A phase II randomized study of tiragolumab plus atezolizumab versus atezolizumab in the treatment of stage II melanoma patients who are ctDNA-positive following resection

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Study Drugs: Atezolizumab (Tecentriq)
Tiragolumab

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Protocol Revision History

Initial Approval Version	**23 Nov 2021**
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Principal Investigator Signature Page

Principal Investigator (printed):

Name of Institution:

PI Signature *Date*

By my signature, I agree to personally supervise the conduct of this study and to ensure its conduct in compliance with the protocol, informed consent, IRB/HRPO procedures, the Declaration of Helsinki, ICH Good Clinical Practices guidelines, and the applicable parts of the United States Code of Federal Regulations or local regulations governing the conduct of clinical studies.

STATEMENT OF COMPLIANCE

The trial will be carried out in accordance with International Conference on Harmonisation Good Clinical Practice (ICH GCP) and the following:

- United States (US) Code of Federal Regulations (CFR) applicable to clinical studies (45 CFR Part 46, 21 CFR Part 50, 21 CFR Part 56, 21 CFR Part 312, and/or 21 CFR Part 812)

National Institutes of Health (NIH)-funded investigators and clinical trial site staff who are responsible for the conduct, management, or oversight of NIH-funded clinical trials have completed Human Subjects Protection and ICH GCP Training.

The protocol, informed consent form(s), recruitment materials, and all participant materials will be submitted to the Institutional Review Board (IRB) for review and approval. Approval of both the protocol and the consent form must be obtained before any participant is enrolled. Any amendment to the protocol will require review and approval by the IRB before the changes are implemented to the study. In addition, all changes to the consent form will be IRB-approved; a determination will be made regarding whether a new consent needs to be obtained from participants who provided consent, using a previously approved consent form.

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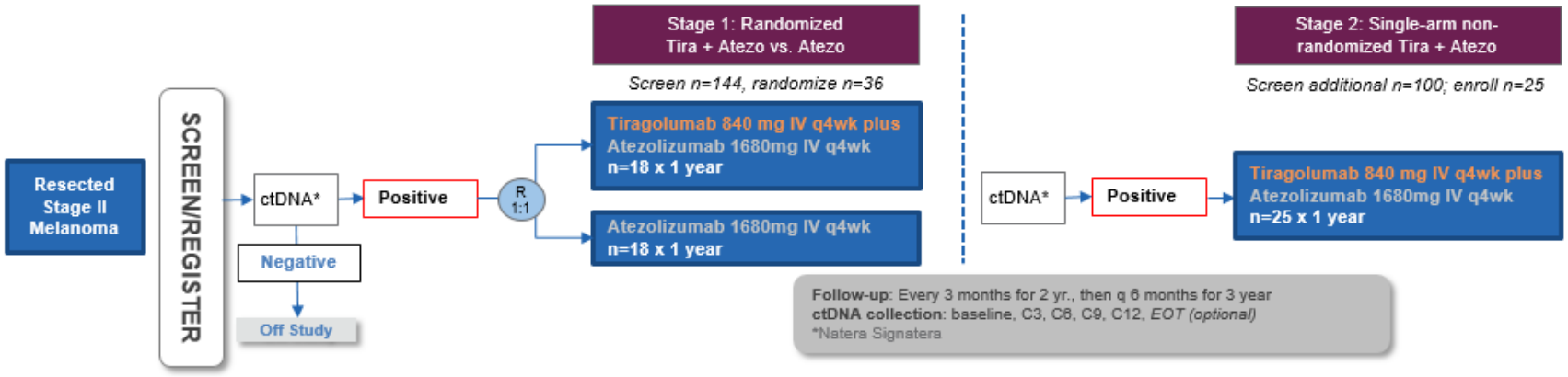
PROTOCOL SUMMARY

Title:	A phase II randomized study of tiragolumab plus atezolizumab versus atezolizumab in the treatment of stage II melanoma patients who are ctDNA-positive following resection
Study Description:	This study's hypothesis is that patients with stage II melanoma who test positive for circulating tumor DNA are at a higher risk for recurrence and therefore adjuvant treatment is justified. In this study, the blood of consenting and eligible patients will be tested for ctDNA and those patients who test positive will be randomized on a 1:1 basis to either treatment with atezolizumab and tiragolumab or atezolizumab alone. Patients who test negative for ctDNA will be observed off protocol.
Objectives:	Primary Objective: To determine the ctDNA clearance rate in patients with stage II melanoma who test positive for ctDNA and are treated with the combination of atezolizumab and tiragolumab. Secondary Objective: To determine the confirmed ctDNA negativity proportion with two consecutive negative results in patients with stage II melanoma who test positive for ctDNA and are treated with atezolizumab and tiragolumab, and those who are treated with atezolizumab alone. To determine the relapse-free survival in patients with stage II melanoma who test positive for ctDNA and are treated with atezolizumab and tiragolumab, and those who are treated with atezolizumab alone. To determine the distant metastasis-free survival in patients with stage II melanoma who test positive for ctDNA and are treated with atezolizumab and tiragolumab, and those who are treated with atezolizumab alone. To determine the overall survival of patients with stage II melanoma. To evaluate the safety and tolerability of atezolizumab and tiragolumab in patients with stage II melanoma. To assess the impact of ctDNA kinetics on outcomes (relapse-free survival, distant metastasis-free survival, and overall survival) in patients with surgically resected stage II melanoma.
Endpoints:	Primary Endpoint: ctDNA clearance rate, defined as the proportion of ctDNA-positive patients having a ctDNA-negative test at C3D1. Secondary Endpoint: Proportion of ctDNA-positive patients having a ctDNA-negative test at two consecutive measures, relapse-free

	survival (RFS), distant metastasis-free survival (DMFS), overall survival (OS), treatment-related grade 3 or greater AEs
Study Population:	During stage 1 of this study, 18 patients with stage II melanoma who have undergone complete resection and who have tested positive for ctDNA will be enrolled in each arm (atezolizumab + tiragolumab (treatment) vs. atezolizumab monotherapy (reference)) for a total of 36 patients randomized. If at least 3 patients in the atezolizumab + tiragolumab arm are shown to be ctDNA negative at C3D1, stage 2 of the study will begin enrollment. Stage 2 consists of 25 patients all enrolled to the atezolizumab + tiragolumab arm (no randomization and no atezolizumab monotherapy arm). With an anticipated ctDNA positive rate of 25%, a total of approximately 144 patients will need to be consented for ctDNA testing in stage 1 and 100 in stage 2.
Phase:	II
Description of Sites / Facilities Enrolling:	This study will be open at the Siteman Cancer Center at Washington University School of Medicine as well as other cancer centers in the United States.
Description of Study Intervention:	Patients who test positive for ctDNA will be randomized on a 1:1 basis during stage 1 to either treatment with atezolizumab and tiragolumab or treatment with atezolizumab alone. If, after the completion of stage 1, it is determined that enrollment will continue in stage 2, patients will not be randomized and all will receive treatment with atezolizumab and tiragolumab. Atezolizumab is given as an IV infusion at a dose of 1680 mg every 4 weeks and tiragolumab is given as an IV infusion at a dose of 840 mg every 4 weeks.
Study Duration:	Study duration is anticipated to be 84 months (24 months for accrual + 12 months for treatment + 36 months for follow-up + 12 months for data analysis). Primary analysis will take place after all patients have completed adjuvant therapy. Additionally, an interim analysis will occur after 18 patients have been accrued to the treatment arm (atezolizumab + tiragolumab).
Participant Duration:	Forty-eight months (12 months for treatment + 36 months for follow-up)

SCHEMA


Resected Stage II Melanoma
SCREEN/REGISTER
ctDNA*
Positive
Negative
Off Study
R 1:1
Stage 1: Randomized Tira + Atezo vs. Atezo
Screen n=144, randomize n=36
Tiragolumab 840 mg IV q4wk plus Atezolizumab 1680mg IV q4wk n=18 x 1 year
Atezolizumab 1680mg IV q4wk n=18 x 1 year
Stage 2: Single-arm non-randomized Tira + Atezo
Screen additional n=100; enroll n=25
ctDNA*
Positive
Tiragolumab 840 mg IV q4wk plus Atezolizumab 1680mg IV q4wk n=25 x 1 year
Follow-up: Every 3 months for 2 yr., then q 6 months for 3 year
ctDNA collection: baseline, C3, C6, C9, C12, EOT (optional)
*Natera Signatera

SCHEDULE OF ACTIVITIES

Cycles are 28 days in duration. Step 2 screening may take place up to 28 days prior to C1D1 except where noted. Safety assessments may take place +/- 3 days from time point.

