MSK PROTOCOL COVER SHEET

A Phase II Trial of Homologous Recombination Repair Status as a Biomarker of Response in Locally Recurrent/Metastatic Triple Negative Breast Cancer Patients Treated with Concurrent Cisplatin and Radiation Therapy

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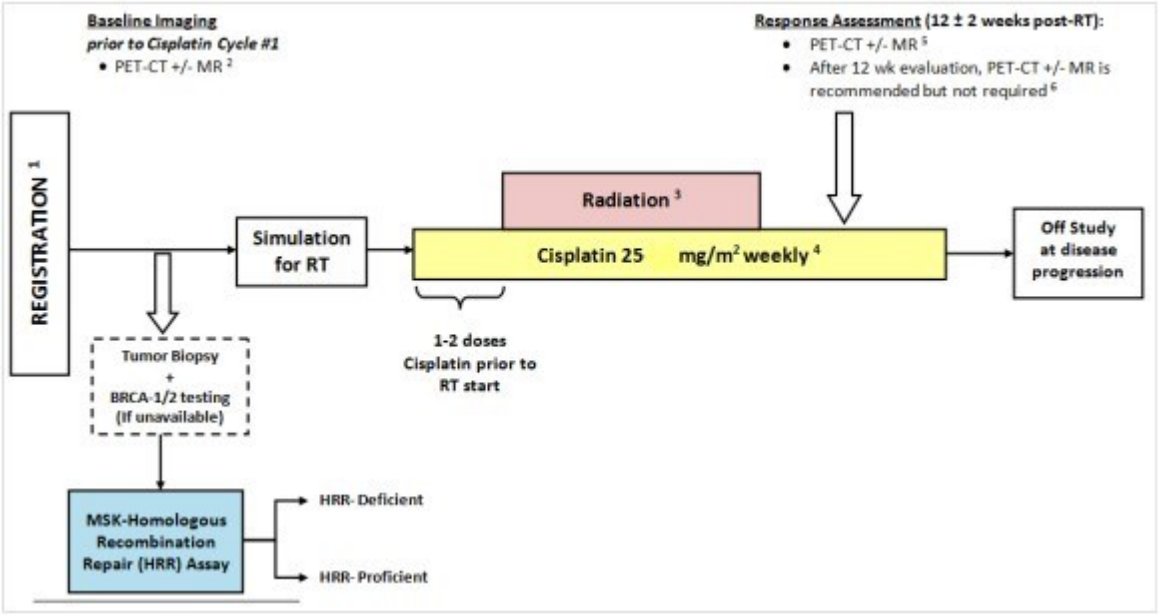
1.0 PROTOCOL SUMMARY AND SCHEMA

Triple negative breast cancer patients with metastatic or unresectable, locally advanced disease who require radiation therapy to a measurable tumor site are eligible. Prior to treatment, all patients will receive a biopsy of the tumor to be irradiated. Utilizing the Memorial Sloan Kettering Homologous Recombination Repair (MSK-HRR) Assay, patients will be identified as either HR-deficient or HR-proficient. Knowledge of the HR repair status prior to treatment is not necessary, since all patients in the study will receive the same treatment, regardless of the knowledge of the tumor- concurrent cisplatin and radiation (RT) to the tumor.

The primary purpose of the trial is to assess whether or not HR repair status is a sensitive biomarker of response, as measured by complete response (CR) rate in patients receiving cisplatin and radiation. Cisplatin will be given once a week(+ 3 days) for 1-2 weeks, followed by concurrent delivery with RT to the tumor. Following the completion of RT, pending no toxicities, but prior to the week 12 post RT evaluable timepoint, patients will continue to receive cisplatin or until POD, based on MD discretion.

Secondary objectives are to assess time to progression and overall response rate (CR+PR+SD) by HRR status. Exploratory objectives are to assess association between tumor infiltrating lymphocyte (TIL) and treatment response and to compare the effectiveness of RECIST 1.1 criteria with PET Response Criteria in metabolically active tumors.

At baseline, all patients will receive PET-CT. An MR will be obtained in addition only if the study radiologist determines that tumor is insufficiently evaluable with PET-CT. At 12 +2 weeks after completion of RT, patients will undergo the same imaging modality used at baseline assessment (PET-CT or MR) in order to assess treatment response. The target accrual is 54 patients, to be achieved within 3.5 years.



[1] Patients with biopsy-proven TNBC requiring locoregional control or palliation with radiation to a measurable tumor.
[2] Obtain study staging only if unavailable 28 days prior to study registration. All patients will receive baseline PET-CT performed at MSKCC. An MR will be added if PET-CT is deemed to be insufficient for tumor evaluation by the study radiologist at baseline evaluation.
[3] Radiation dose will be 250 cGy × 15 fx = 3750 cGy to the metastatic site delivered in 3 weeks for metastatic disease or 200 cGy x 25 fx=5000 cGy delivered in 5 weeks to the chest wall, breast and/or regional lymph nodes , plus an optional 10-14 Gy boost to areas of gross tumor, for non-metastatic/locallyrecurrent disease.
[4] The length of one Cisplatin cycle is 21 days.If cisplatin is withheld, the cycle count for treatment is also held.
[5] Cisplatin will be once a week (±3 days). The dose of cisplatin can be reduced in the event of toxicities defined in Section 4.2.1.
[6] Following the completion of RT, pending no toxicities, but prior to the week 12 post RT evaluable timepoint, patients will continue to receive cisplatin or until POD, based on MD discretion.
[7] At 12±2 weeks after RT, all patients will receive PET-CT for treatment response and/or staging. MR will also be obtained if the patient had an MR at baseline.
[8] PET-CT +/- MR will occur at 12±2 weeks after RT. PET/CT +/- MRI q6 months after RT is recommended but not required.

2.0 OBJECTIVES AND SCIENTIFIC AIMS

The primary objective of this trial is to evaluate the performance of the MSK-HR repair assay as a biomarker in predicting response to concurrent cisplatin and RT in breast cancer patients with measurable metastatic or unresectable, locally advanced disease. Radiographic complete response (CR) of the irradiated tumor will be counted as a positive response to treatment. Radiographic response less than a CR, progression of the irradiated tumor or appearance of a new lesion within the radiated field will be categorized as a failure to respond to treatment. All other sites of metastatic disease that is outside of the irradiated field will be monitored and counted toward disease progression.

Secondary objectives are to assess:

- Partial response, stable disease and progression of disease within the irradiated tumor as well as systemically (in sites distant to the irradiated tumor)
- Overall time to progression
- Toxicities of concurrent cisplatin and radiation

Exploratory objectives are to assess:

- Examine immune correlates to determine the impact of treatment on anti-tumor immunity and as a predictor of treatment response
- Determine the sensitivity of ctDNA detection in patients receiving chemoradiation for metastatic and locally recurrent TNBC
- Compare the efficacy of RECIST 1.1 vs. PET Response Criteria (PRC) as measurement tools for treatment response

3.1 BACKGROUND AND RATIONALE

Triple negative breast cancer (TNBC), defined as estrogen receptor and progesterone receptor levels <1% and HER2-negative (0-1+ by immunohistochemistry or HER2/CEP17 ratio <2 or HER2 signals/cell <4 by in-situ hybridization [1]), comprises 12-17% of breast cancers. TNBC is associated with rapid growth, early metastasis and unfavorable prognosis compared to other subtypes of breast cancer [2]. Since targeted treatment options such as endocrine therapy and HER2-directed therapies are unavailable, chemotherapy is the mainstay of treatment. Although chemotherapy and radiation therapy (RT) are common treatments for palliation and local control in metastatic or recurrent breast cancer, there are no effective biomarkers that can reliably predict the sensitivity of these tumors to these treatments.

TNBCs are characterized by a genomic profile that is similar to those of breast cancer patients who are BRCA1-mutation carriers. This profile is indicative of defects in the BRCA pathway, therefore resulting in a “BRCA-like” phenotype. Although rarely mutated in sporadic breast cancer (<5% of breast cancer patients harbor germline mutations in either gene), BRCA1 expression has been reported to be reduced in a small number of sporadic tumors, particularly in TNBC [3-5]. BRCA1 and BRCA2 are only two of many proteins involved in executing HRR [6]. A subset of sporadic breast cancers may also have defects in HR, in the absence of mutations in either BRCA1 or BRCA2 [7-9].

3.2 Rationale for Homologous Recombination (HR) Repair Testing In the Clinic

BRCA1 and BRCA2 are essential for error-free repair of double strand breaks (DSB) induced by ionizing radiation and certain DNA damaging chemotherapeutic agents, through their activity in the BRCA1-BRCA2-RAD51 DNA repair pathway of homologous recombination (HR). In homologous recombination (HR), both BRCA1 and BRCA2 are required for the recruitment of Rad51 to sites of DSB’s, with BRCA2 interacting directly with Rad51. Rad51 is a vital downstream protein required for homology search and strand invasion, using the sister chromatid as a template for error-free repair of the DSB. In the absence of HR, cells rely on more error-prone mechanisms, including non-homologous end-joining, in which pieces of DNA are pieced together in a quick but error-prone way, resulting in deletions of significant amounts of genetic material and chromosomal instability.

PARP is an important enzyme involved in base-excision repair which repairs endogenous or therapy-induced single strand breaks. Inhibition of that pathway by a PARP inhibitor results in the accumulation of large amounts of single strand breaks which can be converted to double strand

breaks at stalled replication forks. In HR-proficient cells, these DSB's are repaired by homology directed repair which is a Rad51-dependent process. In HR-deficient cells, however, such as cells with loss of BRCA1, BRCA2 or other proteins involved in the complex BRCA1-BRCA2 pathway, the repair machinery of the cell can be overwhelmed, leading to error-prone repair by alternative mechanisms such as non-homologous end joining, leading to chromosomal instability and cell death. Inhibition of homologous recombination or PARPs may be well tolerated in isolation, but combined inactivation of these distinct DNA-repair pathways, it can result in cell death through a process called "synthetic lethality."

In preclinical models, BRCA1 and BRCA2-deficient cells have been shown to be sensitive to PARP inhibitors. However, clinical studies utilizing PARP inhibitors in unselected sporadic TNBC have failed to show benefit of PARP inhibitors in improving progression-free survival [10]. These findings underscore the importance of assaying the function of the HRR pathway in order to better identify TNBC patients who would most likely to benefit from DNA repair-based therapies.

