For Protocol Amendment 8 to: **NRG-DT001**, A Phase IB Trial Of Neoadjuvant AMG 232 (KRT-232) Concurrent With Preoperative Radiotherapy In Wild-Type P53 Soft Tissue Sarcoma (STS)

NCI/Local Protocol #: NRG-DT001

NCI Protocol Version Date: December 01, 2021

Section	**Change**
Global	The protocol version date was updated in the document footer. Formatting and typographical errors were corrected as needed.
Cover pages	This amendment was added to the Document History table.
5.1.1 9.1	• In response to a CTEP Notice to Investigators for studies using AMG 232 (KRT 232), sections 5.1.1 and 9.1 were updated to include changes in the drug supply and pharmaceutical information. • The step 2 dosing time for AMG 232 (KRT) was revised to *"30 minutes to 1 hour before RT"* for clarification in section 5.1.1.
10.5 Appendix IX	• References to the "*ETCTN Biorepository*" were changed to "*EET Biobank*" in 10.5 and Appendix IX. • An email contact was added in Appendix IX.

NRG ONCOLOGY

NRG-DT001
(ClinicalTrials.gov NCT # 03217266***)***

A PHASE IB TRIAL OF NEOADJUVANT AMG 232 (KRT-232) CONCURRENT WITH PREOPERATIVE RADIOTHERAPY IN WILD-TYPE P53 SOFT TISSUE SARCOMA (STS)

This trial is sponsored by the National Cancer Institute (NCI) and will be led by NRG Oncology. (9May2018)

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Agent	Supply	NSC #	IND #	IND Sponsor
AMG 232 (KRT-232)	NCI/PMB	789723	[redacted]	DCTD

Participating Sites (9May2018)

☒ U.S.
☐ Canada
☐ Approved International Member Sites

Document History

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Amendment 8	**December 01, 2021**
Amendment 7	February 22, 2021
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SCHEMA (16NOV2017)

Cohort A: Extremity/body wall
Cohort B: Abdomen/pelvis/retroperitoneum

STEP 1

Biopsy submission for TP53 NGS sequencing

AMG 232 (KRT-232) administration (on week -1 only*) (see escalated dose level)

NOTE: Tumor tissue must be received and NGS sequencing results must be completed before STEP 2 registration can occur.

*Patients can continue on AMG 232 (KRT-232) week 1 dose if there is a delay in NGS sequencing results.

↓

STEP 2

p53 wild-type (data collection for MTD/RP2D)

Continue AMG 232 (KRT-232) week 1 to 5 at assigned dose level,

and RT (Week 1-5), followed by surgery (5-8 weeks after RT completion)

p53 deleted/mutant

Change to standard of care (RT alone) (Week 1-5) followed by surgery (5-8 weeks after RT completion)

Patients enrolled in Cohorts A and B will be given independently escalated AMG 232 (KRT-232) dose regimen as detailed in table below.

AMG 232 (KRT-232) Dose Escalation/De-escalation Table for MTD/RP2D Determination*

Dose Level	Dose Frequency	Dose (mg)
-1	2x/week	120 mg
1**	3x/week	120 mg
2	4x/week	120 mg
3	5x/week	120 mg

* For detailed information on the process of dose escalation/de-escalation to determine MTD/RP2D as well as the DLT definition, please refer to section 14.
** Dose level 1 is the starting dose level.

1. OBJECTIVES

1.1 Primary Objectives

1.1.1 To evaluate the safety and tolerability of AMG 232 (KRT-232) in combination with standard-dose radiotherapy in STS in two separate patient cohorts (A, extremity or body wall; B, abdomen/pelvis/retroperitoneum);

1.1.2 To determine the maximum tolerated dose/recommended phase II dose (MTD/RP2D) of AMG 232 (KRT-232) in combination with radiotherapy.

1.2 Secondary Objectives

1.2.1 To observe and record anti-tumor activity. Although the clinical benefit of AMG 232 (KRT-232) in combination with radiotherapy has not yet been established, the intent of offering this treatment is to provide a possible therapeutic benefit, and thus the patient will be carefully monitored for tumor response and symptom relief in addition to safety and tolerability.

1.2.2 To determine % necrosis and pathologic complete response (pCR) in final surgical resection specimen;

1.2.3 To determine % local failure (LF), disease free survival (DFS) and overall survival (OS) at 2 years;

1.2.4 To determine pharmacodynamics (PD) effects of AMG 232 (KRT-232) when combined with radiotherapy by assessing serial serum Macrophage Inhibitory Cytokine (MIC)-1 levels;

1.2.5 To determine AMG 232 (KRT-232) exposure (pharmacokinetics)-response relationships (PD, toxicity, and efficacy).

1.3 Exploratory Objectives:

1.3.1 To determine tumor volume changes determined by MRI or CT with and without contrast pre- and post-radiotherapy;

1.3.2 To characterize clinical outcomes in patients treated with AMG 232 (KRT-232) by genomic biomarkers;

1.3.3 To determine the correlation between mdm2/4 expression determined by NGS and the protein levels by IHC;

1.3.4 To explore the possibility of identifying tumor genetic mutations in (1) cf ctDNA, (2) DNA/RNA isolated from exosomes, and determine the concordance of these results and that from NGS

2. BACKGROUND

2.1 Soft Tissue Sarcoma (STS)

Soft tissue sarcomas (STSs) are uncommon, with an estimated 12,300 new cases and 5000 deaths each year in the U.S. (Siegel 2016). Of these, 45-50% are located in the extremities, and 15% in the retroperitoneum. Seventy percent of retroperitoneal sarcomas present in abdomen, and 30% occur in the pelvis (Liles 2009). A majority of STS present as large masses, many greater than 5 cm in diameter before they are diagnosed since they could remain asymptomatic for a long time. This is especially true for STS located in the retroperitoneum, abdomen or pelvis. The large tumor size and complex anatomy of the tumor location represent a therapeutic challenge in the management of these tumors as negative surgical margins are difficult to achieve in many patients. With the possible

exception of low-grade, well-differentiated liposarcoma, most studies of retroperitoneal sarcoma have shown the importance of complete macroscopic resection to improve local control and disease-specific survival (Lewis 1998; Stoeckle 2001; Jaques 1990; Hassan 2004; Gronchi 2009; Bonvalot 2009). Recently, Keung (2014) reported median PFS, LRFS, DRFS, and OS were 21.1, 21.5, 45.8, and 59.0 months, respectively, and were significantly worse with R2 resection in 119 patients with retroperitoneal dedifferentiated liposarcoma. Since most patients with retroperitoneal sarcoma die of local recurrence, improving local control may translate into improved overall survival and patient quality of life, since surgical removal of recurrent tumors can be morbid and challenging. In addition, even though STS located in the extremities and body wall have better local control rate, distant failure is still substantial, especially in high grade tumors. Rate of distant metastasis was reported to be in a range of 15% to 37% at 5 years (Lewis 1998; Gronchi 2009; Bonvalot 2009). For example, Bonvalot reported 1-, 3-, and 5-year metastasis rates were 14% (95% CI, 0.11 to 0.19), 29% (95% CI, 0.24 to 0.34), and 34% (95% CI, 0.29 to 0.40), respectively, after curative treatment in his large institutional series (286 patients with retroperitoneal sarcoma). Thus, improving cure rate in STS represents an unmet clinical need, and better therapies conferring better local and distant control are needed.

2.2 p53-MDM2 regulation in STS, radiotherapy, and MDM2 inhibitors (9May2018)

Tumor suppressor gene p53 is one of the most commonly mutated genes in all human cancers; in tumors with WT p53, its activity is principally regulated by its high rate of turnover and degradation mediated by the E3 ubiquitin ligase, MDM2 (mouse double minute 2). MDM2 catalyzes p53 ubiquitination and subsequent degradation by proteasome. Activation of p53 in response to genotoxic stress leads to induction or repression of p53-responsive genes at the transcriptional level, resulting in cell cycle arrest, senescence, or apoptosis. (Wasylishen 2016; Karni-Schmidt 2016) In tumors with WT p53, high levels of MDM2 suppress p53 activity resulting in worse treatment outcome. Conversely, inhibition of MDM2 is an excellent strategy to treat WT p53 tumors. Since majority of STS contain WT p53, STS is an excellent target disease for MDM2 inhibition. (Tabubert 1998). More recent analyses showed that TP53 is altered (deleted or mutated) in 13.5 % soft tissue sarcoma cases (207 cases from MSKCC) (Barrentina 2010) and 44.76% soft tissue sarcomas cases from TCGA project (248 case). (Cancer Genome Atlas Research Network 2017)

MDM2 overexpression is also a good biomarker predictive for response to this class of inhibitors. An analysis of 4000 human tumors samples have previously found that MDM2 is overexpressed in 7% of cancers and its overexpression is mutually exclusive from p53 mutations. It has long been observed that 90% of well-differentiated (WD) and de-differentiated (DD) LPS contain supernumerary rings and/or giant rod chromosomes that contain amplified segments of the 12q13-15 region. Of the genes residing in this amplified chromosomal region, MDM2 (murine double minute) and CDK4 (cyclin-dependent kinase 4) amplifications are used in pathology to help differentiate WD and DD LPS from other STS subtypes and lipoma. (Pollock 1997; Bartel 2001). Many current MDM2 inhibitors, including AMG 232 (KRT-232), function by inhibiting MDM2-p53 protein-protein interactions in order to re-activate p53 function. MDM2/MDM4 accumulation has also been found to be the cause of developing resistance to this class of MDM2 inhibitors, causing tumor regrowth after drug withdrawal (Hoffman-Luca 2015; Aziz 2011). This

accumulation leading to resistance can be abrogated by the use of DNA damage agents, including ionizing radiation. Since RT is a part of standard therapy for STS, combining RT with MDM-2 inhibition to improve efficacy is a rational strategy.

Multiple competitive inhibitors (e.g. nutlin, MI-219, RG7112. MK8242, AMG 232 (KRT-232), RG7338) of the MDM2-p53 interaction have been developed to counter the effect of MDM2 and restore the activity of cellular p53. These MDM2 antagonists seem to function only in the presence of tumors with wild-type p53. Pre-clinical studies that evaluated these inhibitors in the treatment of osteosarcoma and LPS cell lines overexpressing MDM2 demonstrated that these MDM2 antagonists were able to reactivate p53 activity, induce G2 cell cycle arrest and apoptosis *in vitro*. In a prior evaluation of RG7112 against pediatric cancer cell lines, and a variety of p53 wild-type xenografts in immune-deficient mice (Carol 2013), cell sensitivity *in vitro* differed based on p53 mutation status. It was observed that p53-mutant cell lines were ~ 10-fold less sensitive to this MDM2 inhibitor than cells with wild-type p53. In contrast, RG7112 induced regressions in only 5 of 26 solid tumor models that had wild-type p53 by sequencing (Carol 2013). However, despite the relatively modest anti-tumor activity, RG7112 did induce p53 and downstream targets (p21, PUMA), hence clearly inhibited the MDM2:p53 interaction by stabilizing p53. In these "non-responsive" tumors, the increase in p53 was inadequate to induce apoptosis, or cell cycle arrest, but the combination of an MDM2 inhibitor with DNA damage agents is potentially synergistic in stabilizing p53 and inducing cell death. This is the rationale for combining a second generation MDM2 inhibitor, RG7338, with ionizing radiation. *In vivo* studies combining RG7338, using either daily or weekly schedules of administration, with daily fractionated radiation (2Gy fractions) to a total of 10, 20 or 30Gy are completed using childhood sarcoma models transplanted into immune-deficient mice (Phelps 2015). Results of this study demonstrated that the MDM2 inhibitor RG7388 was well tolerated at 80 mg/kg on the daily x 5 schedule (schedule 1) and once weekly x 2 at 200 mg/kg (given as split doses 12 hr apart; Schedule 2). RG7388 was administered 2 hr before XRT (2 Gy) daily for 5 days (Schedule 1) and a split dose given 12 hr and 2 hr pre-irradiation (Schedule 2). RG7388, as a single agent, had no significant effect on the growth of Rh18 xenografts administered on either schedule (P=0.8959 and P=0.5011 for Schedule 1 and 2, respectively). Skin toxicity was not significantly increased in the mice treated with radiation and RG7388 compared with radiation alone (Phelps 2015). RG7388, administered on both schedules, significantly enhanced the effect of daily fractionated radiation (figure 1). In a separate study, AMG 232 (KRT-232) was found to synergize with radiotherapy in a variety of cancer cell lines and xenograft models harboring WT p53 (Werner 2015).

In summary, strong preclinical evidence suggests that MDM-2 inhibitor AMG 232 (KRT-232) and radiotherapy may have additive or synergistic anti-tumor activity in p53 WT STS. Since the majority of STS harbor WT p53 and radiotherapy is a standard treatment modality in STS, we believe this is a very feasible and promising study.

2.3 AMG 232 (KRT-232) (9May2018)

2.3.1 Nonclinical Studies: Mechanism of Action and Activity

AMG 232 (KRT-232) is an orally (PO) bioavailable, selective small molecule inhibitor of MDM2 that blocks the protein-protein interaction between MDM2 and p53 (Investigator's Brochure, 26 February 2018; Rew and Sun, *et al.*, 2014; Sun *et al.*, 2014). [redacted] AMG 232 (KRT-232) effectively inhibited proliferation of SJSA-1 human osteosarcoma cells *in vitro* (which displays MDM2 gene amplification and has $p53^{WT}$) at IC_{50}=9.1 nM, as assessed in a 5-ethynyl-2'-deoxyuridine (EdU) cell proliferation assay (Sun *et al.*, 2014). AMG 232 (KRT-232) demonstrated robust antitumor activity *in vivo*, resulting in dose-dependent reduction in tumor growth with half-maximal effective dose (ED_{50}) of 9.1 mg/kg in the SJSA-1 xenograft mouse model following 14-day PO once daily (QD) dosing. Complete tumor regression was observed in 10 of 12 animals at a dose of 60 mg/kg. Activation of p53 signaling was determined by induction of p21 mRNA, which is a direct transcriptional target of p53 in three $p53^{WT}$ tumor cell lines, *i.e.*, SJSA-1, HCT116 (non-amplified MDM2 colorectal [CRC] cell line), and ACHN (renal carcinoma) (Canon *et al.*, 2015). AMG 232 (KRT-232) increased p21 mRNA levels in these cell lines 34.9-, 9.8-, and 11.5-fold, respectively. AMG 232 (KRT-232)-mediated induction of p21 mRNA was also tested *in vivo* in the SJSA-1 xenograft murine model; animals receiving only a vehicle served as negative control/baseline. [redacted]

Selectivity of AMG 232 (KRT-232) antitumor activity for $p53^{WT}$ cells was examined via a 5-bromo-2'-deoxyuridine (BrdU) cell proliferation assay (Sun *et al.*, 2014). Following 16 h incubation, AMG 232 (KRT-232) substantially inhibited growth of $p53^{WT}$ HCT116 (IC_{50}=10 nM) compared to no growth inhibition observed in p53-deficient ($p53^{-/-}$) cell culture.

In vivo antitumor activity of AMG 232 (KRT-232) was also assessed in xenograft mouse models of various tumor types which have $p53^{WT}$ and non-amplified MDM2, *i.e.*, A375sq2 (mutant $BRAF^{V600E}$ melanoma), HCT116 (mutant KRAS CRC), and NCI-H460 (mutant KRAS non-small cell lung cancer [NSCLC]) (Canon *et al.*, 2015). [redacted]

Nonclinical Pharmacology

After intravenous administration, the estimated mean AMG 232 (KRT-232) systemic clearance (CL) was low in mice, rats, and monkeys ([redacted] but high in dogs [redacted] Ye *et al.*, 2015). Steady-state volume of distribution (V_{ss}) was moderate in mice, rats, and dogs (1.10; 1.11, and 1.53 L/kg, respectively), but high in monkeys [redacted] suggesting a wide distribution of AMG 232 (KRT-232) into tissues. The estimated terminal elimination half-life ($t_{½}$) of AMG 232 (KRT-232) was [redacted]. Apparent bioavailability of AMG 232 (KRT-232) after oral dosing was moderate to high in mice, rats, monkeys ([redacted], respectively) but low in dogs (19%). [redacted]

The *in vitro* plasma protein binding of AMG 232 (KRT-232) was high across species ([redacted] Ye *et al.*, 2015). The unbound fraction of AMG 232 (KRT-232) in the mouse, rat, dog, monkey, and human plasma was [redacted], respectively. The AMG 232 (KRT-232) blood-to-plasma partition ratio in the mouse, rat, dog, monkey, and human was [redacted], respectively.

[redacted] The *in vivo* metabolism study of AMG 232 (KRT-232) in rats, and the *in vitro* metabolism study in hepatocytes from humans and preclinical species indicate that the predominant clearance mechanism was acyl-glucuronidation. The AMG 232 (KRT-232)-acyl glucuronide was the only metabolite detected *in vitro* in hepatocytes from rat, dog, monkey, and human. Formation of AMG 232 (KRT-232)-acyl-glucuronide was catalyzed by human uridine diphosphoglucuronosyl transferases (UGTs) UGT1A1, UGT1A3, and UGT1A4. [redacted]

[redacted]

[redacted]

[redacted]

Based on nonclinical pharmacokinetics (PK), the predicted PK parameter values for AMG 232 (KRT-232) in humans are as follows: CL = 0.15 L/h/kg, V_{ss} = 5.0 L/kg, and $t_{1/2}$=23 h (Ye *et al.*, 2015). Overall, the nonclinical PK and drug metabolism data support the clinical testing of AMG 232 (KRT-232) in human subjects.

Nonclinical Toxicology

[redacted]

[redacted]

AMG 232 (KRT-232) has demonstrated appropriate preclinical PK and acceptable safety profile in rats and monkeys to support clinical development. [redacted]

[redacted]

2.3.2 Effects in Humans

Summary: AMG 232 (KRT-232) is being evaluated in 3 clinical studies. Study 20120106 is a completed phase 1, first in human (FIH) study evaluating the safety, tolerability, PK, PD, and maximum tolerated dose (MTD) of orally administered AMG 232 (KRT-232) in subjects with advanced p53WT solid tumors or multiple myeloma. Study 20120234 is a completed phase 1b study evaluating the safety, tolerability, PK, PD, and MTD of orally administered AMG 232 (KRT-232) as a monotherapy or in combination with trametinib in subjects with relapsed/refractory acute myeloid leukemia (AML). Study 20120238 is a phase 1b/2a ongoing study evaluating the safety, tolerability, PK, PD, and MTD of orally administered AMG 232 (KRT-232) in combination with trametinib and dabrafenib or trametinib in adult subjects with metastatic cutaneous melanoma. A total of [redacted] subjects have been enrolled in 3 studies ([redacted] subjects in Study 20120106, [redacted] subjects in Study 20120234, and [redacted] subjects in Study 20130238). All [redacted] subjects across the 3 studies have received ≥ 1 dose of AMG 232 (KRT-232). Doses ranged from 15 to 480 mg QD for 7 days every 3 weeks (Q3W) (Study 20120106 and 20120238) or every 2 weeks (Q2W) (Study 20120234) [redacted].

Clinical Pharmacokinetics

[redacted]

Clinical Toxicity

A total of [redacted] subjects were enrolled in part 1 (dose escalation) and [redacted] subjects were enrolled in part 2 (dose expansion) of the study. All subjects enrolled in both parts of the study received ≥ 1 dose of investigational product.

[redacted]

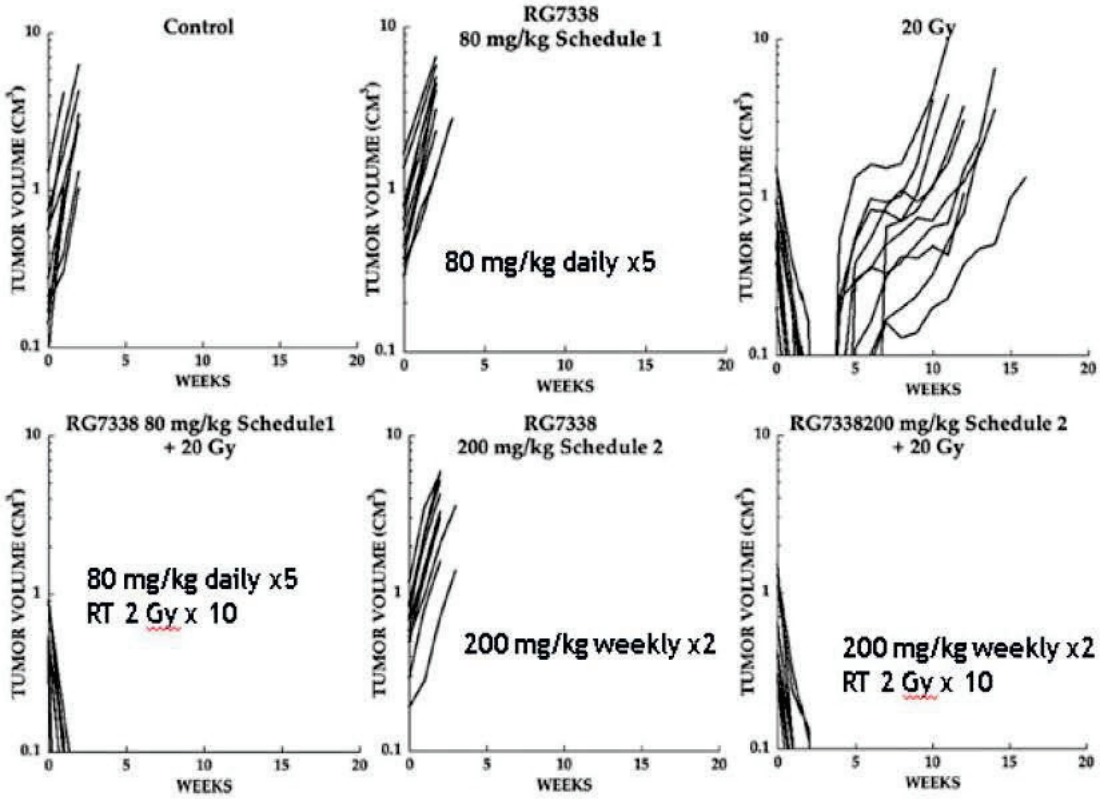

Figure 1, mdm-2 inhibitor RG7338 synergizes radiotherapy in a soft tissue sarcoma xenograft model (Phelps 2015)

2.3.3 Pharmacokinetic/Pharmacodynamics and Potential Drug Interactions

[redacted]

AMG 232 (KRT-232) is not the first-in-class MDM2 inhibitor. When reviewing this class, the majority of clinical trials are assessing multiple potential biomarkers including elevation of p53 and p21 expression, Ki-67 for decreased cell proliferation, TUNEL for

apoptosis, MDM2 mRNA, and serum macrophage inhibitory cytokine-1 (MIC-1). While MIC-1 can be highly variable but has been demonstrated to be a potential biomarker for various cancers (Khaled 2012). Serum MIC-1 levels have been typically measured by the commercially available ELISA kit (e.g., Biovendor, LLC, Asheville, NC). As a biomarker, the serum MIC-1 levels are normalized to the baseline values (an average of 2 baseline samples) in each patient due to the large inter-patient variability. In a clinical trial of RG7112 (an MDM2 inhibitor), elevations in serum MIC-1 (change from baseline) were observed and correlated with drug exposure and were schedule dependent (Patnaik 2006). [redacted] Therefore, an abbreviated pharmacokinetic profile will be obtained on all patients to correlate with pharmacodynamics alterations (primarily MIC-1), adverse events (e.g., thrombocytopenia).