	Step 1 Screening	**Step 2 Screening**	**D1 of each cycle**	**Q12W[1]**	**EOT**	**F/U[2]**
Screening consent	X					
Treatment consent		X				
Review of medical record	X					
Confirmation of resection	X					
H&P, PS, wt		X	X		X	
CBC		X	X			
CMP, LDH, amylase, lipase		X	X			
TSH, free T3 or total T3, free T4		X	X			
INR, aPTT[9]		X				
EBV serology	X[11]					
HIV serology	X[11]					
HBV serology	X[11]					
HCV serology	X[11]					
Pregnancy test[3]		X	X			
Urinalysis reflex to micro and/or culture[9]		X				
ECG		X	X[12]			
Randomization[6]		X				
Blood for ctDNA (Signatera testing)		X[5]	X[8]		X[10]	
Whole blood and tissue for whole exome sequencing (for design of Signatera assay)	X[13]					
Atezolizumab			X[4]			
Tiragolumab[7]			X[4]			
CT scan		X		X[2]		X
AE assessment			Baseline through 90 days after last day of treatment			

1. corresponds with the end of every 3rd cycle; if there are treatment delays, imaging should continue to be done very 12 weeks rather than waiting for the end of every 3rd cycle
2. Q12W during Years 1 and 2 after surgery, Q24W during Year 3 after surgery; depending on when a patient discontinues treatment, follow-up may be Q12W or Q24W; follow-up will discontinue after 3 years have elapsed from date of surgery
3. women of childbearing potential only
4. treatment can continue for up to 13 cycles; refer to Section 5.2 for instructions regarding monitoring of vital signs and post-infusion observation
5. must take place 4-12 weeks after surgery; if ctDNA positive, patients will be randomized to receive either atezolizumab and tiragolumab or atezolizumab monotherapy, and if ctDNA negative, patient will not continue in this study
6. stage 1 only; randomization must occur no more than 16 weeks after date of surgery
7. if randomized to receive tiragolumab during stage 1; all patients during stage 2.
8. prior to dosing on C3D1, C6D1, C9D1, C12D1 – the collection of blood for ctDNA should be discontinued after 2 consecutive negative tests
9. required at Step 2 screening and thereafter only as clinically indicated
10. optional

11. may take place up to 42 days prior to C1D1
12. ECG should be performed at Step 2 screening, C3D1, C6D1, C9D1, and C12D1
13. should be collected around time of resection; blood may be drawn for WES and ctDNA testing at the same time to consolidate blood draws; if the blood draw for WES takes place before 4 weeks post-resection, a separate draw is required for ctDNA testing

1.0 INTRODUCTION

1.1 Stage II Melanoma

Recent clinical trials in melanoma have focused on treating high risk stage III patients with programmed cell death inhibitor-1 (PD-1) inhibitors and combination BRAF and MEK inhibitors in the adjuvant setting to decrease the risk of recurrence and improve distant metastasis-free survival (DMFS) and overall survival (OS) (S1404/KN053, EORTC1325/KN054, CheckMate 238, COMBI-AD). Adult patients with stage III A, B, or C melanoma who received 18 doses (~1 year) of pembrolizumab post-resection in EORTC1325/KN054 had a significantly longer recurrence-free survival (RFS) than placebo [hazard ratio (HR) = 0.57; 98.4% confidence interval (CI), 0.43-0.74; p<0.0001][1]. In CheckMate 238, resected stage IIIB, IIIC and IV melanoma patients have improved RFS and fewer Grade 3 and 4 adverse events (AEs) after one year of adjuvant treatment with nivolumab compared to patients treated with ipilimumab, which was previously approved by the Food and Drug Administration as adjuvant therapy. COMBI-AD has shown RFS benefit with one year of adjuvant treatment using dabrafenib and trametinib in patients whose tumors harbor BRAF V600E or V600K mutations with Stage IIIA, B, and C melanoma.

All stage II patients are at risk of recurrence after complete surgical resection, which is considered standard of care in this patient population. Interferon (IFN) alpha as adjuvant therapy in melanoma has shown modest results at best (consistent effect on RFS with a maintained HR of 0.86 and approximately a 3% absolute survival advantage at 5 years)[2]. IFN alpha is a regimen with well described significant toxicities, particularly at high-dose, and these can profoundly affect quality of life and can even be life threatening. With high-dose IFN, approximately 40% of patients have treatment-related AEs that lead to dose delays or reductions[3]. Adjuvant IFN offers a reduction in RFS but is not universally recommended because of the significant toxicity associated with treatment that can affect quality of life[4]. Pegylated IFN alpha 2b was approved by the Food and Drug Administration on 29-MAR-2011 for adjuvant therapy. Today IFN alpha has a limited role in adjuvant treatment of melanoma and is now recommended only to patients with ulcerated primary melanomas and to patients without access to more modern treatments[1].

Some patients with stage II melanoma have OS outcomes similar to those of stage III patients. Data from the American Joint Committee on Cancer suggest that 94% of stage IIA, 87% of stage IIB, and 82% of stage IIC patients will be alive at 5 years and by 10 years 88, 82, and 75% respectively.

The stage IIIB 5-year OS rate of 83% is comparable to stage IIC OS at 5 years (82%). Similarly, stage IIIA (88%) and IIIB (77%) OS at 10 years are comparable to stage IIA (88%) and IIC (75%) OS rate at 10 years, respectively[5]. These data clearly show that survival outcomes can be as dismal for some stage II patients as for stage III patients.

Despite the increased risk of recurrence, adjuvant therapy is not generally recommended for stage II melanoma because of the associated toxicity and the relatively favorable

prognosis. Therefore, we propose to treat these patients based on a biomarker (circulating tumor DNA) that can enrich for a group of patients with the highest risk of recurrence rather treating all comers and exposing some patients to unnecessary toxicities.

This population of high risk of recurrence thus has an unmet medical need for adjuvant therapy with the goal of preventing disease recurrence and increasing survival by providing treatment with adjuvant atezolizumab and tiragolumab.

1.2 Circulating Tumor DNA and Risk of Relapse

In the evolving paradigm of effective adjuvant therapy in melanoma, it is essential to develop biomarkers identifying patients at high risk of relapse. Currently, features of the primary tumor such as ulceration, Breslow and number of mitoses in addition to nodal classification and disease stage are with a poor outcome in stages I–III melanoma; however, patient numbers in these studies were small and have yet to be confirmed in larger cohorts[6, 7].