3.3 The Memorial Sloan Kettering HR Repair (HRR) Assay

There are two components of the MSK HRR assay. The first, the induction of RAD51 foci, is a functional assay that assesses HRR status in real-time, prior to the initiation of study treatment. The second, the CGH (comparative genomic hybridization) array, validates the RAD51 foci by detecting copy number changes in the genome that are characteristic of HRR status. Fresh tumor tissue is required for the RAD51 foci assay, whereas extracted DNA from the fresh tumor specimens is used for array-CGH studies. The assays will be conducted in the lab of Dr. Simon Powell.

The rationale for using both components of the assay is to detect HRR deficiency as determined by RAD51 foci assembly, and then to validate these results with the CGH read-out of the genomic architecture. We anticipate that validation with the array CGH will be particularly useful in identifying "conversion" patients in whom HRR-deficient status may have converted to HR-proficient status secondary to pre-treated with multiple systemic agents prior to study enrollment.

3.3.1 RAD51 Foci

DNA repair proteins accumulate into foci in the nuclei of cells following DNA damage including irradiation, and these foci can be detected by immunofluorescence and represent sites of ongoing repair. This method has proven to be a robust method of observing HR in cells [11]. Given its downstream location in the BRCA1-BRCA2 pathway of HR, the recruitment of Rad51 into foci correlates well with the ability to carry out HR and has been utilized to assay the integrity of the entire pathway. BRCA1 and BRCA2 deficient cells are unable to form RAD51 foci following exposure to ionizing radiation [12]. Powell et al. has previously reported the feasibility of quantifying ionizing radiation (IR) induced RAD51 and other DNA repair foci formation *ex vivo* in fresh pretreatment breast cancer specimens as a means of assessing the function of the BRCA1-BRCA2-RAD51 pathway[13].

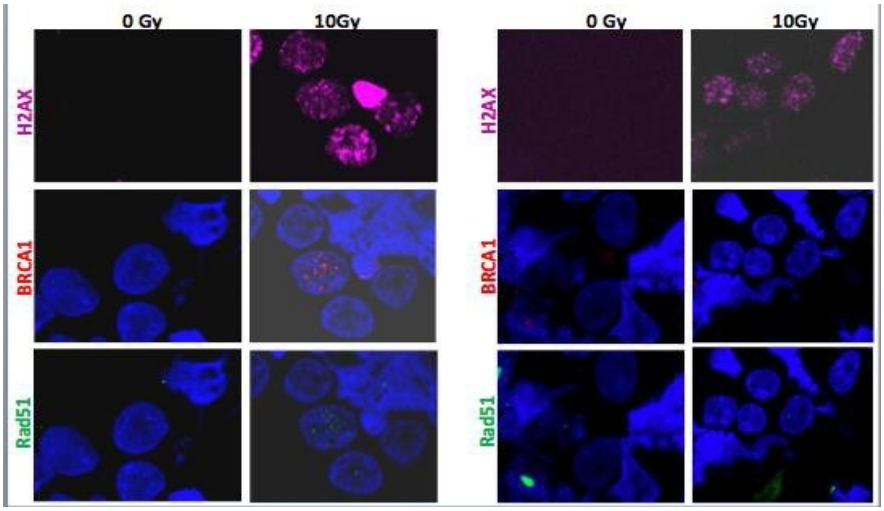


Figure 2: Immunofluorescent representation of the induction of nuclear foci in human tumor cells with and without RT: a) There is prominent phosphorylation of histone H2AX (at serine 139) following 10Gy. H2AX was used as an upstream event in the DNA damage response as a control for cell viability and adequacy of the preparation. In one of the cells shown, there is recruitment of both BRCA1 and RAD51 into foci following 10Gy. At least 100 cells were counted in each condition (no RT vs RT) and cells were considered positive if they have ≥5 foci. The nuclei of the MCF 7 cells are stained blue with DAPI, and on the right following 10Gy, >50% increase in rad51 foci is visible after RT, suggesting an intact BRCA1-BRCA2 pathway b) The nuclei of the MCF 7 cells are stained blue with DAPI, and on the right following 10Gy. There was normal gammaH2AX foci formation upstream, but a failure to recruit BRCA1 or Rad51 into foci following 10Gy. This tumor was therefore deemed to have a defective BRCA1-BRCA2 pathway.

3.3.2 Array-CGH

Array CGH uses microarray technology to assess for copy number changes (genomic gains, heterozygous losses, high-level amplifications, homozygous deletions). DNA from a test sample and normal reference sample are labeled differentially using different fluorophores, and hybridized to several thousand probes. The probes are derived from most of the known genes and non-coding regions of the genome and printed on a glass slide.

The ratio of the fluorescence intensity of the test to that of the reference DNA is then calculated, to measure the copy number changes for a particular location in the genome. Typical CGH genomic profiles from breast cancers in BRCA1 and BRCA2 mutation carriers are characterized by prominent large segment deletions and some gains likely due to aberrant processing of chromatid breaks during mitosis. In contrast, sporadic tumors tend to have one of two types of genome profiles, with either very few larger altered segments or a high number of small copy number alterations throughout the genome, with occasional high-level amplification events.

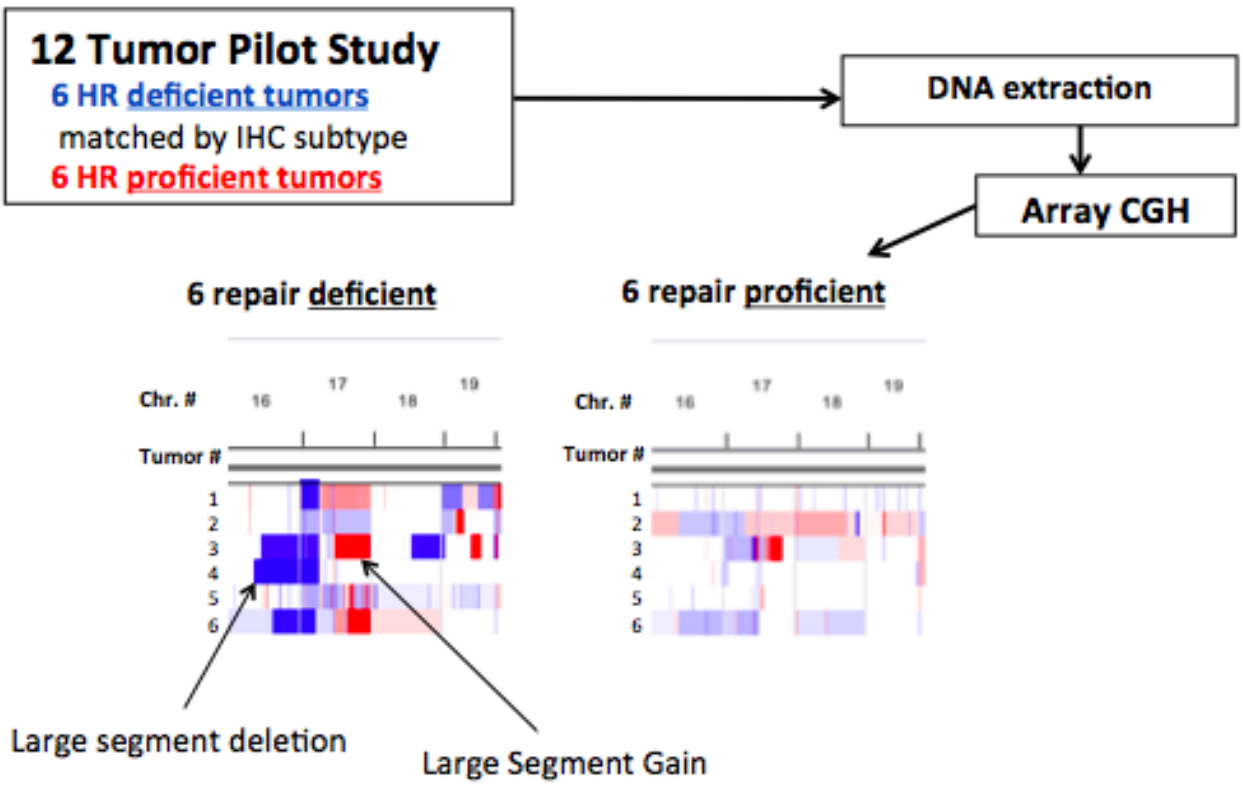


Figure 3: Analysis of genomic profiles by array-CGH of 12 tumors grouped by HRR status: On the left are the 6 HRR-deficient tumors found to have functional defects in the BRCA1-BRCA2 pathway by the RAD51 foci assay. On the right are 6 HRR-proficient tumors. Gains are shown in red and losses are shown in blue. The 6 HRR-deficient tumors characteristically show a greater number of large deletions and some gains, compared to the 6 HRR-proficient tumors, displaying a BRCA-like genomic landscape with significantly more deletions of large segments of genetic material likely due to aberrant processing of chromatid breaks.

3.3.3 Exome Resequencing

To evaluate for homologous recombination pathway alterations in genes other than BRCA1 and BRCA2, we will obtain whole exome resequencing data through the MSK Genomics Core Facility using genomic DNA obtained from core biopsy samples and matched against normal tissue (buffy coat of peripheral blood samples). Exome resequencing of breast cancer samples in our pilot study that were MSK-HRR deficient and BRCA1/2 wild type revealed that 65% had potentially deleterious alterations in other components of the HR pathway (unpublished). Quality control of extracted DNA, and sequencing library preparation will be performed by the MSK Genomics Core Facility. We will use a data-analysis pipeline wherein germline variants will be filtered out of sequencing analyses using matched blood samples. Thus, germline alterations will not become apparent to researchers.

3.3.4 Preliminary Data on the Frequency of HRR Defects by Breast Cancer Subtype

The frequency of BRCA1-BRCA2-RAD51 pathway defects in sporadic breast cancer has been quantified using the MSK HRR assay in 60 invasive breast cancer patients receiving lumpectomy or mastectomy in whom fresh tumor tissue specimens were collected at the time of surgery (IRB protocol #10-093). Overall, seventeen of 60 (28%) tumors displayed defective RAD51 recruitment

following ex-vivo irradiation. HRR-deficiency was seen in all subtypes of breast cancer but was enriched in TNBC: 7 of 33 (21%) ER positive, 4 of 14(29%) HER2 amplified, and 6 of 13 (48%) TNBCs. HRR deficiency was not associated with low BRCA1 mRNA or protein expression. As noted above, HRR-deficient sporadic breast cancers displayed genomic landscapes characterized by large segment gains and losses due to genomic instability from loss of HR [14].