Rationale for the AMG 232 (KRT-232) Schedule

[redacted] nother MDM2 inhibitor, RG7112, exerted clinical benefit and on-target toxicity on a QDx3 or QDx5 regimen while less benefit and toxicity was observed with weekly or chronic daily dosing of lower doses (Andreeff 2016). Therefore, an intermittent schedule of AMG 232 (KRT-232) was selected that is anticipated to be biologically active (120 mg) with the intention to increasing the frequency of dosing from 2 to 5X/week at the 120 mg dose.

Dose Level		Dose (mg)	Total Dose (mg)	Dose intensity (mg/day)	Dose intensity (mg/day over XRT days)
AMG 232 (KRT-232) Alone (MTG) in Solid Tumors Dose Level	7 days on/14 off Dose Freq	240 Dose (mg)	1680 Total 3-wk dose	80 Dose intensity (mg/day)	112 Dose Intensity over XRT days
Increasing frequency with constant doses					
-1	2X/week	120	720	34.3	48
1	3X/week	120	1080	51.4	72
2	4X/week	120	1440	68.6	96
3	5X/week	120	1800	85.7	120

2.4 Pathologic Versus Radiographic Response Assessment

Even though imaging of the primary tumor before and after neoadjuvant therapy (MRI with and without contrast for extremity STS, CT with and without contrast for abdominal/pelvic/retroperitoneal tumors) have been standard imaging modalities for STS, it has not been established whether a significant change in the size of the tumor mass could serve as a meaningful surrogate marker for patient outcomes after neoadjuvant therapy. While some randomized studies comparing chemotherapy regimens for soft tissue sarcoma have shown significant differences in radiologic response rates but no difference in outcome (Antman K 1992, Edmonson 1993), others have described a significant correlation between tumor response and improved survival (Meric 2002, Pezzi 1990). Radiographic response after neoadjuvant radiotherapy or chemoradiotherapy has also been highly variable when only RECIST criteria was utilized.

Treatment-induced tissue necrosis following neoadjuvant therapy has been established as a reliable predictor of outcome in bone sarcomas (Bacci 2005, Picci 1997). For STS, a large retrospective study of 496 patients with intermediate to high-grade extremity STS soft tissue sarcomas showed superior 5- and 10-year local recurrence (LR) rates for patients with ≥95% pathologic necrosis compared to those with <95% pathologic necrosis (Eilber 2001).

Tumor volume change, and in particular, necrosis adjusted tumor volume, is known to be highly correlated with pathologic percent necrosis. Percent necrosis on surgical pathology analysis following neo-adjuvant chemotherapy in sarcoma is a predictive biomarker of long term clinical outcomes. The use of imaging, especially MRI, as a means to predict necrosis as a result of CRT would provide a less costly and less invasive means of assessing this predictive factor in sarcoma patients undergoing standard and experimental types of neoadjuvant CRT.

3. ELIGIBILITY AND INELIGIBILITY CRITERIA (29-MAY-2019)

Note: Per NCI guidelines, exceptions to inclusion and exclusion criteria are not permitted. For questions concerning eligibility, please contact the Biostatistical/Data Management Center (see protocol cover page). For radiation therapy-related eligibility questions, please contact RTQA (see protocol cover page).

3.1 Eligibility Criteria (29-MAY-2019)

A patient cannot be considered eligible for this study unless ALL of the following conditions are met.

Prior to Step 1 Registration:

3.1.1 Patients with pathologically proven diagnosis of grade 2-3 (intermediate or high grade) soft tissue sarcoma with size ≥ 5 cm are eligible to enroll if the intention to treat is curative. They must have sufficient tissue to submit to central laboratory for review as well as for NGS sequencing (see submission requirement). Biopsy should be obtained within 180 days prior to registration.

Availability of tumor tissue is mandatory for study eligibility. The patient must have consented to provide archived formalin-fixed paraffin-embedded tumor tissue for future central pathology review, NGS sequencing and/or translational research. (Refer to Section 10 regarding tissue collection parameters).

3.1.2 Appropriate stage for study entry based on the following diagnostic workup:

- History/physical examination within 30 days prior to registration;
- Imaging of the primary tumor by MRI and/or CT with or without contrast and/or PET/CT within 30 days prior to registration;
- Staging workup evaluated by chest CT and/or PET/CT showing no distant metastasis within 30 days prior to registration

3.1.3 There is a planned definitive surgical resection of the primary tumor;

3.1.4 Age ≥ 18;

3.1.5 ECOG or Zubrod performance status of 0-1 within 30 days prior to registration;

3.1.6 Adequate hematologic function within 30 days prior to registration defined as follows:

- Absolute neutrophil count ≥ 1500/μL
- Platelet count ≥ 100,000/μL
- Hemoglobin: ≥ 10 g/dL (transfuse as necessary to raise levels; no transfusions within 7 days of start)

3.1.7 Adequate renal and hepatic function within 30 days prior to registration defined as follows:

- Calculated Creatinine Clearance ≥ 60 ml/min (by Cockroft-Gault formula) within 30 days prior to registration.
- The patient has an adequate coagulation function as defined by International Normalized Ratio (INR) ≤1.5 x ULN or prothrombin time (PT) ≤1.5 x ULN, and partial thromboplastin time (PTT or aPTT) ≤1.5 x ULN (those receiving anticoagulation therapy except low molecular weight heparin are excluded).
- Total bilirubin ≤ 1.5 x upper limit of normal (ULN) appropriate for age (except for patients with Gilbert's Syndrome, who must have a total bilirubin <3 mg/dL)
- SGOT (AST) or SGPT (ALT) < 2.5 x upper limit of normal (ULN) appropriate for age

3.1.8 Females of child-bearing potential must have a negative serum pregnancy test within 7 days prior to registration.

Exceptions: Females not of child-bearing potential due to surgical sterilization (at least 6 weeks following tubal ligation, hysterectomy, or surgical bilateral oophorectomy with or without hysterectomy) confirmed by medical history; or female after menopause.

- A "postmenopausal woman" is a woman meeting either of the following criteria:
    - spontaneous amenorrhea for at least 12 months, not induced by a medical condition such as anorexia nervosa and not taking medications during the amenorrhea that induced the amenorrhea (for example, oral contraceptives, hormones, gonadotropin releasing hormone, antiestrogens, selective estrogen receptor modulators [SERMs], or chemotherapy)
    - spontaneous amenorrhea for 6 to 12 months and a follicle-stimulating hormone (FSH) level >40 mIU/mL

- Females of child-bearing potential and males must agree to use highly effective contraceptive precautions during the trial and up to 12 months following the last dose of study treatment. A highly effective method of birth control is defined as one that results in a low failure rate (that is, <1% per year) when used consistently and correctly, such as implants, injectables, combined oral contraceptives, some intrauterine contraceptive devices (IUDs), sexual abstinence, or a vasectomized partner.

3.1.9 The patient or a legally authorized representative must provide study-specific informed consent prior to study entry.

Prior to Step 2 registration

3.1.10 TP53 sequencing by NGS performed by central pathology lab (Refer to section 10.1)

3.2 Ineligibility Criteria (22-FEB-2021)

Patients with any of the following conditions are NOT eligible for this study.

Prior to Step 1 Registration:

3.2.1 Well-differentiated liposarcoma or other low grade STS; Kaposi sarcoma, bone sarcomas, cartilage sarcomas and GIST. See Appendix VIII.

3.2.2 Definitive clinical or radiologic evidence of metastatic disease; indeterminate lung nodules less than 8mm are acceptable.

3.2.3 The patient has history of another primary malignancy, with the exception of
a) curatively treated non-melanomatous skin cancer;
b) curatively treated cervical carcinoma in situ;
c) non-metastatic prostate cancer;
d) other primary non-hematologic malignancies or solid tumor treated with curative intent, no known active disease, and no treatment administered during the last 3 years prior to registration.

3.2.4 The patient has a serious cardiac condition, such as congestive heart failure; New York Heart Association Class II/ III/IV heart disease; unstable angina pectoris, cardiac stenting within 6 months of enrollment; myocardial infarction within the last 3 months; valvulopathy that is severe, moderate, or deemed clinically significant; or arrhythmias that are symptomatic or require treatment.

3.2.5 Females who are pregnant or breastfeeding.

3.2.6 Prior systemic chemotherapy for the study cancer (sarcoma); note that prior chemotherapy for a different cancer is allowable. However, unresolved toxicities from prior anti-tumor therapy, defined as not having resolved to Common Terminology Criteria for Adverse Events (CTCAE) version 5.0 grade 0 or 1, or to levels dictated in the eligibility criteria with the exception of alopecia (Grade 2 or 3 toxicities from prior antitumor therapy that are considered irreversible [defined as having been present and stable for > 6 months], such as ifosfamide-related proteinuria, may be allowed if they are not otherwise described in the exclusion criteria AND there is agreement to allow by both the investigator and sponsor).

3.2.7 Prior radiotherapy to the region of the study cancer that would result in overlap of radiation therapy fields.

3.2.8 Clinically significant bleeding within 4 weeks of screening, current use of warfarin, factor Xa inhibitors, and direct thrombin inhibitors unless these medications can be safely discontinued 14 days prior to AMG 232 (KRT-232) administration. Note: Low molecular weight heparin and prophylactic low dose warfarin are permitted. PT/PTT must meet the inclusion criteria. Subjects taking warfarin must have their INR followed closely.

3.2.9 History of allergic reactions attributed to compounds of similar chemical or biologic composition to AMG 232 (KRT-232).

3.2.10 Medication use:

- All subjects must agree to stop the use of all herbal medicines (*e.g.*, St. John's wort), and supplements, within the 14 days prior to receiving the first dose of AMG 232 (KRT-232), and during the protocol AMG 232 (KRT-232) treatment (Weeks 1-5). Subjects may renew the use of the above at week 6. Standard adult multi-vitamin is allowed.
- All subjects must agree to stop the use of any known CYP3A4 substrates with [redacted]
- All subjects are required to submit a list of medications consumed within 14 days prior to receiving the first dose of AMG232 and during the protocol AMG232 treatment (Weeks 1-5).

3.2.11 Patients with GI tract disease causing the inability to take oral medication, malabsorption syndrome, requirement for intravenous alimentation, prior surgical procedures affecting absorption, uncontrolled inflammatory GI disease (e.g., Crohn's disease, ulcerative colitis), therefore could affect the absorption of AMG 232 (KRT-232) at the discretion of treating physician.

3.2.12 Patients with active infection requiring IV antibiotics within 2 weeks of registration.

3.2.13 Patients with known Positive Hepatitis B Surface Antigen (HepBsAg) (indicative of chronic Hepatitis B), positive Hepatitis total core antibody with negative HBsAG (suggestive of occult hepatitis B), or detectable Hepatitis C virus RNA by a polymerase-chain reaction (PCR) assay (indicative of active Hepatitis C – screening is generally done by Hepatitis C Antibody (HepCAb), followed by Hepatitis C virus RNA by PCR if HepCAb is positive).

3.2.14 Patients known to be positive for human immunodeficiency virus (HIV) are NOT excluded from this study, but HIV-positive patients must have:

- A stable regimen of highly active anti-retroviral therapy (HAART)
- No requirement for concurrent antibiotics or antifungal agents for the prevention of opportunistic infections
- A CD4 count above 250 cells/mcL and an undetectable HIV viral load on standard PCR-based test

- HIV testing is not required

3.2.15 Treatment with medications known to cause QTc interval prolongation within 7 days of study day 1 is not permitted unless approved by the sponsor. Use of ondansetron is permitted for treatment of nausea and vomiting.

4. REQUIREMENTS FOR STUDY ENTRY, TREATMENT, AND FOLLOW-UP (29-MAY-2019)

PRE-TREATMENT ASSESSMENTS

Assessments	Prior to Step 1 Registration (calendar days)	Prior to Treatment (calendar days)
Tumor tissue of primary tumor	≤180	
Evaluable disease defined by RECIST	≤30	
History/Physical Exam	≤30	
Height/Weight	≤30	
Concurrent Medications	≤30	
Baseline Toxicity	≤30	
MRI with/without contrast and/or CT with/without contrast and/or PET/CT of the primary tumor evaluation	≤30	
Chest CT and/or PET/CT staging studies	≤30	
ECOG or Zubrod status	≤30	
No major surgery		≤28
ANC, platelet, hemoglobin	≤30	
Calculated creatinine clearance, PT/INR, PTT or aPTT, total bilirubin, SGOT (AST)* or SGPT (ALT), Alk phos	≤30	
SGOT (AST)		≤ 30*
Amylase, lipase		≤ 30
Serum pregnancy test	≤7	≤72 hours
Informed consent	≤30	
Tissue for TP53 NGS sequencing	Must be submitted within 7 days after Step 1 registration	
PK/PD of AMG 232 (KRT-232)	As scheduled in the PK/PD section (see Appendices II, III, and IV)	

* If SGOT (AST) is not done prior to Step 1 registration then it is required within 30 days prior to the first dose of AMG 232 (KRT-232).

ASSESSMENTS DURING TREATMENT

Assessments	Weekly During AMG 232 (KRT-232) alone and AMG 232 (KRT-232)/RT	4 wks post AMG 232 (KRT-232)/RT prior to surgery	At time of surgery
History and physical	X	X	
CBC, ANC, platelets	X	X	
ALT, AST, total bilirubin, alk phos, amylase, lipase	X	X	
Coagulation (PT, PTT, INR)	X	X	
Adverse Events	X	X	
Concurrent Medications	X	X	
PK/PD of AMG 232 (KRT-232)	As scheduled in the PK/PD section (see Appendices II, III, and IV)		
MRI and/or CT with contrast for primary tumor		X	
CT chest for re-staging purpose		X	
Research specimen collection – surgical tissue specimen			X (As specified in Section 10)

NOTE: Final surgical specimen will be collected and sent to Dr. Wakely for central pathology review.

NOTE: If P53 status is wild type, the patient is eligible for final analysis. If p53 status is found to be deleted or mutated, the patient will be informed of the results and protocol treatments [AMG 232 (KRT-232)] will be stopped. Standard of care treatments, however, including preoperative radiotherapy and subsequent surgical resection will remain in place. Clinical data will continue to be collected from these patients but collected data will not be used for primary and secondary end point.

ASSESSMENTS IN FOLLOW UP

Assessments	**Time Point** From end of RT: q3 mos. for 2 yrs. then final follow-up at 2.5 years from end of RT
MRI or CT of primary tumor with/without contrast	X
Chest X-ray, chest CT or PET/CT	X
History and physical (including adverse event evaluation)	X
Adverse Events	**X**
Research specimen collection (optional)	At time of recurrence/progression or of solid second neoplasm occurrence (as specified in Section 10)

Measurement of Effect/Definition of Disease Assessments

Although the clinical benefit of AMG 232 (KRT-232) has not yet been established, the intent of offering this treatment is to provide a possible therapeutic benefit, and thus the patient will be carefully monitored for tumor response and symptom relief in addition to safety and tolerability. Patients with measurable disease will be assessed by standard criteria per the assessment tables above.

All tumor recurrences including local recurrence, regional recurrence, distant metastasis, and second primary tumor must be recorded in this study. All tumor recurrences should be documented on cross-sectional imaging (CT or MRI with and without contrast). Pathological confirmation of recurrence is strongly recommended.

Local tumor recurrence: Any tumor recurrence inside the clinical target volume (CTV) is defined as “in-field recurrence”; any tumor recurrence beyond the CTV to within 3 cm distance from the edge of the CTV is defined as “marginal recurrence”; this marginal recurrence is considered “geographic miss”. Any other local recurrence is defined as “outside-field recurrence”.

Local tumor progression: At least a 20% increase in the maximal dimension of the primary tumor taking as reference the smallest maximal dimension recorded since treatment started;

Regional tumor recurrence: Any nodal metastasis adjacent to the primary soft tissue sarcoma;

Distant tumor metastasis: Any tumor that develops distantly from the primary site of sarcoma;

Second primary tumor: Any different histology of sarcoma or any other type of malignancy within or outside the radiation field

5. TREATMENT PLAN/REGIMEN DESCRIPTION

5.1 AMG 232 (KRT-232) Therapy (01-DEC-2021)

- **Step 1**: All eligible patients will be registered in Step 1 and will provide tissue to the central lab for sequencing. Patients will also start AMG 232 (KRT-232) lead-in treatment on week -1 at the assigned dose level and frequency from the dose escalation table. **Week (-1) is defined as one week before starting RT treatment.** Protocol treatment must begin as soon as possible but within 21 days after Step 1 registration.

- **Step 2**: Based on the sequencing results, those with p53 WT will continue to receive combination treatment of AMG 232 (KRT-232) and RT for a total of 5 weeks. Those who have p53 deletion/mutations will stop taking AMG 232 (KRT-232) but will continue daily radiotherapy, followed by subsequent surgery. Patients can continue on AMG 232 (KRT-232) week 1 dose if there is a delay in NGS sequencing results.

- All data will be collected, but only data from p53 WT will be used for primary and secondary endpoint analysis.

5.1.1 Agent Administration

AMG 232 (KRT-232) is to be given alone during week -1 in the lead-in (Step 1) and for a total of 5 weeks in Step 2 treatment. In step 2, AMG 232 (KRT-232) is to be given with RT starting on day 2 (or per the assigned dose level from the dose escalation table) and approximately 30 minutes to 1 hour before RT.

AMG 232 (KRT-232) is to be taken with a full glass of water with or without food. AMG 232 (KRT-232) should not be crushed, chewed, or dissolved in water. Patients should not be re-dosed with AMG 232 (KRT-232) after vomited doses.

If a dose of AMG 232 (KRT-232) is missed due to reasons other than toxicity and the next dose is in less than 12 hours, take the next dose at the normal time. Do not make up the missed dose. If a dose is missed and the next dose is in 12 hours or more, take the missed dose as soon as possible. Take the next dose at normal time.

Patients will keep a drug diary log book and return the drug diary at each clinic visit.

Radiotherapy will start on week 1 to week 5 to receive a daily dose, day 1-5 for a total of 25 treatments. If RT is held on a day when the drug should be taken, the drug should be held, and the dose will be taken with RT on the following treatment day. Make up doses can occur outside of the 5-week treatment period if RT is to be given in this time frame due to missed or delayed RT treatments.

AMG 232 (KRT-232) Dose Escalation/De-escalation Schedule for MTD/RP2D Determination (See Section 14 for dose escalation/de-escalation rules; dose level 1 is the starting dose)

Dose Level	Dose frequency	Dose (Weekly Total Dose mg)
-1	2x/week (D2,4)	120 mg (240 mg)
1	3x/week (D2-4)	120 mg (360 mg)
2	4x/week (D2-5)	120 mg (480 mg)
3	5x/week (D1-5)	120 mg (600 mg)

D2: Day 2 of weekly radiation
D2, 4: Day 2 and 4 of weekly radiation
D2-4: Day 2, 3 and 4 of weekly radiation
D2-5: Day 2, 3, 4 and 5 of weekly radiation
D1-5: Day 1, 2, 3, 4 and 5 of weekly radiation

AMG to start 7 days prior to radiation therapy (week -1) at the dose frequency of the above table and then for 5 weeks
Radiotherapy to start from week 1 to week 5

5.1.2 The following medications and/or therapies should not be administered within the timeframes specified prior to enrollment and during the study (unless otherwise specified below):

- Antitumor therapy (chemotherapy, antibody therapy, molecular targeted therapy, retinoid therapy, hormonal therapy, or investigational agent) within 28 days prior to receiving the first dose of AMG 232 (KRT-232); concurrent use of hormone deprivation therapy for hormone-refractory prostate cancer or breast cancer and continuation of previous hormone therapy in breast cancer subjects are permitted.
- All herbal medicines (*e.g.*, St. John's wort), vitamins, and supplements consumed by the subject within the 14 days prior to receiving the first dose of AMG 232 (KRT-232), and during the protocol AMG 232 (KRT-232) treatment (Week 1-5). Subjects may renew the use of the above at week 6. Standard adult multi-vitamin is allowed.
- [redacted]
- Treatment with medications known to cause QTc interval prolongation within 7 days prior to receiving the first dose of AMG 232 (KRT-232) unless approved by the sponsor. Use of ondansetron is permitted for treatment of nausea and vomiting.
- Systemic anticoagulation therapy, with warfarin, direct thrombin inhibitors, indirect thrombin inhibitors and direct factor Xa inhibitors, within 14 days or 5 half-lives (whichever is longer) prior to receiving the first dose of AMG 232 (KRT-232). Low

molecular weight heparin and prophylactic low-dose warfarin are permitted. PT/PTT must meet the inclusion criteria. Subjects taking warfarin must have their INR followed closely.

- Clinically significant bleeding within 4 weeks of screening, current use of warfarin, factor Xa inhibitors, and direct thrombin inhibitors unless these medications can be safely discontinued 14 days prior to receiving the first dose of AMG 232 (KRT-232) or surgical resection. Note: Low molecular weight heparin. PT/PTT must meet the inclusion criteria. Subjects taking warfarin must have their INR followed closely.

5.2 Radiation Therapy (22-FEB-2021)

IGRT IS MANDATORY FOR THIS STUDY (Section 5.2.1)

Note: Pre-treatment reviews will be mandatory for **Cohort B only -** the first case from each institution will require a pre-treatment review. The patient cannot begin treatment until complete data from the first case is received, reviewed and approved. **Sites need to allow 3 business days for the pre-treatment review, and if necessary, 3 additional business days if a revision and re-plan is requested by the Reviewer** (See Section 5.2.11).

Protocol treatment must begin within 21 calendar days after Step 1 registration

5.2.1 Treatment Technology

Treatment Modalities

This protocol focuses on the pre-operative external beam radiotherapy. All endpoints will be tracked till week 4 after the pre-operative radiotherapy (prior to surgery). Post-surgery boost treatment is *not* allowed in this protocol. Radiation therapy using megavoltage photons (4-18 MV) and Co-60 energy (MRIdian only) are permitted in this study. For tumors adjacent or included in lung tissue, beam energy should be <10 MV. Proton therapy is not allowed for this protocol.