Lee et al. showed that detecting circulating tumor DNA (ctDNA) in plasma taken within 12 weeks of curative intent surgery is highly predictive of relapse in patients with stage II/III melanoma. The majority of patients with detectable ctDNA relapsed within 1 year of surgery, suggesting that ctDNA in the plasma can reveal occult metastatic disease that is not evident on radiological imaging. Notably, they were able to identify melanoma patients at high risk of both distant metastatic relapse and local recurrence, which is consistent with studies showing that ctDNA can signal micrometastatic disease after neoadjuvant chemotherapy postsurgical resection in breast cancer and following surgery for stage II colorectal cancer[8]

In the same study, ctDNA was independent of standard staging indices in predicting disease recurrence, demonstrating the value of this approach in melanoma.

The ability to predict progression to stage IV disease is extremely important in light of recent findings that immune checkpoint inhibition improves RFS in stage III melanoma. Detection of ctDNA allows identification of a subgroup of patients at high risk of early relapse and inferior survival, allowing stratification of patients to adjuvant regimens associated with higher toxicity but greater potential for efficacy. Taken at a single time-point following surgery, it can add to AJCC staging in informing individual prognosis and therefore discussion regarding risks and benefits of adjuvant therapy, while longitudinal sampling will likely improve the ability to detect disease progression before radiological imaging.

We advocate that the findings from Lee et al. be confirmed in clinical trials investigating treatment responses in this population in order to evaluate whether it is also a predictive biomarker for response to immune or targeted therapy.

Also, in stage IIIA-D, circulating tumor cell (CTC) detection was not associated with sub-stage, or primary tumor characteristics. Multivariable analysis demonstrated that the

detection of ≥1 baseline CTC was significantly associated with decreased 6-month RFS [log-rank, $P < 0.0001$; HR, 3.62, 95% confidence interval (CI), 1.78-7.36; $P < 0.0001$] and 54-month RFS (log-rank, $P = 0.01$; HR, 1.69; 95% CI, 1.13-2.54; $P = 0.01$).[9]

In fact, the use of ctDNA and CTCs has been compared for *KRAS* mutation detection in 82 patients with lung cancer.[10] Results showed that at a diagnostic specificity of 95%, ctDNA had a diagnostic sensitivity (detection rate) of 96%. By contrast, CTCs had an inferior diagnostic sensitivity of 52% and a specificity of 88%.

In addition, the prognostic value of *BRAF* mutation-positive ctDNA was investigated in patients with melanoma.[11] *BRAF* mutations are observed in approximately 50% of the melanoma tumor samples. In this study, 732 patients with *BRAF* V600 mutation-positive melanoma treated mainly with dabrafenib were evaluated. The mutational status determined by BEAMing (Beads, Emulsification, Amplification, and Magnetics) technology and tumor tissue was compared. *BRAF* mutations were detectable in ctDNA in 76%/81% of late-stage patients with *BRAF* V600E/V600K-positive tumors. Patients with *BRAF* mutation-positive tumors but negative for *BRAF*-mutant ctDNA at baseline had longer progression-free survival and overall survival compared to patients with a positive baseline ctDNA result. These results suggest that ctDNA alone seems not to be suitable as the principal screening method for patients with unknown *BRAF* mutation status, but the presence and amount of ctDNA has prognostic value.

Thus, compared to CTCs, ctDNA containing tumor biomarkers is easier to isolate from blood. ctDNA is thought to contain single or double-stranded fragmented (~150 bp) DNA derived from populations of normal and tumor cells. DNA mutations and epigenetic modifications such as DNA methylation derived from ctDNA have been frequently studied in melanoma and its application in identifying MRD can complement detection and enumeration of melanoma CTCs. ctDNA detection of single biomarkers such as *BRAF* or *NRAS* hotspot mutations have been useful in predicting immunotherapy response. Levels of ctDNA mutations have been found to be associated with tumor burden and OS.

Like the current CTC assays, ctDNA assays also face the same issue with need for increased sensitivity for detecting ctDNA biomarkers in early-stage melanoma patients where there is high tumor heterogeneity. One approach to overcome tumor heterogeneity is to utilize Signatera.

Signatera is a personalized, custom-built ctDNA assay for MRD assessment and evaluation of disease recurrence and treatment response, with the ability to detect somatic variants at a variant allele frequency (VAF) of 0.01% in ctDNA isolated from plasma. [12-15] Signatera offers several advantages over static liquid biopsy panels. Given the heterogeneity of cancer, static liquid biopsy panels that target the same set of genes for each patient can track on average ~3 mutations from a given individual's primary tumor. [16-18] Moreover, many mutations identified in these panels may not be tumor derived, making such approaches less specific.[16-18] Also, the targeted mutations may be driver or subclonal mutations which may be susceptible to treatment. Thus, treatment may lead to selective

attrition of cancer cells that contain these susceptible mutations, thereby reducing the ability of that mutation to track overall cancer burden. In this regard, the Signatera assay has an advantage that it prioritizes the detection of clonal, passenger mutations. Clonal variants occur early in tumor evolution and remain present in every cancer cell as the tumor evolves.[19] Most clonal variants are passenger mutations (not driver mutations), i.e. these mutations have no effect on the fitness of a clone but are associated with clonal expansion, and thus are truly representative of the overall tumor burden. These clonal variants are unique to each individual tumor. Identifying and tracking a subset of clonal variants provides an opportunity to detect residual disease with high sensitivity and specificity, irrespective of tumor heterogeneity.

1.3 Signatera Assay – Test Principle

To detect residual disease or to determine cancer recurrence with ctDNA, the Signatera process starts with sequencing the tumor tissue (resection or shave biopsy) and matched normal (whole blood) sample for each patient. Based on the sequencing results, a list of somatic SNVs specific to each patient are identified bioinformatically. Next, 16 SNVs are selected for multiplex PCR primer design based on several factors, including the clonality, and detectability of the mutations identified in the tumor tissue DNA. Regardless of the tumor type, the 16 SNV targets selected are unique to each patient with <1% of patients sharing any SNV target. PCR primers targeting the personalized 16 SNVs are generated and stored for testing. Then, two tubes of blood are drawn from the patient, the plasma is isolated, and the DNA, possibly including some ctDNA, is extracted. A universal library is then generated from plasma ctDNA, wherein the ctDNA is amplified. An aliquot of the universal library is used as an input for amplification of 16 patient-specific targets using multiplex PCR (mPCR). The mPCR product is barcoded, pooled, and sequenced on the Illumina HiSeq 2500. Sequenced data that passes the quality control (QC) goes through ctDNA analysis algorithm followed by issuance of a signed report by a Lab Director that contains information on the analyzed data, indicating the presence or absence of ctDNA.[12,15,17,19]

1.4 Atezolizumab

Atezolizumab is a humanized IgG1 monoclonal antibody that targets PD-L1 and inhibits the interaction between PD-L1 and its receptors, PD-1 and B7-1 (also known as CD80), both of which function as inhibitory receptors expressed on T cells. Therapeutic blockade of PD-L1 binding by atezolizumab has been shown to enhance the magnitude and quality of tumor-specific T-cell responses, resulting in improved anti tumor activity (Fehrenbacher et al. 2016; Rosenberg et al. 2016). Atezolizumab has minimal binding to fragment crystallizable (Fc) receptors, thus eliminating detectable Fc effector function and associated antibody-mediated clearance of activated effector T cells.

Atezolizumab shows antitumor activity in both nonclinical models and cancer patients and is being investigated as a potential therapy in a wide variety of malignancies. Atezolizumab is being studied as a single agent in the advanced cancer and adjuvant therapy settings, as well as in combination with chemotherapy, targeted therapy, and CIT.

Atezolizumab is approved for the treatment of urothelial carcinoma, NSCLC, small-cell lung cancer, and triple-negative breast cancer.

Refer to the Atezolizumab Investigator's Brochure for details on nonclinical and clinical studies.

