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DEPARTMENT OF MEDICAL ONCOLOGY AND EXPERIMENTAL THERAPEUTICS

TITLE: ^{64}Cu-DOTA-trastuzumab Positron Emission Tomography in Women with Advanced *HER2* Positive Invasive Breast Cancer

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Table of Contents

1.0 Background

The term “breast cancer” encompasses a number of different diseases that are clinically defined by hormone receptor status and *HER2* expression. *HER2* is a transmembrane protein in the epidermal growth factor family. *HER2* has tyrosine kinase activity that results in intracellular signaling and activation of genes for cell growth and survival. Women whose cancers overexpress the *HER2* protein have a distinct natural history and are candidates for treatment with trastuzumab. Trastuzumab is at humanized IgG-1 antibody that binds to the ectodomain of *HER2*. When combined with chemotherapy, trastuzumab significantly improves the survival of women with both early stage and advanced disease.[1-3] The pathologic assessment of *HER2* status is made on the primary tumor or metastatic foci. *HER2* overexpression and candidacy for trastuzumab is defined as 3+ staining by immunohistochemistry (IHC) or gene amplification by fluorescence in situ hybridization (FISH).[4, 5] Recently, trastuzumab has been shown to be of benefit also in patients whose cancers are *HER2* negative by IHC and have low level amplification of *HER2* when associated with chromosome 17 polysomy. Additional data from the NSABP B31 trial suggest that some women with *HER2* negative disease benefit from adjuvant trastuzumab.[5, 6] The American Society of Clinical Oncology and American College of Pathology convened a panel of experts to define guidelines for the pathologic assessment of *HER2*. In these recently published guidelines it is noted that when multiple measures of *HER2* are performed at different institutions, the discordance rate (positive versus negative) may be as high as 20%. While assessment of *HER2* by FISH is considered to be the “gold standard” for measuring *HER2*, this panel of experts concluded that FISH (ratio of *HER2* gene signals to chromosome 17 signals) amplification level of less that 1.8 was “negative” and that levels greater than 2.2 were positive. An “indeterminate” level of *HER2* was defined between 1.8 and 2.2. [7] The impact of trastuzumab on disease outcome is considerable. However, the cost, potential risk of cardiac toxicity, and inconvenience of intravenous administration make it imperative that an

accurate assessment of *HER2* be made in all women with breast cancer. We hypothesize that a functional assessment of *HER2* will be more accurate than pathologic assessment.

At initial diagnosis, the *HER2* status of the primary tumor and axillary nodes is generally concordant.[8] Similarly, when *HER2* assessment is performed on multiple sites of metastatic disease in the same patient, the concordance among different sites is 95%. [9-11] Such data suggest that the *HER2* status is consistently positive or negative. However, the majority of women develop metastatic disease after having been treated for early stage breast cancer. Following systemic treatment of early stage disease, the *HER2* status may change from being *HER2* positive to *HER2* negative or from *HER2* negative to *HER2* positive. In women whose disease recurs after treatment for early stage disease, the *HER2* status in the metastases may differ from that of the primary tumor as often as 40%. [8, 11, 12] These data suggest that systemic treatment may have an effect on the *HER2* status. Neoadjuvant chemotherapy plus trastuzumab was administered to 141 women with *HER2* positive breast cancers. A complete pathologic response rate was achieved in 72 (51%) women. The primary tumor was *HER2* negative in one third of patients with residual cancer after neoadjuvant chemotherapy and trastuzumab.[13] Such data support a role for the serial assessment of *HER2* status across the continuum of the breast cancer disease process. The assessment of *HER2* status from a metastatic focus requires a biopsy procedure and the location of the metastasis may be technically difficult to access with a needle biopsy. A less invasive measurement of *HER2* would allow treating physicians to repeatedly assess the status of *HER2* before and after therapeutic interventions. We believe that positron emission tomography (PET) using radiolabeled trastuzumab will provide such an assessment.

Trastuzumab in combination with chemotherapy improves the overall survival for women with early stage and metastatic *HER2* positive breast cancers. However, the activity of trastuzumab as a single agent in metastatic disease is modest. Objective tumor responses are reported in 35% of women treated with trastuzumab as first-line therapy and 18% in those

who have been previously treated. [14, 15] This raises the possibility that trastuzumab is not of benefit to all *HER2* positive breast cancers. Given the cost and potential toxicity of trastuzumab, an assessment of the *HER2* status that predicts for benefit to trastuzumab is needed.

Positron emission tomography (PET) provides a non-invasive way of studying tumor location, metabolic functions, and response to therapy. In breast cancer, PET imaging is used to stage women with advanced disease and to assess response to chemotherapy and endocrine therapy before tumor regression can be documented on clinical or radiological exam [16-18]. The ability to identify patients who may benefit from systemic therapy is critically important to the quality of life of patient; toxicities from ineffective therapies are prevented and medical costs are contained.

Radiolabeled trastuzumab has effectively been used to study *HER2* positive breast cancers in animal models and in humans, in part because trastuzumab has a high rate of internalization, which results in trapping of radiometals within the cell.[19-21] Researchers at City of Hope have utilized ^{111}In-DOTA-trastuzmab to image women with advanced *HER2* positive invasive breast cancer. The agent has variable accuracy in identifying *HER2* positive disease on planar and SPECT scans.[22] In the City of Hope experience, ^{111}In-DOTA-trastuzmab identified areas of known metastasis in 4 of 7 patients.[23] The positron emitting isotope ^{64}Cu has a number of potential advantages over ^{111}In including: improved image resolution with PET and lower doses of radiation to the patient. ^{64}Cu-DOTA-trastuzumab, first developed at City of Hope, accurately identified *HER2* positive breast cancer in nude mice bearing an MCF7 *HER2* overexpressing xenograft.[21, 24] Therefore, it is likely that ^{64}Cu-DOTA-trastuzumab-PET will be a superior imaging agent to ^{111}In-DOTA-trastuzmab. We propose to utilize ^{64}Cu-DOTA-trastuzumab-PET to image women with metastatic *HER2* positive breast cancer. We anticipate that the agent will be well tolerated and that the radiation absorbed doses to normal

tissues using ^{64}Cu-DOTA-trastuzumab will be much lower than the doses incurred with ^{111}In-DOTA-trastuzmab.