For cohort A (Extremity and Body Wall), advanced delivery techniques including intensity modulated radiation therapy (IMRT), volumetric modulated radiation therapy (VMAT), MRIdian, CyberKnife, Vero, and Tomotherapy are recommended. However, 3D- conformal radiation therapy (3DCRT) is also permitted. For cohort B (Abdomen/Pelvis/Peritoneal cavity), advanced delivery technique is mandatory. 3DCRT is not permitted for cohort B.

Image Guidance Devices

Treatment machine with image-guided radiation therapy (IGRT) capability is mandatory for this protocol. IGRT is defined as the use of computer-assisted system that provides detailed information on shifts of the patient support system based on image registration software.

Image guidance may be achieved using any one or more of the following techniques:

- Orthogonal kilovoltage (KV) and/or MV electronic images, e.g., ExacTrac;
- Linear-accelerator mounted kV and MV cone beam CT images (CBCT);
- Linear-accelerator mounted MV CT images (e.g., Tomotherapy);

- Other mechanism, (ex. optical surface tracking and electromagnetic localization) after discussion with the Medical Physics Co-chair.

5.2.2 Localization, Immobilization and Simulation

Patients should be immobilized in stable and comfortable positions to allow accurate repositioning from treatment to treatment and to prevent movement during treatments. A variety of immobilization devises may be utilized, including Vacbag, Alpha Cradle and thermoplastic casts.

Adjustments of patient position should be made accordingly, if needed prior to treatment. Pretreatment images may include 2D or 3D imaging acquired using the techniques described in Section 5.2.1.

Motion Management Technique

Motion management is highly recommended for this protocol especially for tumors located in upper abdomen. Three categories of motion management are allowed on this protocol:

(1) Motion encompass approach: 4DCT and free breathing treatment
(2) Motion control approach: breath hold and abdominal compression
(3) Active motion management approach: gated delivery or motion tracking delivery

Simulation Imaging

Simulation imaging must encompass the entire body part where tumor is located and the adjacent critical structures. Appropriate pre-treatment diagnostic imaging studies, such as the MRI (T1 post contrast and T2 sequences), PET/CT or CT, when possible, must be fused with simulation CT to aid target delineation. Radiotherapy treatment plans will be generated after immobilization and computerized tomography (CT) simulation. Typically contrast is not required for sarcoma simulation but it can be considered if deemed necessary by the treating radiation oncologist. In case the contrast agent is employed, the density of contrast should be overridden to a representative background electron density or a set of non-contrast free-breathing scan can be acquired to prevent the dose calculation uncertainty caused by contrast agent.

Simulation imaging for cohort A: Extremity and Body Wall

Free-breathing CT with slice thickness ≤ 3mm is mandatory as primary data set for simulation and dose calculation. For extremity cases, CT scan should encompass the tumor and the adjacent joints (if applicable). For trunk/chest wall cases, CT scan should encompass the tumor, starting from cricoid cartilage and extending inferiorly to the bottom of ribcage.

Simulation imaging for cohort B: Abdomen/Pelvis/Peritoneal cavity

4DCT is recommended for primary tumors located in the upper abdomen. Free-breathing CT, averaged CT (if 4DCT is employed), or BH-CT (if breath-hold treatment is intended) with slice thickness ≤ 3mm is mandatory as primary data set for simulation and dose calculation. It is desired to cover the tumor and the critical structures from liver to the bottom of pubic symphysis. However, the scanning range can be adjusted and determined by the physician.

5.2.3 Imaging for Structure Definition, Image Registration/Fusion and Follow-up

Pre-treatment diagnostic imaging studies including MRI with and without contrast and/or CT with and without contrast and/or FDG-PET/CT will be required as part of staging and/or to assist in volume delineation in all eligible patients. In order to standardize target volume definition using better tumor and soft tissue definition an MRI of primary site is highly preferred unless patient has contraindications to MRI (e.g., implanted pacemaker, neurostimulator, aneurysm clips, metallic foreign objects, or other contra-indications).

Diagnostic MRI, CT, PET/CT images, when possible, must be fused to the planning CT (free-breathing CT, averaged 4DCT, or BH-CT). Image fusion should be checked by physician prior to contouring. All targets and critical structure should be delineated on the planning CT. When the motion management is utilized for treatment, the motion of target should be properly accounted in the target definition. Site-specific guidance is provided in sections 5.2.4 and 5.2.5.

Note: Diagnostic imaging that is fused with planning CT will be required for submission via TRIAD. Refer to protocol specific page on the NRG website for data submission and section 13.2 in the protocol text.

5.2.4 Definition of Target Volumes and Margins

Note: All structures must be named for digital RT data submission as listed in the respective table below. The structures marked as "Required" in the table must be contoured and submitted with the treatment plan. Structures marked as "Required when applicable" must be contoured and submitted when applicable.

Resubmission of data may be required if labeling of structures does not conform to the standard DICOM name listed. Capital letters, spacing and use of underscores must be applied exactly as indicated.

Target Definition for cohort A: Extremity and Body Wall

Standard Name	Description	Validation **Required/Required when applicable/Optional**
GTV_5000	GTV to receive 50 Gy	Required
CTV_5000	CTV to receive 50 Gy	Required
PTV_5000	PTV to receive 50 Gy	Required

PTV_Eval_5000	PTV minus OARs	Required when applicable

Detailed Specifications
The definition of volumes will be in accordance with the ICRU Report #62: Prescribing, recording and Reporting Photon Beam Therapy (supplement to ICRU Report #50).

Gross Target Volume (GTV_5000): Gross tumor defined by MRI T1 plus contrast images (MRI with contrast is required unless contraindicated). Fusion of MRI and CT is recommended to delineate the GTV for radiotherapy planning, but this is optional.

Clinical Target Volume (CTV_5000) for Intermediate-to-High Grade Tumors ≥ 8 cm: Include gross tumor and clinical microscopic margins. Typically CTV = GTV and suspicious edema (defined by MRI T1 post contrast images unless contraindicated) plus 3 cm margins in the longitudinal (proximal and distal) directions. CTV should also include areas with any suspicious MRI T2 signals. If this causes the field to extend beyond the compartment, the field can be shortened to include the end of a compartment. The radial margin from the lesion should be 1.5 cm including any portion of the tumor not confined by an intact fascial barrier or bone or skin surface.

CTV for All Other Tumors: Include gross tumor and clinical microscopic margins. Typically CTV = GTV and suspicious edema (defined by MRI T1 post contrast images) plus 2 cm margins in the longitudinal (proximal and distal) directions. CTV should also include areas with any suspicious MRI T2 signals. If this causes the field to extend beyond the compartment, the field can be shortened to include the end of compartment. The radial margin from the lesion should be 1 cm including any portion of the tumor not confined by an intact fascial barrier or bone or skin surface.

Planning Target Volume (PTV_5000): Include CTV and error of setup and organ motion. Typically PTV includes CTV plus 5 mm (with daily IGRT). Skin surfaces should not be contoured in CTV or PTV unless these are involved by gross tumor. If the incisional biopsy scar is small and will be resected at the time of surgery, it may not be contoured as CTV at the discretion of the treating radiation oncologist. Use of bolus on the skin surfaces is not encouraged when IMRT is used.

Planning Target Volume for Evaluation (PTV_Eval_5000): PTV volume minus impinging high priority OARs created for dosimetric evaluation. In cases where the PTV_5000 overlaps with critical organs, such as the spinal cord, the PTV_Eval_5000 should be created to limit dose to the OARs. Care should be taken to ensure that the cold spots within the PTV_Eval_5000 do not exist within the GTV.

Target Definitive for cohort B: Abdomen/Pelvis/peritoneal cavity

Standard Name	Description	Validation **Required/Required when applicable/Optional**
GTV_5000	GTV to receive 50 Gy	Required
IGTV_5000	Volume enveloping GTV motion over the course of a respiratory cycle	Required when applicable
*CTV_4500 (target in abdomen/pelvis/retro peritoneal cavity)	CTV to receive a minimum of 45 Gy (while GTV receives 50 Gy)	Required
*PTV_4500 (target in abdomen/pelvis/retro peritoneal cavity)	PTV to receive a minimum of 45 Gy (while GTV receives 50 Gy)	Required
PTV_Eval_4500	PTV minus OARs	Required when applicable

* **Cohort B only**: Prescription dose to CTV and PTV will be 45 Gy while dose to GTV (and IGTV if applicable) will be 50 Gy with simultaneous integrated boost (SIB) technique.

Detailed Specifications

The definition of volumes will be in accordance with the ICRU Report #62: Prescribing, recording and Reporting Photon Beam Therapy (supplement to ICRU Report #50).

Internal margin (IM) definition
For patients scanned with 4DCT, no IM is needed when IGTV is delineated based on individual phase CTs, maximum intensity projection (MIP) of the tumor. Otherwise, proper IM need to be applied to account for the internal motion.

Setup margin (SM) definition: 5 mm as daily IGRT is mandatory for cohort B.

> Gross Target Volume (GTV_5000): gross tumor defined by CT or MRI T1 plus contrast images. Fusion of MRI and CT is recommended to delineate the GTV for radiotherapy planning, but this is optional. Dose to GTV (and IGTV if applicable) will be 50 Gy with simultaneous integrated boost (SIB) technique.

Clinical Target Volume (CTV_4500): typically CTV = GTV plus 10 mm margin, however, the CTV field should not be extended beyond other organ structures, or compartment or intact fascia or bone.

For the retroperitoneal sarcoma extending to thigh through inguinal canal, the inferior margin in the thigh is typically 3 cm below the GTV and radial margin in thigh must be 1.5 cm, but should not beyond the compartment or intact fascia or bone.

Planning Target Volume (PTV_4500): typically includes CTV_4500 + IM + SM. In case the PTV expansion extends outside the skin, toward the spinal cord or into the spinal canal, it can be assumed that tumor motion will not occur in this direction and the PTV margin can be limited up to 5 mm in this direction.

Planning Target Volume (PTV_Eval_4500): PTV volume minus impinging high priority OARs created for dosimetric evaluation. In cases where the PTV_4500 overlaps with critical organs, such as the spinal cord, the PTV_Eval_4500 should be created to limit the dose to the OARs. Care should be taken to ensure that the cold spots within the PTV_Eval_4500 do not exist within the GTV.

5.2.5 Definition of Critical Structures and Margins

Note: All structures must be named for digital RT data submission as listed in the table below. The structures marked as "Required" in the table must be contoured and submitted with the treatment plan. Structures marked as "Required when applicable" must be contoured and submitted when applicable.
Resubmission of data may be required if labeling of structures does not conform to the standard DICOM name listed. Capital letters, spacing and use of underscores must be applied exactly as indicated.

Critical Structure Definition for cohort A: Extremity and Body Wall

1. Organs at Risk for Primary Tumors at Extremities

Structure Name	**Description**	Validation **Required/Required when applicable/Optional**
Femur_R(L)	Right (or left) femur within field	Required when applicable
Joint_R(L)	Right (or left) joint	Required when applicable
Skin	Skin contoured as a 3 mm thick rind	Required
SoftTissue_Sub	A strip of subcutaneous soft tissue; at the discretion of treating physician	Required

1. Organs at Risk for Primary Tumors at Body Wall (Trunk/chest wall)

Structure Name	**Description**	Validation **Required/Required when applicable/Optional**
BrachialPlex_R(L)	Right (or left) brachial plexus	Required
Esophagus	Esophagus	Required
Heart	Heart	Required
Lungs	Lungs	Required
SpinalCord	Spinal cord	Required

Critical Structure Definition for cohort B: Abdomen/Pelvis/peritoneal cavity

Structure Name	**Description**	Validation **Required/Required when applicable/Optional**
Liver	Liver	Required when applicable
Kidney_R(L)	Right (or left) kidney	Required when applicable
Spc_Bowel	Bowel space	Required when applicable
Stomach	Stomach	Required when applicable
Anus	Anus	Required when applicable
Bladder	Bladder	Required when applicable
Rectum	Rectum	Required when applicable
Vulva	Vulva	Required when applicable
Testis_R (L)	Right (or left) Testis; Testes should be defined if fertility preservation desired	Required when applicable
Ovary_R (L)	Right (or left) Ovary; Ovaries should be defined if fertility preservation desired	Required when applicable
SpinalCord	Spinal cord	Required when applicable

5.2.6 Dose Prescription

Note: The information provided in this section can be used for adjusting the dose constraints for treatment planning purposes. This table together with the planning priority table should be used during dose optimization. It is important to remember that ideal plans might not be achievable in all cases. Thus, the Compliance Criteria table could be different than the information given here. Cases will be scored using the Compliance Criteria table.

Dose Prescription for cohort A: Extremity and Body Wall

Target Standard Name	Dose (Gy)	Fraction Size (Gy)	# of Fractions	Frequency	Dose Specification Technique
PTV_5000	50	2	25	Daily	Covering >=95% PTV

Dose Prescription for cohort B: Abdomen/Pelvis/peritoneal cavity

Target Standard Name	Dose (Gy)	Fraction Size (Gy)	# of Fractions	Frequency	Dose Specification Technique
PTV_4500	45	1.8	25	Daily	Covering >=95% PTV
GTV_5000	50	2	25	Daily	Covering >= 95% GTV

5.2.7 Compliance criteria

The compliance criteria listed here will be used to score each case. Given the limitations inherent in the treatment planning process, the numbers given in this section can be different than the prescription table. The Per Protocol and Variation Acceptable categories are both considered to be acceptable. The Per Protocol cases can be viewed as ideal plans, and the Variation Acceptable category can include more challenging plans that do not fall at or near the ideal results. A final category, called Deviation Unacceptable, results when cases do not meet the requirements for either Per Protocol or Variation Acceptable. Plans falling in this category are considered to be suboptimal and additional treatment planning optimization is recommended.

Compliance Criteria for cohort A: Extremity and Body Wall

Normalization of Dose: The plan is normalized such that 95% of the PTV_5000 volume receives prescription dose of 50 Gy.

Note: Deviation Unacceptable occurs when dose limits for Variation Acceptable are not met

Target Volume Constraints:

Name of Structure	Dosimetric parameter*	Per Protocol	Variation Acceptable
PTV_5000 or PTV_Eval_5000	D95%[Gy]	>=50 Gy	>=49 Gy
	D99%[Gy]	> =46.5 Gy	> =45 Gy
	V55Gy[%]	< =20%	< =25%

Normal Structure Constraints for Primary Tumors at Extremities:

Name of Structure	Dosimetric parameter	Per protocol	Variation Acceptable	Toxicity endpoint
Femur	V50Gy[%]	<50%	<=60%	Fracture
Joint	V50Gy[%]	<50%	<=60%	Joint Stiffness
Skin	V20Gy[%]	<50%	<=60%	Poor wound healing/lymphedema

Normal Structure Constraints for Primary Tumors at Body Wall (Trunk/Chest Wall):

Name of Structure	Required DVH metric	Per protocol cumulative dose	Variation Acceptable	Toxicity endpoint
BrachialPlexus	D0.03cc[Gy]	< =63 Gy	<=65Gy	Brachial Plexopathy
Esophagus	Mean[Gy]	<30 Gy	<=33 Gy	Esophagitis
	D0.03cc[Gy]	<70 Gy	<=74 Gy	
Heart	Mean[Gy]	<26 Gy	<=30 Gy	Pericarditis
	V30Gy[%]	<40%	<=50%	
Lungs	Mean[Gy]	<7 Gy	<=8 Gy	Pneumonitis
	V5Gy[%]	<60%	<=65%	
	V20[%]	<30%	<=35%	
SpinalCord	D0.03cc[Gy]	< =45 Gy	<=48 Gy	Myelopathy

Delivery Compliance Criteria

	Per Protocol	Variation Acceptable
RT Start Date	Concurrent with AMG 232 (KRT-232) chemotherapy	0-1 week
Overall Treatment Time	< 37 days	38-42 days
Non-Medically Indicated Treatment Interruption	0-2 days	3-5 days

Compliance Criteria for cohort B: Abdomen/Pelvis/Retroperitoneum

Normalization of Dose: The plan is normalized such that 95% of the PTV_4500 volume receives prescription dose of 45 Gy.

Note: Deviation Unacceptable occurs when dose limits for Variation Acceptable are not met

Target Volume Constraints:

Name of Structure	Dosimetric parameter*	Per Protocol	Variation Acceptable
PTV_4500 or PTV_Eval_4500	D95%[Gy]	>=45 Gy	>=44 Gy
	D99%[Gy]	>=41.9 Gy	>=40.5 Gy
	V55Gy[%]	<=15%	<=25%
GTV_5000	D95%[Gy]	>=50 Gy	>=49 Gy
	D99%[Gy]	>=46.5 Gy	>=45 Gy
	V55Gy[%]	<=20%	<=25%

Normal Structure Constraints:

Name of Structure	Required DVH metric	Per protocol cumulative dose	Variation Acceptable	Toxicity endpoint
Liver	Mean[Gy]	<30 Gy	<=33 Gy	RILD in normal function liver
R&L Kidneys	Mean[Gy]	<14.4 Gy	<=16 Gy	Renal Dysfunction
	V20Gy[%]	<30%	<=33 %	
Bowel Space	V45 Gy[%]	<20%	<=30 %	Grade 3+ toxicity
Stomach	D0.03cc[Gy] D2%[Gy] D25%[Gy]	<52 Gy <=50 Gy <=45 Gy	<=54 Gy <=54 Gy <=54 Gy	Ulceration
Anus	V30Gy[%]	<50%	<= 60%	Grade 3 toxicity
	V50Gy[%]	<20%	<= 25%	
Bladder	V50Gy[%]	<50%	<= 60%	Grade 3 toxicity
	V70Gy[%]	<20%	<= 25%	
Rectum	V50Gy[%]	<50%	<= 60%	Grade 3 toxicity
	V70Gy[%]	<20%	<= 25%	
Vulva	V30Gy[%]	<50%	<= 60%	Moist Desquamation
Testes (if fertility preservation desired)	V1Gy[%]	<50%	<= 60%	Infertility
Ovaries (if fertility preservation desired)	V5Gy[%]	<50%	<= 60%	Infertility
Spinal Cord	D0.03cc[Gy]	45 Gy	<=48 Gy	Myelopathy

Delivery Compliance Criteria

	Per Protocol	Variation Acceptable
RT Start Date	Concurrent with AMG 232 (KRT-232) chemotherapy	0-1 week
Overall Treatment Time	<37 days	38-42 days
Non-Medically Indicated Treatment Interruptions	0-2 days	3-5 days

5.2.8 Treatment Planning Priorities and Instructions

Every attempt should be made to minimize dose to OAR without compromising coverage of the target coverage and to meet the planning goals listed in the above session. The priorities of structures are listed in the order of decreasing importance and to be used when conflict between tumor coverage and OAR dose presents in the optimization.

Critical structure and target priority for cohort A: Extremity and Body Wall

1. Spinal Cord
2. PTV_Eval_5000 (if applicable)
3. PTV_5000
4. Esophagus
5. Lung
6. Other

Critical structure and target priority for cohort B: Abdomen/Pelvis/Retroperitoneum

1. Spinal Cord
2. PTV_Eval_4500 (if applicable)
3. PTV_4500
4. Liver
5. Rectum
6. Other

Dose calculation algorithm and dose reporting

The tissue heterogeneity correction is mandatory for this protocol. Any algorithm used for this study must be credentialed by IROC Houston. Acceptable choices of algorithm are listed at http://rpc.mdanderson.org/RPC/Services/Anthropomorphic_%20Phantoms/TPS%20-%20algorithm%20list%20updated.pdf

For Convolution/Superposition type algorithms, dose should be reported as computed inherently by the given algorithm. For Monte Carlo or Grid Based Boltzmann Solver algorithms, conversion of Dm (dose-to-medium) to Dw (dose-to-water) should be avoided. Dm, computed inherently by these algorithms, should be reported.

Primary dataset
Calculation must take into account tissue heterogeneity and should be performed with CT-based treatment planning. The primary dataset for dose calculation must be a contrast corrected or non-contrasted free-breathing CT, averaged CT when 4D-CT, or breath-hold CT. Maximum Intensity Pixel (MIP) generated images from 4DCTs may not be used as the primary dose calculation dataset. The relative electron density (or the physical density for Monte Carlo) derived from the Hounsfield Units (HU) of CT images should be utilized for dose calculation.

Dose matrix resolution
Dose grid size should be ≤ 3 mm in all directions.

5.2.9 Patient specific QA
Independent patient specific dose/MU QA is mandatory for all plans. For 3D CRT, an independent secondary dose calculation can be used instead of measurement based QA. Point dose or MU difference between plan calculation and secondary dose calculation should be <5%. For IMRT and VMAT, the plan must be verified prior to first fraction of treatment. Measurements in a QA phantom can be sufficient for a check as long as patient's plan can be directly applied to the phantom geometry. QA can also be done in calculation-based approach, which reconstruct the 3D patient dose via a machine recorded delivery parameter file (Dynalog file) or EPID measured images obtained from a dry run IMRT plan delivery. The recommended patient specific QA criteria is for > 90% of the comparison points to pass a 3%/3mm Gamma analysis.

5.2.10 Daily Treatment Localization/IGRT
Image-guided radiation therapy (IGRT) is radiation therapy using imaging to facilitate accuracy and precision throughout its entire process from target and normal tissue delineation, to radiation delivery, to adaptation of therapy to anatomic and biological changes over time in individual patients. In this section we use the terminology IGRT to focus on image-guidance at the time of radiation delivery to ensure its adherence to the planned treatment.

Image Guidance Procedures
The institution's procedure to register treatment day imaging dataset with the reference dataset should comply with the following recommendations:

- The allowed image modalities are listed in session 5.2.1
- Region-of-Interest (ROI) or "clip box" for image registration should be set to encompass the high dose PTV and adjacent bony anatomy;
- If the image registration software allows the user to create an irregular ROI (e.g.,ExacTrac), treatment room objects (e.g., patient support system structure) seen on in-room X-rays should be excluded from the registration;
- Both manual (e.g., based on bony anatomy) and automatic (e.g., based on mutual information) types of registration can be used; the result of the image registration must be visually checked for the alignment of the bony anatomy.
- Institutions are encouraged to include joints in the imaging process in order to increase the information used in the image registration process.
- When orthogonal kV imaging is employed for sites where respiratory motion is expected and not controlled via motion management techniques, care must be taken to ensure

accurate targeting of the ITV within the treatment

- Daily pretreatment images for each case should be archived at each treating institution and made available for central review by NRG Oncology upon request.
- IGRT requirements and tolerance

Location	No Fiducials	With Fiducials**	Tolerance Level
Extremity	Orthogonal 2D	Orthogonal 2D	Residual error after shift -Acceptable: ≤ 3mm -Minor variance: 3-5 mm -Unacceptable: > 5 mm
Lung*	Volumetric 3D	Orthogonal 2D	
Liver*	Volumetric 3D	Orthogonal 2D	
Abdominal-pelvic*	Volumetric 3D	Orthogonal 2D	

*Registration using a soft tissue surrogate for the tumor is recommended.
**Clearly visible anatomical markers are acceptable as fiducials, e.g. inserted radio-opaque markers or Lipiodol from prior TACE treatment.