3.3 Rationale for Concurrent Cisplatin and RT

Biologically, cisplatin is a known radiosensitizer, with both additive and synergistic potentiation of radiation. Platinum potentiates radiation-induced cell injury through a number of putative mechanisms. These agents act via formation of DNA cross links resulting in DNA double-strand breaks. When cisplatin is integrated into DNA adjacent to a radiation-induced single cell strand break, it can make the break more difficult to repair and thus lead to cell death [15,16]. In addition, cisplatin increases cellular platinum uptake [16], and enhance formation of toxic platinum intermediates [17]. The potentiation of radiation-induced cell injury appears under both oxic and hypoxic conditions [18].

Preclinical studies have shown that the combination of cisplatin and RT delivers the maximum ex-vitro stress response needed to stimulate HRR repair, compared to cisplatin or RT when delivered individually. Mouse embryo fibroblast cells with mutated DNA repair pathways were measured for responses after cisplatin, radiation and combination treatments. The combination treatment showed that inhibition of the HRR pathway resulted in super additive effects, compared to cisplatin or RT alone [19].

Concurrent cisplatin and radiation treatment is standard of care in the definitive treatment of advanced head and neck as well as cervical cancer [20-30]. A large meta-analysis has established the survival benefit of concurrent chemoradiation versus RT alone for head and neck cancer [27]. The efficacy and safety profile of concurrent cisplatin and RT in cervical cancer has also been well established in randomized trials [20-24]. The toxicities of concurrent cisplatin and RT are specific to the irradiated site but are generally tolerable. Adverse events among cervical cancer patients who received both cisplatin and RT included 12% grade 3 leukopenia, 1.1% thrombocytopenia, 5.6% other hematologic toxicities and 4.5% grade 3 gastrointestinal toxicities. Grade 2 toxicities included 14.7% leukopenia, 2.3% thrombocytopenia, 15.3% other hematologic toxicities and 15.9% gastrointestinal toxicities. No treatment-related deaths were reported in these trials.

Whereas q3 week dosing (100 mg/m^2) of cisplatin has been shown to result in significant toxicity in patients with laryngeal cancer when delivered concurrently with RT (40), lower weekly doses (30-50 mg/m^2) has been well tolerated [29,30]. Based on these data, we plan to utilize weekly cisplatin ($\pm$ 3 days) at 25 mg/m^2 prior to and after RT. During RT, the dose of cisplatin will be weekly ($\pm$ 3 days) 25 mg/m^2.

Following the completion of RT, pending no toxicities, but prior to the week 12 post RT evaluable timepoint, patients will either continue to receive cisplatin or until POD, based on MD discretion.

3.4 Evidence for Cisplatin and Biomarkers of Response in Triple Negative Breast Cancer

Currently, there is no standard chemotherapy for the treatment of metastatic or recurrent triple negative breast cancer. Generally, these patients receive a single agent chemotherapy, of which there are many options. The standard chemotherapy treatment for breast cancer patients with BRCA1 or BRCA2 mutations are the same as those treatments for sporadic breast cancer patients, which do not include platinum chemotherapy for non-metastatic disease. The "BRCA-ness" features of sporadic TNBC are suggestive of sensitivity to interstrand crosslinking agents such as platinum chemotherapy [31]. There is mounting preclinical as well as early clinical evidence suggesting benefits of platinum chemotherapy in patients with BRCA1 and BRCA2 mutations and TNBC.

Single-agent weekly cisplatin is not routinely used in breast cancer, but there is growing use of cisplatin in triple negative breast cancer because patients with BRCA mutations appear to be uniquely sensitive to cisplatin in non-randomized trials, and the broader array of TNBC patients with HRR defects (but no known BRCA mutations) are thought to be sensitive to cisplatin. Several clinical studies have evaluated the role of platinum chemotherapy among TNBC patients, including those with and without BRCA1 or BRCA2 mutations. A Dutch study demonstrated that patients with BRCA-like genomic profiles observed a greater benefit with high dose platinum chemotherapy vs conventional chemotherapy, compared to patients with non-BRCA-like tumors [32]. A proof-of-concept neoadjuvant study from Poland involving 12 breast cancer patients who were BRCA1 mutation carriers demonstrated a pCR of 83% following treatment with 4 cycles of single-agent cisplatin 75 mg/m^2 [33]. Two studies, both from the Dana-Farber Cancer Institute, evaluated neoadjuvant cisplatin at 75 mg/m^2 for four cycles. Silver et al reported a response rate of 21% in a study of 28 unselected TNBC [34], whereas the larger n=51 patient study by Ryan et al [35], in which patients also received bevacizumab every 3 weeks for three cycles, demonstrated a pCR rate of 15% among patients without BRCA mutations. Factors associated with good cisplatin response in the Silver et al. study included young age (p=0.001), low BRCA1:mRNA expression (p= 0.03), BRCA1 promoter methylation (p=0.04), p53 nonsense or frameshift mutations (p=0.01), and a gene expression signature of E2F3 activation (p=0.03), suggesting that biomarkers other than BRCA1 may be predictive for cisplatin response [34].

A study of preoperative cisplatin in early-stage breast cancer patients with germline BRCA1 mutations demonstrated high rates of pathologic complete response (72%) [36]. Others have shown high rates of pathologic complete response in mutation carriers, including a neoadjuvant trial of gemcitabine, carboplatin and iniparib. Among 17 enrolled women with germline BRCA1 or BRCA2 mutations, 15/17 (88%) achieved a pathologic complete response (pCRs) or had minimal residual disease at the time of surgery [35].

Recently published results from the GepardSixto (GBG 66) trial also suggest a beneficial role for platinum chemotherapy in TNBC. This trial included 595 patients with HER2+ and TNBC. All patients received paclitaxel and non-pegylated-liposomal doxorubicin. HER2+ patients also received trastuzumab and lapatinib, whereas TNBC patients also received bevacizumab. All patients were randomized 1:1 to receive concurrent carboplatin or not, stratified by subtype. Among the TNBC patients, pCR rates increased by 20%; 84 (53.2%) of 158 patients achieved a pCR with carboplatin,

compared with 58 (36.9%) of 157 without (p=0.005). This effect was not seen among the HER2+ patients, 45 (32.8%) of 137 of whom achieved a pCR with carboplatin compared with 50 (36.8%) of 136 without (p=0.58) [37]. The advantage seen with carboplatin was driven by patients with triple negative disease. The regimen was very toxic, with about 50% of patients receiving carboplatin having to discontinue treatment due to adverse events.

The preliminary results of CALGB/Alliance 40603 study (presented at the 2013 San Antonio Breast Cancer Symposium) also support these findings. In this study, 454 patients with stage II/III TNBC were randomized to standard neoadjuvant chemotherapy or chemotherapy plus either carboplatin, bevacizumab, or the combination of carboplatin/bevacizumab. Patients were randomly assigned in a 2×2 schema to receive weekly paclitaxel plus dose-dense anthracycline/cyclophosphamide +/- either the addition of bevacizumab or the addition of carboplatin. For patients receiving carboplatin, 60% achieved a pCR, compared to 46% of those who did not carboplatin - an increase of 76% (p=0.0018). Defined by no disease in the breast or axilla, pCR rates were 54% with carboplatin vs 41% without carboplatin, a 71% increase (p=0.0029). When the combination of carboplatin and bevacizumab were used in addition to chemotherapy, 67% of patients achieved a pCR, however, a significant treatment interaction between the two drugs was not shown. Bevacizumab resulted in a greater frequency of adverse effects, as did the combination of bevacizumab and carboplatin. Patients receiving carboplatin were more likely to experience neutropenia and thrombocytopenia.

Finally, the PrECOG 0105 study included 80 TNBC and BRCA1/2 mutated patients treated with neoadjuvant carboplatin, gemcitabine and iniparib [38]. Pathologic CR was achieved in 36% of the overall population, 47% in the BRCA1/2 mutated population and 56% in triple negative patients who also had BRCA 1/2 mutations. The toxicity profile was more favorable than that of the Gepar Sixto regimen, with myelosuppression (grade 3 or 4 neutropenia in 49%) and elevations in liver enzymes (grade 3 in 24%).

Whereas the aforementioned trials evaluated platinum chemotherapy in the neoadjuvant setting, the efficacy of platinum chemotherapy has also been studied in metastatic TNBC patients. TBCRC 009 was a multicenter single-arm phase II study of single-agent platinum in 86 patients with metastatic TNBC [39]. Patients received cisplatin 75mg/m^2 or carboplatin AUC=6 every 3 wks, by physician choice. The objective response rate (ORR) was 30.2% overall, including 4 CR (4.7%), 22 PR (25.6%). The ORR was 54.5% vs. 19.7% in BRCA1/2 carriers (n=11) vs. non-carriers (n=66); (p=0.02). Six long-term responders remain disease-free at median follow up of 4 yrs, 3 of whom stopped treatment after 6 cycles. Exploratory subgroup analysis of RR: Cisplatin 37%, Carboplatin 23%; 1st line therapy 31.7%, 2nd line therapy 20%. 33% of pts progressed by 6 weeks, but 33% remained on study for ≥ 6 mos. 51 treatment-related grade 3/4 toxicities occurred. 2 patients had grade 4 toxicities (neutropenia (1) and hypertension (1)), while the others were grade 3 (fatigue (8), neutropenia (6), anemia (5), hyponatremia (4)).

In a subsequent analysis presented at the ASCO 2014 Annual Meeting, predictors of response to platinum chemotherapy were assessed, including p63/p73 expression by RT-PCR as a predictor of response. Tumor-based exploratory studies included gene expression (GE) profiling, PIK3CA and p53 mutational status, and homologous recombination deficiency (HRD) assays correlating with

BRCA1/2 inactivation. All biomarkers *except* the HRD assays failed to predict responses to platinum chemotherapy. Types of HRD assays performed included Loss of Heterozygosity (LOH), Telomere Allelic Imbalance (TAI), and Large-scale State Transition (LST). Among the small group of 22 patients with available tissue, scores in all of the HRD assays were higher in BRCA1/2 carriers than non-carriers, and in responders than non-responders (HRD-LST responders vs. nonresponders, p=0.0016).