1.5 Tiragolumab

Tiragolumab is a fully human IgG1/kappa mAb that binds TIGIT (T-cell Immunoreceptor with Immunoglobulin and Immunoreceptor Tyrosine-Based Inhibition Motif [ITIM] domains) and prevents its interaction with PVR (Poliovirus Receptor). The available nonclinical and clinical data provide a strong rationale for evaluating the potential clinical benefit of tiragolumab in cancer patients. Blockade of TIGIT by tiragolumab represents a novel strategy for cancer therapy because its mechanism of action is complementary to that of other checkpoint inhibitors, such as anti-PD-L1/PD-1. Consequently, tiragolumab as a single-agent or in combination with anti-cancer therapies, may enhance the magnitude and quality of tumor-specific T-cell responses, which may result in meaningful anti-tumor activity in solid tumors and in hematological malignancies.

In the Phase II GO40290 study (CITYSCAPE), the primary analysis (clinical cutoff of 30 June 2019) showed that the combination of tiragolumab plus atezolizumab improved ORR and PFS compared to placebo plus atezolizumab in the ITT population. ORR for tiragolumab plus atezolizumab was 31.3% (95% CI: 19.5, 43.2) compared to placebo plus atezolizumab which was 16.2% (95% CI: 6.7, 25.7). Investigator-assessed PFS for tiragolumab plus atezolizumab was 5.4 months (95% CI: 4.2, not reached) compared to placebo plus atezolizumab which was 3.6 months (95% CI: 2.7, 4.4), with a HR of 0.57 (95% CI: 0.37, 0.90).

Based on the above findings and results, the combination of atezolizumab and tiragolumab is of interest to us to investigate in enriched group of stage II melanoma with positive ctDNA with the intent to maximize the benefit and render the toxicity of treatment only to those who are in most need for therapy.

1.6 Background on Blockade of the TIGIT Pathway in Cancer as a Potential Anti-Cancer Therapy

TIGIT is a novel immune inhibitory receptor that is a member of the Ig super family (Yu et al. 2009; Manieri et al. 2017). TIGIT expression is elevated in the tumor microenvironment in many human tumors, is coordinately expressed with other checkpoint immune receptors such as PD 1 and is associated with impaired T cell function and anti tumor immunity (Johnston et al. 2014). Activation of TIGIT on T cells and NK cells limits cellular proliferation, effector cytokine production, and killing of target tumor cells (TCs) (Stanietsky et al. 2009; Yu et al. 2009; Johnston et al. 2014; Wang et al. 2015; Manieri et al. 2017).

TIGIT is expressed in a wide variety of human tumors and is highly correlated with T cell

infiltration and PD-1 expression (Johnston et al. 2014). Fluorescence activated cell sorting analysis of fresh tumor samples showed that TIGIT and PD-1 are also co expressed on tumor infiltrating T cells. TIGIT expression ranges from 30% to 80% and from 50% to 80% on tumor infiltrating CD4+ and CD8+ T cells, respectively (Johnston et al. 2014).

Therefore, TIGIT is a potential target for therapeutic intervention aimed at restoring the immune response against the tumor. Agents that inhibit the activity of TIGIT may relieve an important source of tumor associated immune suppression and may enhance the activity of other immune based therapies, such as atezolizumab, an inhibitor of PD-L1. Early nonclinical results using genetically deficient mice and blocking antibodies reveal a key role for TIGIT in regulating T cell responses. Together the data support the hypothesis that anti-TIGIT in combination with anti-PD-L1 may reactivate anti tumor immunity to provide clinical benefit to patients.

1.7 Combined Inhibition of the TIGIT and PD-L1/PD-1 Pathways as Potential Anti-Cancer Therapy

The inhibitory immunoreceptor TIGIT has been shown to limit the effector function of tumor associated lymphocytes. Activation of TIGIT on T cells and NK cells limits proliferation, effector cytokine production, and killing of target TCs. Therefore, in the context of the tumor microenvironment, TIGIT acts to limit anti-tumor immune responses. Interference with TIGIT-PVR interaction may enhance the magnitude and quality of the tumor specific T cell responses through increased expansion of T cells as well as improved T cell priming and/or effector function. Because TIGIT and PD 1 are co-expressed by infiltrating T cells in several human tumors, inhibition of the TIGIT/PVR pathway may complement and potentiate the anti-tumor activity of a PD L1 pathway inhibitor such as atezolizumab.

The combined inhibition of the TIGIT and PD-L1/PD-1 pathways by tiragolumab and atezolizumab, respectively, has demonstrated promising clinical activity in the Phase I study GO30103 and the Phase II study GO40290 (hereafter referred to as CITYSCAPE). Study GO30103 is a first-in-human, combined Phase Ia/Phase Ib, open-label, dose escalation, multicenter study. The study evaluated the safety, tolerability, immunogenicity, pharmacokinetics, exploratory pharmacodynamics, and preliminary evidence of biologic activity of tiragolumab administered as a single agent (Phase Ia) or in combination with atezolizumab (Phase Ib) to patients with locally advanced or metastatic malignancies. As of December 2, 2019, 171 patients with multiple tumor types, including NSCLC, had been enrolled in the Phase Ib portion of the study. Objective responses, including complete responses (CR) in 4 patients, and partial responses (PR) in 23 patients, have been observed.

No maximum tolerated dose (MTD), no dose-limited toxicities (DLTs), and no clear dose-related trends in the incidence or severity of adverse events have been determined for single-agent tiragolumab (2-1200 mg Q3W) or tiragolumab (2-1200 mg Q3W) in combination with atezolizumab (1200 mg Q3W) in Study GO30103.

Tiragolumab was further evaluated in patients with PD-L1 selected advanced NSCLC

(tumor proportion score [TPS] ≥ 1%) in the Phase II, global, randomized, double-blind, placebo-controlled CITYSCAPE study. As of the clinical cutoff date of Dec 2, 2019, the confirmed ORR in the intent-to-treat (ITT) population was higher in the tiragolumab combined with atezolizumab arm (37%) than in the placebo combined with atezolizumab arm (21%). Investigator assessed PFS was also improved with a stratified HR of 0.58 (95% CI: 0.38 to 0.89), with a median PFS not estimable and 3.9 months in the tiragolumab combined with atezolizumab arm compared to the placebo combined with atezolizumab arm, respectively. Responses to tiragolumab in combination with atezolizumab were observed in patients with both squamous and non-squamous histology (Rodriguez-Abreu et al, 2020).

As of the clinical cutoff date of December 2, 2019 in the CITYSCAPE study, there were 135 safety evaluable patients. The safety profile was comparable between the tiragolumab combined with atezolizumab arm and the placebo combined with atezolizumab arm in terms of all-grade adverse events (99% vs 96%), Grade ≥ 3 adverse events (48% vs 44%), Grade 5 adverse events (4.5% vs 7.4%), serious adverse events (37% vs 35%), and adverse events leading to study treatment withdrawal (10.4% vs 8.8%). Study treatment related adverse events occurred at a higher frequency in the tiragolumab combined with atezolizumab arm (82%) compared to the placebo combined with atezolizumab arm (72%).

Analyzed with a comprehensive medical concepts strategy, immune-mediated adverse events were reported with a higher frequency in the tiragolumab combined with atezolizumab arm (69%) compared to the placebo combined with atezolizumab arm (47%). The difference (≥ 10% difference between arms) was predominantly attributed to events of immune mediated rash (preferred terms of rash, rash maculopapular, dermatitis, erythema, eczema, pruritic rash, folliculitis and skin ulcer) (40% vs 15%) and infusion related reactions (preferred term of infusion related reaction) (30% vs 10%).

Tiragolumab is being investigated in clinical studies as a potential therapy against various tumor types. Refer to the Tiragolumab Investigator's Brochure for details on the nonclinical and clinical studies for tiragolumab.