5.2.11 Case Review

The Principal Investigators, Meng Xu Welliver, MD, PhD, and Dian Wang, MD, PhD, will perform RT Quality Assurance Review after cases enrolled have been received at IROC Philadelphia-RT. The scoring mechanism is: **Per Protocol, Variation Acceptable, Deviation Unacceptable.**

Note: Pre-treatment reviews will be mandatory for Cohort B only - the first case from each institution will require a pre-treatment review. The patient cannot begin treatment until complete data from the first case is received, reviewed and approved. **Sites need to allow 3 business days for the pre-treatment review, and if necessary, 3 additional business days if a revision and re-plan is requested by the Reviewer.** The goal of the review is to evaluate protocol compliance.

5.3 Imaging Requirement for Primary Tumor Site (28-JAN-2020)

5.3.1 For STS located at extremities or body wall (cohort A), MRI with and without contrast or CT with and without contrast is required at baseline, 4 weeks after neoadjuvant AMG 232 (KRT-232)/RT treatment prior to surgery and after surgery.

- If possible, try to use the same imaging facility for the below 4 scans;
- Slice thickness and matrix per institutional norms. Recommend maximum slice thickness of 4 mm MRI sequences should include: (1) Axial (short axis) T1 and T2 (fat saturation encouraged) or STIR imaging; (2) Coronal (long axis) T1 and T2 fat saturated (or STIR) imaging; (3) Axial (short axis) post contrast T1 fat saturated and either coronal or sagittal T1 fat saturated imaging. Fat saturation required; (4) if able axial diffusion imaging with b50 and b800 with ADC map; use same FOV and slice thickness and above axial scans if possible
- Local extremity coils should be used for distal appendicular imaging (elbows through wrist/hand), knees through ankle/foot. Shoulder coil for upper arm imaging encouraged if entire tumor can be encompassed. Otherwise, torso array coil recommended.

5.3.2 For STS located at abdomen/pelvis/retroperitoneum (cohort B), MRI with and without contrast or CT with and without contrast is required at baseline, 4 weeks after neoadjuvant AMG 232 (KRT-232)/RT treatment prior to surgery and after surgery

- CT imaging should use slice thickness of 5 mm or less.

- T1 and T2 axial imaging should include slice thickness of 5mm. Post gadolinium T1 axial imaging should include fat saturation. Remainder of MR imaging should be per local institutional norms based on anatomic region of the tumor.

5.3.3 Tumor volume measurements: Primary tumor size should be included in the radiology report. Tumor volume should be estimated utilizing ellipsoid approximation (L * W * D * 0.52). This estimate will be made by site radiologists and reviewed/audited by Dr. Alan Rogers.

5.3.4 **MRI or CT images for the primary tumor at baseline and 4 weeks after neoadjuvant treatment (prior to surgery) and at first local progression should be submitted for central review. Other imaging studies are not required for central review.**

5.4 **Surgery** (9-OCT-2018)

5.4.1 Planned surgical resection must occur within week 5-8 after completion of radiation therapy.

5.4.2 Initial Biopsy for Diagnosis and Tissue Submission.

For extremity or truncal tumors (cohort A), core needle biopsy or an incisional biopsy are preferred. Fine needle aspiration (FNA) biopsy is not acceptable to establish the diagnosis. A minimum of 6-8 core needle biopsies are recommended. Care must be taken to prevent penetration of the needle through the tumor into unaffected soft tissues. If the grade and subtype of the tumor cannot be determined from the core biopsy, then an incisional biopsy is required. Either biopsy technique should be done in such a way as to permit excision of the biopsy site at the time of definitive wide resection. Based upon the staging imaging, the tumor should be sampled in a manner to allow for adequate pathologic assessment of heterogeneous areas.

For retroperitoneal/pelvic/peritoneal tumors (cohort B), image guided core needle biopsy is the preferred technique. Fine needle aspiration biopsy is not acceptable to establish the diagnosis. Surgical incisional biopsy should be avoided unless absolutely necessary to make the diagnosis to reduce the risk for peritoneal seeding/sarcomatosis. In that situation, surgically guided core needle biopsies would be preferred. A minimum of 6-8 core needle biopsies are recommended. Based upon the staging imaging, the tumor should be sampled in a manner to allow for adequate pathologic assessment of heterogeneous areas.

5.4.3 It is strongly recommended that any eligible patient is seen by the surgeon, medical oncologist and radiation oncologist prior to instituting pre-operative therapy. The surgeon should determine and document a high possibility of both primary tumor resection and anticipation of a limb preservation approach (if applicable for cohort A) after preoperative radiation. For cohort A, every effort should be made to perform limb preservation surgery after preoperative radiation for extremity tumors unless there is documented evidence of tumor progression during or after the course of radiation that would require amputation for an appropriate negative margin resection. At the discretion of the treating physician(s), plastic and/or vascular surgeons may be consulted.

- Upon removal, the tumor specimen should be immediately submitted to the pathologist. It should not be bisected or cut into pieces prior to pathologic evaluation. If there will be

any delay in transferring the specimen to the pathologist, it should be transported in cytogenetics medium. The surgeon should document both the time of tumor removal from the patient as well as the time that the specimen was sent to pathology.

- The resection should be done with the goal of trying to achieve negative pathologic margins. Quality assurance for surgical resection will be performed by both a review of the operative note and assessment of the specimen by surgical pathology. A negative margin (R0) resection will be defined as microscopic absence of tumor on the inked margins.

5.4.4 For protocol purposes, the adequacy of surgical resection will be determined after an assessment of the formal operative note by one of the study surgical co-chairs and pathologic evaluation of the resected specimen. The definitions include:

- Amputation: Margin status will still be assessed and categorized if a limb preservation approach was not possible after the preoperative radiation.
- Limb sparing surgery with the following margin status:
    - R0: No residual tumor and microscopically negative margins;
    - R1: Microscopic positive margin(s) but no gross tumor;
    - R2: Macroscopic gross/residual tumor. It is strongly encouraged that any patient with an R2 margin status should be assessed for surgical re-excision, to try to achieve an R0 or R1 margin.

5.4.5 Definitive Surgical Procedure

The surgical procedure necessary to resect the tumor with negative margins should be used. The definitions, as noted above, will be recorded in the surgical form. Boost radiotherapy will not be allowed in this protocol. Therefore all patients who underwent an R2 resection should be encouraged to undergo re-resection to obtain R0/R1 margins.

- For cohort A, any incisional biopsy site should be excised en bloc with the definitive surgical specimen. Core needle biopsy tracts should also be excised unless doing so would result in excessive morbidity.
- Resectability will depend upon the judgment of the operating surgeon. The goal of all surgery for extremity tumors should be limb preservation, if possible, within the realm of an appropriate oncologic resection. Every effort should be made to perform limb preservation surgery. However, some extremity tumors may require amputation to obtain even grossly negative margins. Only amputation due to treatment complications (not primary intended surgical therapy) will be considered a surgical complication.
- Dissection should always be done through grossly normal tissue planes and ideally should be done beyond the fascial plane adjacent to the tumor. If the tumor is close to or displaces major vessels or nerves, these need not be removed if the tissues immediately adjacent to the structures are removed and the underlying neurovascular structures are not involved with gross tumor. Dissection should include the periosteum if the tumor is adjacent to the bone but not invading. Radical excision or entire anatomic compartment resection is not recommended for patients on this study.
- In general, lymph node dissection is not recommended, but primary tumors overlying major lymph node stations may be treated with surgical resection to include the adjacent lymph nodes.
- To facilitate the assessment of surgical margins, the surgeon should take care to manipulate the specimen as little as possible to preserve anatomic relationships present at

the time of resection; to mark and orient the margins for the pathologist; and to avoid bisecting, bivalving, or cutting the specimen into separate specimens.

- Secondary to the preoperative radiation, closed suction drainage should be strongly considered for all postoperative dead spaces. The drains should exit the skin close to the edge of the surgical incision and ideally in line with the incision.
- As all patients will have received preoperative radiation, special attention must be given to the skin flaps. Use of muscle flaps, pedicled myocutaneous flaps, and even free flaps are encouraged to fill dead space and provide well-vascularized tissue. These flaps should be used if there is any concern regarding the viability of the skin flaps.
- It is strongly encouraged that the surgeon clearly state in the operative note what type of surgical resection was intended (R0, R1, or R2) and from where any frozen sections of the margins were taken, if applicable. The final margin status (R status) should be based on the permanent pathology assessment (not frozen section).
- The surgeon may state in the operative note that the intent of the procedure was wide resection to obtain a negative oncologic margin. However, the term "radical" should not be used to describe the procedure unless the intent of the surgery was truly to do a complete extracompartmental resection.
- In general, the following principles should be followed in postoperative management of these patients: Maintain staples or skin sutures per surgeon preference, recognizing that all patients will have received preoperative radiation. Leave drains in place until the drainage meets the surgeon's criteria for removal. Begin rehabilitation per surgeon's discretion.
- If the postoperative pathologic evaluation reveals positive soft tissue margins other than bone (periosteum), nerve or major blood vessels, surgical re-resection to obtain negative margins should strongly be considered if it will not have a major impact upon the patient's functionality. If the margin on bone, major blood vessel or nerve is microscopically positive, additional radiation boost will not be used in this protocol.

5.4.6 Surgical Adverse Events

- Wound Complications. Major wound complications (eg, secondary operations, re-admissions, anastomotic dehiscence, and/or invasive procedures for wound complications: deep wound packing and prolonged dressing changes) will be reported using the relevant CTCAE version 5.0 criteria.

5.4.7 Surgical Quality Assurance Reviews. The Surgical Oncology Co-Chairs from NRG Oncology will perform a Quality Assurance Review on a rolling basis once data is received at the Statistics and Data Management Center (SDMC). The final cases will be reviewed within 3 months after this study has reached the target accrual or as soon as complete data for all cases enrolled has been received at NRG SDMC, whichever occurs first.

- Surgical quality assurance review will include examination of the formal operative report for the surgical resection, the treating institution's final pathology report (including gross, frozen section, and microscopic pathologic descriptions), and the central pathology review. The reported resection (R) status from the operative note will be assessed and re-evaluated based upon the final pathology report and the central pathology review. The surgical quality assurance review will then assign an official protocol resection status based upon this information. From a clinical treatment and protocol standpoint, an R0 or

R1 resection (especially following an attempt to preserve a major neurovascular, visceral, or bony structure) would be considered an adequate surgical resection.

- Although every effort should be made to perform limb preservation surgery, some extremity tumors may require amputation to obtain negative margins if a limb preservation approach is not possible after preoperative radiation. In this scenario, amputation would still be considered adequate surgical therapy and margin status will be assessed or categorized based upon the R criteria for limb sparing surgery.

5.5 General Concomitant Medication and Supportive Care Guidelines

5.5.1 Permitted Supportive/Ancillary Care and Concomitant Medications

All supportive therapy for optimal medical care can be given during the study period at the discretion of the attending physician(s). Concomitant medications must be reported on each site's source document, and these concomitant drugs are not limited to:

- Anticonvulsants
- Antiemetics
- Anticoagulants
- Antidiarrheals
- Analgesics
- Hematopoietic Growth Factors
- Nutritional supplementation
- Highly active antiretroviral therapy (HAART)

5.5.2 Prohibited Therapies

- [redacted]
- Medications known to cause QTc interval prolongation (see https://www.crediblemeds.org/research-scientists/why-lists/)

5.5.3 Participation in other trials is not allowed.

5.6 Duration of Therapy

In the absence of treatment delays due to adverse event(s), treatment may continue as specified in the above treatment modality sections or until one of the following criteria applies:

- Disease progression,
- Intercurrent illness that prevents further administration of treatment,
- Unacceptable adverse event(s), as described in Section 6
- Patient decides to withdraw consent for participation in the study, or
- General or specific changes in the patient's condition render the patient unacceptable for further treatment in the judgment of the investigator
- Once patients have undergone surgery or have completed the planned RT but are no longer candidates for surgery.

6. TREATMENT MODIFICATIONS/MANAGEMENT (29-MAY-2019)

DLT includes all grade 4-5 toxicities attributable to AMG 232 (KRT-232) or combined AMG 232 (KRT-232) with radiotherapy up to 4 weeks after the completion of AMG 232 (KRT-232)+RT treatment. Any grade 3 AE at least possibly attributable to AMG 232 (KRT-232) or combined AMG 232 (KRT-232) with radiotherapy will also be considered DLT if any of the two following situations occur: a delay of >2 weeks due to the grade 3 toxicity, or ≥ two dose reductions due to the grade 3 toxicity.

For all toxicity management:

- All toxicities are per NCI Common Terminology Criteria for Adverse Events (CTCAE) version 5.0.
- Patients requiring a delay of >2 weeks must discontinue AMG 232 (KRT-232).
- Patients requiring > two dose frequency reductions must discontinue AMG 232 (KRT-232).
- Toxicities include nausea, vomiting, diarrhea, febrile neutropenia, thrombocytopenia or anemia.

Table 6A. AMG 232 (KRT-232) "Dose" Reduction by Changing Frequency
AMG 232 (KRT-232) will be dose reduced per the toxicity tables detailed below

Dose Level	Dose (Weekly Total Dose mg)	First Dose Frequency Reduction by 1 dose/week (Weekly Total Dose mg)	Second Dose Frequency Reduction by 2 doses/ week (Weekly Total Dose mg)
-1	120 mg (240 mg) (Days 2 and 4 only)	120 mg (120mg) (Day 2 only)	Discontinue AMG 232 (KRT-232) (Off study)
1	120 mg (360 mg) (Days 2, 3, and 4 only)	120 mg (240 mg) (Days 2 and 4 only)	120 mg (120mg) (Day 2 only)
2	120 mg (480 mg) (Days 2, 3, 4, and 5 only)	120 mg (360 mg) (Days 2, 3, and 4 only)	120 mg (240 mg) Days 2 and 4 only
3	120 mg (600 mg) (Days 1, 2, 3, 4 and 5 only)	120 mg (480 mg) (Days 2, 3, 4, and 5 only)	120 mg (360 mg) (Days 2, 3, and 4 only)

Nausea	**Management/Next Dose for AMG 232 (KRT-232)**	**Management/Next Dose for *Radiotherapy***
≤ Grade 1	No change in dose	Continue treatment
Grade 2	No change in dose with appropriate antiemetics and best supportive care.	Continue treatment
Grade 3	Hold* until ≤ Grade 2. Resume at next	Hold* until ≤ Grade 2.

Nausea	**Management/Next Dose for AMG 232 (KRT-232)**	**Management/Next Dose for *Radiotherapy***
	lower dose level according to table 6A above with appropriate antiemetics and best supportive care.	
Grade 4	Off protocol therapy	
Recommended management: * antiemetics.		

Vomiting	**Management/Next Dose for AMG 232 (KRT-232)**	**Management/Next Dose for *Radiotherapy***
≤ Grade 1	No change in dose	Continue treatment
Grade 2	No change in dose with appropriate antiemetics and best supportive care	Continue treatment
Grade 3	Hold until ≤ Grade 2. Resume at next lower dose level according to table 6A above with appropriate antiemetics and best supportive care.	Hold until ≤ Grade 2.
Grade 4	Off protocol therapy	Hold until ≤ Grade 2.
Recommended management: antiemetics.		

Diarrhea	**Management/Next Dose for AMG 232 (KRT-232)**	**Management/Next Dose for *Radiotherapy***
≤ Grade 1	No change in dose	Continue treatment
Grade 2	No change in dose with appropriate medical management below and best supportive care	Continue treatment
Grade 3	Hold until < Grade 2. Resume at next lower dose level according to table 6A above with appropriate medical management below and best supportive care.	Hold until ≤ Grade 2.
Grade 4	Off protocol therapy	Hold until ≤ Grade 2.
Recommended management: Loperamide antidiarrheal therapy Dosage schedule: [redacted] at first onset, followed by [redacted] with each loose motion until diarrhea-free for 12 hours (maximum dosage: [redacted] Adjunct anti-diarrheal therapy is permitted and should be recorded when used.		

Febrile Neutropenia	**Management/Next Dose for AMG 232 (KRT-232)**	**Management/Next Dose for *Radiotherapy***
≤ Grade 1	No change in dose	Continue treatment
Grade 2	No change in dose	Continue treatment
Grade 3	Hold until < Grade 3. Resume at next lower dose level according to table 6A above.	Hold until < Grade 3.
Grade 4	Off protocol therapy	Hold until < Grade 3

Thrombocytopenia	**Management/Next Dose for AMG 232 (KRT-232)**	**Management/Next Dose for *Radiotherapy***
≤ Grade 1	No change in dose	Continue treatment
Grade 2	Hold until ≤ Grade 1. Resume at same dose level.	Continue treatment
Grade 3	Hold until < Grade 3. Resume at next lower dose level according to table 6A above with best supportive care.	Hold until <Grade 3.
Grade 4	Off protocol therapy	Hold until < Grade 3

Anemia	**Management/Next Dose for AMG 232 (KRT-232)**
≤ Grade 1	No change in dose
Grade 2	No change in dose
Grade 3	Hold until ≤ Grade 2. Resume at next lower dose level according to table 6A above with best supportive care.
Grade 4	Off protocol therapy

Other toxicities related to AMG 232 (KRT-232)	**Management/Next Dose for AMG 232 (KRT-232)**
≤ Grade 1	No change in dose
Grade 2	No change in dose
Grade 3	Hold until ≤ Grade 2. Resume at next lower dose level according to table 6A above with best supportive care.
Grade 4	Off protocol therapy

6.1 Hepatotoxicity Stopping and Re-challenge Rules

Subjects with abnormal hepatic laboratory values (*e.g.*, ALP, AST, ALT, total bilirubin, or INR) or signs/symptoms of hepatitis may meet the criteria for withholding of investigational product. Withholding is either permanent or conditional depending upon the clinical circumstances discussed below (as specified in the Guidance for Industry Drug-Induced Liver Injury: Premarketing Clinical Evaluation, July 2009).

6.1.1 Criteria for Permanent Withholding of AMG 232 (KRT-232) Due to Potential Hepatotoxicity

AMG 232 (KRT-232) should be permanently withheld and the subject should be followed according to the recommendations in if ALL of the criteria below are met:

- Increased AST or ALT from the relevant baseline value as specified below:
    - Baseline AST/ALT level is < 1 × ULN and AST/ALT is elevated > 3 × ULN
      AND
    - Total bilirubin level is > 2 × ULN or INR is > 1.5
      AND
    - No other cause for the combination of the above laboratory abnormalities is immediately apparent; important alternative causes for elevated AST/ALT and/or elevated total bilirubin values include, but are not limited to:
        - Hepatobiliary tract disease
        - Viral hepatitis (*e.g.*, hepatitis A/B/C/D/E, Epstein-Barr virus (EBV), cytomegalovirus (CMV), herpes simplex virus (HSV), varicella, toxoplasmosis, and parvovirus)
        - Right sided heart failure, hypotension or any cause of hypoxia to the liver causing ischemia
        - Exposure to hepatotoxic agents/drugs including herbal and dietary supplements, plants, and mushrooms,
        - Heritable disorders causing impaired glucuronidation (*e.g.*, Gilbert's syndrome, Crigler-Najjar syndrome) and drugs that inhibit bilirubin glucuronidation (*e.g.*, indinavir and atazanavir)
        - Alpha-one antitrypsin deficiency
        - Alcoholic hepatitis
        - Autoimmune hepatitis
        - Wilson's disease and hemochromatosis
        - Nonalcoholic steatohepatitis (NASH) or other "fatty liver disease"
        - Non-hepatic causes (*e.g.*, rhabdomyolysis and hemolysis).

6.1.2 Criteria for Conditional Withholding of AMG 232 (KRT-232) due to Potential Hepatotoxicity

For subjects who do not meet the criteria for permanent withholding of AMG 232 (KRT-232) outlined above and with no underlying liver disease and eligibility criteria requiring normal transaminases and total bilirubin level at baseline or subjects with underlying liver disease and baseline abnormal transaminases, the following rules are recommended for conditional withholding of AMG 232 (KRT-232) and other protocol-required therapies:

- Elevation of either AST/ALT according to the following schedule:
    - AST/ALT baseline value is any AND AST/ALT is elevated to any of the following values:
        - >8 × ULN at any time
        - >5 × ULN but <8 × ULN for ≥ 2 weeks
        - >5 × ULN but < 8 × ULN and unable to adhere to enhanced monitoring schedule > 3 × ULN with clinical signs or symptoms that are consistent with hepatitis (such as right upper quadrant pain/tenderness, fever, nausea, vomiting, jaundice, rash, or eosinophilia (> 5%).

        OR
    - Total bilirubin is > 3 × ULN at any time

        OR
    - Alkaline phosphatase is > 8 × ULN at any time

AMG 232 (KRT-232) should be withheld pending investigation into alternative causes of drug-induced liver injury. If AMG 232 (KRT-232) is withheld, the subject should be followed according to recommendations for possible drug-induced liver injury in Appendix V. Rechallenge may be considered if an alternative cause for impaired liver tests (*i.e.*, ALT, AST, alkaline phosphatase) and/or elevated total bilirubin level is discovered and the laboratory abnormalities resolve to normal or baseline.

7. ADVERSE EVENTS REPORTING REQUIREMENTS

7.1 Protocol Agents

Investigational Agent

The investigational agent administered in NRG-DT001, AMG 232 (KRT-232), is being made available under an IND sponsored by CTEP.

7.2 Adverse Events and Serious Adverse Events (9-OCT-2018)

7.2.1 This study will utilize the NCI Common Terminology Criteria for Adverse Events (CTCAE) version 5.0 for CTEP-AERS (CTEP Adverse Event Reporting System) CAERs reporting of adverse events (AEs), located on the CTEP web site, http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm. All appropriate treatment areas should have access to a copy of the CTCAE version 5.0.

7.2.2 Definition of an Adverse Event (AE)

Any untoward medical occurrence associated with the use of a drug in humans, whether or not considered drug related. Therefore, an AE can be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal (investigational) product, whether or not considered related to the medicinal (investigational) product (attribution of unrelated, unlikely, possible, probable, or definite). (International Conference on Harmonisation [ICH], E2A, E6).

For multi-modality trials, adverse event reporting encompasses all aspects of protocol treatment including radiation therapy, surgery, device, and drug.

For AMG 232 (KRT-232) determination of whether an adverse event meets expedited reporting criteria, see the reporting table in section 7.4 of the protocol.

Due to the risk of intrauterine exposure of a fetus to potentially teratogenic agents, the pregnancy of a study participant must be reported via CTEP-AERS in an expedited manner.

