinically plausible. The event should be final from a pharmacological point of view, using, if necessary, a conclusive re-exposure procedure.
- **Probable/Likely**: a clinical event, including alterations in the laboratory tests manifesting with a reasonable temporal sequence related to the administration of the drug, which is unlikely to be attributed to the current disease, or other drugs and substances, and which when releasing the drug (dechallenge) a clinically reasonable response appears. No information on re-exposure (rechallenge) is required to assign this definition.
- **Possible**: a clinical event, including alteration in the laboratory tests manifesting with a reasonable temporal sequence related to the administration of the drug, but can also be explained by the concurrent disease, or by other drugs and substances. The information regarding the release of the drug may be missing or unclear.

- **Unlikely**: a clinical event, including alterations in the laboratory tests manifesting with an improbable temporal sequence related to the administration of the drug, and can be explained in a more plausible way by the concurrent disease, or by other drugs or substances.
- **Conditional/Unclassified**: a clinical event, including alterations in the laboratory tests, notified as an adverse reaction, of which it is essential to obtain more data in order to make a proper evaluation, or the additional data are under examination.
- **Non evaluable/Unclassifiable**: a notification that suggests an adverse reaction, but cannot be judged because the information is insufficient or contradictory and cannot be verified or completed in its data.

AEs Recording

All the undesirable and unexpected AEs that follow the administration of the drug will be accurately recorded in the medical record of the patient and in the corresponding section of the CRF. The event description should be recorded, as well as the temporal sequence regarding to the administration of the drug, its duration, the procedures performed for the diagnosis if appropriate, the results of the repeated exposure and the qualification made by the Researcher as regards its severity and its relation to the administered drug.

AE Reporting Responsibilities

Principle Investigator Responsibilities

- The investigator must report any SAE/R, which results in death or is life-threatening, to the sponsor and IEC within the maximum of 24 hours by fax, email or etc.
- The investigator must immediately and not later than seven calendar days report those SAE/Rs which are not life-threatening or do not result in death but may put subjects in danger or require intervention (medical or surgical) to prevent life-threatening outcomes. These SAE/R need to be reported to the sponsor and IEC as soon as possible but not later than seven calendar days of having taken notice of the SAE/R.
- The investigator should report to the sponsor and IEC all predictable adverse events of investigational medicinal product including for example injection site reactions, in case of patient withdrawal from study or adverse event with a greater frequency than expected.

Sponsor Responsibilities

- SUSARs which result in death, or are life-threatening; need to be reported by the sponsor as soon as possible but not later than seven calendar days, to IEC and IFDA, using CIOMS form (Suspect Adverse Reaction Report Form). A follow-up report is to be submitted within 15 calendar days.
- The sponsor must report SUSARs which are not life-threatening or do not result in death, but may put subjects in danger or require intervention (medical or surgical) to prevent life-threatening outcomes, as soon as possible but not later than 15 calendar days, to IEC and IFDA, using CIOMS form (Suspect Adverse Reaction Report Form). The follow-up report considering the relation between the investigational medicinal product and an adverse event is to be submitted as soon as possible.

- All the reports and follow-up results of SAE/R need to be reported to IFDA within maximum 15 calendar days after sponsor awareness of SAE/R.
- If the severity or frequency of predictable adverse events, for example, injection site reactions, results in patient withdrawal from the study, or in case of higher incidence than expected, the sponsor must report it to IFDA within maximum 15 calendar days.
- The sponsor must report all the information regarding the SAE and serious ADR that are reported during the course of the study and recommendations of investigators related to increasing of study risk for subjects to Food and Drug Administration of Iran within 15 calendar days.
- The sponsor must report to IFDA all recommendations from investigators about possible increased risk of adverse events or participants, within maximum 15 calendar days.

Follow-up

- The Investigator should take all appropriate measures to ensure the safety of the patients, notably he/she should follow up the outcome of any Adverse Events (clinical signs, laboratory values or other, etc.) until the return to normal or consolidation of the patient's condition;
- In case of any Serious Adverse Event, the patient must be followed up until clinical recovery is complete and laboratory results have returned to normal, or until progression has been stabilized. This may imply that follow-up will continue after the patient has left the study and that additional investigations may be requested by the Monitoring Team
- In case of any SAE brought to the attention of the Investigator at any time after cessation of Investigational Product and considered by him/her to be caused by the Investigational Product with a reasonable possibility, this should be reported to the Monitoring Team.