1.8 Study Rationale and Benefit/Risk Assessment

1.8.1 Study Rationale

TIGIT is an inhibitory immunoreceptor that can limit the effector function of tumor associated lymphocytes. Unlike other inhibitory co receptors, TIGIT is often coordinately expressed with PD-1 on tumor-infiltrating T cells in multiple tumors. In the Phase Ib portion of Study GO30103, evaluating tiragolumab in combination with atezolizumab, partial responses (PRs) occurred in patients with metastatic cancers, with varying degrees of PD-L1 and/or TIGIT expression (Bendell et al. 2020). The combination of atezolizumab with tiragolumab was tolerated in the Phase Ib portion of the study, with a safety profile consistent with prior observations of atezolizumab.

In the primary analysis of the Phase II CITYSCAPE study, in all randomized patients whose lung cancer had a PD-L1 tumor proportion score (TPS) ≥ 1% (n = 135), the confirmed objective response rate (ORR) was higher in the tiragolumab plus atezolizumab group (31.3%) than in the placebo plus atezolizumab group (16.2%) (Rodriguez-Abreu et al. 2020). Furthermore, investigator-assessed progression-free survival (PFS) was improved in the tiragolumab plus atezolizumab group (n = 67) relative to placebo plus atezolizumab group (n = 68) (stratified HR = 0.57; 95% CI: 0.37 to 0.90; median PFS 5.4 vs. 3.6 months, respectively). The greatest efficacy was observed in patients whose tumors had high PD-L1 expression (TPS ≥ 50%; n = 58) with clear separation of the Kaplan-Meier curve (Rodriguez-Abreu et al. 2020) for whom the confirmed ORR was 55.2% in the tiragolumab plus atezolizumab group and 17.2% in the placebo plus atezolizumab group (stratified HR = 0.33; 95% CI: 0.15 to 0.72; median PFS not yet reached vs. 3.88 months, respectively).

Consistent with the Phase Ib portion of Study GO30103, the combination of atezolizumab with tiragolumab was tolerated in the Phase II study (for more details, refer to the Tiragolumab Investigator's Brochure). Atezolizumab plus tiragolumab demonstrated an overall safety profile similar to that of atezolizumab alone in terms of all Grade adverse events, Grade 3 and 4 adverse events, serious adverse events and adverse events leading to study treatment discontinuation. While adverse events related to any study treatment and adverse events leading to dose interruption of any study treatment were higher in the tiragolumab plus atezolizumab arm, there was no increase in Grade 5 adverse events (see the Tiragolumab Investigator's Brochure). Adverse events with potentially immune-mediated causes have been observed with a higher frequency for tiragolumab in combination with atezolizumab in the CITYSCAPE study. However, the imbalance was mostly attributed to Grade 1 and 2 rashes and infusion-related reactions (IRRs). Grade 3 and 4 immune-mediated adverse events were similar between the tiragolumab plus atezolizumab treatment group compared with atezolizumab treatment alone. To date, immune-mediated adverse events have been manageable with standard medical practice supplemented with corticosteroids, immunosuppressive agents, and/or hormone replacement therapy.

1.8.2 Rationale for Atezolizumab / Tiragolumab Dose and Schedule

Atezolizumab will be administered at a fixed dose of 1680 mg Q4W (1680 mg on Day 1 of each 28-day cycle), which is an approved dosage for atezolizumab, as outlined in the prescribing information. The average concentration following the 1680 mg Q4W dosage is expected to be equivalent to that of 1200 mg Q3W. Anti-tumor activity has been observed across doses ranging from 1 mg/kg to 20 mg/kg Q3W. In Study PCD4989g, the maximum tolerated dose of atezolizumab was not reached and no DLTs were observed at any dose. The fixed dose of 1200 mg Q3W (equivalent to an average body weight-based dose of 15 mg/kg Q3W) was selected on the basis of both nonclinical studies (Deng et al. 2016) and available clinical pharmacokinetic, efficacy, and safety data (refer to the Atezolizumab Investigator's

Brochure for details).

Tiragolumab will be administered to patients at a fixed dose of 840 mg Q4W by IV infusion on Day 1 of each 28-day cycle. The 840-mg Q4W dosing regimen is supported by results from PK modeling and simulation and exposure-safety analyses. Briefly, the average and trough concentrations following the 840-mg Q4W dosing regimen are predicted to be similar to those of the 600-mg Q3W dosing regimen, which was evaluated in Studies GO30103 and GO40290. The predicted maximum serum concentration (Cmax) of the 840-mg Q4W dosing regimen is 28% higher at steady state relative to the predicted Cmax of the 600-mg Q3W dosing regimen, but falls within the range of observed exposure of the highest administered dose (1200 mg Q3W in Study GO30103). A preliminary analysis of the tiragolumab exposure-safety relationship based on data from Study GO30103 (tiragolumab doses of 2-1200 mg Q3W administered as monotherapy or in combination with 1200 mg atezolizumab Q3W) suggests that tiragolumab does not appear to have an exposure-safety relationship. In summary, the 840-mg Q4W dosing regimen is expected to have comparable efficacy and safety as the 600-mg Q3W dosing regimen given that the predicted exposure is within the range of observed efficacious exposures and tiragolumab does not appear to have an exposure-safety relationship.

The fixed tiragolumab dose of 600 mg IV Q3W was selected for Study GO40290 on the basis of available PK, safety, and preliminary efficacy data from Study GO30103, in which patients received single-agent tiragolumab or tiragolumab plus atezolizumab. In Study GO30103, the MTD was not reached, and no DLTs were observed with tiragolumab monotherapy or in combination with 1200 mg Q3W atezolizumab (tiragolumab doses of 2-1200 mg Q3W). In addition, development of ADAs to tiragolumab was observed in < 5% of evaluable patients receiving tiragolumab (doses of 2-1200 mg Q3W) in combination with atezolizumab. Complete occupancy of peripheral TIGIT receptors on CD4+ CD8+ T cells and NK cells was observed beginning at the 30 mg Q3W dose of tiragolumab and remained sustained at all higher doses. Anti-tumor activity (as demonstrated by radiographic PRs) was observed at tiragolumab doses of 30-600 mg Q3W when given in combination with 1200 mg atezolizumab Q3W.

1.8.3 Benefit/Risk Assessment

The toxicities of atezolizumab alone and the combination of atezolizumab plus tiragolumab are expected to be similar. Immune-mediated adverse events, although reported at a higher frequency for the atezolizumab plus tiragolumab arm in the Phase II study GO40290, are generally mild, transient, monitorable, and manageable in nature.

2.0 OBJECTIVES AND ENDPOINTS

Objectives	Endpoints	Justification for Endpoints
Primary		
To determine the ctDNA clearance rate in patients with stage II melanoma who test positive for circulating tumor DNA and are treated with atezolizumab and tiragolumab.	ctDNA clearance rate, defined as the proportion of ctDNA-positive patients having a ctDNA-negative test at C3D1.	Immune checkpoint blockade (ICB) provides clinical benefit to a subset of patients with cancer. However, existing biomarkers do not reliably predict treatment response across diverse cancer types. Bratman et al showed how serial circulating tumor DNA (ctDNA) testing may perform as a predictive biomarker in patients receiving ICB. Authors ran a prospective phase II clinical trial to assess ctDNA in five distinct cohorts of patients (including melanoma) with advanced solid tumors treated with pembrolizumab (NCT02644369). They applied bespoke ctDNA assays to 316 serial plasma samples obtained at baseline and every three cycles from 94 patients. Baseline ctDNA concentration correlated with progression-free survival, overall survival, clinical response and clinical benefit. This association became stronger when considering ctDNA kinetics during treatment. All 12 patients with ctDNA clearance during treatment were alive