7.3 Comprehensive Adverse Events and Potential Risks list (CAEPR) for KRT-232 (AMG 232, NSC 789723) (22-FEB-2021)

The Comprehensive Adverse Events and Potential Risks list (CAEPR) provides a single list of reported and/or potential adverse events (AE) associated with an agent using a uniform presentation of events by body system. In addition to the comprehensive list, a subset, the Specific Protocol Exceptions to Expedited Reporting (SPEER), appears in a separate column and is identified with bold and italicized text. This subset of AEs (SPEER) is a list of events that are protocol specific exceptions to expedited reporting to NCI (except as noted below). Refer to the 'CTEP, NCI Guidelines: Adverse Event Reporting Requirements' http://ctep.cancer.gov/protocolDevelopment/electronic_applications/docs/aeguidelines.pdf for further clarification. *Frequency is provided based on 247 patients.* Below is the CAEPR for KRT-232 (AMG 232).

NOTE: Report AEs on the SPEER **ONLY IF** they exceed the grade noted in parentheses next to the AE in the SPEER. If this CAEPR is part of a combination protocol using multiple investigational agents and has an AE listed on different SPEERs, use the lower of the grades to determine if expedited reporting is required.

Version 2.3, December 16, 2020[1]

Adverse Events with Possible Relationship to KRT-232 (AMG 232) (CTCAE 5.0 Term) [n= 247]			Specific Protocol Exceptions to Expedited Reporting (SPEER)
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
BLOOD AND LYMPHATIC SYSTEM DISORDERS			
Anemia			***Anemia (Gr 2)***
GASTROINTESTINAL DISORDERS			
	Abdominal pain		***Abdominal pain (Gr 2)***
	Constipation		
Diarrhea			***Diarrhea (Gr 2)***
Nausea			***Nausea (Gr 2)***
Vomiting			***Vomiting (Gr 2)***
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS			
	Edema limbs		
Fatigue			***Fatigue (Gr 2)***
INVESTIGATIONS			
	Neutrophil count decreased		***Neutrophil count decreased (Gr 2)***
Platelet count decreased			***Platelet count decreased (Gr 2)***
METABOLISM AND NUTRITION DISORDERS			
	Anorexia		***Anorexia (Gr 2)***
MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS			
	Myalgia		
NERVOUS SYSTEM DISORDERS			
	Dysgeusia		***Dysgeusia (Gr 2)***

[1]This table will be updated as the toxicity profile of the agent is revised. Updates will be distributed to all Principal Investigators at the time of revision. The current version can be obtained by contacting PIO@CTEP.NCI.NIH.GOV. Your name, the name of the investigator, the protocol and the agent should be included in the e-mail.

Adverse events reported on KRT-232 (AMG 232) trials, but for which there is insufficient evidence to suggest that there was a reasonable possibility that KRT-232 (AMG 232) caused the adverse event:

BLOOD AND LYMPHATIC SYSTEM DISORDERS - Blood and lymphatic system disorders - Other (pancytopenia); Blood and lymphatic system disorders - Other (hematocrit increased); Febrile neutropenia; Leukocytosis
CARDIAC DISORDERS - Palpitations; Sinus tachycardia
GASTROINTESTINAL DISORDERS - Ascites; Duodenal ulcer; Gastrointestinal disorders - Other (feces discolored); Gastrointestinal disorders - Other (duodenitis); Upper gastrointestinal hemorrhage
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS - Edema face; Fever; General disorders and administration site conditions - Other (general physical health deterioration)
INFECTIONS AND INFESTATIONS - Infections and infestations - Other (gastroenteritis); Lung infection; Sepsis; Sinusitis; Upper respiratory infection; Urinary tract infection
INVESTIGATIONS - Activated partial thromboplastin time prolonged; Alanine aminotransferase increased; Alkaline phosphatase increased; Blood bilirubin increased; Blood lactate dehydrogenase increased; CPK increased; Creatinine increased; INR increased; Investigations - Other (blood bicarbonate increased); Investigations - Other (blood creatinine decreased); Investigations - Other (blood urea increased); Investigations - Other (pH urine increased); Investigations - Other (platelet count increased); Investigations - Other (reticulocyte count increased); Lipase increased; Lymphocyte count decreased; Serum amylase increased; Weight loss; White blood cell decreased
METABOLISM AND NUTRITION DISORDERS - Dehydration; Hyperglycemia; Hyperkalemia; Hyperuricemia; Hypokalemia; Hypomagnesemia; Hyponatremia; Hypophosphatemia

MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS - Arthritis; Back pain; Bone pain; Joint range of motion decreased; Pain in extremity
NEOPLASMS BENIGN, MALIGNANT AND UNSPECIFIED (INCL CYSTS AND POLYPS) - Myelodysplastic syndrome; Tumor pain
NERVOUS SYSTEM DISORDERS - Anosmia; Dizziness; Headache; Nervous system disorders - Other (hemorragic stroke); Somnolence; Syncope
RENAL AND URINARY DISORDERS - Acute kidney injury; Dysuria; Proteinuria
REPRODUCTIVE SYSTEM AND BREAST DISORDERS - Pelvic pain
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS - Cough; Dyspnea; Epistaxis; Hypoxia; Respiratory failure
SKIN AND SUBCUTANEOUS TISSUE DISORDERS - Alopecia; Hyperhidrosis; Pruritus
VASCULAR DISORDERS - Flushing; Hypertension

Note: KRT-232 (AMG 232) in combination with other agents could cause an exacerbation of any adverse event currently known to be caused by the other agent, or the combination may result in events never previously associated with either agent.

7.4 Expedited Reporting of Adverse Events (28-JAN-2020)

All adverse events (AEs) are submitted for expedited reporting protocol-specific rules evaluation using the Medidata Rave data management system. All AEs will be evaluated by the Cancer Therapy Evaluation Program Adverse Event Reporting System (CTEP-AERS) to determine whether expedited reporting is recommended based on a set of programmed expedited reporting rules. AEs identified as meeting the programmed expedited reporting requirements can then be submitted in CTEP-AERS. A deep link in Rave will take the user directly to CTEP-AERS where the expedited report may be completed and submitted via CTEP-AERS.

All serious adverse events that meet expedited reporting criteria defined in the reporting table below will be reported via the CTEP Adverse Event Reporting System, CTEP-AERS, accessed via the link in RAVE. CTEP-AERS is also accessed via the CTEP web site, **but all expedited reports must be initiated in RAVE** https://eapps-ctep.nci.nih.gov/ctepaers/pages/task?rand=1390853489613.

See Section 13.3 for additional details.

Submitting a report via CTEP-AERS satisfies NRG requirements for expedited adverse event reporting.

In the rare event when Internet connectivity is disrupted, a 24-hour notification must be made to CTEP by telephone at 301-897-8497 and to the NRG Oncology by phone at 1-215-574-3191**.** An electronic report must be submitted immediately upon re-establishment of the Internet connection.

7.4.1 Expedited Reporting Methods

- Per CTEP NCI Guidelines for Adverse Events Reporting, a CTEP-AERS 24-hour notification must be submitted within 24 hours of learning of the adverse event. Each CTEP-AERS 24-hour notification must be followed by a complete report within 5 days.

- Supporting source documentation is requested by NRG as needed to complete adverse event review. Supporting source documentation should include the protocol number, patient ID number, and CTEP-AERS ticket number on each page, and contact NRG Oncology at 1-215-574-3191 for instructions to submit source documents. Supporting source documentation should also be faxed to CTEP at 301-230-0159.
- A serious adverse event that meets expedited reporting criteria outlined in the AE Reporting Tables but is assessed by the CTEP-AERS as "an action *not* recommended" must still be reported to fulfill NRG safety reporting obligations. Sites must bypass the "NOT recommended" assessment; the CTEP-AERS allows submission of all reports regardless of the results of the assessment.

7.4.2 Expedited Reporting Requirements for Adverse Events

Phase 1 and Early Phase 2 Studies: Expedited Reporting Requirements for Adverse Events that Occur on Studies under an IND/IDE within 30 Days of the Last Administration of the Investigational Agent/Intervention [1, 2]

FDA REPORTING REQUIREMENTS FOR SERIOUS ADVERSE EVENTS (21 CFR Part 312)

NOTE: Investigators **MUST** immediately report to the sponsor (NCI) **ANY** Serious Adverse Events, whether or not they are considered related to the investigational agent(s)/intervention (21 CFR 312.64)

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

1) Death
2) A life-threatening adverse event
3) An adverse event that results in inpatient hospitalization or prolongation of existing hospitalization for ≥ 24 hours
4) A persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
5) A congenital anomaly/birth defect.
6) 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 subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition. (FDA, 21 CFR 312.32; ICH E2A and ICH E6).

ALL SERIOUS adverse events that meet the above criteria MUST be immediately reported to the NCI via CTEP-AERS within the timeframes detailed in the table below.

Hospitalization	Grade 1 and Grade 2 Timeframes	Grade 3-5 Timeframes
Resulting in Hospitalization ≥ 24 hrs	10 Calendar Days	24-Hour 5 Calendar Days

Not resulting in Hospitalization ≥ 24 hrs	Not required	

NOTE: Protocol specific exceptions to expedited reporting of serious adverse events are found in the Specific Protocol Exceptions to Expedited Reporting (SPEER) portion of the CAEPR.

Expedited AE reporting timelines are defined as:

- "24-Hour; 5 Calendar Days" - The AE must initially be reported via CTEP-AERS within 24 hours of learning of the AE, followed by a complete expedited report within 5 calendar days of the initial 24-hour report.
- "10 Calendar Days" - A complete expedited report on the AE must be submitted within 10 calendar days of learning of the AE.

[1]Serious adverse events that occur more than 30 days after the last administration of investigational agent/intervention and have an attribution of possible, probable, or definite require reporting as follows:

Expedited 24-hour notification followed by complete report within 5 calendar days for:

- All Grade 3, 4, and Grade 5 AEs

Expedited 10 calendar day reports for:

- Grade 2 AEs resulting in hospitalization or prolongation of hospitalization

[2]For studies using PET or SPECT IND agents, the AE reporting period is limited to 10 radioactive half-lives, rounded UP to the nearest whole day, after the agent/intervention was last administered. Footnote "1" above applies after this reporting period.

Effective Date: May 5, 2011

Additional Protocol-Specific Instructions or Exceptions to Expedited Reporting Requirements:

To facilitate appropriate monitoring for signals of drug-induced liver injury (DILI), cases of concurrent AST or ALT and total bilirubin and/or INR elevation according to the criteria specified in Section 6.1 require a 24 hour notification/5 day report. See Appendix V for additional requirements.

7.4.3 Reporting to the Site IRB/REB

Investigators will report serious adverse events to the local Institutional Review Board (IRB) or Research Ethics Board (REB) responsible for oversight of the patient according to institutional policy.

7.4.4 Secondary Malignancy

A secondary malignancy is a cancer caused by treatment for a previous malignancy (e.g., treatment with investigational agent/intervention, radiation or chemotherapy). A secondary malignancy is not considered a metastasis of the initial neoplasm.

CTEP requires all secondary malignancies that occur during or subsequent to treatment with an agent under an NCI IND/IDE be reported via CTEP-AERS. In addition, secondary malignancies following radiation therapy must be reported via CTEP-AERS. Three options are available to describe the event:

- Leukemia secondary to oncology chemotherapy (e.g., acute myelocytic leukemia [AML])
- Myelodysplastic syndrome (MDS)
- Treatment-related secondary malignancy

Any malignancy possibly related to cancer treatment (including AML/MDS) should also be reported via the routine reporting mechanisms outlined in each protocol.

Second Malignancy:
A second malignancy is one unrelated to the treatment of a prior malignancy (and is NOT a metastasis from the initial malignancy). Second malignancies require ONLY routine AE reporting unless otherwise specified.

8. REGISTRATION AND STUDY ENTRY PROCEDURES

8.1 Food and Drug Administration (FDA) Regulations and National Cancer Institute (NCI) Policy (22-FEB-2021)

Food and Drug Administration (FDA) regulations and National Cancer Institute (NCI) policy require all individuals contributing to NCI-sponsored trials to register and renew their registration annually. To register, all individuals must obtain a Cancer Therapy Evaluation Program (CTEP) Identity and Access Management (IAM) account (https://ctepcore.nci.nih.gov/iam). In addition, persons with a registration type of Investigator (IVR), Non-Physician Investigator (NPIVR), or Associate Plus (AP) must complete their annual registration using CTEP's web-based Registration and Credential Repository (RCR) at https://ctepcore.nci.nih.gov/rcr.

RCR utilizes five person registration types.

- IVR — MD, DO, or international equivalent;
- NPIVR — advanced practice providers (e.g., NP or PA) or graduate level researchers (e.g., PhD);
- AP — clinical site staff (e.g., RN or CRA) with data entry access to CTSU applications, such as the Roster Update Management System [RUMS], OPEN, Rave, acting as a primary site contact, or with consenting privileges;
- Associate (A) — other clinical site staff involved in the conduct of NCI-sponsored trials; and
- Associate Basic (AB) — individuals (e.g., pharmaceutical company employees) with limited access to NCI-supported systems.

RCR requires the following registration documents:

Documentation Required	IVR	NPIVR	AP	A	AB
FDA Form 1572	✔	✔			
Financial Disclosure Form	✔	✔	✔		
NCI Biosketch (education, training, employment, license, and certification)	✔	✔	✔		
GCP training	✔	✔	✔		
Agent Shipment Form (if applicable)	✔				
CV (optional)	✔	✔	✔		

An active CTEP-IAM user account and appropriate RCR registration is required to access all CTEP and CTSU (Cancer Trials Support Unit) websites and applications. In addition, IVRs and NPIVRs must list all clinical practice sites and IRBs covering their practice sites on the FDA Form 1572 in RCR to allow the following:

- Addition to a site roster
- Assign the treating, credit, consenting, or drug shipment (IVR only) tasks in OPEN
- Act as the site-protocol Principal Investigator (PI) on the IRB approval

In addition, all investigators acting as the Site-Protocol PI (investigator listed on the IRB approval) or consenting/treating/drug shipment investigator in OPEN must be rostered at the enrolling site with a participating organization.

Additional information can be found on the CTEP website at < https://ctep.cancer.gov/investigatorResources/default.htm >. For questions, please contact the **RCR Help Desk** by email at < RCRHelpDesk@nih.gov >.

8.2 CTSU Registration Requirements (22-FEB-2021)

This study is supported by the NCI Cancer Trials Support Unit (CTSU).

IRB Approval

For CTEP and Division of Cancer Prevention (DCP) studies open to the National Clinical Trials Network (NCTN) and NCI Community Oncology Research Program (NCORP) Research Bases after March 1, 2019, all U.S.-based sites must be members of the NCI Central Institutional Review Board (NCI CIRB). In addition, U.S.-based sites must accept the NCI CIRB review to activate new studies at the site after March 1, 2019. Local

IRB review will continue to be accepted for studies that are not reviewed by the CIRB, or if the study was previously open at the site under the local IRB. International sites should continue to submit Research Ethics Board (REB) approval to the CTSU Regulatory Office following country-specific regulations.

Sites participating with the NCI CIRB must submit the Study Specific Worksheet for Local Context (SSW) to the CIRB using IRBManager to indicate their intent to open the study locally. The NCI CIRB's approval of the SSW is automatically communicated to the CTSU Regulatory Office, but sites are required to contact the CTSU Regulatory Office at CTSURegPref@ctsu.coccg.org to establish site preferences for applying NCI CIRB approvals across their Signatory Network. Site preferences can be set at the network or protocol level. Questions about establishing site preferences can be addressed to the CTSU Regulatory Office by email or calling 1-888-651-CTSU (2878).

Sites using their local IRB or REB, must submit their approval to the CTSU Regulatory Office using the Regulatory Submission Portal located in the Regulatory section of the CTSU website. Acceptable documentation of local IRB/REB approval includes:

- Local IRB documentation;
- IRB-signed CTSU IRB Certification Form; and/or
- Protocol of Human Subjects Assurance Identification/IRB Certification/Declaration of Exemption Form.

In addition, the Site-Protocol Principal Investigator (PI) (i.e. the investigator on the IRB/REB approval) must meet the following criteria in order for the processing of the IRB/REB approval record to be completed:

- Holds an active CTEP status;
- Rostered at the site on the IRB/REB approval and on at least one participating roster;
- If using NCI CIRB, rostered on the NCI CIRB Signatory record;
- Includes the IRB number of the IRB providing approval in the Form FDA 1572 in the RCR profile; and
- Holds the appropriate CTEP registration type for the protocol.

Additional Requirements for Protocol NRG-DT001 Site Registration

Additional requirements to obtain an approved site registration status include:

- An active Federal Wide Assurance (FWA) number;
- An active roster affiliation with the Lead Protocol Organization (LPO) or a Participating Organization (PO); and
- Compliance with all protocol-specific requirements (PSRs)
- IROC Credentialing Status Inquiry (CSI) Form – this form is submitted to IROC to begin the modality credentialing process

- This is a study with a radiation and/or imaging (RTI) component and the enrolling site must be aligned to an RTI provider. To manage provider associations or to add or remove associated providers, access the Provider Association page from the Regulatory section on the CTSU members' website at https://www.ctsu.org/RSS/RTFProviderAssociation. Sites must be linked to at least one Imaging and Radiation Oncology Core (IROC) credentialed provider to participate on trials with an RTI component. Enrolling sites are responsible for ensuring that the appropriate agreements and IRB approvals are in place with their RTI provider. An individual with a primary role on any roster is required to update provider associations, though all individuals at a site may view provider associations. To find who holds primary roles at your site, view the Person Roster Browser under the RUMS section on the CTSU website.

To complete protocol-specific credentialing, the RTI provider or enrolling site should follow instructions in protocol section 8.3 to submit documentation or other materials to the designated IROC Quality Assurance (QA) center. Upon the IROC QA center approving the RTI provider for the study modality, IROC will notify your institution when all credentialing requirements have been met and the institution is credentialed to enter patients onto this study. The credentialing notification document (email) must be uploaded by the site to the CTSU Regulatory Submission Portal for RSS to be updated.

Upon site registration approval in RSS, the enrolling site may access OPEN to complete enrollments. The enrolling site will select their credentialed provider treating the subject in the OPEN credentialing screen and may need to answer additional questions related to treatment in the eligibility checklist.

Submitting Regulatory Documents:

Submit required forms and documents to the CTSU Regulatory Office using the Regulatory Submission Portal on the CTSU website.

To access the Regulatory Submission Portal log in to the CTSU members' website, go to the Regulatory selection and select Regulatory Submission.

Institutions with patients waiting that are unable to use the Regulatory Submission Portal should alert the CTSU Regulatory Office immediately at 1-866-651-2878 in order to receive further instruction and support.

Checking Site's Registration Status:

Site registration status may be verified on the CTSU members' website.

- Click on *Regulatory* at the top of the screen;
- Click on *Site Registration; and*
- Enter the site's 5-character CTEP Institution Code and click on Go.
    - Additional filters are available to sort by Protocol, Registration Status, Protocol Status, and/or IRB Type.

Note: The status shown only reflects institutional compliance with site registration requirements as outlined within the protocol. It does not reflect compliance with protocol requirements for individuals participating on the protocol or the enrolling investigator's status with NCI or their affiliated networks.

8.3 RT-Specific Pre-Registration Requirements (22-FEB-2021)

For detailed information on the specific technology requirement required for this study, please refer to the table below and utilize the web link provided for detailed instructions. The check marks under the treatment modality columns indicate whether that specific credentialing requirement is required for this study. Specific credentialing components may require you to work with various QA centers; however, the IROC Houston QA center will notify your institution and NRG Headquarters when all credentialing requirements have been met and the institution is RT credentialed to enter patients onto this study.

RT Credentialing Requirements	**Web Link for Procedures and Instructions: http://irochouston.mdanderson.org**	
	Treatment Modality	**Key Information**
	Photons	
Facility Questionnaire	✓	The IROC Houston electronic facility questionnaire (FQ) should be completed or updated with the most recent information about your institution. To access this FQ go to http://irochouston.mdanderson.org
Credentialing Status Inquiry Form	✓	Please complete a "Credentialing Status Inquiry Form" found under Credentialing on the IROC Houston QA Center website (http://irochouston.mdanderson.org).
Phantom Irradiation	✓	An IROC Houston anthropomorphic phantom must be successfully completed (if the institution has not previously met this credentialing requirement) if the institution plans to deliver IMRT. Flattening-filter-free (FFF) photon beam delivery, Tomotherapy, MRIdian, and Cyberknife treatment delivery modalities must be credentialed individually. Instructions for requesting and irradiating the phantom are available on the IROC Houston website under credentialing

		(http//irochouston.mdanderson.org).
Credentialing Notification Issued to:		
Institution		Institution will be credentialed for the treatment modality that they intend to use on all patients. IROC Houston QA Center will notify the site that all desired credentialing requirements have been met. The site will need to upload a PDF of the approval email from IROC Houston to the CTSU Regulatory Portal for RSS to be updated.

8.3.1 Digital RT Data Submission to NRG Using TRIAD

Transfer of Images and Data (TRIAD) is the American College of Radiology's (ACR) image exchange application. TRIAD provides sites participating in clinical trials a secure method to transmit images. TRIAD anonymizes and validates the images as they are transferred.

TRIAD Access Requirements:

- A valid CTEP-IAM account.
- Registration type of: Associate (A), Associate Plus (AP), Non-Physician Investigator (NPIVR), or Investigator (IVR) registration type. Refer to the CTEP Registration Procedures section for instructions on how to request a CTEP-IAM account and complete registration in RCR.
- TRIAD Site User role on an NCTN or ETCTN roster.

All individuals on the Imaging and Radiation Oncology Core provider roster have access to TRIAD and may submit images for credentialing purposes, or for enrollments to which the provider is linked in OPEN.

TRIAD Installation:

To submit images, the individual holding the TRIAD Site User role will need to install the TRIAD application on their workstation. TRIAD installation documentation is available at https://triadinstall.acr.org/triadclient/.

This process can be done in parallel to obtaining your CTEP-IAM account and RCR registration.

For questions, contact TRIAD Technical Support staff via email TRIAD-Support@acr.org or 1-703-390-9858.

8.4 **Patient Enrollment** (22-FEB-2021)

Patient registration can occur only after evaluation for eligibility is complete, eligibility criteria have been met, and the study site is listed as 'approved' in the CTSU RSS. Patients must have signed and dated all applicable consents and authorization forms.

8.4.1 Oncology Patient Enrollment Network (OPEN)

The Oncology Patient Enrollment Network (OPEN) is a web-based registration system available on a 24/7 basis. OPEN is integrated with CTSU regulatory and roster data and with the LPOs registration/randomization systems or the Theradex Interactive Web Response System (IWRS) for retrieval of patient registration/randomization assignment. OPEN will populate the patient enrollment data in NCI's clinical data management system, Medidata Rave.