Withdrawal of patients

Participants may be withdrawn from the study for any of the following reasons:

- Withdrawal of consent by the patient
- Noncompliance, including refusal of study medical requirements, refusal of procedures as stated in the study protocol, or use of prohibited medications
- In the case of suspected pregnancy, a pregnancy test for Beta hCG will be requested, and if the test is positive, the patient will be excluded from the study
- Any hematologic, hepatic or renal AEs which needs cessation of study treatment
- The occurrence of an undesirable event that causes the investigator to consider the patient's exclusion from the study
- Not possible to follow the patient's condition (Loss to follow-up)
- In case patients' treatment need to be changed or use of prohibited drug according the protocol
- Progress od disease based on the RECIST criteria
- In case patients recive less than half of the study doses (less than 3 cycle)

The reason(s) for withdrawal should be stated clearly in the eCRF.

Patient Admission Criteria

Since all injections during the study are performed under the supervision of the nurse/nurses who have been trained, we will ensure that patients who are present at all injection days and whose injections are approved in the CRF, has an admission to the treatment.

Ethics and Dissemination

Research Ethics Approval

- Ethics committee approval is mandatory for start of this study
- No patient will be recruited to this study without a signed informed consent.
- Patients will be informed that they can leave the study anytime they desire with no need for any explanation.
- To ensure the confidentiality in case a form is lost, the name and surname of the patients will not appear on any forms.
- Adverse effect report forms will be evaluated after every visit. The research team is responsible for dealing with the immediate aftermath of any adverse event regardless of the event being directly related to the medication that is being studied.
- Before initiation of the trial, it will be reviewed with IFDA. The protocol, CRF, information for patients and informed consent form will be submitted to the ethics committee of Tehran university of medical sciences and Isfahan University of medical sciences, for review and approval according to international regulatory guidelines

Consent

The investigator will thoroughly explain the purpose of the study to patient. The patient will be provided with an information sheet and will be given sufficient time and opportunity to inquire about the details of the study and to decide whether or not to participate in the study, e.g. to give permission to use their data for investigative purposes, knowing their information will remain confidential. The informed consent form should be signed and dated by the patient and the person with whom they discuss the information regarding the consent form. The investigator will explain that the patient is completely free to refuse to give permission for his/her data to be used or to withdraw from the trial at any time and for any reason. Similarly, the investigator and/or sponsor will be free to withdraw the patient at any time for administrative reasons. Any other requirements necessary for the protection of the human rights of the patient will also be explained, according to GCP guidelines, declaration of Helsinki and local regulation for clinical trials.

Confidentiality

All study-related information will be stored securely at the study site.

To ensure confidentiality, randomization codes will be used on all the reports, gathered data, information regarding the study progress and administrative forms.

All records that contain names or other personal identifiers, such as subject identification form and informed consent forms, will be stored separately from study records.

All local databases will be secured with password-protected access systems.

Forms, lists, logbooks, appointment books, and any other lists which link participant's randomization code to other identifying information will be stored in a separate, locked file in an area with limited access.

All laboratory and other test results will be kept strictly confidential. All counseling and blood sampling will be conducted in private rooms, and study staff will be required to preserve the confidentiality of all participants.

Amendment

Any modifications to the protocol which may impact on the conduct of the study, the potential benefit of the patient or may affect patient safety, including changes to study objectives, study design, patient population, sample size, study procedures, or significant administrative aspects will require a formal amendment to the protocol. Such amendment will be agreed upon by Principal Investigator and sposnor and should be approved by IFDA and the Ethics Committee prior to implementation. Administrative changes to the protocol are minor corrections and/or clarifications that have no effect on the way the study is to be conducted. These administrative changes will be agreed upon by Principal Investigator and sponsor and will be documented in a memorandum.

Declaration of Interests

The presence or absence of any kind of financial and or nonfinancial relationship between the sponsor and principal investigator and investigators (with the exemption of this study contract) should be officially declared to IFDA as a written conflict of interest letter.

Funding Organization

All expenses of this study including patients' treatment and medicines, study conduct and performing and research related injuries compensation will be provided by Sponsor.

Access to Data

Only Principal investigator will have access to full dataset.

Ancillary and Post-Trial Care

Ancillary care (related to trial) will be provided by investigators under sponsor support for participants.

Dissemination Policy

No other publication is allowed before the primary publication. Any subsequent presentation or publication (including the sub-studies) by a study team member must be approved by the steering committee and chief Investigator and the primary publication should be cited. The final decision to publish any manuscript/ abstract/ presentation will be made by principal Investigator and the sponsor after prior notice to the Trial CRO for their review and comments.