		with median 25 months follow up. This study demonstrates the potential for broad clinical utility of ctDNA-based surveillance in patients treated with ICB.[22]
Secondary		
To determine the confirmed ctDNA negativity proportion with two consecutive negative results in patients with stage II melanoma who test positive for circulating tumor DNA and are treated with atezolizumab and tiragolumab, and those who are treated with atezolizumab alone.	Proportion of ctDNA-positive patients having a ctDNA-negative test at two consecutive measures	
To determine the relapse-free survival in patients with stage II melanoma who test positive for circulating tumor DNA and are treated with atezolizumab and tiragolumab, and those who are treated with atezolizumab alone	Relapse-free survival (RFS), defined as the duration of time from the date of randomization to the date of earliest disease relapse or death, whichever occurs first.	RFS is an acceptable measure of clinical benefit for a late stage study that demonstrates superiority of a new antineoplastic therapy, especially if the magnitude of the effect is large and the therapy has an acceptable risk/benefit profile.
To determine the distant metastasis-free survival in patients with stage II melanoma who test positive for circulating tumor DNA and are treated with atezolizumab and tiragolumab, and those who are treated with atezolizumab alone (both circulating tumor DNA positive and negative).	Distant-metastasis free survival (DMFS), defined as the duration of time from the date of positive ctDNA being confirmed to the date of appearance of a distant metastasis or death, whichever occurs first.	The secondary efficacy objective of this study is to compare DMFS between the two treatment arms
To determine the overall survival of patients with stage II melanoma.	Overall survival (OS), defined as the duration of time from the date of positive ctDNA being confirmed to death from any cause.	The secondary efficacy objective of this study is to compare OS between the two treatment arms
To evaluate the safety and tolerability of atezolizumab	Defined as the number of treatment-related grade 3 or	Safety parameters commonly used for

and tiragolumab in patients with stage II melanoma.	greater AEs and discontinuations due to treatment-related AEs.	evaluating investigational systemic anticancer treatments are included as safety endpoints including, but not limited to, the incidence of, causality, and outcome of AEs/serious adverse events (SAEs); and changes in vital signs and laboratory values. Adverse events will be assessed as defined by CTCAE, Version 5.0.
To assess the impact of ctDNA kinetics on outcomes (relapse-free survival, distant metastasis-free survival, and overall survival) in patients with surgically resected stage II melanoma.		

3.0 STUDY POPULATION

3.1 Step One (Signatera Assay Development) Eligibility

3.1.1 Inclusion Criteria

1. Surgically resected and histologically/pathologically confirmed stage II cutaneous melanoma. No more than 16 weeks may elapse between final surgical resection and randomization. Treatment should start only after complete wound healing from the surgery.

2. Participants must not have been previously treated for melanoma beyond complete surgical resection.

3. At least 18 years of age.

4. Ability to understand and willingness to sign an IRB approved written informed consent document (or that of legally authorized representative, if applicable).

3.1.2 Exclusion Criteria

1. A history of other malignancy with the exception of malignancies for which all treatment was completed at least 2 years before registration and the patient has no evidence of disease or malignancies in situ (such as DCIS), basal cell

carcinoma, or localized cutaneous squamous cell carcinomas.

2. Currently receiving any other investigational agents.

3. Prior history of pneumonitis.

4. A history of allergic reactions attributed to compounds of similar chemical or biologic composition to atezolizumab and tiragolumab or other agents used in the study.

5. Significant cardiovascular disease (such as New York Heart Association Class II or greater cardiac disease, myocardial infarction, or cerebrovascular accident) within 3 months prior to initiation of study treatment, unstable arrhythmia, or unstable angina.

6. Active or history of autoimmune disease or immune deficiency, including, but not limited to, myasthenia gravis, myositis, autoimmune hepatitis, systemic lupus erythematosus, rheumatoid arthritis, inflammatory bowel disease, antiphospholipid antibody syndrome, Wegener granulomatosis, Sjögren syndrome, Guillain-Barré syndrome, or multiple sclerosis, with the exceptions listed below:
   a. Patients with a history of autoimmune-related hypothyroidism who are on thyroid replacement hormone are eligible for the study.
   b. Patients with controlled Type 1 diabetes mellitus who are on an insulin regimen are eligible for the study.
   c. Patients with eczema, psoriasis, lichen simplex chronicus, or vitiligo with dermatologic manifestations only (e.g., patients with psoriatic arthritis are excluded) are eligible for the study provided all of following conditions are met:
      i. Rash must cover < 10% of body surface area
      ii. Disease is well controlled at baseline and requires only low-potency topical corticosteroids
      iii. No occurrence of acute exacerbations of the underlying condition requiring psoralen plus ultraviolet A radiation, methotrexate, retinoids, biologic agents, oral calcineurin inhibitors, or high potency or oral corticosteroids within the previous 12 months

7. Active tuberculosis.

8. Prior allogeneic stem cell or solid organ transplantation

9. Positive hepatitis B surface antigen (HBsAb) at screening.

10. Positive hepatitis C virus (HCV) antibody test at screening (unless followed by a negative HCV RNA test).

11. Current treatment with anti-viral therapy for HBV

12. Positive Epstein-Barr virus (EBV) viral capsid antigen immunoglobulin M (IgM) test at screening. An EBV PCR test should be performed as clinically indicated to screen for acute infection or suspected chronic active infection. Patients with a positive EBV PCR test are excluded.

13. Prior treatment with CD137 agonists or immune checkpoint blockade therapies, including anti-CTLA-4, anti-PD-1, anti-TIGIT, and anti-PD-L1 therapeutic antibodies.

14. History of severe allergic anaphylactic reactions to chimeric or humanized antibodies or fusion proteins

15. Patients with HIV are eligible unless their CD4+ T-cell counts are < 350 cells/mcL or they have a history of AIDS-defining opportunistic infection within the 12 months prior to registration. Concurrent treatment with effective ART according to DHHS treatment guidelines is recommended.

3.2 Step Two (Randomization and Treatment) Eligibility

3.2.1 Inclusion Criteria

1. Positive ctDNA test.

2. Normal bone marrow and organ function as defined below:
   a. Absolute neutrophil count ≥ 1,500/mcL
   b. Lymphocyte count ≥ 500/mcL
   c. Platelets ≥ 100,000/mcL
   d. Hemoglobin ≥ 9.0 g/dL (patients may be transfused to meet this criterion)
   e. Total bilirubin ≤ 1.5 x IULN or direct bilirubin ≤ IULN for participants with total bilirubin levels > 1.5 x IULN; patients with known Gilbert disease must have total bilirubin ≤ 3 x IULN
   f. AST(SGOT)/ALT(SGPT) ≤ 2.5 x IULN
   g. Alkaline phosphatase ≤ 2.5 x IULN
   h. Creatinine clearance ≥ 45 mL/min by Cockcroft-Gault
   i. Serum albumin ≥ 2.5 g/dL
   j. INR or PT ≤ 1.5 x IULN unless participant is receiving anticoagulant therapy as long as PT or aPTT is within therapeutic range of intended use of anticoagulants
   k. aPTT ≤ 1.5 x IULN unless participant is receiving anticoagulant therapy as long as PT or aPTT is within therapeutic range of intended use of anticoagulants

3. ECOG performance status ≤ 1 (see Appendix A)

4. The effects of atezolizumab and tiragolumab on the developing human fetus are unknown. For this reason, women of childbearing potential and men must agree to use adequate contraception (hormonal or barrier method of birth control, abstinence) prior to study entry and for the duration of study participation, for 90 days after the final dose of tiragolumab, and for 5 months after the final dose of atezolizumab. Should a woman become pregnant or suspect she is pregnant while participating in this study, she must inform her treating physician immediately. Men treated or enrolled on this protocol must also agree to use adequate contraception prior to the study, for the duration of the study, and 90 days after completion of the study

3.2.2 Exclusion Criteria

1. Pregnant and/or breastfeeding. Women of childbearing potential must have a negative pregnancy test within 14 days of study entry.