HER2 is expressed on a number of normal tissues such as the heart and kidney. Some investigators have reported that ^{111}In-trastuzumab uptake in the heart predicts for potential cardiac toxicity from trastuzumab, while others have not. [22, 25]

Because normal tissues that express *HER2* will bind the radioabeled (hot) antibody, administration of a "cold dose" of antibody might improve the tumor specificity of the radiographic image. The studies that have utilized radiolabeled trastuzumab as an imaging agent have consistently administered a large "cold dose" of trastuzumab prior to radiographic imaging. We hypothesize that the dose of "cold antibody" is critical for image quality. We believe that too high a dose of "cold antibody" will reduce tumor uptake and thereby compromise the quality of the radiographic image. In the clinical trial of ^{111}In-DOTA-trastuzmab at City of Hope, a high dose of "cold" trastuzumab (4mg/kg) was administered prior to imaging. This may explain why ^{111}In-DOTA-trastuzmab failed to image disease in 3 of 7 women with metastatic *HER2* positive breast cancer.

The primary objective of this pilot study is to determine the optimal dose of cold antibody required for ^{64}Cu-DOTA-trastuzumab PET to produce a high image quality in women with metastatic *HER2* positive breast cancer. In addition, we will assess the impact of the cold antibody dose on cardiac uptake of ^{64}Cu-DOTA-trastuzumab. To establish the optimal dose of cold antibody, we will image women with documented metastatic breast cancer with 3 different protein (trastuzumab) doses - 5 mg, 50 mg, and 4 mg/kg.
As of February 10, 2012, we have imaged 7 women; two received the 5 mg dose and 5 with 50 mg. The 50 mg trastuzumab dose had a marked effect on liver uptake of ^{64}Cu [SUV = 23±3 (mean±sd) and 6.3±0.6 for 5 and 50 mg tras, respectively; $p < 0.05$]. 72 CT+ lesions (32 bone, 28 nodal, 9 liver, 2 breast, 1 lung) were included in Day 1 and/or Day 2 scans. Of these,

66 of 72 (92%), 48 of 63 (76%) and 36 of 44 (82%) were detected on FDG, D1 and D2 scans, respectively. Lesion SUV_{max} was 9±5 (n=37, range 3-23) on D1 and 11±6 (n=27, range 1-29) on Day 2. ^{64}Cu-DOTA-trastuzumab-to-^{18}FDG SUV_{max} ratios were 1.0±0.8 (range 0.2-4.3) and 1.3±1.1 (range 0.2-3.5) for Days 1 and 2. The uptake by ^{64}Cu trended to be greatest in the liver, then bones, then lymph nodes.

^{64}Cu-DOTA-trastuzumab PET has been studied by investigators at Memorial Sloan Kettering Cancer Center. Women with advanced HER2 positive breast cancer, who were receiving either trastuzumab 2 mg/kg weekly or 6 mg/kg every 21 days were considered for the imaging study. The data is only published in abstract form with data from the first 9 women studied. The authors reported that the imaging was feasible and safe. However, they identified a low sensitivity of tumor detection with Cu-64 trastuzumab and hypothesize that the concurrent use of therapeutic doses of trastuzumab may have saturated tumor sites and compromised the image quality. Given this experience, we do not believe that the highest planned dose of cold trastuzumab (4 mg/kg) in our initial study design would be a wise use of patient resources. We have demonstrated that the 50 mg preimaging dose of cold trastuzumab overcomes non-specific tissue binding of HER2 and provides excellent image quality. To test the superiority of a slightly higher dose of cold trastuzumab would require many additional patients. We are thus satisfied by the image quality obtained with the pre-imaging dose of 50 mg of trastuzumab, Aim 1. We will therefore proceed to the second series of experiments which will test whether tumor uptake on ^{64}Cu-DOTA-trastuzumab PET imagine correlated with tumor expression of HER2. To accomplish this goal, we will perform ^{64}Cu-DOTA-trastuzumab PET imaging in an additional 15 patients with advanced cancer that are HER2 1+ (5 patients), HER2 2+ (5 patients), and HER2 3+. (5 patients).

In preclinical models, resistance to trastuzumab is associated with cellular changes that amplify the PI3K pathway. For example, low levels of PTEN, increased IGF1 and increased *HER2* heterodimerization have been shown to activate the PI3K pathway and decrease the

effectiveness of trastuzumab.[24] Because of the importance of the EGFR pathway in preclinical models of trastuzumab resistance, we will perform an exploratory analysis of biomarkers for the PI3K/Akt pathway. We know that ^{64}Cu-DOTA-trastuzumab is internalized. Therefore we hypothesize that a negative ^{64}Cu-DOTA-trastuzumab PET in a patient whose tumor is *HER2* positive suggests resistance to trastuzumab. In this setting, biomarkers that are consistent with amplification of the EGFR pathway provide support for trastuzumab resistance.

We believe that functional imaging with ^{64}Cu-DOTA-trastuzumab PET has the potential to improve the accuracy of *HER2* assessment. Future studies will address tumor biomarker assessment with serial ^{64}Cu-DOTA-trastuzumab PET and [^{18}F]FDG/PET before and after treatment with trastuzumab to determine which patients are likely to benefit from the drug.

2.0 Study Objectives

2.1 Determine the dose of pre-administered cold antibody that optimizes image quality of ^{64}Cu-DOTA-trastuzumab PET without increasing the radiation dose to the heart in women with metastatic *HER2* positive breast cancer.

2.2 Determine whether tumor uptake on ^{64}Cu-DOTA-trastuzumab PET correlates with tumor expression of *HER2* in women with metastatic disease.

2.3 Perform an exploratory analysis of the relationship between uptake on ^{64}Cu-DOTA-trastuzumab PET, *HER2* overexpression, and inactivation of the PI3K/Akt pathway.

3.0 Research Design

This is a prospective clinical trial in two parts. The initial study enrolled 7 women with metastatic *HER2* positive breast cancer who will receive one of 3 different protein doses of trastuzumab to determine the dose that provides the optimal ^{64}Cu-DOTA-trastuzumab PET image quality. The optimal “cold dose” of trastuzumab has been determined to be 50 mg. This dose will be used to image an additional 15 women whose cancers stain 1+, 2+, or 3+ by IHC.

Five women with each level of IHC staining will undergo ^{64}Cu-DOTA-trastuzumab PET imaging and the intensity of ^{64}Cu-DOTA-trastuzumab PET uptake will be correlated with the IHC measurement of *HER2*.

4.0 Eligibility Part I (Determination of the cold dose)

4.1 Participants must be women who have histological confirmation of metastatic invasive breast cancer that has metastasized outside the region of the primary tumor and axilla. Biopsy must be obtained within 28 days prior to study. Patients must have metastatic disease in lung, liver, soft-tissue or bone to qualify for the study (more than one site is permissible).