Appendixes

Appendix 1: Informed Consent Form

Informed Consent Form

A Phase III, randomized, two armed, parallel, triple-blind, active controlled, equivalency clinical trial to determine the therapeutic efficacy and safety between Pertuzumab® (Manufactured by CinnaGen Co.) compared with active control group treated by Perjeta® (Pertuzumab, the reference drug, Manufactured by Genentech) in neoadjuvant treatment of HER 2 positive Breast Cancer patients

Mrs./Miss ……

We invite you to participate in the above-mentioned clinical study. Information about this research is provided in this sheet, and you are free to join this research or not.

You do not have to make an immediate decision and you are given a deadline to announce your opinion after consulting the research team and anyone you desire, about participation and your involvement in this research is entirely voluntarily. Before signing this consent, make sure that you understand all the information in this form and all of your questions are answered.
Investigator:

1) I know that purposes of this research are evaluation of the efficacy and safety of Pertuzumab (Manufactured by CinnaGen Co.) compared to Perjeta® (Manufactured by Genentech)

2) I know that my participation in this study is voluntary and I am not obligated to participate in this research. I was assured that if I were not willing to participate in this research, I would not be deprived of routine diagnostic and therapeutic care and my therapeutic relationship with the health center and my physician will not get affected.

3) I know that even after agreeing to participate in the research, I can resign freely at any time after informing the researcher, without giving any reason and my withdrawal from the research will not deprive me of the usual treatment services.

4) This is how my participation is in this study:
Whenever the physician diagnosed that I need to receive Pertuzumab to improve my clinical conditions, and I consented to receive it, treatment regimen according to the study protocol of will

be started and I will allocated in one of the treatment group (one of the treatment group receive Pertuzumab (Manufactured by CinnaGen Co.) and the next group will receive Perjeta® (Manufactured by Genentech)). During the study, I have to go to trial site in order to get my treatment drugs, laboratory test and other interventions of visits. The activity of visits of study include:

Duration of study is 18-20 weeks with pathological sampling test after surgery. With considering screening visit, I will have 8 visits (each visit is about 15 min without considering time of drug administration) in this study. The difference between screening and first visit (treatment visit) is maximum 2 weeks and the difference between other visits is 3 weeks. In screening visit, my health condition, medical history, height, weight and vital sign will be evaluated by physician. In addition, one blood sampling (3-5 mm) for general and specific laboratory test (CBC diff,. BUN, Cr, Bilirubin, ALP, AST, ALT, NA, K, Mg, Ca, P, Cl, Albumin, Glucose, LDH, INR, PTT/aPTT, HIV, HCV, HBs-Ag, assessment of antibody formation against drug, Unrine analysis, and pregnancy test if it is required) will be drawn. Other evaluation include Echocardiography, MRI, and ECG. If all of my lab results, imaging and clinical examination were confirmed by physician and I passed all eligibility criteria of the study, I can enrolled in the study after signing this consent form. During the study I will receive these drugs: in each treatment visits I will receive Carboplatin, Docetaxel, Trastuzumab and one of the Pertuzumab drugs. I know that my allocation to each treatment group will be randomly. My physician informed me that this blinding of the treatment is scientific and is to evaluate the efficacy of the drugs. My physician informed me that all of the treatments and interventions will be based on the routine treatment procedure and I do not need to have any extra visit of clinic. Regardless of treatment group, I will receive all of the required interventions and whenever participating in this study will considered not safe for me, I can discontinue the study and will receive appropriate treatment after my quit. I know that all of the drugs of this study between two treatment arms are similar and the only difference is related to Pertuzumab group.

In my next visits (visit 1-6), in each one of them, my health condition will be assessed by physician (investigator). In each visit and before drug administration (every 3 weeks), one blood sampling will be drawn in specified laboratory and these test include: CBC diff,. BUN, Cr, Bilirubin, ALP, AST, ALT, NA, K, Mg, Ca, P, Cl, Albumin, Glucose, LDH, assessment of antibody formation against drug (this test will be done with other lab test and there is no need to give extra blood sample).

In every 6 weeks interval (visit 3, 5, 7), echocardiography test is planned for me. In addition, in first visit and last visit (visit 7) or any other visit that physician consider necessary, MRI test is planned for me and there will be laboratory test in last visit which includes similar laboratory test with previous visits.