2. Treatment with a live, attenuated vaccine within 4 weeks prior to initiation of study treatment, or anticipation of need for such a vaccine during study treatment, within 90 days after the final dose of tiragolumab, or within 5 months after the final dose of atezolizumab

3. Treatment with systemic immunosuppressive medication (including, but not limited to, corticosteroids, cyclophosphamide, azathioprine, methotrexate, thalidomide, and anti-tumor necrosis factor-α [TNF-α] agents) within 2 weeks prior to initiation of study treatment, or anticipation of need for systemic immunosuppressive medication during study treatment, with the following exceptions:
   a. Patients who received acute, low-dose systemic immunosuppressant medication or a one-time pulse dose of systemic immunosuppressant medication (e.g., 48 hours of corticosteroids for a contrast allergy) are eligible for the study
   b. Patients who received mineralocorticoids (e.g., fludrocortisone), corticosteroids for chronic obstructive pulmonary disease (COPD) or asthma, or low-dose corticosteroids for orthostatic hypotension or adrenal insufficiency are eligible for the study.

4. Major surgical procedure within 4 weeks prior to initiation of study treatment, or anticipation of need for a major surgical procedure during the study.

5. Severe infection within 4 weeks prior to initiation of study treatment, including, but not limited to, hospitalization for complications of infection, bacteremia, or severe pneumonia, or any active infection that, in the opinion of the investigator, could impact patient safety

6. Treatment with therapeutic oral or IV antibiotics within 2 weeks prior to initiation of study treatment. Patients receiving prophylactic antibiotics (e.g.,

to prevent a urinary tract infection or chronic obstructive pulmonary disease exacerbation) are eligible for the study.

7. Treatment with systemic immunostimulatory agents (including, but not limited to, interferon and IL-2) within 4 weeks or 5 drug-elimination half-lives (whichever is longer) prior to initiation of study treatment.

3.3 Inclusion of Women and Minorities

Both men and women and members of all races and ethnic groups are eligible for this trial.

4.0 REGISTRATION PROCEDURES

Patients must not start any protocol intervention prior to registration through the Siteman Cancer Center.

Registration for this trial will be a two-step process. Step One will be screening and confirmation of eligibility for Signatera assay development. Step Two will be screening and confirmation of eligibility for randomization (if applicable) and treatment, which must occur no more than 16 weeks after resection.

Step One screening may occur as soon as pathology has confirmed that the patient has stage II cutaneous melanoma. Step One enrollment will be "On Study" in the OnCore database. Step Two screening will occur after the Signatera assay has been developed. Blood for ctDNA testing must be drawn and sent for analysis between 4 and 12 weeks after resection. It is possible for blood for assay development and blood for ctDNA testing to be drawn at the same time provided that time point is 4 to 12 weeks after resection. Step Two enrollment will be "On Treatment" in the OnCore database.

The following steps must be taken before registering patients to this study:

1. Confirmation of patient eligibility by Washington University
2. Registration of patient in the Siteman Cancer Center database
3. Assignment of unique patient number (UPN)

Once the patient has been entered in the Siteman Cancer Center OnCore database, the WUSM coordinator will forward verification of enrollment and the UPN via email.

4.1 Confirmation of Patient Eligibility

Confirm patient eligibility by collecting the information listed below and scanning and emailing it to the research coordinator listed in the *Siteman Cancer Center Clinical Trials Core Protocol Procedures for Secondary Sites* packet at least two business days prior to registering patient:

1. Your name and contact information (telephone number, fax number, and email address)
2. Your site PI's name, the registering MD's name, and your institution name
3. Patient's race, sex, and DOB
4. Three letters (or two letters and a dash) for the patient's initials
5. Currently approved protocol version date
6. Copy of signed consent form (patient name may be blacked out)
7. Planned date of enrollment
8. Completed eligibility checklist, signed and dated by a member of the study team
9. Copy of appropriate source documentation confirming patient eligibility

4.2 Patient Registration in the Siteman Cancer Center OnCore Database

Registrations may be submitted Monday through Friday between 8am and 5pm CT. Urgent late afternoon or early morning enrollments should be planned in advance and coordinated with the Washington University research coordinator. Registration will be confirmed by the research coordinator or his/her delegate by email within two business days. Verification of eligibility and registration should be kept in the patient chart.

All patients at all sites must be registered through the Siteman Cancer Center OnCore database at Washington University.

4.3 Assignment of UPN

Each patient will be identified with a unique patient number (UPN) for this study. Patients will also be identified by first, middle, and last initials. If the patient has no middle initial, a dash will be used on the case report forms (CRFs). All data will be recorded with this identification number on the appropriate CRFs. UPN will be formatted as Site No. (XXX) – Patient No. (XXX). Site No. will be assigned by the Siteman Cancer Center and is site-specific; patient no. will be assigned by the Siteman Cancer Center and is patient-specific.

4.4 Screen Failures

Screen failures are defined as participants who consent to participate in the clinical trial but are not subsequently entered in the study (i.e. who do not subsequently undergo ctDNA testing). A minimal set of screen failure information is required to ensure transparent reporting of screen failure participants, to meet the Consolidated Standards of Reporting Trials (CONSORT) publishing requirements and to respond to queries from regulatory authorities. Minimal information includes demography, screen failure details, eligibility criteria, and any serious adverse event (if applicable).

4.5 Measures to Minimize Bias: Randomization

After enrollment to stage 1, patients who are determined to be ctDNA positive will be randomized to receive either atezolizumab and tiragolumab (treatment group) or atezolizumab monotherapy (reference group) on a 1:1 basis. The study will use a simple

randomization method. This method is equivalent to tossing a coin (where the probability of a head is 0.5) to determine treatment assignment (i.e., heads - atezolizumab and tiragolumab, tails – atezolizumab). Using the computer, a sequence of random numbers will be generated which are uniformly distributed between 0 and 1 and independent of each other. For the i-th patient entering the trial, if the i-th random number ≤ 0.5, a combined treatment of atezolizumab and tiragolumab will be assigned; if the i-th random number > 0.5 then atezolizumab monotherapy will be assigned.

If it is determined (after analysis of stage 1) that enrollment will continue to stage 2, randomization will discontinue. All stage 2 patients will receive atezolizumab + tiragolumab.

4.6 Screening and Enrollment Considerations

The screening and enrollment targets outlined below should be considered in order to maintain adequate momentum towards trial completion. If at any time a screening or enrollment milestone cannot be met, the sponsor may elect to employ additional screening enhancement strategies upon discussion with Genentech/Roche, and consider stopping the study altogether due to feasibility.

	Screening Target	Sub-Site Participation	Enrollment Target
6-month milestone	20% of Stage 1 patients screened (n>28)	1 additional sub-site added	n>5 enrolled
12-month milestone	40% of Stage 1/II patients screened (n>97)	1 additional sub-site added	n>19 enrolled
18-month milestone	60% of Stage 1/II patients screened (n>144)	1 additional sub-site (optional)	n>29 enrolled
24-month milestone	80% of Stage I/II patients screened (n>195)	--	n>39 enrolled

5.0 TREATMENT PLAN

5.1 Study Intervention Description

Atezolizumab is a PD-L1 blocking antibody which is indicated for the treatment of:

- some adult patients with locally advanced or metastatic urothelial carcinoma
- adult patients with metastatic non-squamous non-small cell lung cancer with no EGFR or ALK genomic tumor aberrations (in combination with bevacizumab, paclitaxel, and carboplatin)
- adult patients with metastatic non-small cell lung cancer who have disease

progression during or following platinum-containing chemotherapy
- adult patients with extensive-stage small cell lung cancer (in combination with carboplatin and etoposide)

Tiragolumab is a fully human IgG1/kappa mAb that binds TIGIT (T-cell Immunoreceptor with Immunoglobulin and Immunoreceptor Tyrosine-Based Inhibition Motif [ITIM] domains) and prevents its interaction with PVR (Poliovirus Receptor).