Birback et al. have also suggested that the number of subchromosomal regions with allelic imbalance extending to the telomere [N(tAI)] is predictive for pathologic response to preoperative cisplatin treatment in sporadic TNBC patients. They found an inverse relationship between BRCA1 expression and N(tAI), suggesting that N(tAI) could be a genomic measure of DNA repair pathway defects and thereby identify patients who could benefit from cisplatin therapy[40]

The results from a clinical trial performed at Washington University provide the most compelling preliminary evidence for this proposed study [41]. In this trial, neoadjuvant pre-operative cisplatin + RT were administered for locally advanced triple-negative breast cancer patients, all of whom received modified radical mastectomy as definitive surgical treatment. Response was analyzed by HRR status, utilizing the functional HRR assay developed by Powell et al. Among the 10 patients enrolled to date, 6/10 show pathologic CR, of whom 5/6 (83%) were HR-defective. Given the small number of patients accrued to this trial, further validation of the results are required, and is therefore the aim of this proposed protocol.

3.5 Immune Correlates

A large body of research using experimental animal models indicates that the immune system controls the development of cancer through immune surveillance mechanisms[42]. Histologic analyses of solid malignancies reveal that they are infiltrated by various cells of the innate and adaptive immune system [43]. Therapeutic modulation of the immune system for clinical benefit in cancer patients has been demonstrated by numerous modalities, including antibody blockade of inhibitory molecules, adoptive T cell transfer, and autologous cell-based vaccines [44-47]. A recent large-scale meta-analysis of the correlation of tumor infiltrating T cells with survival clearly establishes that a relative high density of T_H1 and cytotoxic memory T cells correlated with a favorable prognosis in the majority of cancers [48].

Tumor infiltrating lymphocytes (TILs) may be a useful histological predictor of benefit from chemotherapy in breast cancer patients, although it has not been established whether or not response to chemotherapy would be specific to a class of chemotherapy. Specific to breast cancer, the evaluation of TILs has been shown in the GeparSixto (GBG 66) trial to be relevant for response to chemotherapy in triple negative and Her2-positive early breast cancers [37,49]. In this analysis, a lymphocyte predominant pattern of infiltrate was predictive of a complete pathologic response to neoadjuvant chemotherapy. Similarly, the association of TILs and response to trastuzumab and chemotherapy (epirubicin/cyclophosphamide with docetaxel +/- capecitabine) was evaluated in 156 HER2+ patients from the neoadjuvant GeparQuattro trial [50]. TILs were associated with higher responses to the trastuzumab and chemotherapy combination, and tumor-mediated immunosuppression was evident in the lymphocytic infiltrate, with PD-1 and IDO1 significantly

predictive of trastuzumab benefit.

The prognostic value of TILs has also been validated in a combined analysis of two Phase III randomized adjuvant breast cancer clinical trials [37,51-54]. In these studies, the percentage of intratumoral and stromal lymphocytes in pre-treatment tumor biopsies was found to be an independent predictor of pCR in both the training and validation cohorts. Higher TIL pathologic scores were associated with a better prognosis: for every 10% increment in stromal TIL, a 14% reduction of risk for recurrence or death was observed (p=0.02), and for every 10% increment in intratumoral TIL, a 28% risk reduction was seen (p=0.06). Multivariate analysis confirmed stromal TIL to be an independent prognostic marker of disease-free survival.

Collectively, these data suggest that the natural course of breast cancer as well as the response to therapy is linked to the degree by which the tumor is recognized by the immune system. Response to radiotherapy may be also be predicted by the level of TIL, however data supporting this in breast cancer is scant. In squamous cell carcinoma of the head and neck as well as rectal cancer however, TILs can predict response to combination chemoradiotherapy [55,56]. The induction of an immune response by radiotherapy may also have important consequences. Several studies have shown the various mechanisms by which RT stimulates the immune system. One effect of localized tumor irradiation is the exposure of a large amount of tumor antigens in the form of necrotic and apoptotic tumor cells [57]. The contexts in which these antigens are presented to the immune system are also impacted by tumor irradiation. Tumor radiotherapy leads to an inflammatory tumor microenvironment by inducing the expression of several proinflammatory cytokines [5858]. In addition, upregulation of major histocompatibility molecules and costimulatory molecules can potentiate effector cytolytic T cell responses [59].

The pretreatment biopsy planned in this study will allow for the comprehensive assessment of TIL which will be correlated to response. This will provide important insight into the role of the tumor immunity in the response to chemoradiotherapy. In addition the assessment of homologous recombination can also factor into the endogenous response of the immune system to the tumor as well as response to therapy. The effect of homologous recombination on tumor immunity is not well characterized, however the induction of microsatellite instability as a pathway of carcinogenesis has clearly been linked to tumor immune response [60]. Tumor cells with deficient DNA mismatch repair develop microsatellite instability and the resulting tumors are densely infiltrated by TILs [61]. It is suggested that mismatch repair deficiency leads to numerous insertion/deletions that ultimately translate to the generation of tumor-specific neopeptides that can then be recognized by the immune system [62]. Radiation administered to a tumor with an underlying homologous recombination defect may lead to significant “epitope spreading” resulting in the induction of an immune response against newly recognized tumor associated antigens. This study will begin to address the immune consequences of tumor genomic instability.

3.6 Circulating Tumor DNA (ctDNA) Sensitivity

In a number of clinical settings, circulating tumor DNA(ctDNA) has emerged as a promising biomarker of disease burden that in some cases provides greater sensitivity than imaging. Furthermore, ctDNA in patients with very high disease burdens can also be used to track emergence of treatment resistant subclones[63,64]. However, ctDNA in the setting of cisplatin and radiation for recurrent/metastatic breast cancer remains entirely uninvestigated and the sensitivity of detection is unknown.

When feasible, we will collect 1 tube of blood (8-10mL) in EDTA or Streck tubes[65] for ctDNA analysis at the scheduled time periods of blood sample collection as outlined in Section 10.0. The plasma will be isolated through centrifugation (820xg x 10 minutes, then 16,000g x 10 minutes) and stored at -80C for analysis. Using known somatic mutations obtained through exome resequencing data of tumor samples, mutant alleles in ctDNA will be assessed through amplification of loci from extracted cell free DNA and massively parallel sequencing or detected through digital droplet polymerase chain reaction (ddPCR). We will use a data-analysis pipeline wherein incidentally sequenced germline variants will be filtered out of sequencing analyses using matched DNA from buffy coat DNA. Thus, germline alterations will not become apparent to researchers.

4.1 OVERVIEW OF STUDY DESIGN/INTERVENTION

4.2 Design

This is a single-arm Phase II trial testing the efficacy of concurrent cisplatin and radiation in breast cancer patients with measurable metastatic or locally recurrent tumor, based on HRR status as determined by the MSK-HRR assay. Prior to study enrollment, all patients will require assessment by the radiation oncologist and medical oncologist. Patients will receive a baseline PET-CT +/- MR for evaluation of the tumor to be irradiated and to assess extent of disease systemically. If unavailable within the time specifications of the study, the PET-CT +/- MR will be performed after study registration. Biopsy of the tumor to be irradiated will be performed. Fresh tissue specimens will be sent to the laboratory of Dr. Simon Powell for performance of the MSK-HRR Assay. Results from the functional assay will be utilized towards the primary endpoint. Additional unutilized tissue will be sent to the Immune Monitoring Facility Laboratories for TIL analyses to be conducted by Dr. George Plitas. Bloodwork will be sent to the lab of Dr. Daniel Higginson for ctDNA analyses. After the biopsy, patients will undergo simulation for palliative radiation to the tumor site. Following simulation, patient will initiate a 25 mg/m^2 infusion of cisplatin once a week $\pm$ 3 days. The starting dose will depend on baseline laboratory results within 28 days prior to registration. Following 1-2 weeks of cisplatin, RT will be initiated. Weekly cisplatin ($\pm$ 3 days) will be continued and delivered concurrently with RT. After the completion of RT, weekly cisplatin ($\pm$ 3 days) will be continued until disease progression or until assessment of treatment response at Week 12 post RT (+/- 2 weeks), based on MD discretion. Patients whose HRR status is found to be inevaluable by the MSK assay will not be included in the study and will be replaced by eligible patients.

At Week 12 (12$\pm$2 weeks after RT; with strong recommendation at 12 weeks), repeat imaging with the same imaging methods used for baseline assessment will be performed. Treatment response will be

determined by either PRC or RECIST 1.1.

We estimate accrual will occur at the rate of 15 patients per year. It will take 3.5 years to accrue the total of 54 patients. Patients will be followed up to 24 weeks after completion of RT to determine all study endpoints. Clinical follow-up will continue until death.

4.3 Intervention

4.3.1 Cisplatin Administration

Cisplatin treatment should be initiated following study registration. The starting dose of cisplatin is established as 25 mg/m^2 ivpb once a week (+ 3 days) for 1-2 weeks prior to radiation. Dose modification following the first dose of cisplatin is subject to the discretion of the treating physician. Dosing calculation guidelines will follow MSKCC policies described on the website: http://mskweb5.mskcc.org/intranet/shared/pharmacy/guidelines/Chemo_Guidelines/Manuals/Cisplatin.html

Cisplatin may be initiated immediately following simulation but prior to first radiation fraction. The rationale is so that the patient may initiate a form of treatment while waiting 1-2 weeks for radiation treatment planning to be completed.

Cisplatin may cause myelosuppression or other known non-hematologic toxicities. Following the completion of RT, pending no toxicities, but prior to the week 12 post RT evaluable timepoint, patients will continue to receive cisplatin based on MD discretion, or until POD.

The starting (first) dose of cisplatin will be 25 mg/m^2 ivpb once a week (+ 3 days). Following the first dose of cisplatin, a CBC will be drawn once a week (+ 3 days) prior to treatment to ensure treatment parameters are met. A comprehensive panel and Mg++ level will be obtained on D15 (+3 days) of every cycle of cisplatin; these values will inform the dose for the following cycle of cisplatin. The length of one cycle of Cisplatin is 21 days. If cisplatin is withheld, the cycle count for treatment is also held. Based on the laboratory values drawn from the prior week's bloodwork, if the following parameters are met, cisplatin should be held as directed.

- If ANC is <1,000 mcL and/or platelets are <100,000 mcL, withhold cisplatin therapy for one week and repeat measurements. Physicians have the option of administering growth factor support during the time when ANC<1,000 mcL or platelets are <100,000 mcL. The treating MD may modify the subsequent cisplatin doses at their discretion. If there is a delay of 3 consecutive weeks of treatment due to toxicity, then the patient must discontinue protocol therapy and be removed from the study.