Requirements for OPEN access:

- A valid CTEP-IAM account;
- To perform enrollments or request slot reservations: Must be on an LPO roster, ETCTN corresponding roster, or participating organization (PO) roster with the role of Registrar. Registrars must hold a minimum of an Associate Plus (AP) registration type;
- Have an approved site registration for the protocol prior to patient enrollment.

To assign an Investigator (IVR) or Non-Physician Investigator (NPIVR) as the treating, crediting, consenting, drug shipment (IVR only), or receiving investigator for a patient transfer in OPEN, the IVR or NPIVR must list the IRB number used on the site's IRB approval on their Form FDA 1572 in RCR.

Prior to accessing OPEN, site staff should verify the following:

- Patient has met all eligibility criteria within the protocol stated timeframes; and
- All patients have signed an appropriate consent form and HIPAA authorization form (if applicable).

Note: The OPEN system will provide the site with a printable confirmation of registration and treatment information. You may print this confirmation for your records.

Access OPEN at https://open.ctsu.org or from the OPEN link on the CTSU members' website. Further instructional information is in the OPEN section of the CTSU website at https://www.ctsu.org or https://open.ctsu.org. For any additional questions, contact the CTSU Help Desk at 1-888-823-5923 or ctsucontact@westat.com.

In the event that the OPEN system is not accessible, participating sites can contact NRG web support for assistance with web registration: websupport@acr.org or call the NRG Registration Desk at (215)-574-3191, Monday through Friday, 8:30 a.m. to 5:00 p.m. ET. The registrar will ask the site to fax in the eligibility checklist and will need the registering individual's e-mail address and/or return fax number. This information is required to assure that mechanisms usually triggered by the OPEN web registration system (e.g. drug shipment and confirmation of registration) will occur.

8.4.2 Summary of Registration Procedures

This is a 2-step registration study.

- All eligibility criteria (except central p53 review) must be met prior to Step 1 registration.

- All patients will be registered to Step 1.
  - Primary tissue submission is required for the central p53 review after Step 1 registration has been completed and a case number assigned (refer to section 10 for details on tissue submission).
    - NRG Oncology will notify the sites via an e-mail once the p53 results have been received. Sites can complete Step 2 registration at this time.

9. DRUG INFORMATION

9.1 AMG 232 (KRT-232) (NSC 789723) (01-DEC-2021)

Chemical Name: 2-((3R,5R,6S)-5-(3-chlorophenyl)-6-(4-chlorophenyl)-1-((S)-1-(isopropylsulfonyl)-3-methylbutan-2-yl)-3-methyl-2-oxopiperidin-3-yl) acetic acid

International Non-Proprietary Name (INN): Navtemadlin

Classification: MDM2 inhibitor

Molecular Formula: $C_{28}H_{35}Cl_2NO_5S$ **M.W.:** 568.55

Mode of Action: AMG 232 (KRT 232) is a small molecule, cytotoxic chemotherapeutic agent that binds to murine double minute chromosome 2 (MDM2) and inhibits the MDM2/tumor protein 53 protein-protein interaction.

Description: Is a slightly hygroscopic, anhydrous, and crystalline white to off-white powder.

How Supplied: Kartos Therapeutics supplies, and the Pharmaceutical Management Branch distributes AMG 232 (KRT 232) as an immediate release tablet formulated as either an uncoated tablet with a low drug load (LDL tablet) or a film-coated tablet with a high drug load (HDL tablet).

[redacted]

Route(s) of Administration: Oral.

Method of Administration: [redacted]

Potential Drug Interactions:

[redacted]

[redacted]

[redacted]

Patient Care Implications

Advise women study participants of reproductive potential to use effective contraception while receiving AMG 232 (KRT 232) and 1 month + 1 week (or 5 weeks) after the last dose of AMG 232 (KRT 232). Men study participants must continue to use contraception for 3 months and 1 week (or 13 weeks) after the last dose of AMG 232 (KRT 232). Refer to the protocol document for specific guidance.

9.2 Supply (28-JAN-2020)

This study will be conducted under an IND to be held by NCI and will require FDA submission and approval as part of the IND. AMG 232 (KRT-232) is provided by Kartos Therapeutics and distributed by the DCTD/NCI. AMG 232 (KRT-232) will be supplied to patients on study free of charge.

Agent Ordering and Agent Accountability

NCI-supplied agents may be requested by eligible participating Investigators (or their authorized designee) at each participating institution. The CTEP-assigned protocol number must be used for ordering all CTEP-supplied investigational agents. The eligible participating investigators at each participating institution must be registered with CTEP, DCTD through an annual submission of FDA Form 1572 (Statement of Investigator), NCI Biosketch, Agent Shipment Form, and Financial Disclosure Form (FDF). If there are several participating investigators at one institution, CTEP-supplied investigational agents for the study should be ordered under the name of one lead participating investigator at that institution.

Confirmation of the patient enrollment is required for the initial drug shipment. Submit agent requests through the PMB Online Agent Order Processing (OAOP) application. Access to OAOP requires the establishment of a CTEP Identity and Access Management (IAM) account and the maintenance of an "active" account status, a "current" password, and active person registration status. For questions about drug orders, transfers, returns, or accountability, call or email PMB any time. Refer to the PMB's website for specific policies and guidelines related to agent management.

Agent Inventory Records – The investigator, or a responsible party designated by the investigator, must maintain a careful record of the receipt, dispensing and final disposition of all agents received from the PMB using the appropriate NCI Investigational Agent (Drug) Accountability Record (DARF) available on the CTEP forms page. Store and maintain separate NCI Investigational Agent Accountability Records for each agent, strength, formulation and ordering investigator on this protocol.

Useful Links and Contacts

- CTEP Forms, Templates, Documents: http://ctep.cancer.gov/forms/
- NCI CTEP Investigator Registration: RCRHelpDesk@nih.gov
- PMB policies and guidelines: http://ctep.cancer.gov/branches/pmb/agent_management.htm
- PMB Online Agent Order Processing (OAOP) application: https://ctepcore.nci.nih.gov/OAOP
- CTEP Identity and Access Management (IAM) account: https://ctepcore.nci.nih.gov/iam/
- CTEP IAM account help: ctepreghelp@ctep.nci.nih.gov
- IB Coordinator: IBCoordinator@mail.nih.gov
- PMB email: PMBAfterHours@mail.nih.gov
- PMB phone and hours of service: (240) 276-6575 Monday through Friday between 8:30 am and 4:30 pm (ET)

10. PATHOLOGY/BIOSPECIMEN

10.1 Mandatory Submission of Specimens for NGS sequencing (See Section 10.5 for submission logistics) (9-OCT-2018)

TP53 DNA sequencing will be an integral biomarker and only patients with wild-type (WT) p53 status will be evaluable for dose-escalation decisions, determination of the MTD/RP2D, and studied for other exploratory biomarkers.

10.2 Mandatory Submission of Specimens for Retrospective Central Pathology Review (See Section 10.5 for submission logistics)

10.2.1 Aims

- To determine whether the addition of AMG 232 (KRT-232) to neoadjuvant radiotherapy improves pathologic response in STS.
- To determine if pathologic response is predictive of outcome in STS.
- To assess whether tumor response based on diagnostic imaging accurately reflects the pathologic response to neoadjuvant therapy in STS.
    - To confirm that only patients with STS with g2-3 are enrolled on this clinical trial.
    - All patients enrolled on this clinical trial will submit tumor tissue for

central pathology review. The diagnosis of STS and the histologic subtype will be confirmed during central pathology review of the percent pathologic necrosis rate.
    - To avoid delays, no rapid central review of the sarcoma histologic subtype will be required prior to enrollment or treatment initiation.

10.2.2 Guidelines

- Diagnostic Specimen: No rapid central review of the sarcoma histologic subtype will be required prior to enrollment or treatment initiation. The original pretreatment specimen and the definitive surgical specimen will be reviewed together. The diagnostic specimen will be centrally reviewed by an expert panel of pathologists to determine the histologic subtype of soft tissue sarcoma. Each tumor also will be diagnosed based on the 2013 WHO sarcoma classification and be graded according to the guidelines of the FNCLCC grading systems (Appendix VIII). FNCLCC Grade 1, 2 and 3 will be considered low grade, intermediate grade and high grade, respectively. Please see the 2013 WHO sarcoma classification and the FNCLCC grading in Appendix VII.
- Final surgical resection specimen: The definitive resection specimen will be evaluated for pathologic response. This assessment will include a formal evaluation of percent necrosis according to the guidelines established for osteosarcoma. The tumor response as assessed by central pathology review also will be evaluated to determine whether it predicts clinical outcomes such as local and distant disease control and survival.
- Tumor Progression/Recurrence Specimens (Optional): The specimen obtained at the time of tumor progression/recurrence will be reviewed to confirm that the diagnosis is the same as at study entry and to document changes in pathologic appearance.
- Second Malignant Neoplasm Specimens (Optional): The specimen obtained at the time of diagnosis of a second malignant neoplasm will be examined to confirm the diagnosis and to ensure that the new diagnosis differs from the one assigned at the time of original diagnosis.

10.3 Mandatory Submission of Specimens for AMG 232 (KRT-232) Plasma Pharmacokinetic Profile (See Section 10.5 for submission logistics)

10.3.1 Background: This assay will be utilized to determine AMG 232 (KRT-232) plasma concentrations and ultimately the drug exposure. There was an exposure-response correlation observed between changes in serum MIC-1 levels and AMG 232 (KRT-232) plasma exposure (both maximal (C_{max}) and total exposure (AUC) (Amgen Inc., 2015).

10.3.2 Method: AMG 232 (KRT-232) blood levels will be measured by LC-MS/MS. The bioanalytical method for determining AMG 232 (KRT-232) in human plasma will be developed and validated utilizing liquid chromatography coupled with tandem mass spectrometry (LC-MS/MS) techniques. The analytical method will be validated as recommended by the FDA Guidance for Industry: Bioanalytical Method Validation (FDA 2013).

10.3.3 Analysis: For AMG 232 (KRT-232), single dose PK parameters and steady-state trough

concentrations will be calculated. The plasma concentration-time data will be will be analyzed by standard noncompartmental or compartmental analysis as implemented in the program Phoenix WinNonlin (Pharsight, A Certara™ Company, Cary, NC). Pharmacokinetic parameters such as C_{max}, T_{max}, AUC, Cl/F, Vd/F and $T_{1/2}$ will be calculated and reported. Additional compartmental and/or pharmacometric analyses may be explored to elucidate inter- and intra-patient variability. Exploratory correlative studies with pharmacodynamic (biological endpoints, toxicity and efficacy) will be analyzed using nonparametric statistics. Significance for comparisons will be at the $p<0.05$ level.

10.4 Mandatory Submission of Specimens for Serum Macrophage Inhibitory Cytokine-1 (MIC-1) (See Section 10.5 for submission logistics)

10.4.1 Background: MIC-1, a secreted protein that is strongly induced by activated p53, can be detected in the peripheral blood following MDM2 inhibition (Yang 2013). Therefore, MIC-1 could have utility as a pharmacodynamic biomarker for AMG 232 (KRT-232). MIC-1 level can be determined by a sensitive ELISA test and if correlated with dose or other pharmacodynamic effects can serve as a serum biomarker for p53 activation. We anticipate that most patients treated with AMG 232 (KRT-232) will have an induction in MIC-1 and the level MIC-1 induction will be correlated with the dose of AMG 232 (KRT-232). A commercially available ELISA kit (e.g., Biovendor, LLC, Asheville, NC) will be utilized to measure serum MIC-1 levels. This is for an integrated assay aimed at assessing drug response in patients on the clinical trial and will be utilized to determine serum MIC-1 concentrations and ultimately the drug response.

10.4.2 Method: A commercially available ELISA kit (e.g., Biovendor, LLC, Asheville, NC) will be utilized to measure serum MIC-1 levels.

10.4.3 Analysis: For serum MIC-1 levels, each individual level will be normalized to the baseline level (an average of 2 samples) for that patient. Since MIC-1 levels can be variable, we plan to obtain two baseline blood work (one at the registration and one right before AMG 232 (KRT-232) treatment) in order to obtain a more accurate level of MIC-1 level. In preliminary analyses of clinical trial data, there was an exposure-response correlation observed between changes in serum MIC-1 levels and AMG 232 (KRT-232) exposure (both maximal (Cmax) and total exposure (AUC)).

10.5 Biospecimen Submission Tables (01-DEC-2021)

10.5.1 Mandatory Specimen Submission for p53 Testing (for eligibility determination)
From 10.1 above

Sites are responsible for providing their own supplies and shipping labels for the NGS specimen portion of this protocol.

Sites must submit the specimen for p53 testing within 7 days after Step 1 registration.

Time Point	Specimen	Send Specimens To:
Archival		
	• 1 H&E stained slide at 5 microns [1,2,3] • 10-20 (10 micron) unstained,	EET Biobank

	uncharged, air dried slides cut from one block.[1,2,3,4] • With prior permission from the chairs, submission of specimen in blocks is permitted, but please be aware results may be delayed.	

[1] Include a copy of the corresponding anatomic pathology report with the shipment of tissue.
[2] All slides must be cut sequentially from one block and numbered, starting with the H&E.
[3] Include a copy of the Specimen Transmittal form for TP53 sequencing.
[4] Prefer 20 slides, but minimal 10 slides. If testing of the sample fails due to insufficient amount of tissue submitted, additional tissue will be requested.

Archival Formalin-Fixed Paraffin-Embedded (FFPE) Tumor Specimen

The following criteria must be met for archival tissue:

- Tissue must have been collected ≤ 180 days prior to registration
- Formalin-fixed paraffin-embedded tumor tissue slides must be submitted. The optimal block used for sectioning is at least 70% tumor. Specimen size requirement is as follows:
  - Surface area: 25 mm^2 is optimal. Minimum is 5 mm^2.
  - Volume: 1 mm^3 optimal. Minimum volume is 0.2 mm^3, however the success of DNA extraction decreases at suboptimal tissue volume.

The following are mandatory and must be cut and numbered sequentially from one block:
- One (1) H&E slide
- Ten (10) to twenty (20) 10-µm unstained air-dried uncharged slides. Twenty slides are preferable, but a minimal of ten slides are required. If testing of the sample fails due to insufficient amount of tissue submitted, additional tissue will be requested.
- With prior permission from the chairs, submission of specimen in blocks is permitted, but please be aware results may be delayed.
- If a block is submitted, it will be forwarded to the NRGBB-SF for the mandatory central review after testing is complete.

Specimen Labeling

Tissue Specimen Labels

Include the following on all tissue specimens or containers:
- Protocol Number
- Patient initials
- Patient Study ID
- Time point
- Tissue type (P for primary or M for metastatic)
- Collection date

- Surgical pathology ID (SPID) number from the corresponding pathology report
- Block number from the corresponding pathology report

Shipping Specimens from Clinical Site to the EET Biobank

The Specimen Transmittal Form (see Appendix) and the corresponding anatomical clinical pathology report are required with specimen submission. The pathology report must state the disease diagnosis made by the reviewing pathologist.

Specimen Shipping Instructions

Slides may be shipped on Monday through Friday, but should not be shipped for Saturday delivery.

Ship specimens via overnight courier to the address below. FedEx Priority Overnight is strongly recommended to prevent delays in package receipt. Include a cold pack during warm weather.

Shipping Address

Ship to the address below. Do not ship specimens the day before a holiday.

EET Biobank
The Research Institute at Nationwide Children's Hospital
700 Children's Drive, WA1340
Columbus, Ohio 43205
PH: (614) 722-2865
FAX: (614) 722-2897
Email: BPCBank@nationwidechildrens.org

FedEx Priority Overnight service is very strongly preferred.

NOTE: The EET Biobank FedEx Account will not be provided to submitting institutions.

Contact Information for Assistance

For all queries regarding shipping specimens to the EET Biobank, please use the contact information below:

EET Biobank
Toll-free Phone: (800) 347-2486
E-mail: BPCBank@nationwidechildrens.org

Mandatory Specimen Submission for Retrospective Central Pathology Review

From 10.2 above

Forms: ST forms must be submitted with all submissions. Pathology reports must be submitted with all pathology submissions and must include study and case#, date of procedure and pathology accession number. Remaining PHI information must be fully redacted.
Kits: Punch kits can be provided to sites when requested to NRGBB@ucsf.edu
Shipping: Ship to NRGBB-SF. Sites are responsible for shipping costs for FFPE submissions

Ship specimens by overnight courier to:
NRG Oncology Biospecimen Bank – San Francisco
UCSF Department of Radiation Oncology –Box 1800
2340 Sutter Street- Room S341
San Francisco, CA 94115
415-476-7864/Fax 415-476-5271
Email: NRGBB@ucsf.edu

The NRG Biospecimen Bank will forward all material to Dr. Wakely for central pathology review at the end of the study.

For questions about central pathology review, contact:
Paul E. Wakely, Jr. MD; 614-292-2064; paul.wakely@osumc.edu

Mandatory Specimen Types	Collection Time Points	Collection Information and Requirements/ Instructions for Site	Shipping
Representative formalin-fixed paraffin blocks and 2 corresponding H&E slides from each block. If blocks are unavailable, submit 2 H&E section of all available blocks and one-two 3mm punches as an acceptable alternative to FFPE blocks.	Pre-Treatment: Initial diagnostic specimen at time of Step 1 registration NOTE: For cases where there is only one tumor FFPE block and there is no tissue remaining in the block after sectioning for p53 analysis, please note this on the ST form. The H&E slides should still be submitted for central review. The H&E slide with final pathology report should be submitted for central review.	Submit within 4 weeks of surgery date along with the final surgical resection specimen *If site is submitting punches instead of original block the punches should be from the same block and the H&E being submitted. Sites should embed the punches and create a new H&E to be submitted.	Ship by overnight carrier to NRG BB – San Francisco (use cold packs during warm weather)
Representative formalin-fixed paraffin blocks and 2 corresponding H&E slides from each block. If blocks are unavailable, submit 2 H&E section of all available blocks and one-two 3mm punches as an acceptable alternative to FFPE blocks.	Definitive Surgical Specimen - Mandatory	Submit within 4 weeks of surgery date. If the patient undergoes more than one operation, specimens should be submitted from all procedures. (Mandatory) *If site is submitting punches instead of original block the punches should be from the same block and the H&E being submitted. Sites should embed the punches and create a	Ship by overnight carrier to NRG BB – San Francisco (use cold packs during warm weather)

		new H&E to be submitted.	
Representative formalin-fixed paraffin blocks and 2 corresponding H&E slides from each block. If blocks are unavailable, submit 2 H&E section of all available blocks and one-two 3mm punches as an acceptable alternative to FFPE blocks.	At the time of tumor progression/recurrence (optional, strongly encouraged)	Only at the time of tumor progression or recurrence and when location for recurrence safe for biopsy. (optional) *If site is submitting punches instead of original block the punches should be from the same block and the H&E being submitted. Sites should embed the punches and create a new H&E to be submitted.	Ship by overnight carrier to NRG BB – San Francisco (use cold packs during warm weather)
Representative formalin-fixed paraffin blocks and 2 corresponding H&E slides from each block. If blocks are unavailable, submit 2 H&E section of all available blocks and one-two 3mm punches as an acceptable alternative to FFPE blocks.	At the time of development of a solid second neoplasm (optional, strongly encouraged)	Only when a secondary tumor was found near the treatment field.(optional) *If site is submitting punches instead of original block the punches should be from the same block and the H&E being submitted. Sites should embed the punches and create a new H&E to be submitted	Ship by overnight carrier to NRG BB – San Francisco (use cold packs during warm weather)

If sites are unable to embed the FFPE punches they can send the punches to the biospecimen bank in the punch tools. The NRGBB-SF will embed the punches.

Mandatory Specimen Submission for Pharmacokinetics and Pharmacodynamics from Serum and Plasma Samples
From 10.3 and 10.4 above

Kits: The NRGBB-SF will provide kits that contain the following for these studies. All kit requests must be emailed to the lab at NRGBB@ucsf.edu.

- 100 cryovials and shipping materials for ONE dry ice batch shipment per case.
- Note: Sites are responsible for providing all blood draw tubes
- One Fed Ex priority overnight shipping label per case for one dry ice batch shipment per case from sites to the Baltimore lab. Sites unable to batch ship will be responsible for any additional shipping container and shipping costs.

For any questions regarding the PK collections please contact: email onc-pharmacology@lists.johnshopkins.edu here.

Shipping:
Analytical Pharmacology Core Laboratory*
Attn: NRG-DT001 AMG 232 (KRT-232) Study Samples

1650 Orleans St. CRB1 Rm 184
Baltimore, MD 21231-1000**
Phone: 410-502-7192 or 410-955-1129
Email: onc-pharmacology@lists.johnshopkins.edu

**This zip code is for FedEx shipments. If UPS will be utilized, please ship to the following zip code: 21287

NOTE: Please notify the lab by email at onc-pharmacology@lists.johnshopkins.edu at least 24 hours prior to shipment.

Mandatory Specimen Types	Collection Time Points	Collection Information and Requirements/ Instructions for Site	Shipping to Analytical Pharmacology Core Lab
Please refer to Appendix II for Plasma Pharmacokinetics **Please refer to Appendix III for Serum Macrophage Inhibitory Cytokine-1 (MIC-1)** **Please refer to Appendix IV Sample Combined PK/PD Blood Draw Schedules**			*All samples (but not all aliquots) should be batch shipped via overnight express courier in insulated containers with enough dry ice to maintain the samples in a frozen state. *Overnight shipments should occur on Monday through Wednesday (Tuesday is the preferred day) except when the following day is a holiday.

11. SPECIAL STUDIES (NON-TISSUE)

Not applicable

12. MODALITY REVIEWS

12.1 Radiation Therapy Quality Assurance Reviews

The Radiation Oncology Co-Chairs, Meng Xu Welliver, MD, PhD, and Dian Wang, MD, PhD, will perform ongoing remote RT Quality Assurance Review after cases enrolled have been received at IROC-Philadelphia-RT. The scoring mechanism is: **Per Protocol, Variation Acceptable, Deviation Unacceptable.**

Note: Pre-treatment reviews will be mandatory for Cohort B only - the first case from each institution will require a pre-treatment review. The patient cannot begin treatment until complete data from the first case is received, reviewed and approved.

Sites need to allow 3 business days for the pre-treatment review, and if necessary, 3 additional business days if a revision and re-plan is requested by the Reviewer. The goal of the review is to evaluate protocol compliance.

12.2 Medical Oncology Modality Quality Assurance Reviews

The Medical Oncology Co-Chairs, Gary Schwartz, MD, and Brian Van Tine, MD, will perform a AMG 232 (KRT-232) targeted therapy Assurance Review of all patients who receive or are to receive AMG 232 (KRT-232) targeted therapy in this trial. The goal of the review is to evaluate protocol compliance. The review process is contingent on timely submission of chemotherapy treatment data as specified in Section 12.1. The scoring mechanism is: **1) Per Protocol, 2) Acceptable Variation, 3) Unacceptable Deviation, and 4) Not Evaluable**.