4.2 At least 1 non-hepatic site of metastasis $\geq$ 2 cm in mean diameter must be identified in addition to the site that was biopsied.

4.3 The cancer must over express HER2 as determined by IHC and FISH.

4.4 Patients may have received trastuzumab in the adjuvant, neoadjuvant, or metastatic setting, but cannot have received the drug within the prior 2 months.

4.5 Participants must have normal cardiac ejection fraction.

5.0 Eligibility Part 2 (correlation of HER2 expression with PET uptake)

5.1 Participants must be women who have histological confirmation of metastatic invasive breast cancer that has metastasized outside the region of the primary tumor and axilla. Biopsy must be obtained within 28 days prior to study. Patients must have metastatic disease in lung, liver, soft-tissue or bone to qualify for the study (more than one site is permissible).

5.2 At least 1 non-hepatic site of metastasis site $\geq$ 2 cm in mean diameter must be identified in addition to the site that was biopsied.

5.3 Participants with HER2 1+, 2+ and 3+ by IHC are eligible.

5.4 Patients may have received trastuzumab in the adjuvant, neoadjuvant, or metastatic setting, but cannot have received the drug within the prior 2 months.

5.5 Participants must have normal cardiac ejection fraction.

6.0 Ineligibility

6.1 Participants who have received trastuzumab and/or lapatinib within the prior2 months

6.2 Participants who are not considered candidates for trastuzumab

6.3 Metastatic disease in a single site

6.4 No metastatic site $\geq$ 2 cm

6.5 Concurrent malignancy other than skin cancer

6.6 Inability to provide informed consent

6.7 Participants who are pregnant

7.0 Recruitment Process

7.1 Participants will be recruited by the treating Medical Oncologists from patients receiving their breast cancer treatment at City of Hope.

7.2 Recruitment letters or advertisements will be reviewed and approved by the IRB prior to their use to recruit potential study subjects.

8.0 Informed Consent Process

8.1 The Principal Investigator or IRB approved named designate will explain the nature, duration, purpose of the study, potential risks, alternatives and potential benefits, and all other information contained in the informed consent document as well as their rights as a research subject (Experimental Subjects Bill of Rights) and the HIPAA research authorization form. Research participants will be informed that they may withdraw from the study at any time and for any reason without jeopardizing, include as applicable, their future care or their employment at City of Hope or any relationship they have with City of Hope. After signing the study consent form, HIPAA authorization form and the Experimental Subject's Bill of Rights, research subjects will undergo an assessment of their comprehension of the study by the Research Subject Advocate, followed by eligibility testing. Should sufficient doubt be raised regarding the adequacy of comprehension, further clarifications will be made and the consent comprehension assessment may be repeated until a satisfactory result is obtained. Prospective research subjects who cannot adequately comprehend the fundamental aspects of the research study with a reasonable amount of discussion, education and proctoring will be ineligible for enrollment. Following this procedure, the protocol management team will review the results of eligibility testing and determine if the research subject is a candidate for study enrollment.

9.0 Study Procedures/Research Interventions

9.1 Treatment Plan for determination of the "Cold dose"

9.1.1 Fifteen women with metastatic breast cancer that is HER2 positive by IHC and FISH will be recruited from the Medical Oncology Clinics at City of Hope.

9.1.2 As patients will ultimately receive trastuzumab-based therapy, a baseline cardiac ejection fraction, by MUGA or ECHO cardiogram, will be performed.

9.1.3 All participants will undergo a history and physical exam and radiographic staging workup, including whole body CT [^{18}F]FDG/PET.

9.1.4 Five women will receive a total protein dose of trastuzumab of 5 mg, five will receive a total protein dose of 50 mg of trastuzumab and five will receive a total protein dose of 4 mg/kg of trastuzumab. Assignment of dose will be by stratification by site of metastatic disease (bone, lung, liver) so that an approximate balance in metastatic sites is achieved for each group (see statistics).

9.1.5 A PET scan of the regions of interest will be performed 24 and 48 hours after injection of ^{64}Cu-DOTA-trastuzumab.

9.1.6 Systemic therapy with trastuzumab-based therapy may be initiated one week after completion of the ^{64}Cu -DOTA-trastuzumab PET imaging,

9.1.7 Participants will be observed for toxicity for 1 year.

9.1.8 Antibodies to DOTA and trastuzumab will be assessed at 1, 3, 6, and 12 months after imaging.

As of February 10, 2012, part I of this study has been prematurely terminated early as the optimal dose of cold antibody was determined to be 50 mg of trastuzumab.

9.2 Treatment Plan to correlate HER2 positivity by IHC with of ^{64}Cu-DOTA-trastuzumab PET uptake

9.2.1 Fifteen women with metastatic breast cancer that is HER2 1+, 2+, and 3+ positive by will be recruited from the Medical Oncology Clinics at City of Hope.

9.2.2 As patients will ultimately receive trastuzumab-based therapy, a baseline cardiac ejection fraction, by MUGA or ECHO cardiogram, will be performed.

9.2.3 All participants will undergo a history and physical exam and radiographic staging workup, including whole body CT [18F]FDG/PET.

9.2.4 We will image 5 women whose metastatic cancer is 1+; 5 whose metastatic cancer is 2+, and 5 whose metastatic cancer is 3+ by IHC.

9.2.5 A PET scan of the regions of interest will be performed 24 and 48 hours after injection of ^{64}Cu -DOTA-trastuzumab.

9.2.6 Systemic therapy with trastuzumab-based therapy may be initiated one week after completion of the ^{64}Cu -DOTA-trastuzumab PET imaging.

9.2.7 Participants will be observed for toxicity for 1 year.

9.2.8 Antibodies to DOTA and trastuzumab will be assessed at 1, 3, 6, and 12 months after imaging.

9.3 Handling of specimens: All specimens will be handled in accordance with RSC 09001 and OSBC 90029

10.0 Study Calendar

	≤ 28 days	**≤14 days Prior to imaging**	**Day 0**	**Day 1**	**Day 2**	**Day 7+**	**Day 14**	**1, 3, 6 mo**	**1 year**
Biopsy of metastasis for diagnosis and *HER2* assessment	**X**								
History and Physical Exam		**X**							
Cardiac ejection fraction (by MUGA or ECHO)	**X****								

Pregnancy Test			**X[$]**						
Bone Scintigraphy	**X**								
[^{18}F]FDG/PET/CT		**X**							
Cold trastuzumab			**X[+]**						
^{64}Cu-DOTA-trastuzumab			**X***						
PET imaging				**X**	**X**				
Trastuzumab-based therapy						**X**			
Toxicity assessment**						**X**	**X**	**X**	**X**
CBC						**X**	**X**		
Antibodies to DOTA and trastuzumab			**X**					**X**	**X**
Pharmacokinetics[#]			**X**	**X**	**X**				