In addition, in each visit, questions related to my health condition, other used drugs will be asked. I know that laboratory and imaging test can be repeated based on the physician discretion.

I am assured that if in any time of the study, I had progress, I can discontinue the study and another treatment will start for me based on the physician discretion. In addition, if I received less than 3

doses of pertuzumab (less than 3 cycle of 6 cycle) due to my condition, I will be withdrawan form the study and new treatment based on the physician decision will be started for me.

By considering my health condition, I accept that during the study duration and 6 months after that, I will not try to be pregnant and if there is a chance that I became pregnant or had delay in my menstrual period, I will forthwith inform my physician (To be sure about pregnancy, there is pregnancy test in screening, visit 3 and 6)

5) The possible benefits of my participation in this study are as follows:

Pertuzumab is one of the novel drug which has FDA and EMA approval for breast cancer indication. The efficacy of Pertuzumab in breast cancer before and after surgery is confirmed and addition of this drug to chemotherapy regimen can increase significantly clinical response without increasing AE frequency. By participating in this study, I can receive pertuzumab while this drug is not easily accessable in my country and I do not need to pay any cost for this drug. In addition, I will be examined and evaluated by the physicians with greater precision and sensitivity regarding my condition and the side effects of drugs. Also, the effects of my treatment will be carefully evaluated

6) The possible harms and adverse events of my participation in this research are as follows:

It is possible to experience these adverse events after participation in this study, including diarrhea, cardia issue, hematologic issue, dermatologic issue, gastrointestinal issue, or reduce of white blood cells, alopecia, nausea, vomiting (most commonly related to chemotherapy regimen drugs and not necessarily related to Pertuzumab). The mentioned adverse events, can be occurred in both treatment groups and can be observed by other chemotherapy regimen too. I have to remember that these adverse events are temporary and after cessation of the tretamnet, all of these adverse events will be disappear after specific time. In addition, if any AEs were occurred, I or my family should inform the physician as soon as possible. This AE can be not related to the investigational drug of the study, but should be reported to the physician of the study.

7) In the case of unwillingness to participate in the research, the usual services (therapeutic, diagnostic, etc.) for me will be provided. My treatment will be based on the physician decision in case of unwillingness to participate in the research

8) I know that the researchers of this study will keep all of my information confidential and are only allowed to publish the overall and collective results of this research without mentioning my name and my profile.

9) I know that the Ethical Committee in my study is allowed to have access to my information to monitor my rights.

10) I know that I should not pay any costs of the trial drugs and tests including Chemotherapy regimen drugs, Trastuzumab drugs, Pertuzumab drugs, Pegagen drugs, laboratory test, MRI, Echocardiography, ECG, pathology test

11) Physician of the study and Mr. Siavash Bakhshian (sponsor representative) has been introduced to me for answering my questions, and I was told that during the study any time a health problem occurred to me or if I had a question regarding participation in this research, I can share with him and ask for guidance.

12) I know that if during and after the research any physical and mental problems arose because of my participation in this research, it will be the responsibility of the physician to treat the complications and the related damages.

13) I know that if I have a problem or objection to executors of the research or the research process, I can contact the ethic Committee of the Tehran University of Medical Sciences at the address of: Room 605, 6th Floor, Central headquarters of Tehran University of Medical Sciences, Qods Street, Keshavarz Blvd., Tehran, Iran. Telephone: 009821-81633626 or ethic committee of Guilan reseach center at the address of: Research and technology building, west beheshti blv, Rasht, Guilan and present my problem either verbally or in writing.

14) This form of information and informed consent is provided in two copies and will be signed by the physician and me. A signed copy will be given to me and a signed copy will be given to the physician.

This section should be completed by patient

I have read and understood the explanations mentioned above, and based on that, I declare my informed consent to participate in this research.

Participant signature:

This section should be completed by investigator (physician) of the study

I consider myself bound to comply with the obligations of the executor in the above provisions, and I undertake to work on the rights and safety of people participating in this research.