- If the ANC is still <1,000 mcL or platelets are 75,000-99,000 mcL, then hold weekly cisplatin again until the ANC is >1,000 mcL and platelets are >100,000 mcL. The treating MD may modify the subsequent cisplatin doses at their discretion.

- If serum creatinine is >1.5 mg/dL, delay treatment, provide intravenous hydration and check 12 or 24 hour creatinine clearance, serum creatinine and electrolytes after 24 hours. If the serum

creatinine is ≤1.5 mg/dL and creatinine clearance ≥60 cc/min, may give full dose of cisplatin. If serum creatinine is 1.6-1.9 and creatinine clearance is between 30-60 cc/min, cisplatin can be given at the discretion of the treating physician. If serum creatinine is ≥2 OR creatinine clearance is <30 cc/min, hold cisplatin.

- If the patient develops any non-hematologic grade 3-4 toxicity, cisplatin will be held until toxicity is reduced to grade 2 or less. Dose reduction is permitted at the discretion of the treating physician. If a patient experiences grade 2 toxicity (with exception of alopecia) that lasts >2 weeks, cisplatin will be held and dose reduction recommended at discretion of the treating physician. Cisplatin will be discontinued if greater than grade 2 toxicity persists following 2 dose reductions.

- If the patient develops an intercurrent illness (e.g. infection) that, in the opinion of the treating medical oncologist, mandates a delay of cisplatin therapy, the intercurrent illness must be resolved within a time frame such that no more than 2 weekly doses of cisplatin are withheld. If therapy must be withheld for a longer period of time, the patient will be removed from the study.
- Following the completion of RT but prior to the week 12 post RT evaluable timepoint, patients will continue receiving cisplatin based on MD discretion. The dose and schedule of cisplatin after the completion of RT will be at the discretion of the treating MD. If there is a delay of 3 consecutive weeks of treatment, then the patient must discontinue protocol therapy and will be removed from study. Continuation of cisplatin and the dose and schedule of cisplatin after Week 12 post RT assessment will be at the discretion of the treating MD.
- Note: Disruption to the cisplatin treatment schedule for 3 weeks due to unforeseen circumstances, holidays or the patient being on vacation is permitted and will be reviewed on a case-by-case basis.

4.3.2 Radiation Therapy

Radiation therapy to the target tumor will be delivered with external beam radiation therapy. In metastatic cases where the intent is palliative, the dose of radiation will be 3750 cGy delivered in 15 daily fractions, 4-5 days a week. In locoregionally recurrent or progressive cases where the intent is to enhance locoregional control, the dose of radiation will be 5000 cGy delivered in 25 fractions, 4-5 days a week plus an optional 10-14 Gy boost to areas of gross tumor. In cases where patients have had prior radiation and there is overlap between current and prior fields, normal tissue constraints will be followed as per departmental guidelines.

Radiation therapy will not be delivered on holidays for which the Department of Radiation Oncology is closed. When radiation treatment is missed because of a holiday or unforeseen circumstances, the missed dose will be made up at the end of RT.

5.1 THERAPEUTIC/DIAGNOSTIC AGENTS

5.2 Cisplatin Availability, Preparation and Storage

Commercially available cisplatin injection (1 mg/ml): Each ml of sterile, unpreserved solution contains 1mg of cisplatin with 9mg of sodium chloride and 1mg of mannitol in water for injection. Hydrochloric acid is added to adjust the pH. Single-dose glass vials of 10 and 50 ml contain 10 and 50 mg of cisplatin, respectively. None of the injection components such as IV needles, syringes and sets should have aluminum components, because aluminum is incompatible with cisplatin. A black precipitate forms (platinum) when exposed to aluminum and loss of potency will occur. Vials of cisplatin injection, USP, are stored at room temperature between 15-25 degrees Celsius. Please refer to the FDA-approved package insert for complete product information.

5.3 Tumor Biopsy

4 cores biopsies are required, with 2 optional cores, will be obtained from the tumor to be irradiated, with image-guidance as necessary. The safety and feasibility of the biopsy will be determined by the study Interventional Radiologist. Clip placement in target lesion is recommended if CT or MR-guided biopsy is performed. Skin metastases will be biopsied by an MSKCC surgeon, dermatologist or a radiation oncologist. The minimum core length is 6 mm. One out of the 4 cores will be fixed in 10% neutral buffered formalin and sent to Pathology for routine diagnostic confirmation of triple negative breast cancer, prior to irradiation. The remainder of the cores will be placed in a 1 mL freezer vial in DMEM standard medium at room temperature with regular ice on top of the specimens and transported directly to Dr. Powell's laboratory for the MSK-HRR assay. If there is any remaining tissue left after the MSK-HRR assay, DNA will be extracted and stored in Dr. Powell's lab for possible future analysis. The estimated turn-around time for functional assay, from receipt of tissue to interpretation, is 5 business days. However, the results of the assay (determining HR-proficient vs HR-deficient) status are not necessary for patients in order to receive the study treatment and will only be shared with the study statistician and primary investigator, for the purposes of confirming which patients can remain on study (i.e. they must have a definitive HRR repair status). Patients who are unable to obtain a definitive HRR status will be excluded from analysis of the primary endpoint of the study, but will be counted towards the secondary and exploratory objectives of the study.

5.4 MSK-HRR Assay

The specimens will be transported to the Laboratory of Molecular Radiation Biology in the Zuckerman Research Building, Room 419, led by Dr. Simon Powell and managed by Rob Delsite. The time from tumor harvest to initiation of laboratory processing should ideally not exceed 2 hours. Tumor specimens will be divided into 2 separate and equally sized parts; one part will be irradiated to 10 Gy (gamma-rays from the Shepard Mark-I ^{137}Cs irradiator, Model 68, SN643), in the SKI Radiation Core Facility and the second part will be sham-treated. Following irradiation or sham treatment, the specimens will be incubated in humidified cell culture incubators at 37°C with 5% CO2 for 4 hours. One part of each specimen will then be snap-frozen in optimal cutting temperature (OCT) compound (Sigma-Aldrich), with a view to preparing histological sections in the Molecular Cytology Core Facility

(MCCF). Tumor will be snap frozen in optimal cutting temperature compound and stored at -80°C. RNA was extracted using standard Trizol extraction. The DNeasy kit (Quiagen, Valencia, CA) was used to isolate genomic DNA. The quality of the DNA was verified on a 1% agarose gel. RNA quality was assessed using the Agilent Bioanalyzer 2100.

Hematoxylin and eosin staining of representative sections will also be performed by the MCCF. The second part will be disaggregated by scalpel mincing, with or without collagenase, to develop a cell suspension which can be used in a cytospin followed by formaldehyde fixation and cytologic analysis. Thin sections or cytospin preparations will be stained using the published immunofluorescence protocols. Immunofluorescence microscopy will be carried out in the Laboratory of Molecular Radiation Biology on a confocal microscope. Briefly, cryoslides or cytospin slides will be fixed and permeabilized, then blocked and probed with anti-RAD51, anti-BRCA1, and anti-FANCD2 antibodies. Slides will then be washed and incubated with fluorescently labeled secondary antibodies. Images will be captured and subnuclear foci will then be quantified.

Cell nuclei will be analyzed for subnuclear foci formation of phosphohistone H2AX (γH2AX), BRCA1, and RAD51 in both the irradiated and non-irradiated states using a Zeiss LSM 510 confocal microscope. In order to quantify foci formation, over 200 nuclei will be counted for both the irradiated and non-irradiated conditions. A nucleus will be scored as positive if it contains greater than 5 foci. The formation of γH2AX foci is an upstream event in the DNA damage response and indicates the presence of a double strand break [66]. γH2AX staining will be used as a control for cell viability at the time of fixation and adequacy of the staining procedures. In addition, the mean number of positive BRCA1 and RAD51 nuclei for each condition will be quantified as a function of the percent of nuclei with γH2AX foci following IR in order to normalize for differences in cell viability across various tumor specimens.

Comparative genomic hybridization (CGH) will be performed at the Genomics Core Laboratory at Memorial Sloan-Kettering Cancer Center by cy5 labeling tumor DNA to 1M aCGH human microarrays from Agilent (Santa Clara, CA) with cy3 labeled genomic DNA from Roche (Indianapolis, IN). The arrays will be scanned with Agilent high-resolution microarray scanner (G2538C). Data will be extracted with Agilent feature extraction software (v10.7). Data analysis will be performed using the RAE algorithm [67].

Any remaining tissue from the tumor biopsies will be flash frozen in liquid nitrogen and stored at -80°C and banked in Dr. Powell's lab for future analyses, including BRCA promotor methylation status.

If BRCA status is not known at the time of study registration, patients will either be referred to the MSKCC Genetics department for evaluation of BRCA status, or this information may be obtained from analysis of the fresh tumor biopsy in Dr. Powell's laboratory.

5.5 Immune Correlates

In order to evaluate the association between intratumoral and stromal lymphocytes and response to treatment [65-67], the pre-treatment tumor biopsy slides (formalin fixed paraffin embedded, FFPE) will be reviewed by the two study pathologists, using the standardized methodology for evaluating TILs based on H&E sections of core biopsies [68]. Intratumoral lymphocytes are defined as intraepithelial

mononuclear cells within tumor cell nests or in direct contact with tumor cells and will be reported as the percentage of the tumor epithelial nests that contain infiltrating lymphocytes [65]. Stromal lymphocytes are defined as the percentage of tumor stroma area that contains a lymphocytic infiltrate without direct contact to tumor cells. In addition, immunohistochemical analyses including but not limited to CD3, CD4, CD8, FOXP3 and CD20 will be performed in order to further characterize TILs.