[$]Pregnancy test should be done may be done one day prior to the scan
[+]Research participants on 5 mg dose will not get additional cold dose herceptin
*Injection only
** If research participant has history of cardiac disease or possible cardiac toxicity, a MUGA should be obtained within 14 days of treatment.
[#] Participants will have one PK just prior to the Cu-OTA injection, one hour after the Cu-DOTA injection, one at the time of the first scan and one at the time of the second scan. (one red top tube to be delivered to Nicole in the Beckman Building, room 5212. No special handling needed)

11.0 Protocol Drug and Radiolabeled Imaging Agent

11.1 Trastuzumab

11.1.1 Drug description: Trastuzumab is a recombinant humanized monoclonal antibody that binds to the extracellular domain of the HER2. Trastuzumab is a sterile, white to pale yellow, preservative-free lyophilized powder for intravenous (IV) administration. The nominal content of each trastuzumab vial is 440 mg trastuzumab, 440 mg α,α-trehalose dehydrate, 9,9 mg L-histidine HCl, 6.4 mg L-histidine, and 1.8 mg polysorbate 20, USP. Reconstitution with 20 mL of the supplied Bacteriostatic Water for Injection (BWFI), USP, containing 1.2% benzyl alcohol as a preservative, yields a multi-dose solution containing 21 mg/mL trastuzumab, at a pH of approximately 6.

11.1.2 Procurement of trastuzumab: Trastuzumab will be supplied free of charge as part of the study to the patients receiving doses less than the conventional loading dose of 4 mg/kg. Lower doses will be charged to the grant.

11.1.3 Storage/stability: Vials of trastuzumab are stable at 2°-8° C (36°-46° F) prior to reconstitution. Do not use beyond the expiration date stamped on the vial. A vial of trastuzumab reconstituted with BWFI, as supplied, is stable for 28 days after reconstitution when stored refrigerated at 2°-8° C (36°-46° F), and the solution is preserved for multiple use. The 50mg dose will be taken from one 440mg vial into WebIDS (drug accountability system) at the time that a patient is treated vial, and the remainder of the vial will be used for non-study patients. Discard any remaining multi-dose reconstituted solution after 28 days. If unpreserved Sterile Water for Injection (SWFI) (not supplied) is used, the reconstituted trastuzumab solution should be used immediately and any unused portion must be discarded. Do not freeze trastuzumab that has been reconstituted. The solution of trastuzumab for infusion diluted in polyvinylchloride or polyethylene bags containing 0.9% Sodium Chloride

Injection, USP, may be stored at 2° -8° C (36 ° -46 ° F) for up to 24 hours at room temperature 2 °-25 ° C. However, because diluted trastuzumab contains no effective preservative, the reconstituted and diluted solution should be stored refrigerated 2°-8 ° C.

11.1.4 Reconstitutions and administration

11.1.4.1 Reconstitution

11.1.4.1.1 The diluent provided has been formulated to maintain the stability and sterility of trastuzumab for up to 28 days. Other diluents have not been shown to contain effective preservatives for trastuzumab. Each vial of trastuzumab should be reconstituted with 20 mL of BWFI, USP, 1.1% benzyl alcohol preserved as supplied, to yield a multi-dose solution containing 21 mg/mL trastuzumab.

11.1.4.1.2 Immediately upon reconstitution with BWFI, the vial of trastuzumab must be labeled in the area marked “Do not use after:” with the future date that is 28 days from the date of reconstitution.

Note: When administering trastuzumab to a patient with a known hypersensitivity to benzyl alcohol, trastuzumab must be reconstituted with SWFI, and only one dose per trastuzumab vial should be used. Trastuzumab which has been reconstituted with SWFI must be used immediately and any unused portion must be discarded. Use of other reconstitution diluents should be avoided.

Shaking the reconstituted trastuzumab or causing excessive foaming during the addition of diluent may result in problems with dissolution and the amount of trastuzumab that can be withdrawn from the vial. Use appropriate aseptic technique when performing the following reconstitution steps:

- Using a sterile syringe, slowly inject the 20 mL of diluent into the vial containing the lyophilized cake of trastuzumab. The stream of diluent should be directed into the lyophilized cake.
- Swirl the vial gently to aid reconstitution. Trastuzumab may be sensitive to shear-induced stress, e.g., agitation or rapid expulsion from a syringe. Do not shake.
- Slight foaming of the product upon reconstitution is not unusual. Allow the vial to stand undisturbed for approximately 5 minutes. The solution should be essentially free of visible particulates, clear to slightly opalescent and colorless to pale yellow.

11.1.4.2 Administration

The recommended loading dose of trastuzumab of 4 mg/kg is administered intravenously over 90 minutes. For participants in the arm assigned 50 mg, the trastuzumab will be administered over 15 min. For those assigned 5 mg, trastuzumab will be administered with the ^{64}Cu -DOTA-trastuzumab. The cold trastuzumab will be injected prior to the injection of ^{64}Cu-DOTA-trastuzumab. Trastuzumab will be administered by the research nurse in the Phase I unit of the hospital. Patients will be

monitored by the study nurse for any symptoms suggestive of an infusion reaction. If a patient experiences a reaction, the infusion is immediately discontinued and the treating physician is contacted. Benadryl and/or hydrocortisone may be administered before resuming the infusion at a slower infusion rate. If a patient continues to have reactions despite these interventions, the trastuzumab will be discontinued and the patient will be taken off study.

11.1.5 Drug accountability: Trastuzumab will be purchased by the City of Hope Pharmacy, which will store and control the drug. It will be prepared for administration in the outpatient pharmacy.

11.1.6 Discard of unused agent: Unused trastuzumab will be discarded in the chemotherapy discard containers within the outpatient pharmacy after 28 days.

11.1.7 Warnings and Contraindications

11.1.7.1 Cardiotoxicity: Administration of trastuzumab can result in the development of ventricular dysfunction and congestive heart failure. Signs and symptoms of cardiac dysfunction, such as dyspnea, increased cough, paroxysmal nocturnal dyspnea, peripheral edema, S3 gallop, or reduced ejection fraction, have been observed in patients treated with trastuzumab. Congestive heart failure associated with trastuzumab therapy may be severe and has been associated with disabling cardiac failure, death, and mural thrombosis leading to stroke.