Investigator signature:

Appendix 2: Classification of Breast Cancer

Tumor classification	Lymph Node classification	Anatomic Staging	Breast Cancer type
T2	N0	IIA	Operable (Early Stage)
T2	N1	IIB	Operable (Early Stage)
T3	N0	IIB	Operable (Early Stage)
T2	N2	IIIA	Locally Advanced
T3	N1	IIIA	Operable (Early Stage)
T3	N2	IIIA	Locally Advanced
T4a	N0	IIIB	Locally Advanced
T4b	N0	IIIB	Locally Advanced
T4c	N0	IIIB	Locally Advanced
T4d	N0	IIIB	Inflammatory
T4a	N1	IIIB	Locally Advanced
T4b	N1	IIIB	Locally Advanced
T4c	N1	IIIB	Locally Advanced
T4d	N1	IIIB	Inflammatory
T4a	N2	IIIB	Locally Advanced
T4b	N2	IIIB	Locally Advanced
T4c	N2	IIIB	Locally Advanced
T4d	N2	IIIB	Inflammatory
T2	N3	IIIC	Locally Advanced
T3	N3	IIIC	Locally Advanced
T4a	N3	IIIC	Locally Advanced
T4b	N3	IIIC	Locally Advanced
T4c	N3	IIIC	Locally Advanced
T4d	N3	IIIC	Inflammatory

Appendix 3: RECIST 1.1

New response evaluation criteria in solid tumours: Revised RECIST guideline (version 1.1)

Evaluation of target lesions

Complete Response (CR): Disappearance of all target lesions. Any pathological lymph nodes (whether target or non-target) must have reduction in short axis to <10 mm.

Partial Response (PR): At least a 30% decrease in the sum of diameters of target lesions, taking as reference the baseline sum diameters.

Progressive Disease (PD): At least a 20% increase in the sum of diameters of target lesions, taking as reference the smallest sum on study (this includes the baseline sum if that is the smallest on study). In addition to the relative increase of 20%, the sum must also demonstrate an absolute increase of at least 5 mm. (Note: the appearance of one or more new lesions is also considered progression).

Stable Disease (SD): Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum diameters while on study.