12.3 Surgical Quality Assurance Reviews

The Surgical Oncology Co-Chairs, Dr. John Kane and Dr. Raphael Pollock, will perform a Quality Assurance Review after NRG Headquarters has received complete data for the first 20 cases enrolled.

12.4 Standard of Care Imaging Quality Assurance Reviews

The Diagnostic Imaging Corelab staff of Imaging Analysts and Imaging Technologists will perform ongoing remote Image Quality Assurance reviews after cases enrolled have been received at IROC-Philadelphia-DI. IROC DI QA center staff will monitor for timeliness and completeness of imaging submissions form enrolling centers. QA of imaging submission would include assaying adherence to the recommended protocol requirements, and the overall image quality. Regular feedback to enrolling centers via RAVE will provide for rapid communication of incomplete/non-compliant submissions.

13. DATA AND RECORDS

13.1 Data Management/Collection (22-FEB-2021)

Medidata Rave is a clinical data management system being used for data collection for this trial/study. Access to the trial in Rave is controlled through the CTEP-IAM system and role assignments.

Requirements to access Rave via iMedidata:

- A valid CTEP-IAM account; and
- Assigned a Rave role on the LPO or PO roster at the enrolling site of: Rave CRA, Rave Read Only, Rave CRA (LabAdmin), Rave SLA, or Rave Investigator.
- Rave role requirements:
    - Rave CRA or Rave CRA (Lab Admin) role must have a minimum of an Associate Plus (AP) registration type;
    - Rave Investigator role must be registered as a Non-Physician Investigator (NPIVR) or Investigator (IVR); and
    - Rave Read Only role must have at a minimum an Associates (A) registration type.

Refer to https://ctep.cancer.gov/investigatorResources/default.htm for registration types and documentation required.

Upon initial site registration approval for the study in Regulatory Support System (RSS), all persons with Rave roles assigned on the appropriate roster will be sent a study invitation e-mail from iMedidata. To accept the invitation, site staff must log in to the Select Login (https://login.imedidata.com/selectlogin) using their CTEP-IAM username and password, and click on the *accept* link in the upper right-corner of the iMedidata page. Site staff will not be able to access the study in Rave until all required Medidata and study specific trainings are completed. Trainings will be in the form of electronic learnings (eLearnings) and can be accessed by clicking on the link in the upper right pane of the iMedidata screen. If an eLearning is required and has not yet been taken, the link to the eLearning will appear under the study name in iMedidata instead of the *Rave EDC* link; once the successful completion of the eLearning has been recorded, access to the study in Rave will be granted, and a *Rave EDC* link will display under the study name.

Site staff that have not previously activated their iMedidata/Rave account at the time of initial site registration approval for the study in RSS will receive a separate invitation from iMedidata to activate their account. Account activation instructions are located on the CTSU website in the Data Management section under the Rave resource materials (Medidata Account Activation and Study Invitation Acceptance). Additional information on iMedidata/Rave is available on the CTSU members' website in the Data Management > Rave section at www.ctsu.org/RAVE/ or by contacting the CTSU Help Desk at 1-888-823-5923 or by e-mail at ctsucontact@westat.com.

13.2 Summary of Data Submission (29-MAY-2019)

Adverse event data collection and reporting, which are required as part of every clinical trial, are done to ensure the safety of patients enrolled in the studies as well as those who will enroll in future studies using similar agents. Adverse events are reported in a routine manner at scheduled times during the trial using Medidata Rave®. Additionally, certain adverse events must be reported in an expedited manner for more timely monitoring of patient safety and care. See Section 7.4 for information about expedited and routine reporting.

Summary of Data Submission: Refer to the protocol-specific page on the NRG website.

See Section 8.3.1 for TRIAD account access and installation instructions.

RT Digital Data Submission Requirements
Summary of Dosimetry Digital Data Submission

DICOM DIGITAL DATA	DICOM CT IMAGE SET	TRIAD submission time point = RT DIGITAL PLAN Due within 1 week of the start of
	DICOM RT STRUCTURE	

	DICOM RT DOSE	RT ***Cohort B only*** – **mandatory pre-treatment review required for the first case from each institution; see section 5.2 and 5.2.11 for details on time allotment for review.** TRIAD submission time point = RT DIGITAL PLAN
	DICOM RT PLAN	
	*DICOM PET (Required when Applicable) *DICOM PET/CT (Required when Applicable) *DICOM MRI (Required when Applicable)	
*All image data sets used for structure delineation must be submitted with RT data. (section 5.2.3)		
All required structures MUST be labeled per the tables in Section 5.2		
Upon submission of the Digital Data via TRIAD, complete an online Digital Data Submission Information Form (DDSI) https://www.irocqa.org/Resources/TRIAD-for-RT-QA		
NOTE: ALL SIMULATION AND PORTAL FILMS AND OR DIGITAL FILM IMAGES WILL BE KEPT BY THE INSTITUTION AND ONLY SUBMITTED IF REQUESTED.		

13.3 Data Quality Portal (28-JAN-2020)

The Data Quality Portal (DQP) provides a central location for site staff to manage unanswered queries and form delinquencies, monitor data quality and timeliness, generate reports, and review metrics.

The DQP is located on the CTSU members' website under Data Management. The Rave Home section displays a table providing summary counts of Total Delinquencies and Total Queries. DQP Queries, DQP Delinquent Forms and the DQP Reports modules are available to access details and reports of unanswered queries, delinquent forms, and timeliness reports. Review the DQP modules on a regular basis to manage specified queries and delinquent forms.

The DQP is accessible by site staff that are rostered to a site and have access to the CTSU website. Staff that have Rave study access can access the Rave study data using a direct link on the DQP.

To learn more about DQP use and access, click on the Help icon displayed on the Rave Home, DQP Queries, and DQP Delinquent Forms modules.

Note: Some Rave protocols may not have delinquent form details or reports specified on the DQP. A protocol must have the Calendar functionality implemented in Rave by the Lead Protocol Organization (LPO) for delinquent form details and reports to be available on the DQP. Site staff should contact the LPO Data Manager for their protocol regarding questions about Rave Calendaring functionality.

13.4 Rave-CTEP-AERS integration (22-FEB-2021)

The Rave Cancer Therapy Evaluation Program Adverse Event Reporting System (CTEP-AERS) Integration enables evaluation of post-baseline Adverse Events (AE) entered in Rave to determine whether they require expedited reporting, and facilitates entry in CTEP-AERS for those AEs requiring expedited reporting.

All AEs that occur after baseline are collected in Medidata Rave using the Adverse Event form, which is available for entry at each treatment or reporting period and used to collect AEs that start during the period or persist from the previous reporting period. The CRA will enter AEs that occur prior to the start of treatment on a baseline form that is not included in the Rave-CTEP-AERS integration. AEs that occur prior to enrollment must begin and end on the baseline Adverse Events form and should not be included on the standard Adverse Events form that is available at treatment unless there has been an increase in grade.

Prior to sending AEs through the rules evaluation process, site staff should verify the following on the Adverse Event form in Rave:

- The reporting period (course/cycle) is correct; and
- AEs are recorded and complete (no missing fields) and the form is query free.

The CRA reports AEs in Rave at the time the Investigator learns of the event. If the CRA modifies an AE, it must be re-submitted for rules evaluation.

Upon completion of AE entry in Medidata Rave, the CRA submits the AE for rules evaluation by completing the Expedited Reporting Evaluation form. Both NCI and protocol-specific reporting rules evaluate the AEs submitted for expedited reporting. A report is initiated in CTEP-AERS using information entered in Medidata Rave for AEs that meet reporting requirements. The CRA completes the report by accessing CTEP-AERS via a direct link on the Medidata Rave Expedited Reporting Evaluation form.

In the rare occurrence that internet connectivity is lost, a 24-hour notification is to be made to CTEP by telephone at 301-897-7497. Once internet connectivity is restored, the 24-hour notification that was phoned in must be entered immediately into CTEP-AERS using the direct link from Medidata Rave.

Additional information about the CTEP-AERS integration is available on the CTSU website:

- Expedited Safety Reporting Rules Evaluation user guide: Resources > CTSU Operations Information> User Guides & Help Topics.

NCI requirements for SAE reporting are available on the CTEP website:

- NCI Guidelines for Investigators: Adverse Event Reporting Requirements is available at https://ctep.cancer.gov/protocolDevelopment/electronic_applications/docs/aeguidelines.pdf

13.5 Global Reporting/Monitoring (28-JAN-2020)

This study will be monitored by the Clinical Data Update System (CDUS) version 3.0. Cumulative CDUS data will be submitted quarterly to CTEP by electronic means. Reports are due January 31, April 30, July 31, and October 31.

This study has been assigned to CDUS-Complete reporting, **all** adverse events (both routine and expedited) that have occurred on the study and meet the mandatory CDUS reporting guidelines must be reported via the monitoring method identified above.

14. STATISTICAL CONSIDERATIONS

14.1 Study Design

This is a phase I study investigating the safety and tolerability of AMG 232 (KRT-232) in combination with standard-dose radiotherapy in STS in two separate patient cohorts. Patients will be enrolled separately into two cohorts based on STS type (extremity or body wall vs. abdomen/pelvis/retroperitoneal cavity). Within each cohort patients will be assigned to the following dose escalation schedule: increasing frequency with constant doses sequentially. The classic 3+3 design will be used to test the safety of each dose level, and testing will be started with dose level 1. All p53 wild type patients who receive at least one dose of AMG 232 (KRT-232) are considered evaluable for DLT, should a DLT occur. In the absence of DLT, patients must have received at least one fraction of radiation therapy and have completed the DLT observation period to be evaluable for DLT. Patients considered inevaluable will be replaced to ensure the minimum evaluable patients per arm. The minimum sample size is 12 evaluable patients and the maximum sample size is 46 evaluable patients for the two cohorts combined.

14.2 Study Endpoints (9May2018)

14.2.1 Primary Endpoint

MTD/RP2D at 4 weeks after treatment completion (AMG 232 (KRT-232) + radiotherapy).

14.2.2 Secondary Endpoints

- Percent of necrosis and pCR in final surgical resection specimen;
- Local failure (LF),
- Disease free survival (DFS), and
- Overall survival (OS) at 2 years;
- Serial serum Macrophage Inhibitory Cytokine (MIC)-1 levels;
- To correlate AMG 232 (KRT-232) exposure-response relationships (PD, toxicity, and efficacy).

14.2.3 Tertiary Endpoints

Tumor volume changes by cohort and dose levels.
Clinical outcomes by genomic biomarkers
Correlation between mdm2/4 expression vs. protein levels.
tumor genetic mutations in (1) cf ctDNA, (2) DNA/RNA isolated from exosomes

14.3 Primary Objectives Study Design

14.3.1 Primary Hypothesis and Endpoints: Preoperative combined AMG 232 (KRT-232) and radiotherapy treatment is safe and well tolerated, and shows preliminary efficacy in STS

14.3.2

There are two patient cohorts based on tumor locations therefore dose escalation is done separately with these two cohorts.

DLT evaluation will occur 4 weeks after treatment completion prior to surgery, but toxicity data will be collected and monitored till surgery completion and during the follow up period.

Patients must have sufficient tissue for NGS sequencing to be included on this trial. Patients with wild type p53 will continue on study drug.

The rate of DLTs and 95%CI will be summarized using proportions for binary outcome per cohort.

14.3.3 Sample Size and Power Calculations: Based on the dose escalation rule, for each of the two patient cohorts, the classic 3+3 design will be used to make the decision on dose escalation/de-escalation at each dose level, and dose level 1 will be tested first. See section 14.6.1 for the definition of DLTs. At dose level 1 for each cohort, among the first 3 evaluable patients, if there is no DLT, then we escalate to next higher dose level; if one DLT is observed, 3 more patients will be accrued to the same dose level; if there is more than one DLT, we will then stop the accrual to this dose level and de-escalate the dose to dose level -1. Under the condition that there is one DLT among the first 3 patients and 6 evaluable patients are accrued to this dose level still with one DLT observed among these 6, we will escalate the dose to the next higher dose level, dose level 2; if there is one DLT among the first 3 patients and 6 evaluable patients are accrued to dose level 1 with more than one DLT observed among these 6, we will de-escalate the dose to dose level -1. If dose level 1 is proved to be not safe, dose level -1 will be tested based on the classic 3+3 design. If dose level 1 is proved to be safe, dose level 2 will be tested similarly based on the classic 3+3 design. If dose level 2 is proved to be not safe, dose level 1 will be considered the MTD and dose escalation will be stopped: if dose level 2 is proved to be safe, dose level 3 will be tested using the classic 3+3 design. If dose level 3 is proved to be safe, this dose level will be considered the MTD; if not, dose level 2 will be considered the MTD. Five additional patients will be accrued to the MTD to ensure safety. The minimum sample size is 12 evaluable patients and the maximum sample size is 46 evaluable patients for the two cohorts combined.

14.4 Study Monitoring of Primary Objectives

(Interim Analysis)

Interim Analysis to Monitor Study Progress

Interim reports will be prepared twice each year until the final analysis has been accepted for presentation or publication. In general, these reports will contain information about the accrual rate with projected completion date for the accrual phase, exclusion rates, pretreatment characteristics of patients accrued, and the frequency and severity of AEs. This study will be monitored by the Clinical Data Update System (CDUS), version 3.0. Cumulative CDUS data will be submitted quarterly by electronic means.

Evaluable patients (all patients started treatment) will be included in the final analysis. The usual components of this analysis are:
• Tabulation of all cases entered, and any excluded from analysis with reasons for exclusion;
• Patient accrual rate;
• Distribution of important baseline prognostic variables;
• Frequency and severity of adverse events;
• Observed results with respect to the endpoints described above.

After the start of the accrual, the study team, including the study chairs, study statisticians, data managers and protocol administrator, will hold regular conference calls to review the overall conduct of the study. Data on treatment dose delivery, adverse events reported, patient demographics and eligibility will be assembled and reviewed. When a decision on dose de-escalation/escalation must be made, the study team will hold a conference call to review information including the categorization and grading of reported adverse events, determination of the dose-limiting toxicities, etc. The decision to de-escalate or escalate is made by consensus of the study team in accordance with the decision rules outlined in the protocol. At each meeting, consideration is also given to the rate of accrual. Brief minutes of each meeting will be written by the study chairs to document the review of information and any decision made. The meeting minutes will be submitted to the NRG early phase protocol monitoring oversight committee for review.

Number of Patients with DLT at a Given Dose Level	Escalation Decision Rule
0 out of 3	Enter 3 patients at the next higher dose level.
≥2 out of 3	Dose escalation will be stopped. This dose level will be declared the maximally administered dose (highest dose administered).
1 out of 3	Enter 3 more patients at this dose level. • If 0 of these 3 patients experience DLT, proceed to the next higher dose level. • If 1 or more of this group suffer DLT, then dose escalation will be stopped, and this dose is declared the maximally administered dose.

14.5 Accrual/Study Duration Considerations (9May2018)

Based on prior studies (RTOG 9514, RTOG 0630 and ARST 1321) we project a monthly accrual rate of 1.5 patients. Minimal accrual is projected for the first 3 to 6 months after study activation.

14.6 Dose Level Guidelines: See Section 6
(Escalation or de-escalation):

14.6.1 Definition of Dose-Limiting Toxicity

DLT includes all grade 4-5 toxicities attributable to AMG 232 (KRT-232) or combined AMG 232 (KRT-232) with radiotherapy up to 4 weeks after the completion of AMG 232 (KRT-232)+RT treatment.

Any grade 3 AE at least possibly attributable to AMG 232 (KRT-232) or combined AMG 232 (KRT-232) with radiotherapy will also be considered DLT if any of the two following situations occur:

- A delay of >2 weeks due to the grade 3 toxicity.
- ≥ two dose reductions due to the grade 3 toxicity.

Management and dose modifications associated with the above adverse events are outlined in Section 6.

14.6.2 Dose Expansion Cohorts

Once the RP2D is reached, an additional *5 evaluable patients* will be treated at this dose. For the expansion cohort, patients will continue to be monitored for occurrence of DLT. If ≥2 of the -5 patients experience DLT, the Principal Investigator will discuss with all study investigators and with CTEP whether further addition of patients is needed to re-assess the RP2D.

14.7 Secondary or Exploratory Endpoints (including correlative science aims) (9May2018)

Descriptive statistics will be provided for selected demographic, safety, PK, and PD data by dose, p53 status and time as appropriate.

Local failure: local recurrence or progression as defined in RTOG 0630 that occurs after surgery will be considered a local failure; local recurrence or progression prior to surgery will not be considered a local failure. Amputation for treatment complications or recurrence/progression will be considered a failure at the date of surgery; amputation due to any other reason will not be considered a local failure. Any patient that does not continue on to surgery will be considered to have local failure; failure time will be the minimum time to surgery as calculated from the patients that do continue on to surgery

Disease-free survival (DFS) defined as local, regional, or distant failure, or death due to any cause, and overall survival (OS, Death due to any cause) will be described with Kaplan-Meier plots and local failure by cumulative incidence plots or descriptive statistics, as data allow. Percent of necrosis and pCR will be summarized using binomial proportions. Tumor volume changes will be summarized and described by cohort and by dose levels. Pearson's correlation will be used to assess relationships between mdm2/4 expression and protein levels.

A full pharmacokinetic profile of AMG 232 (KRT-232) will be proposed in this LOI to assess exposure-response relationships with various PD endpoints (i.e., MIC-1 changes,

toxicity, efficacy). AMG 232 (KRT-232) concentrations in these samples will be quantitatively measured using liquid chromatography/tandem mass spectrometric (LC/MS/MS) method that will be developed by the Analytical Pharmacology Core Laboratory at the Sidney Kimmel Comprehensive Cancer Center (SKCCC) at Johns Hopkins. For AMG 232 (KRT-232), single dose PK parameters and steady-state trough concentrations will be calculated. The individual PK parameters will be estimated for C_{max}, AUC, $T_{1/2}$, apparent Cl/F, and apparent V/F using non-compartmental or compartmental PK methods with the software WinNonlin. For serum MIC-1 levels, each individual level will be normalized to the baseline level (the average of 2 baseline samples) for that patient. Advanced population PK methods may be employed to assess the link between drug exposure and biological effects and efficacy. The PK variables and changes in MIC-1 will be tabulated and descriptive statistics (e.g., geometric means and coefficients of variation) calculated for each dose level. PK parameters (i.e., T½, Cl, and AUC) and MIC-1 changes will be compared across dose level using nonparametic statistical testing techniques. Exploratory correlative studies with pharmacodynamic (biological endpoints, toxicity and efficacy) will be analyzed using nonparametric statistics. Significance for comparisons will be at the $p<0.05$ level.

14.8 Gender/Ethnicity/Race Distribution

Racial Categories	DOMESTIC PLANNED ENROLLMENT REPORT				
	Ethnic Categories				
	Not Hispanic or Latino		Hispanic or Latino		Total
	Female	Male	Female	Male	
American Indian/Alaska Native	0	1	0	0	1
Asian	0	0	0	0	0
Native Hawaiian or Other Pacific Islander	0	0	0	0	0
Black or African American	1	1	0	1	3
White	20	20	1	1	42
More Than One Race	0	0	0	0	0
Total	21	21	1	3	46

Racial Categories	INTERNATIONAL (including Canadian participants) PLANNED ENROLLMENT REPORT				
	Ethnic Categories				
	Not Hispanic or Latino		Hispanic or Latino		Total
	Female	Male	Female	Male	
American Indian/Alaska Native	0	0	0	0	0

Asian	0	0	0	0	0
Native Hawaiian or Other Pacific Islander	0	0	0	0	0
Black or African American	0	0	0	0	0
White	0	0	0	0	0
More Than One Race	0	0	0	0	0
Total	0	0	0	0	0

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APPENDIX I: COLLABORATIVE AGREEMENTS LANGUAGE

The agent(s) supplied by CTEP, DCTD, NCI used in this protocol is/are provided to the NCI under a Collaborative Agreement (CRADA, CTA, CSA) between the Pharmaceutical Company(ies) (hereinafter referred to as "Collaborator(s)") and the NCI Division of Cancer Treatment and Diagnosis. Therefore, the following obligations/guidelines, in addition to the provisions in the "Intellectual Property Option to Collaborator" (http://ctep.cancer.gov/industryCollaborations2/intellectual_property.htm) contained within the terms of award, apply to the use of the Agent(s) in this study:

1. Agent(s) may not be used for any purpose outside the scope of this protocol, nor can Agent(s) be transferred or licensed to any party not participating in the clinical study. Collaborator(s) data for Agent(s) are confidential and proprietary to Collaborator(s) and shall be maintained as such by the investigators. The protocol documents for studies utilizing Agents contain confidential information and should not be shared or distributed without the permission of the NCI. If a copy of this protocol is requested by a patient or patient's family member participating on the study, the individual should sign a confidentiality agreement. A suitable model agreement can be downloaded from: http://ctep.cancer.gov.

2. For a clinical protocol where there is an investigational Agent used in combination with (an)other Agent(s), each the subject of different Collaborative Agreements, the access to and use of data by each Collaborator shall be as follows (data pertaining to such combination use shall hereinafter be referred to as "Multi-Party Data"):

    a. NCI will provide all Collaborators with prior written notice regarding the existence and nature of any agreements governing their collaboration with NCI, the design of the proposed combination protocol, and the existence of any obligations that would tend to restrict NCI's participation in the proposed combination protocol.

    b. Each Collaborator shall agree to permit use of the Multi-Party Data from the clinical trial by any other Collaborator solely to the extent necessary to allow said other Collaborator to develop, obtain regulatory approval or commercialize its own Agent.

    c. Any Collaborator having the right to use the Multi-Party Data from these trials must agree in writing prior to the commencement of the trials that it will use the Multi-Party Data solely for development, regulatory approval, and commercialization of its own Agent.

3. Clinical Trial Data and Results and Raw Data developed under a Collaborative Agreement will be made available to Collaborator(s), the NCI, and the FDA, as appropriate and unless additional disclosure is required by law or court order as described in the IP Option to Collaborator (http://ctep.cancer.gov/industryCollaborations2/intellectual_property.htm). Additionally, all Clinical Data and Results and Raw Data will be collected, used and disclosed consistent with all applicable federal statutes and regulations for the protection of human subjects, including, if applicable, the *Standards for Privacy of Individually Identifiable Health Information* set forth in 45 C.F.R. Part 164.

4. When a Collaborator wishes to initiate a data request, the request should first be sent to the NCI, who will then notify the appropriate investigators (Group Chair for Cooperative Group studies, or PI for other studies) of Collaborator's wish to contact them.

5. Any data provided to Collaborator(s) for Phase 3 studies must be in accordance with the guidelines and policies of the responsible Data Monitoring Committee (DMC), if there is a DMC for this clinical trial.