11.1.7.2 Hypersensitivity reactions including anaphylaxis; severe hypersensitivity reactions have been infrequently reported in patients treated with trastuzumab. Signs and symptoms include anaphylaxis, urticaria, bronchospasm, angioedema, and/or hypotension. In some

cases, the reactions have been fatal. Trastuzumab infusion should be interrupted in all patients with severe hypersensitivity reactions. In the event of a hypersensitivity reaction, appropriate medical therapy should be administered, which may include epinephrine, corticosteroids, diphenhydramine, bronchodilators, and oxygen. Patients should be evaluated and carefully monitored until complete resolution of signs and symptoms.

11.1.7.3 Infusion reactions: In the postmarketing setting, rare occurrences of severe infusion reactions leading to fatal outcome have been associated with the use of trastuzumab. In clinical trials, infusion reactions consisted of a symptom complex characterized by fever and chills, and on occasion included nausea, vomiting, pain, headache, dizziness, dyspnea, hypotension, rash, and asthenia. These reactions were usually mild to moderate in severity. However, in postmarketing reports, more severe adverse reactions to trastuzumab infusion were observed and included bronchospasm, hypoxia, and severe hypotension. These severe reactions were usually associated with the initial infusion of trastuzumab and generally occurred during or immediately following the infusion. However, the onset and clinical course were variable. Delayed post-infusion events with rapid clinical deterioration have also been reported. Rarely, severe infusion reactions culminated in death within hours or up to one week following an infusion.

11.1.7.4 Pulmonary events: Severe pulmonary events leading to death have been reported rarely with the use of trastuzumab in the postmarketting setting. Signs, symptoms, and clinical findings include dyspnea, pulmonary infiltrates, pleural effusions, non-

cardiogenic pulmonary edema, pulmonary insufficiency and hypoxia, and acute respiratory distress syndrome. These events may or may not occur as a sequelae of infusion reactions. Patients with symptomatic intrinsic lung disease resulting in dyspnea at rest, may be at greater risk of severe reactions.

11.1.7.5 Other severe events reported rarely in the postmarketing setting include pneumonitis and pulmonary fibrosis.

11.2 Preparation of ^{64}Cu -DOTA-trastuzumab

11.2.1 Trastuzumab will be purchased from the City of Hope pharmacy and conjugated with active ester of DOTA (1,4,7,10-tetraazadodecane-1,4,7,10-tetracetic acid) in the COH biologics production facility (CBG) under cGMP compliant conditions. Each lot of trastuzumab-DOTA will undergo testing for sterility, potency, purity and lack of pyrogenicity. Vialing of the conjugated materials will be done in COH biologics production facility Fill and Finish area. An IND application will be filed with the FDA. Radiolabeling with ^{64}Cu will be carried out in the City of Hope Radiopharmacy under the direction of David Colcher, PhD. The ^{64}Cu will be purchased from the Mallinckrodt Institute of Radiology at the Washington University School of Medicine, which is preparing the radiolabel for clinical use. Labeling will be accomplished by incubating conjugated antibody with the ^{64}Cu for 45 minutes at 43^{o} C, followed by a chase with DTPA and subsequent purification on a size exclusion preparative grade Superdex-200 column. Appropriate fractions will be pooled and filtered to make up the patient dose, which will be formulated with human serum albumin. Patients will be injected via a peripheral vein with 15 mCi of ^{64}Cu-DOTA-trastuzumab; the protein dose of t he radiolabel given to the patient will be approximately 5 mg. The total trastuzumab content per ^{64}Cu -DOTA-

trastuzumab injected dose is less than 5 mg. All procedures will be done as specified in an FDA-approved IND.

We have estimated radiation dose from ^{64}Cu -DOTA-trastuzumab based on our previous work with ^{111}In-DOTA-trastuzumab [23] and anticipating that the normal tissue biodistribution will be the same for the ^{64}Cu radiolabel as for ^{111}In. The study protocol also requires an ^{18}F-fluordeoxyglucose (FDG) scan. Patients will be imaged once and twice following injection of ^{18}F-FDG and ^{64}Cu -DOTA-trastuzumab, respectively, for a total of three PET-CT scans. We have used estimated radiation doses from ^{18}F-FDG and CT to arrive at the following estimates of overall radiation absorbed dose:

	Estimated Total Absorbed Dose (cGy)*			
Organ/Tissue	**^{64}Cu-Trastuzumab** (15 mCi)	**^{18}F-FDG** (15 mCi)	**CT** 3 scans	**Total**
Heart wall	12.6	0.6	0.8	14
Spleen	10.5	0.6	0.8	12
Liver	9	1	1	11
Bladder wall	1	9	1	11
Kidneys	6.9	0.6	0.8	8
Red marrow	2.8	0.6	0.8	4
Other	$\leq$ 3.5	0.6	0.8	$\leq$ 5

*Values for ^{18}F-FDG and CT (specific for PET-CT to be used in the proposed study) were obtained from [ref].

Brix G, et al. Radiation exposure to patients undergoing whole-body dual-modality ^{18}F-FDG PET/CT examinations. J Nucl Med 2005;46:608-613.

12.0 Positron Emission Tomography (PET)

12.1 Imaging will be performed on a GE Discovery 16 Ste PET-CT scanner (axial field of view 15.4 cm). PET images will be performed in 3D mode (septa retracted) and corrected for tissue attenuation based on co-registered CT acquired during the same examination. PET images will be reconstructed with spatial resolution of approximately 9 mm full-width-at-half maximum (FWHM) using an iterative algorithm (OSEM).

12.2 ^{18}F-Fluorodeoxyglucose ([^{18}F]FDG). Standard [**^{18}**F]FDG PET-CT examinations will be performed prior to initiation of treatment. Patients will be injected via a peripheral vein

with 15mCi of [^{18}F]FDG. Large-area (eyes to mid-thigh) PET-CT scans will be obtained beginning at 1 hour post-injection; time per bed position during the PET scan will be 3 minutes.