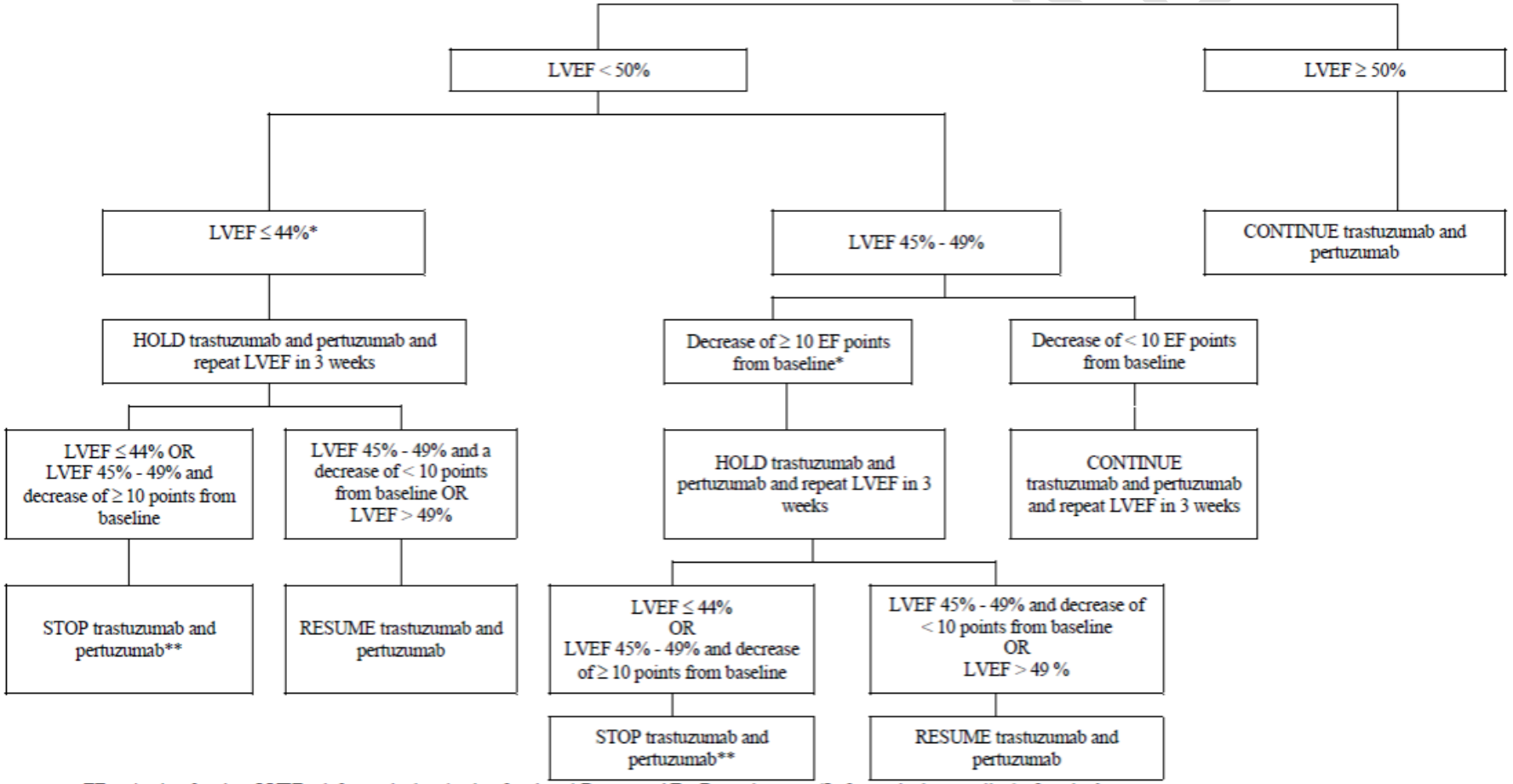
Evaluation of non-target lesions

Complete Response (CR): Disappearance of all non-target lesions and normalisation of tumour marker level. All lymph nodes must be non-pathological in size (<10 mm short axis).

Non-CR/Non-PD: Persistence of one or more non-target lesion(s) and/or maintenance of tumour marker level above the normal limits.

Progressive Disease (PD): Unequivocal progression of existing non-target lesions. (Note: the appearance of one or more new lesions is also considered progression).

Appendix 4: Discontinution Algorithm of Trastuzumab and Pertuzumab based on heart function



EF = ejection fraction; LVEF = left ventricular ejection fraction. * Report as AE – Reporting term 'Left ventricular systolic dysfunction'

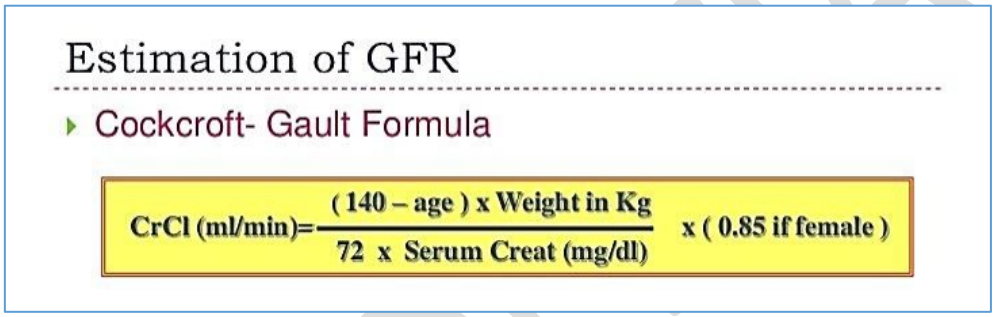
** Report as AE (eCRF AE eform) and Non-Serious Event of Special Interest (SAE form) Reporting term 'Left ventricular systolic dysfunction'

Appendix 5: Carboplatin Dosing

Carboplatin Dose (mg) = Target AUC * (GFR + 25)

AUC* = 6 mg/mL x min

AUC* or area under curve based on the kidney function and Calvert Formula, calculated to required dose of patient according to mg. According to Calvert Formula, the final dose (mg) is multiplication of AUC and GFR+25. In case of application of Serum Creatinine, the maximum limit of GFR will be 125 mL/min.



The below table is based on the above formula and is for easy of application.

GFR	20.0-22.4	22.4- 27.4	27.4-32.4	32.4-38.2	38.2-45.7	45.7-54.1
Dose	270 mg	300 mg	330 mg	360 mg	400 mg	450 mg

GFR	54.1-63.2	63.2-74.0	74.0-85.7	85.7-98.9	98.9-114.8	114.8-132.2
Dose	500 mg	560 mg	630 mg	700 mg	790 mg	890 mg

Appendix 6: Heart Evaluation Questionnaire

In each visit and before any drug interventions, patient's heart condition should be evaluated based on this questionnaire. Please record the result of this evaluation in related part of eCRF.

- Have you recently experienced shortness of breath with physical activity or walking?
- Have you recently feel palpitation while you are in rest or in activity?
- Have you experienced cough or shortness of breath while you are resting in lying down position?
- Have you recently felt swelling in your hands and feet, or tightness of shoes or a ring?

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