6. Any manuscripts reporting the results of this clinical trial must be provided to CTEP by the Group office for Cooperative Group studies or by the principal investigator for non-Cooperative Group studies for immediate delivery to Collaborator(s) for advisory review and comment prior to submission for publication. Collaborator(s) will have 30 days from the date of receipt for review. Collaborator shall have the right to request that publication be delayed for up to an additional 30 days in order to ensure that Collaborator's confidential and proprietary data, in addition to Collaborator(s)'s intellectual property rights, are protected. Copies of abstracts must be provided to CTEP for forwarding to Collaborator(s) for courtesy review as soon as possible and preferably at least three (3) days prior to submission, but in any case, prior to presentation at the meeting or publication in the proceedings. Press releases and other media presentations must also be forwarded to CTEP prior to release. Copies of any manuscript, abstract and/or press release/ media presentation should be sent to:

Email: ncicteppubs@mail.nih.gov

The Regulatory Affairs Branch will then distribute them to Collaborator(s). No publication, manuscript or other form of public disclosure shall contain any of Collaborator's confidential/ proprietary information.

APPENDIX II: PLASMA PHARMACOKINETICS (29-MAY-2019)

Collection of Specimen(s)

Blood samples (~6 mL) will be taken into K2 EDTA tubes at designated time points, and immediately placed on ice. Please note that there is a window of ±5 mins from start to 8 hours and a window of ±1 hour for the 24 hour post dose.

AMG 232 (KRT-232) through 4X/week:
AMG 232 (KRT-232) alone (6 samples): Wk-1 first dose: predose, 1, 3, 5, 8, and 24 hours post dose
AMG 232 (KRT-232) + RT (6 samples): Wk1D2, Wk3D2, and Wk5D2: predose and 24 hours post dose
AMG 232 (KRT-232) 5X/week:
AMG 232 (KRT-232) alone (6 samples): Wk-1 first dose: predose, 1, 3, 5, 8, and 24 hours post dose
AMG 232 (KRT-232) + RT (6 samples): Wk1D1, Wk3D1, and Wk5D1: predose and 24 hours post dose

*See Appendix IV for Sample Combined PK Blood Draw Schedule
**If AMG 232 (KRT-232) is stopped because of p53 gene results or for any other reason PK blood draw will not be required

Handling of Specimens(s)

At each time point, ~6 mL of peripheral blood will be collected in a purple-top EDTA vacutainer (Becton Dickinson Catalog # 367863 or 367899, Franklin Lakes, NJ).

- Obtain venous blood by standard phlebotomy technique from a peripheral access point. NOTE: Suggest using a minimum 18G needle to avoid sample hemolysis.
- Fill-up the tubes as much as possible until blood flow stops.
- GENTLY invert each tube several times (8-10 times) immediately after collection to avoid sample hemolysis.
- Place samples immediately **on ice** after collection; samples must be processed **within 30 minutes**.

Processing instructions

1. Invert sample 8-10 times immediately before processing.
2. Centrifuge at ~1300 xg for 10 minutes in swinging bucket (SW) or 15 minutes in a

fixed angel (FA) rotor at 4ºC in a refrigerated centrifuge. Make sure that the centrifuge reaches speed and is maintained throughout the entire spin.
3. Carefully remove tube from centrifuge.
4. Using a pipette, transfer equal aliquots of plasma into 2-3 labeled 2 mL cryovials (e.g., Corning #430659 external thread vial), not exceeding 1.5 mL per cryovial.
5. Label samples as AMG 232 (KRT-232) PK, including protocol number (NRG-DT001), unique patient ID assigned at registration, initials, date of collection, draw time, and time point.
6. Store plasma samples at -70ºC or below until shipment or transfer to Johns Hopkins.

Shipping of Specimen(s)

Specimens should be stored through the duration of AMG 232 (KRT-232) correlative studies and shipped as a batch by participant (more than one participant/shipment is acceptable if the site has >1 participant on-study). A participant's samples should be shipped to the APC lab within 1 month of the last sample's collection date. (i.e., if sample is collected on 1/1/2017, all of that participant's samples should be at the APC lab by 2/1/2017). The APC lab may contact the study team to request shipment off-schedule. The MIC-1 and AMG 232 (KRT-232) pharmacokinetic specimens can be shipped in the same shipment.
Please ship 1-2 aliquots to the APC laboratory. Once receipt is confirmed, the back-up aliquot may be shipped at that time or it can also be shipped later (e.g., with the next scheduled shipment).

Preparing the shipment

*Samples should be stored in cardboard boxes (5 1/8” x 5 1/8” x 2”, LxWxH).
*Please organize the samples by Patient and Time point in the box.
*Do not store in plastic bags (they break on dry-ice and labels will detach).
*A copy of each of the pharmacokinetic sample collection forms for the respective patients or a sample list should be included with each shipment. To prevent problems with illegible writing on tubes, consider numbering them (in addition to sample label) and numbering samples on the sample sheet.
*Note the study number, PI, and the drugs used/to be measured.
*A name, phone number and email address should be included with samples so that receipt can be acknowledged.
*Please notify the lab by email (onc-pharmacology@lists.johnshopkins.edu) at least 24 hours prior to shipment.

Shipping

*All samples should be shipped via overnight express courier in insulated containers with enough dry ice to maintain the samples in a frozen state.
*Overnight shipments should occur on Monday through Wednesday (Tuesday is the preferred day) except when the following day is a holiday.

Analytical Pharmacology Core Laboratory*
Attn: NRG-DT001 AMG 232 (KRT-232) Study Samples
1650 Orleans St. CRB1 Rm 184
Baltimore, MD 21231-1000**
Phone: 410-502-7192 or 410-955-1129
Email: onc-pharmacology@lists.johnshopkins.edu

**This zip code is for FedEx shipments. If UPS will be utilized, please ship to the following zip code: 21287

NOTE: Please notify the lab by email (onc-pharmacology@lists.johnshopkins.edu) at least 24 hours prior to shipment.

Site(s) Performing Correlative Study
Dr. Michelle Rudek, The Analytical Pharmacology Core Laboratory at the Sidney Kimmel Comprehensive Cancer Center at Johns Hopkins (SKCCC)

APPENDIX III: SERUM MACROPHAGE INHIBITORY CYTOKINE-1 (MIC-1) (29-MAY-2019)

Collection of Specimen(s)

Blood samples (~6 mL) will collected be in red-topped vacutainer tube at designated time points. Serum samples from the peripheral blood will be collected for MIC-1 assay on the following schedule:

A baseline MIC-1 level will be determined by the average of two blood samples obtained at or between the time of step 1 registration and prior to the first AMG 232 (KRT-232) dose.

Please note that there is a window of ±1 hour for the 24 hour post dose.

AMG 232 (KRT-232) through 4X/week:
AMG 232 (KRT-232) alone (3 samples): At or after Step 1 registration, and before Wk-1 first dose: predose (x2), and 24 hours post dose
AMG 232 (KRT-232) + RT (6 samples): Wk1D2, Wk3D2, and Wk5D2: predose and 24 hours post dose
AMG 232 (KRT-232) 5X/week:
AMG 232 (KRT-232) alone (3 samples): At or after Step 1 registration, and before Wk-1 first dose: predose (x2), and 24 hours post dose
AMG 232 (KRT-232) + RT (6 samples): Wk1D1, Wk3D1, and Wk5D1: predose and 24 hours post dose

*See Appendix IV for Sample Combined PD Blood Draw Schedule

**If AMG 232 (KRT-232) is stopped because of p53 gene results or for any other reason PD blood draw will not be required

Handling of Specimens(s)

At each time point, ~6 mL of peripheral blood will be collected in a red-topped vacutainer (Becton Dickinson Catalog #367815, Franklin Lakes, NJ).

- Obtain venous blood by standard phlebotomy technique from a peripheral access point. NOTE: Suggest using a minimum 18G needle to avoid sample hemolysis.
- Fill-up the tubes as much as possible until blood flow stops.
- GENTLY invert each tube several times (8-10 times) immediately after collection to avoid sample hemolysis.

- Transport the tube(s) as soon as possible to the laboratory. Allow the blood to clot in an upright position for at least 30 minutes but not longer than 1 hour before centrifugation.

  Processing instructions:
  1. Centrifuge at ~1200 xg for 15 minutes in swinging bucket (SW) or 15 minutes in a fixed angel (FA) rotor. Make sure that the centrifuge reaches speed and is maintained throughout the entire spin.
  2. Carefully remove tube from centrifuge.
  3. Using a pipette, transfer equal aliquots of serum into ~4-6 labeled 2 mL cryovials (e.g., Corning #430659 external thread vial), with a minimum volume of 0.5 mL per cryovial.
  4. Label samples as MIC-1, including study number (NRG-DT001), unique patient ID assigned at registration, initials, date of collection, draw time, and time point.
  5. Store serum samples at -70ºC or below until shipment or transfer to Johns Hopkins.

Shipping (Please refer to APPENDIX II for further details on the specifics of how to package and when to ship)

*All samples should be shipped via overnight express courier in insulated containers with enough dry ice to maintain the samples in a frozen state.
*Overnight shipments should occur on Monday through Wednesday (Tuesday is the preferred day) except when the following day is a holiday.

Analytical Pharmacology Core Laboratory*
Attn: NRG-DT001 AMG 232 (KRT-232) Study Samples
1650 Orleans St. CRB1 Rm 184
Baltimore, MD 21231-1000**
Phone: 410-502-7192 or 410-955-1129
Email: onc-pharmacology@lists.johnshopkins.edu

**This zip code is for FedEx shipments. If UPS will be utilized, please ship to the following zip code: 21287

NOTE: Please notify the lab by email (onc-pharmacology@lists.johnshopkins.edu) at least 24 hours prior to shipment.

Site(s) Performing Correlative Study
Dr. Michelle Rudek, The Analytical Pharmacology Core Laboratory at the Sidney Kimmel Comprehensive Cancer Center at Johns Hopkins (SKCCC)

APPENDIX IV: SAMPLE COMBINED PK/PD BLOOD DRAW SCHEDULES (28-JAN-2020)

PK Blood Draw Schedule (2x-4x/week dosing)**						
	Predose	Post Dose, 1 hour	Post Dose, 3 hours	Post Dose, 5 hours	Post Dose, 8 hours	Post Dose, 24 hours
AMG 232 (KRT-232) Alone (Week -1, Day 2)	X	X	X	X	X	X
AMG 232 (KRT-232) + RT, (Week 1, Day 2)	X					X
AMG 232 (KRT-232) + RT, (Week 3, Day 2)	X					X
AMG 232 (KRT-232) + RT, (Week 5, Day 2)	X					X
PD Blood Draw Schedule (2x-4x/week dosing)**						
	Predose					Post Dose, 24 hours
AMG 232 (KRT-232) Alone (Week -1, Day 2)	XX (two samples anytime from registration to prior to first dose)					X
AMG 232 (KRT-232) + RT, (Week 1, Day 2)	X					X
AMG 232 (KRT-232) + RT, (Week 3, Day 2)	X					X
AMG 232 (KRT-232) + RT, (Week 5, Day 2)	X					X

**Please note that there is a window of ±5 mins from start to 8 hours and a window of ±1 hour for the 24 hour post dose.

PK Blood Draw Schedule (5x/week dosing)**						
	Predose	Post Dose, 1 hour,	Post Dose, 3 hours,	Post Dose, 5 hours,	Post Dose, 8 hours,	Post Dose, 24 hours
AMG 232 (KRT-232) Alone (Week -1, Day 1)	X	X	X	X	X	X
AMG 232 (KRT-232) + RT, (Week 1, Day 1)	X					X
AMG 232 (KRT-232) + RT, (Week 3, Day 1)	X					X
AMG 232 (KRT-232) + RT, (Week 5, Day 1)	X					X
PD Blood Draw Schedule (5x/week dosing)**						
	Predose					Post Dose, 24 hours
AMG 232 (KRT-232) Alone (Week -1, Day 1)	XX (two samples anytime from registration to prior to first dose)					X
AMG 232 (KRT-232) + RT, (Week 1, Day 1)	X					X
AMG 232 (KRT-232) + RT, (Week 3, Day 1)	X					X
AMG 232 (KRT-232) + RT, (Week 5, Day 1)	X					X

**Please note that there is a window of ±5 mins from start to 8 hours and a window of ±1 hour for the 24 hour post dose.

APPENDIX V: DRUG-INDUCED LIVER INJURY (DILI) REPORTING & ADDITIONAL ASSESSMENTS REPORTING

To facilitate appropriate monitoring for signals of drug-induced liver injury (DILI), cases of concurrent AST or ALT and TBL and/or INR elevation according to the criteria specified in Section 6.1 require the following:

- The event is to be reported to CTEP as a serious adverse event within 24 hours of discovery or notification of the event (*i.e.*, before additional etiologic investigations have been concluded)
- The appropriate CRF (*e.g.*, Adverse Event CRF) that captures information necessary to facilitate the evaluation of treatment-emergent liver abnormalities is to be completed and sent to CTEP.
- Other events of hepatotoxicity and potential DILI are to be reported as serious adverse events if they meet the criteria for a serious adverse event defined in Section 7.

Additional Clinical Assessments and Observation

All subjects in whom investigational product(s) or protocol-required therapies is/are withheld (either permanently or conditionally) due to potential DILI as specified in Section 6.1 or who experience AST or ALT elevations ≥ 3 × ULN are to undergo a period of "close observation" until abnormalities return to normal or to the subject's baseline levels. Assessments that are to be performed during this period include:

- Repeat AST, ALT, ALP, bilirubin (total and direct), and INR within 24 hours
- In cases of TBL > 2 × ULN or INR > 1.5, retesting of liver tests, BIL (total and direct), and INR is to be performed every 24 hours until laboratory abnormalities improve.

Testing frequency of the above laboratory tests may decrease if the abnormalities stabilize or the investigational product(s) or protocol-required therapies has/have been discontinued AND the subject is asymptomatic.

- Initiate investigation of alternative causes for elevated AST or ALT and/or elevated TBL:
  - Obtain complete blood count (CBC) with differential to assess for eosinophilia
  - Obtain serum total immunoglobulin IgG, Anti-nuclear antibody (ANA), Anti Smooth Muscle Antibody, and Liver Kidney Microsomal antibody 1 (LKM1) to assess for autoimmune hepatitis
  - Obtain serum acetaminophen (paracetamol) levels
  - Obtain a more detailed history of:
    - Prior and/or concurrent diseases or illness
    - Exposure to environmental and/or industrial chemical agents
    - Symptoms (if applicable) including right upper quadrant pain, hypersensitivity-type reactions, fatigue, nausea, vomiting and fever
    - Prior and/or concurrent use of alcohol, recreational drugs and special diets

        - Concomitant use of medications (including non-prescription medicines and herbal and dietary supplements), plants, and mushrooms
    - Obtain viral serologies
    - Obtain CPK, haptoglobin, LDH, and peripheral blood smear
    - Perform appropriate liver imaging if clinically indicated
- Obtain appropriate blood sampling for pharmacokinetic analysis if this has not already been collected
- Obtain hepatology consult (liver biopsy may be considered in consultation with an hepatologist)
- Follow the subject and the laboratory tests (ALT, AST, TBL, INR) until all laboratory abnormalities return to baseline or normal. The "close observation period" is to continue for a minimum of 4 weeks after discontinuation of all investigational product(s) and protocol-required therapies.

The potential DILI event and additional information such as medical history, concomitant medications and laboratory results must be captured in corresponding CRFs.

APPENDIX VI: [redacted]

[redacted]

[redacted]

APPENDIX VII: THE FNCLCC SARCOMA GRADING SYSTEM

Tumour differentiation • Score 1: Sarcomas closely resembling normal adult mesenchymal tissue and potentially difficult to distinguish from the counterpart benign tumor (eg, well-differentiated liposarcoma, well-differentiated leiomyosarcoma) • Score 2: Sarcomas for which histologic typing is certain (eg, myxoid, liposarcoma, myxofibrosarcoma) • Score 3: Embryonal and undifferentiated sarcomas, synovial sarcomas, sarcomas of doubtful type, synovial sarcomas
Mitotic count (established on 10 high-power field – HPF; a HPF measures 0.1734 mm^2) • Score 1: 0 to 9 mitoses per 10 HPF • Score 2: 10 to 19 mitoses per 10 HPF • Score 3: ≥ 20 mitoses per 10 HPF
Tumour necrosis • Score 0: no necrosis • Score 1: < 50% of tumour necrosis • Score 2: ≥ 50% of tumour necrosis
Histologic grade • Grade 1: total score 2, 3 • Grade 2: total score 4, 5 • Grade 3: total score 6, 7, 8

REFERENCE:

Guillou L, Coindre JM, Bonichon F, et al: Comparative study of the National Cancer Institute and French Federation of Cancer Centers Sarcoma Group grading systems in a population of 410 adult patients with soft tissue sarcoma. J Clin Oncol 15:350-62, 1997

Trojani M, Contesso G, Coindre JM, et al: Soft-tissue sarcomas of adults; study of pathological prognostic variables and definition of a histopathological grading system. Int J Cancer 33:37-42, 1984

APPENDIX VIII: WORLD HEALTH ORGANIZATION CLASSIFICATION OF SOFT TISSUE TUMOURS: INTERMEDIATE (RARELY METASTASTIZING) AND MALIGNANT TUMOURS, 2013

Adapted from Fletcher CD, et al. World Health Organization Classification of Tumours, Pathology, and Genetics. Tumours of Soft Tissue and Bone. IARC Press, Lyon, 2013

	Malignant
	ADIPOCYTIC TUMORS
Atypical lipomatous tumour / Well differentiated liposarcoma	
	Dedifferentiated liposarcoma
	Myxoid liposarcoma
	Pleomorphic liposarcoma
	Liposarcoma, not otherwise specified
	FIBROBLASTIC/MYOFIBROBLASTIC TUMORS
	Adult fibrosarcoma
Solitary fibrous tumour	Malignant solitary fibrous tumour
Inflammatory myofibroblastic tumour	Myxofibrosarcoma
Dermatofibrosarcoma protuberans*	Low grade fibromyxoid sarcoma*
Fibrosarcomatous dermatofibrosarcoma protuberans*	
Pigmented dermatofibrosarcoma protuberans*	
Low grade myofibroblastic sarcoma	Sclerosing epithelioid fibrosarcoma
Myxoinflammatory fibroblastic sarcoma	
Atypical myxoinflammatory fibroblastic tumour	
Infantile fibrosarcoma*	
	SMOOTH MUSCLE TUMORS
	Leiomyosarcoma
	PERICYTIC (PERIVASCULAR) TUMORS
	Malignant glomus tumour
	SKELETAL MUSCLE TUMORS
	Embryonal rhabdomyosarcoma*
	Alveolar rhabdomyosarcoma*
	Pleomorphic rhabdomyosarcoma*
	Spindle cell/sclerosing rhabdomyosarcoma*
	Botryoid rhabdomyosarcoma*

	Mixed rhabdomyosarcoma*
	Rhabdomyosarcom**a, insufficient sample to categorize*
VASCULAR TUMORS	
Retiform hemangioendothelioma*	Epithelioid hemangioendothelioma*
Papillary intralymphatic angioendothelioma*	Angiosarcoma of soft tissue
Composite hemangioendothelioma*	
Pseudomyogenic (epithelioid sarcoma-like) hemangioendothelioma*	
Kaposi sarcoma*	

	CHONDRO-OSSEOUS TUMORS
	Mesenchymal chondrosarcoma
	Extraskeletal osteosarcoma *
	NERVE SHEATH TUMORS
	Malignant peripheral nerve sheath tumor
	Epithelioid malignant peripheral nerve sheath tumour
	Malignant Triton tumour
	Malignant granular cell tumor
	Ectomesenchymoma*
	TUMORS OF UNCERTAIN DIFFERENTIATION
Atypical fibroxanthoma	
Angiomatoid fibrous histiocytoma	Synovial sarcoma
Ossifying fibromyxoid tumor	Malignant ossifying fibromyxoid tumour
	Epithelioid sarcoma
Mixed Tumour NOS	Malignant mixed tumour
Myoepithelioma	Myoepithelial carcinoma
Phosphaturic mesenchymal tumour	Malignant phosphaturic mesenchymal tumour
	Alveolar soft part sarcoma
	Clear cell sarcoma of soft tissue
	Extraskeletal myxoid chondrosarcoma
	Extraskeletal Ewing sarcoma*
	Desmoplastic small round cell tumor*
	Extra-renal rhabdoid tumor*
	Neoplasms with perivascular epithelioid cell differentiation (PEComa)
	Intimal sarcoma
	VISCERAL TUMORS
	Embryonal sarcoma (undifferentiated sarcoma) of the liver
	UNDIFFERENTIATED SARCOMAS
	Undifferentiated round cell sarcoma

	Undifferentiated pleomorphic sarcoma
	Undifferentiated epithelioid sarcoma
	Undifferentiated spindle cell sarcoma
	Undifferentiated sarcoma NOS

*These tumor types are NOT eligible for the NRG-DT001 protocol.

APPENDIX IX: NRG-DT001 SPECIMEN TRANSMITTAL CLINICAL TESTING: TP53 SEQUENCING (01-DEC-2021)

The information on this form is required for results reporting from the CLIA laboratory. This form MUST be completed and submitted with the corresponding pathology report and the specimens for TP53 Testing. Failure to submit this form or the pathology report will result in delays in tissue processing and in the reporting of results to the ordering physician. FAX transmissions and hard copies of laboratory results will be sent ONLY to the ordering physician, as specified below.

PATIENT INFORMATION

Patient Study ID: _____ **Patient Initials:** _____ (First Last)

Patient Gender: ☐ Male ☐ Female

Diagnosis: _____

INSTITUTION INFORMATION

Treatment Institution: _____

Referring Physician: _____ **Physician NPI#:** _____

Physician Phone Number: _____ **Physician Fax Number:** _____

Physician Email: _____

Physician Address: _____

_____

_____

Contact Person Name: _____ **Contact Phone Number:** _____

Note: Contact Person listed must be someone who can answer questions about the specimens and/or shipment.

Contact Fax Number: _____

Contact Email: _____

Contact Address: _____
(if different from above)

SPECIMEN INFORMATION

Specimen Type: ☐ Primary ☐ Metastatic

Time Point: ☐ Archival ☐ Other _____

	Record number of each type of specimen present in shipment.				
	# of Unstained Slides	*# of Stained Slides*	*Surgical Path ID (SPID)*	*Block #*	*Date Collected*
Primary Tissue					
Metastatic Tissue *Specify Site:*					

Ship specimens to:

EET Biobank
The Research Institute at Nationwide Children's Hospital
700 Children's Drive, WA1340
Columbus, Ohio 43205
PH: (614) 722-2865
Email: BPCBank@nationwidechildrens.org