12.3 ^{64}Cu -DOTA-trastuzumab. For patient convenience, the ^{64}Cu-antibody (^{64}Cu half life 12.7 h) will be injected immediately following the baseline [^{18}F]FDG-PET scan (18F half life 1.8 h). Patients will be injected via a peripheral vein with 15 mCi of ^{64}Cu-DOTA-trastuzumab. In order to allow the ^{18}F to decay and the antibody to accumulate in tumor, PET-CT scanning of ^{64}Cu will be delayed until 18 to 24 hours post injection (Day 1). A second scan will be obtained 42-48 hours post injection (Day 2). Because of the limited amount of activity to be injected and the fact that only 20% of ^{64}Cu decays produce a positron, the count rates will be low. To compensate, time per bed position will be relatively long and the axial field of view relatively short. (Day 1: 2-3 bed positions encompassing known tumors; 30 min per bed position if 2 bed positions, 20 min per bed position if 3 bed positions; Day 2: 1 bed position, 60 min). Based on prior phantom studies, we expect the scanning protocol defined above to yield adequate tumor visualization as well as measurements of tumor uptake and tumor: adjacent background activity concentration ratios ("tumor:bdg contrast") for tumors of at least 2 cm in diameter and average SUV (= tumor activity concentration/injected activity per unit body weight) of at least 3 in body regions for which tumor:bdg contrast is at least 4. The precision (coefficient of variation) of tumor SUV and tumor:bgd contrast measurements is expected to be about 10% and 15%, respectively.

13.0 Image Analysis

The PET-CT examinations will be analyzed and interpreted by the City of Hope Image Response Assessment Team (IRAT). Tumors will be selected for analysis on the basis of the pretreatment [^{18}F]FDG PET-CT examination. “PET –positive” lesions, defined as those clearly visualized by [^{18}F]FDG-PET, will be selected for analysis by the IRAT radiologist. Due to

possible non-specific antibody uptake secondary to biopsy, the tumor site biopsied for pathologic confirmation of HER2 positivity will not be used as an index lesion. The $^{Cu}64$ SUVs will be evaluated in tumors, adjacent non-tumor tissue and selected non-tumor organs and tissues (heart, extracardiac mediastinum, liver, skeletal muscle). Tumor sizes (product of maximum mutually perpendicular transaxial diameters as well as maximum axial diameter) will be estimated from coregistered CT. Tumor uptake of ^{64}Cu-DOTA-trastuzumab will be parameterized in terms of single-voxel maximum values SUVmax and whole-tumor volumes of interest (SUVwhtum) as defined from the coregistered CT images. Ratios of tumor to non-tumor activity concentration will also be calculated for adjacent tissue, extracardiac mediastinum, liver, and skeletal muscle. Receiver-operator curve (ROC) analysis will be performed to estimate optimal cutoff values of SUVmax, SUVwhtum, tumor:background and tumor:organ ratios for classifying tumors as “HER2 positive” or “HER2 negative.” Ratios of tumor ^{64}Cu to ^{18}F SUVs will also be computed and used to examine the relative effects of receptor expression and cellular viability as determinants of ^{64}Cu-DOTA-trastuzumab uptake in tumors. The selected cold antibody dose will be chosen to minimize the dose to the heart with the best ratio of tumor to heart. If multiple doses meet that criterion, (simulations suggest all doses will meet that criterion), the selected dose will be one with the highest image quality as defined by the ratio of tumor SUV to heart SUV. Specifically, for each of the five patients, the median tumor SUV (or the tumor SUV if there is only one non-biopsied lesion under consideration), will be divided by the heart SUV. This ratio will be obtained for all 5 patients. The main comparison will be the median of the five ratios (one value per dose level), compared across dose levels, to select the cold dose for future patients on this protocol.

14.0 Laboratory Evaluation

14.1 Antibody assessment will be performed in the laboratory of Dr. Colcher. Left over serum from the antibody assessment will be stored in the Translational Research Core of the Cancer Center for future research. Biomarker assessment: A core biopsy or an

excisional biopsy will be performed to confirm recurrent disease within 28 days of study initiations. After the diagnosis and HER 2 status have been determined, additional tumor will be stored in pathology for future assessment of PI3K, PTEN, IGF1-R, EGFR, and phospho-S6 by immunohistochemical staining. The DNA Sequencing Core will assess mutations in PI3K and PTEN. A pathologic assessment of HER2 is required for study participation. To ensure that the patient is HER2 positive, assessment of HER2 status will be performed by both IHC and FISH. The analyses of biomarkers of the EGFR pathway are not included in the grant proposal. Left over tumor from the required biopsy will be stored in pathology for future analysis.

14.2 Pharmacokinetic samples will be brought to Beckman Research Building, room 5212 and given to Nicole for processing.

15.0 Statistical Analysis

15.1 During study part I, no new patients will be enrolled until the previous patient has been imaged and observed for one week post ^{64}Cu-DOTA-trastuzumab PET imaging. If any adverse event (other than a grade 1 injection site reaction) is observed that is possibly related to the ^{64}Cu-DOTA-trastuzumab injection, the study will halt pending review at the DSMB. After each patient, a decision will be made regarding modification of the suggested scan times based on pharmacokinetics, biodistribution, and image quality.

15.2 Study Part I: Fifteen HER2 positive patients with metastatic disease will be enrolled, with 5 patients treated with 5mg of antibody, 5 patients treated with 50mg of antibody, and 5 patients treated with 4mg/kg of antibody. This will provide initial estimates of the biodistribution of the ^{64}Cu-DOTA-trastuzumab and the preferred dose of cold antibody. Patients will be assigned to a cold antibody dose group, with assignment based on dynamic balancing to minimization of unbalance in site of metastatic disease between the different doses. Patients with multiple sites will be considered in the strata that allows for best balancing (and as such can be re-classified). The information so derived

will also provide initial guidance on scan times and utility of ^{64}Cu-DOTA-trastuzumab as an imaging agent in HER2 positive invasive breast cancer.

15.3 Study part 2 The HER2 expression (biopsy) must be obtained on a different lesion than is evaluated with the ^{64}Cu -DOTA-trastuzumab PET, and, in fact, the ^{64}Cu -DOTA-trastuzumab PET may be discordant with the pathology findings. It is however, hypothesized that the mean PET score for HER2+ pathology tumors will be higher than HER2- pathology tumors. 5 HER2+ (3+) patients, 5 HER2(+/-) 2+ (may be HER2+ or HER2- depending on FISH), and 5 HER2 1+ patients (generally considered HER2-). will be enrolled. As HER2+ patients (approximately 30% in the COH patient population) have a disproportionate incidence of metastatic disease to the liver, we will require at least 3 patients in each group to have liver involvement to help insure that differences in PET imaging between the different HER2 status groups are not confounded with site of disease. Patient selection will be used to ensure that the three groups are similar with respect to sites of metastasis. Peak SUV value will be plotted (y-axis) against HER2+ status. With 9 patients, there is approximately 80% power to detect a linear correlation coefficient of 0.7, with a type I error of 5% (1-sided) assuming normally distributed covariates. Using a one-way ANOVA, there is 80% power to detect an effect size of 1.8 with a type I error of 5%. Finally, by considering all patients, with approximately 8 patients in HER2- group, and 7 in the HER+ group, there is 80% power to detect an effect size difference of 1.36 (1.36 x the common standard deviation in PET scores), using a two group t-test with a 0.05 one-sided significance level. We will also be examining for a trend in PET scores across the HER2 IHC scoring range for all patients.