Peripheral blood will be drawn before treatment and at two time points following completion of the cisplatin/RT to evaluate epitope spreading by seromic analysis for tumor-associated antigens. In addition, the peripheral blood mononuclear cells will also be preserved for evaluation of T cell immunophenotyping. Immunophenotyping will be determined using flow cytometry to determine the effect on individual lymphocyte subsets in peripheral blood mononuclear cells (PBMC). Flow cytometric assessment of the lymphocyte subsets $CD4^+$, $CD8^+$, $CD16^+$, $CD19^+$ and CD56+ will be examined as percent and absolute number. $FOXP3^+$ and $FOXP3^-$ lymphocytes with the following markers will also include but not limited to: $CD4^+$, CD25, CD45RA, CD45RO, LAG3, GARP, PD1, and CTLA4. Activation markers such as CD44 and CD69 will also be examined. The blood samples will be delivered to the Immune Monitoring Core Facility where the processing and storage will occur. The IMF may also perform ELISPOT, flow tetramer staining, and/or intracellular polyfunctional cytokine staining, TCR Vβ Repertoire analysis and Spectratyping to further characterize tumor-antigen specific $CD4^+$ and $CD8^+$ T cell response before and after treatment. PBMCs will be archived for all patients to support complete analysis of the profile. Archived serum may also be assayed for levels of chemokines, cytokines, and tumor-associated soluble proteins by techniques that may include but are not limited to, ELISA or multiplex assays. Analyses may include markers of inflammation, immune activation, host tumor growth factors, and tumor-derived proteins. These assays will also take place in the Immune Monitoring Facility (IMF).

6.1 CRITERIA FOR SUBJECT ELIGIBILITY

6.2 Subject Inclusion Criteria

- Histologically-confirmed invasive triple negative breast cancer (ER <1%, PR <1%, her-2-neu 0-1+ by IHC or FISH-negative) or as determined by MD discretion
- Radiation to the recurrent or metastatic site is clinically indicated and would be considered standard care for palliation or for locoregional control
- Age ≥18 years
- Tumor to be irradiated is measurable by RECIST 1.1 or PRC (see Appendix 1 and Section 12.0)
- Willingness to undergo tumor biopsy prior to initiation of treatment
- Life expectancy greater than 6 months
- ECOG performance status 0-2
- Any prior chemotherapy is allowed including prior treatment with platinum-containing chemotherapy
- Prior treatment with FDA-approved or investigational biologics or novel molecularly target therapies, including oral or IV formulations, are permitted. Patients must be off prior targeted therapy for at least 14 days prior to study biopsy.

- Use of an effective means of contraception in women of child-bearing potential
- Ability to comprehend and sign informed consent
- Adequate organ and marrow function within 14 days prior to study entry, defined as:
  - Absolute neutrophil count (ANC)>1000/mm^3
  - Hemoglobin >9 gm/dl
  - Platelets >100,000/mm^3
  - Serum creatinine <1.5 mg/dl OR creatinine clearance of $\geq$50 cc/min
  - SGOT/SGPT<2.5X institutional ULN (<5X ULN if known liver metastases)

6.3 Subject Exclusion Criteria

- Unmeasurable target tumor site by RECIST 1.1 or PRC (ex: lesions <2 cm on CT or MR scan, leptomeningeal disease, ascites, pleural/pericardial effusion, lymphangitis, non-FDG-avid skin lesions)
- Brain metastases requiring focal or whole brain radiation will be excluded, as these lesions cannot be biopsied and can have life expectancies <6 months.
- Inability to obtain a biopsy of the tumor as deemed by the study Interventional Radiologist
- Prior chemotherapy completed <7 days prior to planned study entry
- Prior RT is allowed and must have been completed more than 7 days before planned study entry.
  Note: For re-irradiation cases, standard departmental guidelines should be followed so as to not exceed normal tissue
- Life expectancy less than 6 months
- Intercurrent illness or other major medical condition or comorbid condition that might affect study participation (uncontrolled renal, pulmonary or hepatic dysfunction or infection)
- Renal dysfunction for which cisplatin dose would be considered unsafe.
- Women on study must be neither pregnant nor nursing nor expected to become pregnant during therapy. For premenopausal women, negative pregnancy test within 14 days of RT is required.
- Concurrent active malignancy other than non-melanomatous skin cancer or carcinoma in-situ of the cervix, unless treatment for the previous cancer was completed >2 years prior to study entry and patient has remained disease-free.

7.0 RECRUITMENT PLAN

Potential research subjects will be identified by either a breast medical oncologist, breast surgeon or breast radiation oncologist at MSKCC. Eligible patients include women with breast cancer with measurable disease requiring radiation treatment. These include locoregionally progressive or recurrent disease in the breast/chest wall and/or regional lymph nodes, or sites of metastatic disease requiring local palliative treatment. The patients will be referred to the Department of Radiation Oncology, where informed consent will be obtained. This study is open to recruitment all Regional sites, however biopsies of the target tumor for the study must be performed at the Main Campus.

MSKCC has filed forms HHS 441, 641 and 639-A. We have taken notice of NIH/ADAMHA policies concerning the inclusion of women and minorities in clinical research populations. We expect that the study population will be fully representative of the range of patients seen at MSKCC without exclusion to age (≥18 years) or ethnic background.

Potential research subjects will be identified by a member of the patient's treatment team, the protocol investigator, or research team at Memorial Sloan-Kettering Cancer Center (MSKCC). If the investigator is a member of the treatment team, s/he will screen their patient's medical records for suitable research study participants and discuss the study and their potential for enrolling in the research study. Potential subjects contacted by their treating physician will be referred to the investigator/research staff of the study.

The principal investigator may also screen the medical records of patients with whom they do not have a treatment relationship for the limited purpose of identifying patients who would be eligible to enroll in the study and to record appropriate contact information in order to approach these patients regarding the possibility of enrolling in the study.

During the initial conversation between the investigator/research staff and the patient, the patient may be asked to provide certain health information that is necessary to the recruitment and enrollment process. The investigator/research staff may also review portions of their medical records at MSKCC in order to further assess eligibility. They will use the information provided by the patient and/or medical record to confirm that the patient is eligible and to contact the patient regarding study enrollment. If the patient turns out to be ineligible for the research study, the research staff will destroy all information collected on the patient during the initial conversation and medical records review, except for any information that must be maintained for screening log purposes.

In most cases, the initial contact with the prospective subject will be conducted either by the treatment team, investigator or the research staff working in consultation with the treatment team. The recruitment process outlined presents no more than minimal risk to the privacy of the patients who are screened and minimal PHI will be maintained as part of a screening log. For these reasons, we seek a (partial) limited waiver of authorization for the purposes of (1) reviewing medical records to identify potential research subjects and obtain information relevant to the enrollment process; (2) conversing with patients regarding possible enrollment; (3) handling of PHI contained within those records and provided by the potential subjects; and (4) maintaining information in a screening log of patients approached (if applicable).

8.1 PRETREATMENT EVALUATION

- Within 28 days prior to study registration:

- Evaluation by Radiation Oncology
- Evaluation by Medical Oncology
- PET-CT +/- MR performed at MSKCC
- Complete Blood Count
- Comprehensive Metabolic Panel with Magnesium
- Serum Pregnancy Test for Women of Child Bearing Potential
- Biopsy of target tumor

- After study registration but before cisplatin weekly dose #1:
  - PET-CT +/- MR (only if imaging was not done prior to study registration or not done at MSKCC)
  - Biopsy of target tumor (if not completed prior to registration)
  - Peripheral blood sample for immunologic correlate studies, ctDNA, and/or BRCA mutation status (if unknown prior to study registration).

9.0 TREATMENT/ INTERVENTION PLAN

During radiation therapy, patients will undergo a physical exam and evaluation by a radiation oncologist once a week for toxicities in a status check visit as per current standard practice.

While receiving cisplatin, patients will be followed by a medical oncologist, who will obtain complete blood count once a week and serum chemistries with Mg^{2+} on D15 ($\pm$3 days) of every cycle of cisplatin therapy. The medical oncologist will evaluate the patient on D1 of every cycle of cisplatin therapy. The length of one cycle of Cisplatin is 21 days.

Following the completion of RT, pending no toxicities, but prior to the week 12 post RT evaluable timepoint, patients will continue to receive cisplatin or until POD, based on MD discretion.

At Week 12 (12$\pm$2 weeks after completion of radiation therapy), patients will be seen either by a radiation oncologist or a medical oncologist, and then every 6 months for routine follow-up visits either by a radiation oncologist or medical oncologist, until removal from study or death. Peripheral bloods and blood banking research samples (ctDNA) be collected at baseline, at the end of RT and at the 12 $\pm$2 week assessment point. Follow-up imaging studies (PET-CT +/- MRI) will be obtained at 12$\pm$2 weeks after RT. PET/CT +/- MRI of the target lesion q6 months afterwards will be recommended but not required.

10.0 EVALUATION DURING TREATMENT/INTERVENTION

The Table below illustrates the interventions and treatments required at specific time intervals for the study. All evaluations can be performed as frequently as clinically indicated.

Study Requirement	Within 28 days prior to Study Registration	After Study Registration/Before Cisplatin Dose #1	During RT	D1 of Cisplatin Cycle	D8 of Cisplatin Cycle	D15 of Cisplatin Cycle	12±2 weeks after RT	q6 months after RT
Evaluation by Radiation Oncology	X[11]		X[10]				X[4,11]	X[3,4,11]
Evaluation by Medical Oncology[11]	X			X[8]			X[4]	X[3,4]
PET-CT +/- MRI	X	X[1]					X	X[3]
Biopsy of Target Tumor	X	X[12]						
CBC[5]	X		X	X	X	X	X	X
Mg++, CMP[5]	X					X		
Pregnancy Test	X							
Peripheral blood sample for immunologic correlate studies, ctDNA, and/or BRCA mutation status		X[2]	X[9]				X	
Cisplatin Administration[7]			X[6]	X	X	X		
Toxicity Assessment			X[13]	X[13]				

[1] Only if imaging was not done prior to study registration or not performed at MSK
[2] Obtain only if BRCA 1 or 2 mutation status is unknown at baseline. BRCA 1 or 2 mutation status may also be obtained from the tissue biopsy at baseline. Bloodwork for immune correlates and ctDNA will be obtained at baseline.
[3] Recommended but not required.
[4] May be performed by either radiation oncologist or medical oncologist, but both visits are not required.
[5] Except at baseline, CBCs will be checked once a week (± 3 days) during cisplatin but prior to administration. Mg2+ and serum chemistries will be obtained on D15 (±3 days) of every cisplatin cycle.

[6]Radiation will begin following 1-2 weeks of cisplatin. Weekly cisplatin (+ 3 days) will be continued and delivered concurrently with RT.