16.0 Toxicity Assessment

16.1 Antibodies to DOTA and trastuzumab will be assessed at 1, 3, 6, and 12 months after imaging

16.2 Participants may begin trastuzumab-based therapy as early as 7 days after injection of ^{64}Cu -DOTA-trastuzumab. Patients will be monitored for 1 year. Side effects that are unrelated to trastuzumab or concurrent chemotherapy will be recorded.

16.3 Participants will be examined by the treating physician on days 7 and 14 and a complete blood count will be obtained. If there are any clinical concerns about cardiac toxicity, a MUGA will be obtained.

17.0 Data Management

17.1 Methods used for data collection:

17.2 Volunteer Identification: Participants identity will be linked to unique patient numbers (UPN)

17.3 Confidentiality: This research will be conducted in compliance with federal and state of California requirements relating to protected health information (PHI). The study will record individual imaging results and any side effects, and this will be linked to the subject's identity using a UPN. The Protocol Management Team (PMT) consisting of the PI (Joanne Mortimer), Andrew Raubitschek MD, statistician (Paul Frankel PhD), and study nurses (Mary Carroll RN, Lois Wright RN, Phyllis Broene RN) and representatives of the USAMRMC are eligible to review research records, but all information will be treated confidentially. No identifiers will be used in any publication of the study results.

17.4 Data Reporting

17.4.1 Confidentiality of Records

The original data collection forms will be stored at the originating institution. At COH, the forms will be kept in secure cabinets by Clinical Research Informatics Management CRIM. All records will be retained for a minimum of 2 years.

When results of this study are reported in medical journals or at meetings, identification of those taking part will be withheld. Medical records of patients

will be maintained in strictest confidence, according to current legal requirements. However, they will be made available for review, as required by the FDA or other authorized users such as the NCI, under the guidelines established by the Federal Privacy Act.

17.4.2 Patient Consent Form

At the time of registration, the original signed and dated patient's Informed Consent with the Experimental Subject's Bill of Rights (for the medical record) and three copies (for the patient, the research record, and the Coordinating Center) must be available. All Institutional, NCI, Federal, and State of California regulations concerning the Informed Consent form will be fulfilled. ^{64}Cu-DOTA trastuzumab is administered under IND 109,971 which is approved by and subject to the regulations of the FDA.

17.4.3 Data Collection Forms and Submission Schedule

All data will be collected using COH Biostatistics data collection forms. Copies of the completed forms will be sent to the City of Hope Department of Biostatistics for data entry and stored in a secure location. The original data collection forms will reside at the originating institution in secure location.

17.4.3.1 The Eligibility Checklist must be completed by a protocol nurse or clinical research associate and signed by a participating investigator prior to registering the patient.

17.4.3.2 Within two weeks of registration, the clinical research associate will submit Prior Therapy forms (Prior Therapy Summary – COH1987 and supplemental forms, as necessary) and On-Study forms (On-Study Hematology/Other – COH2658 and supplemental forms, as necessary).

17.4.3.3 Within 1 week of imaging, the clinical research associate will submit the following forms:

- Protocol Treatment – Drug Agent (COH2698)
- Monitoring & Follow-Up Summary (COH2001)
- Adverse Events Collection Form (COH2000)

17.4.4 Results Reporting

The COH, as sponsor of IND 109,971, will submit reports annually to the FDA within 60 days of the anniversary date that the IND went into effect (Month DD) in accordance with 21 CFR 312.33.

17.5 Sharing results with participants: Information from the ^{64}Cu-DOTA trastuzumab PET image will be shared with the patient and the treating physician if that information will impact on the patient's medical care.

17.6 Continuing Review and Final Report: As soon as approval notification is available from the IRB, a copy of the approved continuing review report with approval will be submitted to the HRPO for review.

18.0 Data Safety Monitoring Plan

18.1 Definition of Risk Level: This is a Risk Level 4 study, as defined in the "City of Hope Data and Safety Monitoring Plan", http://www.coh.org/dsmc/Pages/forms-and-procedures.aspx involving COH as IND holder and: investigational imaging.

18.2 : **Monitoring and Personnel Responsible for Monitoring** The Protocol Management Team (PMT) consisting of the PI, Collaborating Investigator, CRA/protocol nurse, and statistician is responsible for monitoring the data and safety of this study, including implementation of the stopping rules for safety and efficacy.

Table 1: City of Hope PMT Reporting Timelines for the DSMC

Risk Level	Phase	Standard Reporting Requirement
RL 1, RL2, and Compassionate Use Studies	No reports required	
3	I	Every 3 months from activation date, as indicated in MIDAS
3	Pilot, Feasibility, II-IV	Every 6 months from activation date, as indicated in MIDAS
4	Pilot, Feasibility, I-IV	Every 3 months from activation date, as indicated in MIDAS

Data and safety will be reported to the COH DSMC using the PMT report and submitted quarterly from the anniversary date of activation. Protocol specific data collection will include the following items**: antibody levels to DOTA and trastuzumab**.

18.3 Definitions

Adverse event (AE) - An adverse event is any untoward medical experience or change of an existing condition that occurs during or after treatment, whether or not it is considered to be related to the protocol intervention.

Unexpected Adverse Event [21 CFR 312.32 (a) – An adverse event is unexpected if it is not listed in the investigator's brochure and/or package insert; is not listed at the specificity or severity that has been observed; is not consistent with the risk information described in the protocol and/or consent; is not an expected natural progression of any underlying disease, disorder, condition, or predisposed risk factor of the research participant experiencing the adverse event.

Expected Adverse Event - Any event that does not meet the criteria for an unexpected event OR is an expected natural progression of any underlying disease, disorder, condition, or predisposed risk factor of the research participant experiencing the adverse event

Serious Adverse Event (SAE) [21 CFR 312.32] is defined as *any expected or unexpected adverse event* that results in any of the following outcomes:

- Death
- Is life-threatening experiences (places the subject at immediate risk of death from the event as it occurred)
- Unplanned hospitalization equal or greater than 24 hours)) or prolongation of existing hospitalization
- A persistent or significant disability/incapacity

- A congenital anomaly/birth defect
- Secondary Malignancy
- Any other adverse event that, based upon appropriate medical judgment, may jeopardize the subject's health and may require medical or surgical intervention to prevent one of the outcomes listed above (examples of such events include allergic bronchospasm requiring intensive treatment in the emergency room or at home, blood dyscrasisas of convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse).

Unanticipated problem (UP) – Any incident, experience or outcome that **meets all three** of the following criteria:

1. Unexpected (in term nature, severity, or frequency) given the following: a) the research procedures described in the protocol-related documents such as the IRB approved research protocol, informed consent document or Investigator Brochure (IB); and b) the characteristics of the subject population being studied; **AND**
2. Related or possibly related to participation in the research (possibly related means there is a reasonable possibility that the incident, experience, or outcomes may have been caused by the drugs, devices or procedures involved in the research); **AND**
3. Suggests that the research places subjects or others at greater risk of harm (including physical, psychological, economic, or social harm) than previously known or recognized.

18.4 Reporting of Unanticipated Problems and Adverse Events

Unanticipated Problems: Most unanticipated problems must be reported to the COH DSMC and IRB **within 5 calendar days** according to definitions and guidelines at http://www.coh.org/hrpp/Pages/hrpp-policies.aspx. Any unanticipated problem that occurs during the study conduct will be reported to the DSMC and IRB by submitting electronically in iRIS (http://iris.coh.org).

Serious Adverse Events - All SAEs occurring during this study, whether observed by the physician, nurse, or reported by the patient, will be reported according to definitions and guidelines at http://www.coh.org/hrpp/Pages/hrpp-policies.aspx and Table 2 below. Those SAEs that require expedited reporting will be submitted electronically in iRIS (http://iris.coh.org/).

Adverse Events - Adverse events will be monitored by the PMT. Adverse events that do not meet the criteria of serious OR are not unanticipated problems will be reported only in the continuation reports and PMT reports (see Table 2 below).

Table 2: City of Hope Adverse Event and Unanticipated Problem Reporting Timelines for the DSMC and IRB

Required Reporting Timelines to DSMC for AE/SAEs
Investigator Initiated Studies

Required Reporting Timeframe to DSMC		
Attribution	**UNEXPECTED**	**EXPECTED**
	Death while on active treatment or within 30 days of last day of treatment	
Possibly, Probably, Definitely	5 calendar days	
Unlikely, Unrelated		
	Death after 30 days of last active treatment/therapy	
Possibly, Probably, Definitely	5 calendar days	No reporting required
Unlikely, Unrelated	No reporting required	No reporting required
	Grades 3 and 4 AND meeting the definition of "serious"	
Possibly, Probably, Definitely	5 calendar days	10 calendar days
Unlikely, Unrelated	5 calendar days	10 calendar days
	Grades 1 and 2 AND resulting in "hospitalization"	
Possibly, Probably, Definitely	5 calendar days	10 calendar days
Unlikely, Unrelated	10 calendar days	10 calendar days

Externally Sponsored Studies

Required Reporting Timeframe to DSMC		
Attribution	**UNEXPECTED[1]**	**EXPECTED**
	Death while on active treatment or within 30 days of last day of treatment	
Possibly, Probably, Definitely	No DSMC reporting required - IRB reporting may be necessary	
Unlikely, Unrelated		
	Death after 30 days of last active treatment/therapy	
Possibly, Probably, Definitely	No DSMC reporting required - IRB reporting may be necessary	
Unlikely, Unrelated		
	Grades 3 and 4 AND meeting the definition of "serious"	
Possibly, Probably, Definitely	No DSMC reporting required - IRB reporting may be necessary	
Unlikely, Unrelated		
	Grades 1 and 2	
Possibly, Probably, Definitely	No DSMC reporting required - IRB reporting may be necessary	

An event determined by the IRB of record to be an Unanticipated Problem (UP) will be communicated to the Investigator and COH DSMC through the COH IRB Operations Director. The DSMC will review the case and make a determination as to whether the study will be suspended, terminated, amended, or allowed to continue without amendment.

Required Reporting Timeframe to IRB of Record		
Attribution	**UNEXPECTED**	**EXPECTED**
	Death	
Possibly, Probably, Definitely	5 calendar days	Annual
Unlikely, Unrelated	Annual	Annual
	Grades 3 and 4 AND meeting the definition of a UP	
Possibly, Probably, Definitely	5 calendar days	Annual
Unlikely, Unrelated	Annual	Annual
	Grade 1 and 2 AND meeting the definition of a UP	
Possibly, Probably, Definitely	5 calendar days	Annual
Unlikely, Unrelated	Annual	Annual

ADDITIONAL REPORTING REQUIREMENTS

SAEs meeting the requirements for expedited reporting to the FDA, as defined in 21 CFR 312.32, will be reported as an IND safety report using the MedWatch Form FDA 3500A for Mandatory Reporting which can found at: http://www.fda.gov/Safety/MedWatch/HowToReport/DownloadForms/default.htm

The PI or designee will be responsible for contacting the Office of IND Development and Regulatory Affairs (OIDRA) at COH to ensure prompt reporting of safety reports to the FDA. OIDRA will assist the PI with the preparation of the report and submit the report to the FDA in accordance with the following:

- any unexpected fatal or life threatening adverse experience associated with use of the drug must be reported to the FDA no later than 7 calendar days after initial receipt of the information [21 CFR 312.32(c)(2)];

- any adverse experience associated with use of the drug that is both serious and unexpected must be submitted no later than 15 calendar days after initial receipt of the information [21 CFR 312.32(c)(1)]

- any follow-up information to a study report shall be reported as soon as the relevant information becomes available. [21 CFR 312.32(d)(3)]

19.0 Protocol Deviations

Protocol deviations using the Protocol Deviation Form in IRIS must be submitted by the protocol nurse or the clinical research associate. The clinical research associate at COH will also submit copies to the Protocol Management Team and the COH Data and Safety Management Board.

19.1 In accordance with the COH Policy on Clinical Research Protocol Deviation, there will be a “no deviations” rule for this protocol. However, for subject safety or unforeseen scheduling problems, planned deviations from this protocol will be permitted with approval from the IRB in the form of a Single Subject Amendment. In addition, the sponsor (the CDMRC) must also approve any planned deviations.

19.2 All unplanned deviations will be reported to the COH DSMB.

19.3 If there is a dispute among the persons involved in the provision of research treatment, in regard to whether a treatment deviates from the protocol, the facts of the case will be reported to the DSMB which will be serve as the arbiter of whether a deviation exists.

19.4 Study attrition: Participants who withdraw from study before completion of both imaging procedures will be replaced by another study participant.

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