[7] The length of one Cisplatin cycle is 21 days. Cisplatin is administered on D1, D8 and D15 (+3 days) of every cycle. Following the completion of RT, pending no toxicities, but prior to the week 12 post RT evaluable timepoint, patients will continue to receive cisplatin or until POD, based on MD discretion.
[8] The medical oncologist will evaluate the patient on D1 of each cisplatin cycle. The patient will be assessed for toxicities and will be recorded on a ClinDoc Toxicity form, complete with grade, attribution, start, and stop dates.
[9] Bloodwork will be obtained during the last week of RT for immune correlates and ctDNA.
[10]The radiation oncologist will evaluate the patient weekly during RT at status checks. The patient will be assessed for toxicities and will be recorded on a standard radiation oncology toxicity form, complete with grade, attribution, start, and stop dates.
[11]Evaluations by both Radiation and Medical oncologists will include: medical history (baseline only), physical exam, ECOG/KPS, review of prior and concurrent medications and vital signs (blood pressure, height, weight, temperature, respiratory rate and heart rate).
[12]If not completed prior to registration
[13] Required weekly during radiation therapy in either the clinic of medical oncology or radiation oncology and then on D1 of each cycle after the completion of radiation therapy.

11.1 TOXICITIES/SIDE EFFECTS

A severe toxicity will be defined as any grade 3 toxicity, attributed to radiation or chemotherapy, which can be observed within 2 months of completion of protocol radiation. Toxicities will be assessed using the CTEP Version 4.0 of the NCI Common Terminology Criteria for Adverse Events (CTCAE version 4.0) at the website: http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm. Patients will be assessed for toxicities until progression or death. Only adverse events that are possibly, probably or definitely related to protocol therapy will be documented on the CTCAE toxicities form and will be collected for data entry. Only hematologic toxicities that are considered Grade ≥2, per CTCAE version 4.0 will be captured for this trial.

11.2 Toxicities of Cisplatin

Nephrotoxicity

A major toxicity of cisplatin is cumulative nephrotoxicity. Tubular necrosis of both proximal and distal renal tubules has been noted in 28-36% of subjects treated with a single dose of 50 mg/m^2. It is first noted 14 days after a dose and is manifested by elevations in BUN and creatinine, serum uric acid and/or a decrease in creatinine clearance. Renal toxicity becomes more prolonged and severe with repeated courses of the drug. Renal function must return to normal before another dose of cisplatin can be given. Nephrotoxicity can be reduced by IV hydration and mannitol diuresis.

Nausea and Vomiting

Cisplatin causes moderate to severe nausea and vomiting in almost all subjects treated. Nausea and vomiting usually begin within 1 to 4 hours after treatment and lasts up to 24 hours. Various degrees of vomiting, nausea and/or anorexia may persist up to 1 week after the treatment. Delayed nausea and vomiting (beginning 24 hours or more after chemotherapy) has occurred with complete emetic control on the day of cisplatin therapy. The use of prophylactic and continuing antiemetic medication reduces these adverse effects. Subjects should receive prophylactic anti-nausea medications according to ASCO guidelines for highly emetogenic chemotherapy regimens.

Hypomagnesemia
Hypomagnesemia has been reported in subjects treated with cisplatin and is probably related to renal tubular damage. It may become sever enough to cause tetany. Generally, serum electrolytes return to normal levels when cisplatin is discontinued and supplemental electrolytes are administered.

Ototoxicity
Ototoxicity has been observed in up to 31% of subjects treated with a single dose of cisplatin (50 mg/m2) and is manifested by tinnitus and/or hearing loss in the high frequency range (4000-8000 Hz). Decreased ability to hear normal conversational tones may occur occasionally. Ototoxicity can be more severe in children than in adults and more frequent and severe with repeated administration. Hearing loss can be unilateral or bilateral and is usually not reversible.

Myelosuppression
Myelosuppression occurs in 25-30% of subjects treated with cisplatin. The nadirs in circulating platelets and leukocytes occur between days 18 and 23, with most subjects recovering by day 39. Leukopenia and thrombocytopenia are more pronounced at higher doses (>50 mg/m^2) than will be administered in this study. Anemia (a decrease in hemoglobin of 2gm.100 ml) occurs at approximately the same frequency and with the same timing as leukopenia and thrombocytopenia.

Neurotoxicity
Cisplatin neurotoxicity is characterized by peripheral neuropathies, which are sensory in nature but can also include motor difficulties such as reduced deep-tendon reflexes and leg weakness. The symptoms usually occur after prolonged therapy (4-7 months). Cisplatin therapy should be discontinued when serious neuropathy develops (defined in Section 4.2.1). The neuropathy, however, may progress further even after discontinuation of treatment.

11.3 Dose Delays and Modifications for Cisplatin

See section 4.2.1 for guidelines for dose modification of cisplatin during RT or after RT.

Following the completion of RT, pending no toxicities, but prior to the week 12 post RT evaluable timepoint, patients will continue to receive cisplatin or until POD, based on MD discretion.

11.4 Toxicities of Radiation

Toxicities may occur from radiation treatment. The type and risk of toxicity will depend on the presence of normal tissue structures in close proximity to the target. These normal tissue doses will be constrained by the treatment plan to deliver doses no more than those recommended in our departmental guidelines:
(http://teamshare/dept/medphys/ebtp/Public%20Documents/Forms/Public%20View.aspx)

The estimated risk of each type of toxicity is noted below, and include:

1. Skin (erythema, dry or moist desquamation, patchy ulceration) - 40-50%
2. Non-debilitating fatigue- 50%

3. Brachial plexopathy- 5-7%
4. Bowel (bowel perforation, obstruction or hemorrhage) - 2-5%
5. Peripheral nerve (neuropathy or severe neuropathic pain unresponsive to medications)-2-5%
6. Bladder (hemorrhagic cystitis, chronic cystitis manifesting in frequency and urgency) - 2-5%
7. Rectum (injury including ulceration or perforation)-2-5%
8. Osteonecrosis or bone fracture- 5-10%

12.0 CRITERIA FOR THERAPEUTIC RESPONSE/OUTCOME ASSESSMENT

Response to radiation and chemotherapy treatments will be using the Reviewed Response Evaluation Criteria in Solid Tumors (RECIST) guidelines version 1.1 . However there are limitations in using anatomic response criteria alone, particularly when evaluating bone metastases without an extra-osseous component, which is common in breast cancer patients. “PERCIST” (PET Response Criteria in Solid Tumors), is a commonly proposed method for measuring treatment response based on metabolic response criteria with PET [68]. A limitation of PERCIST is that it only takes into account the single hottest lesion and it relies upon SUV peak measurements, which are not easily reproducible in lesions with low FDG-avidity, as are often found in breast cancer. Since there are no standardized methods for quantifying FDG-tracer avidity in individual tumor lesions, we will use a modification of PERCIST criteria called “PET Response Criteria” (PRC) that is currently being utilized in other cancer protocols at MSK requiring metabolic assessment. In brief, the SUVmax of the tumor to be irradiated will be measured and recorded. On subsequent studies, the same target lesion will be measured and recorded. Lesions where SUVmax is no greater than background will be recorded as zero.

Response or progression will be graded as follows:

CR: All lesions no greater than background SUVmax

PR: SUVmax decreased by ≥30% greater than background

SD: Lesions that do not fit criteria for response or progression

PD: SUM SUVmax increased by ≥30% or development of a new lesion within the radiated field

Radiological review will be conducted by the study radiologists. PET-CT will be obtained in all patients at baseline. In general, PRC will be preferred over RECIST 1.1 for response assessment, except in cases where the tumor target is not clearly defined on PET-CT (e.g. lesions obscured by areas of physiologic FDG-avidity). In such cases, the study radiologists will decide when baseline images are available whether or not to use PRC or RECIST 1.1. If PET-CT evaluation is determined to be insufficient for tumor evaluation, MR will be recommended. The same imaging study (PET-CT or MR) or response criteria (PRC or RECIST 1.1) used at baseline assessment will be utilized for the follow-up assessments of the irradiated tumor.

We anticipate that the most common discrepancy between PRC and RECIST 1.1 will be when there is a CR on PRC, but PR on RECIST 1.1. In these cases, PRC will be utilized for response

assessment. If PRC will be used, RECIST 1.1 is not required for the primary endpoint. However, for the secondary endpoints of comparing PRC to RECIST 1.1, RECIST 1.1 measurements will be done on the CT portion of the PET/CT.

13.0 CRITERIA FOR REMOVAL FROM STUDY

Participants will be removed from the protocol for any of the following reasons:

- Development of disease progression that warrants a change in the radiation or chemotherapy treatment plan or an alternative therapy.
- Any grade 4 non-hematologic toxicity, except in cases where the treating physician believes the patient is deriving benefit from remaining on the study.
- Chemotherapy treatment delay of 3 consecutive weeks for any reason
  Note: Disruption to the cisplatin treatment schedule for 3 weeks due to unforeseen circumstances, holidays or the patient being on vacation is permitted and will be reviewed on a case-by-case basis.
- For safety reasons, the investigator considers it to be in the best interest of the participant that they be withdrawn
- The patient wishes to withdraw from the study
- HRR status is unable to be obtained from the core biopsy taken at the beginning of the study.

14.0 BIOSTATISTICS

The proposed study is a prospective Phase II trial testing the efficacy of concurrent cisplatin and radiation in TNBC patients with measurable metastatic or locally recurrent tumor, based on HRR status. All patients (regardless of HRR status) who meet the eligibility criteria and agree to be part of the trial will be enrolled. That is, knowledge of HRR status is not necessary at the start of the study, as all patients will receive the study therapy of concurrent cisplatin and radiation. The primary objective of this trial is to evaluate whether patients with HRR-deficiency have higher *complete response* (CR) rates than patients who are HRR-proficient. For the primary endpoint, response will be measured at 12+/- 2 weeks after the completion of RT.

Preliminary data from our institution has demonstrated the incidence of HRR deficiency in non-metastatic TNBC patients to be 48% [13]. However, because these studies were performed in non-metastatic breast cancer patients and cannot be fully extrapolated to metastatic patients, we conservatively estimated that the incidence of HRR deficiency in our proposed study population of patients with locally recurrent or metastatic TNBC to be 40%. Based on preliminary data from Washington University in which HRR-deficient patients had a greater than 4-fold pathologic CR rate compared to HRR-proficient patients [39], we conservatively assumed that HRR-deficient patients would achieve approximately 2.5x higher CR rate than HRR-proficient patients. We estimated that the CR rate of in HRR-proficient patients following concurrent cisplatin and RT to be 30%, and that the CR rate in HRR-deficient patients would be increased to 65%. This corresponds to an absolute difference in CR rates of 35%. These estimates were based on response rates after platinum

chemotherapy observed in metastatic TNBC patients in the TBCRC009 study [39] and adjusted for the additional cytotoxic effect of radiation therapy on the target tumor. The below table provides power calculations for varying proportions of HRR deficiency (30% to 40%), assuming a one-sided type I error of 5% and for a sample size of **54**.

Proportions of estimated HRR-deficiency	Power for absolute CR difference of 35%, n=54 Power Calculations
40%	85%
35%	83%
30%	80%

However, we will plan to accrue a total of **62** patients in order to allow for early dropout due to disease progression/death prior to response evaluation, or inability to obtain HRR status from the core biopsy. For the primary analysis, all patients for whom HRR status can be ascertained will be included in the denominator. Patients who progress or die prior to the response evaluation at 12 +/- 2 weeks post-RT will be included as failures. This includes patients with disease progression outside of the irradiated tumor. Patients for whom HRR status cannot be ascertained or who left the study for nondisease related reasons will be replaced.

A secondary analysis of interest is to evaluate the CR rate among the subset of patients who are able to complete the planned study treatment and have a scan at the 12 +/- 2 week post-RT time point. Radiographic complete response (CR) of the irradiated tumor will be counted as a positive response to treatment. Radiographic response less than a CR, progression of the irradiated tumor or appearance of a new lesion within the radiated field will be categorized as a failure to respond to treatment. All other sites of metastatic disease that is outside of the irradiated field wil be monitored and counted toward disease progression.

Based on prior experience with the HRR assay, we estimate that approximately 10% of patients will either 1) not have a definitive HRR status, or 2) will not complete the planned study treatment or 3) not have a response assessment at 12-14 week time point. To ensure we have 80% power for both the primary and secondary analyses we have added 8 patients to the sample size for a total of 62.The accrual number has been adjusted to 54 patients.

We expect to accrue 1 to 2 patients per month, or approximately 18 patients per year. Thus, accrual for the trial will be completed in 3.5 years. Based on previous data on the tolerability of cisplatin and radiation extrapolated from head and neck and gynecologic malignancies, we do not anticipate toxicities associated with the study treatment and as such, we have not incorporated an early stopping rule.

Secondary endpoints include overall response rates (CR+PR+stable disease), time to progression and CTCAE v4.0 toxicities. These endpoints will be compared by HRR status using the log-rank test for time-to-event endpoints and Fisher's exact test for binary endpoints.

Exploratory objectives are to evaluate the association of tumor-infiltrating lymphocytes (TIL), other immune correlates within the tumor biopsy specimen and circulating tumor DNA as stated in section 3.6 and 5.4 as a predictor of treatment response. This will be done using a Wilcoxon rank-sum test. In a second exploratory objective, in patients who have both RECIST 1.1 and PRC assessments, agreement will be calculated and if there is a sufficient sample size, a McNemar's test will be done.

15.1 RESEARCH PARTICIPANT REGISTRATION AND RANDOMIZATION PROCEDURES

15.2 Research Participant Registration

Confirm eligibility as defined in the section entitled Inclusion/Exclusion Criteria. Obtain informed consent, by following procedures defined in section entitled Informed Consent Procedures. During the registration process registering individuals will be required to complete a protocol specific Eligibility Checklist. The individual signing the Eligibility Checklist is confirming whether or not the participant is eligible to enroll in the study. Study staff are responsible for ensuring that all institutional requirements necessary to enroll a participant to the study have been completed. See related Clinical Research Policy and Procedure #401 (Protocol Participant Registration)

15.3 Randomization

There is no randomization.

16.1 DATA MANAGEMENT ISSUES

A Clinical Research Coordinator (CRC) from the Department of Radiation Oncology (Breast Service) will be assigned to the study. The responsibilities of the CRC include project compliance, data collection, abstraction and entry, data reporting, regulatory monitoring, problem resolution and prioritization, and coordinating the activities of the protocol study team.

The data collected for this study will be entered into a secure database (Clinical Research Database – CRDB). Source documentation will be available to support the computerized patient record.

16.2 Quality Assurance

Regular registration reports will be generated to monitor patient accruals and completeness of registration data. Routine data quality reports will be generated to assess missing data and inconsistencies. Accrual rates and extent and accuracy of evaluations and follow-up will be monitored periodically throughout the study period and potential problems will be brought to the attention of the study team for discussion and action. Random-sample data quality and protocol compliance audits may be conducted by the study team, at a minimum of once per year, more frequently if indicated.

16.3 Data and Safety Monitoring

The Data and Safety Monitoring (DSM) Plans at MSKCC were approved by the NCI in September 2001. The plans address the new policies set forth by the NCI in the document entitled "Policy of the

National Cancer Institute for Data and Safety Monitoring of Clinical Trials", which can be found at http://cancertrials.nci.nih.gov/researchers/dsm/index.html. The DSM Plans at MSKCC were established and are monitored by the Office of Clinical Research. The MSKCC DSM Plans can be found on the MSKCC Intranet athttps://one.mskcc.org/sites/pub/clinresearch/Documents/MSKCC%20Data%20and%20Safety%20Monitoring%20Plans.pdf . There are several different mechanisms by which clinical trials are monitored for data, safety, and quality. There are institutional processes in place for quality assurance (e.g. protocol monitoring, compliance and data verification audits, therapeutic response, and staff education on clinical research QA) and departmental procedures for quality control, plus there are two institutional committees that are responsible for monitoring the activities of our clinical trials programs. The committees: Data and Safety Monitoring Committee (DSMC) for Phase I and II clinical trials, and the Data and Safety Monitoring Board (DSMB) for Phase III clinical trials, report to the Center's Research Council and Institutional Review Board. During the protocol development and review process, each protocol will be assessed for its level of risk and degree of monitoring required. Every type of protocol (e.g., NIH sponsored, in-house sponsored, industrial sponsored, NCI cooperative group, etc.) will be addressed and the monitoring procedures will be established at the time of protocol activation.

17.1 PROTECTION OF HUMAN SUBJECTS

Describe in detail the information that would be relevant to an IRB reviewer as to how the study will protect the rights of human subjects. This should include, but not limited to the following: risks, benefits, toxicities/side effects, alternatives/options for treatment, financial costs/burdens. Privacy and confidentiality, adverse event reporting, voluntary nature of the study, and more.

17.2 Privacy

- MSK's Privacy Office may allow the use and disclosure of protected health information pursuant to a completed and signed Research Authorization form. The use and disclosure of protected health information will be limited to the individuals described in the Research Authorization form, including members of research teams at participating sites such as Alice Ho, MD. The following will be shared:
    - Patient identifiers including full name, date of birth, and date of assessments
    - Data collected during protocol visits and assessments
- .A Research Authorization form must be completed by the Principal Investigator and approved by the IRB and Privacy Board (IRB/PB).

- The consent indicates that individualized de identified information collected for the purposes of this study may be shared with other qualified researchers. Only researchers who have received approval from MSK will be allowed to access this information which will not include protected health information, such as the participant's name, except for dates. It is also stated in the Research Authorization that their research data may be shared with other qualified researchers.

17.3 Serious Adverse Event (SAE) Reporting

An adverse event is considered serious if it results in ANY of the following outcomes:

- Death
- A life-threatening adverse event
- An adverse event that results in inpatient hospitalization or prolongation of existing hospitalization
- A persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
- A congenital anomaly/birth defect
- Important Medical Events (IME) that may not result in death, be life threatening, or require hospitalization may be considered serious when, based upon medical judgment, they may jeopardize the patient or participant and may require medical or surgical intervention to prevent one of the outcomes listed in this definition

Note: Hospital admission for a planned procedure/disease treatment is not considered an SAE.

SAE reporting is required as soon as the participant starts investigational treatment/intervention. SAE reporting is required for 30-days after the participant's last investigational treatment/intervention. Any event that occur after the 30-day period that is unexpected and at least possibly related to protocol treatment must be reported. Please note: Any SAE that occurs prior to the start of investigational treatment/intervention and is related to a screening test or procedure (i.e., a screening biopsy) must be reported.

Please note: Any SAE that occurs prior to the start of investigational treatment/intervention and is related to a screening test or procedure (i.e., a screening biopsy) must be reported.

All SAEs must be submitted in PIMS. If an SAE requires submission to the HRPP office per IRB SOP RR-408 'Reporting of Serious Adverse Events', the SAE report must be submitted within 5 calendar days of the event. All other SAEs must be submitted within 30 calendar days of the event.

The report should contain the following information:

- The date the adverse event occurred
- The adverse event
- The grade of the event
- Relationship of the adverse event to the treatment(s)
- If the AE was expected
- Detailed text that includes the following
    - o An explanation of how the AE was handled
    - o A description of the participant's condition
    - o Indication if the participant remains on the study

- If an amendment will need to be made to the protocol and/or consent form
- If the SAE is an Unanticipated Problem

17.2.1

Any additional SAE reporting information required by the sponsor or drug supplier should be included in this section.

17.2.2 Abnormal Laboratory Values

An abnormal laboratory value is considered to be an AE if the abnormality:

- Results in discontinuation from the study
- Requires treatment, modification/interruption of the chemotherapy dose or any other therapeutic intervention
- Or is judged to be of significant clinical importance. Clinically insignificant symptoms or findings will not be collected in the assessment note, or be graded or attributed on the toxicities forms.
- Considered Grade $\geq$2, per CTCAE version 4.0

18.0 INFORMED CONSENT PROCEDURES

Before protocol-specified procedures are carried out, consenting professionals will explain full details of the protocol and study procedures as well as the risks involved to participants prior to their inclusion in the study. Participants will also be informed that they are free to withdraw from the study at any time. All participants must sign an IRB/PB-approved consent form indicating their consent to participate. This consent form meets the requirements of the Code of Federal Regulations and the Institutional Review Board/Privacy Board of this Center. The consent form will include the following:

1. The nature and objectives, potential risks and benefits of the intended study.
2. The length of study and the likely follow-up required.
3. Alternatives to the proposed study. (This will include available standard and investigational therapies. In addition, patients will be offered an option of supportive care for therapeutic studies.)
4. The name of the investigator(s) responsible for the protocol.
5. The right of the participant to accept or refuse study interventions/interactions and to withdraw from participation at any time.

Before any protocol-specific procedures can be carried out, the consenting professional will fully explain the aspects of patient privacy concerning research specific information. In addition to signing the IRB Informed Consent, all patients must agree to the Research Authorization component of the informed consent form.

Each participant and consenting professional will sign the consent form. The participant must receive a copy of the signed informed consent form.

19.0 REFERENCES

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