5.2 Study Intervention Administration

Consenting and eligible patients will undergo a blood draw to test for ctDNA 4 to 12 weeks after surgery. Twenty mL of blood will be drawn into 2 10-mL Streck tubes as described in Section 9.0.

During stage 1 of the study, patients who are ctDNA-positive will be randomized on a 1:1 basis either to receive atezolizumab + tiragolumab or atezolizumab alone. If, after analysis of stage 1 data, it is determined that enrollment will continue to stage 2, all patients will receive atezolizumab + tiragolumab.

Atezolizumab is given as an IV infusion every 4 weeks at a dose of 1680 mg over 60 minutes (+/- 15 minutes). If this rate of infusion is tolerated without an infusion-related reaction, subsequent infusions of atezolizumab may be given over 30 minutes (+/- 10 minutes). Otherwise, the rate of infusion remains 60 minutes (+/- 15 minutes). After the first infusion of atezolizumab, the patient begins a 60-minute observation period during which vital signs should be recorded at 30 (+/-10 minutes) after infusion. If the patient tolerated the first infusion well without infusion-associated adverse events, the observation period after the next and following infusions may be reduced to 30 minutes. If the patient experienced infusion-associated adverse events in the previous infusion, the observation period should be 60 minutes. If clinically indicated, vital signs should be recorded at 30 (+/- 10) minutes after the infusion of atezolizumab.

Tiragolumab is given as an IV infusion every 4 weeks at a dose of 840 mg over 60 minutes (+/- 15 minutes). If this rate of infusion is tolerated without an infusion-related reaction, subsequent infusions of tiragolumab may be given over 30 minutes (+/- 10 minutes). Otherwise, the rate of infusion remains 60 minutes (+/- 15 minutes). After the first infusion of tiragolumab, the patient begins a 60-minute observation period during which vital signs should be recorded at 30 (+/-10 minutes) after infusion. If the patient tolerated the first infusion well without infusion-associated adverse events, the observation period after the next and following infusions may be reduced to 30 minutes. If the patient experienced infusion-associated adverse events in the previous infusion, the observation period should be 60 minutes. If clinically indicated, vital signs should be recorded at 30 (+/- 10) minutes after the infusion of tiragolumab.

Treatment can continue for up to 13 cycles. For patients receiving both drugs, atezolizumab will be given first, followed by tiragolumab. If either study drug is delayed for a related toxicity, it is recommended that the other study drug is also delayed (if

applicable) since the safety profiles for atezolizumab and tiragolumab are similar.

All patients will receive CT scans every 12 weeks for the first two years after initiation of treatment, then every 24 weeks for the next year; frequency of scans will not be adjusted in the event of a treatment delay.

Administration of First and Subsequent Infusions of Atezolizumab and Tiragolumab

Study drug	First Infusion	Subsequent Infusions
Atezolizumab infusion	• No premedication is permitted prior to the atezolizumab infusion. • Vital signs (pulse rate, respiratory rate, blood pressure, and temperature) should be recorded within 60 minutes prior to the infusion. • Atezolizumab should be infused over 60 (± 15) minutes. • If clinically indicated, vital signs should be recorded every 15 (± 5) minutes during the infusion.	• If the patient experienced an IRR with any previous infusion of atezolizumab, premedication with an antihistamine and/or antipyretic medication may be administered for subsequent doses at the discretion of the investigator. • Vital signs should be recorded within 60 minutes prior to the infusion. • Atezolizumab should be infused over 30 (± 10) minutes if the previous infusion was tolerated without an IRR or 60 (± 15) minutes if the patient experienced an IRR with the previous infusion. • If the patient experienced an IRR with the previous infusion, or if clinically indicated, vital signs should be recorded during the infusion.
Observation period after infusion of atezolizumab	• After the infusion of atezolizumab, the patient begins a 60-minute observation period. • Vital signs should be recorded at 30 (± 10) minutes after the infusion of atezolizumab. • Patients will be informed about the possibility of delayed post-infusion symptoms and instructed to contact their study physician if they develop such symptoms.	• If the patient tolerated the previous atezolizumab infusion well without infusion-associated adverse events, the observation period after the next and following infusions may be reduced to 30 minutes. • If the patient experienced infusion-associated adverse events in the previous infusion, the observation period should be 60 minutes. • If clinically indicated, vital signs should be recorded at 30 (± 10) minutes after the infusion of atezolizumab.
Infusion of tiragolumab	• No premedication is permitted prior to the tiragolumab infusion. • Vital signs (pulse rate, respiratory rate, blood pressure, and temperature) should be recorded within 60 minutes prior to the infusion.	• If the patient experienced an IRR during any previous infusion of tiragolumab, premedication with an antihistamine and/or antipyretic may be administered for subsequent doses, at the discretion of the investigator. • Vital signs should be recorded within 60 minutes prior to the tiragolumab infusion.

Study drug	First Infusion	Subsequent Infusions
	• Tiragolumab should be infused over 60 (± 15) minutes. • Vital signs should be recorded every 15 (± 5) minutes during the infusion.	• Tiragolumab should be infused over 30 (± 10) minutes if the previous infusion was tolerated without an infusion-related reaction, or 60 (± 15) minutes if the patient experienced an infusion-related reaction with the previous infusion. • Vital signs should be recorded during the infusion if clinically indicated.
Observation period after infusion of tiragolumab	• After the infusion of tiragolumab, the patient begins a 60-minute observation period. • Vital signs should be recorded at 30 (± 10) minutes after the infusion of tiragolumab. ● Patients will be informed about the possibility of delayed post-infusion symptoms and will be instructed to contact their study physician if they develop such symptoms.	• If the patient tolerated the previous infusion of tiragolumab well without infusion-associated adverse events, the observation period may be reduced to 30 minutes. • If the patient experienced an infusion-associated adverse event in the previous infusion, the observation period should be 60 minutes. • If clinically indicated, vital signs should be recorded at 30 (± 10) minutes after the infusion of tiragolumab. ● Patients will be informed about the possibility of delayed post-infusion symptoms and will be instructed to contact their study physician if they develop such symptoms.

IRR = infusion-related reaction.

5.3 Definitions of Evaluability

In order to be evaluable for the primary objective (ctDNA negativity at C3D1), a patient must have blood drawn to assess ctDNA at that time point. Patients in Stage 1 who discontinue treatment prior to C3D1 will be replaced; patients in Stage 2 who discontinue treatment prior to C3D1 will not be replaced.

All patients who receive any study treatment are evaluable for toxicity. Patients are evaluated from first receiving study treatment until a 90-day follow up after the conclusion of treatment or death.

All patients are evaluable for disease response unless they discontinue participation prior to having any disease assessment.

5.4 Concomitant Therapy and Supportive Care Guidelines

If a patient experiences an infusion-related reaction with any previous infusion of atezolizumab or tiragolumab, premedication with an antihistamine and/or antipyretic medication may be administered for subsequent doses at the discretion of the investigator.

5.4.1 Permitted Therapy

Patients are permitted to use the following therapies during the study:

- Oral contraceptives with a failure rate of < 1% per year
- Hormone-replacement therapy
- Prophylactic or therapeutic anticoagulation therapy (such as warfarin at a stable dose or low-molecular-weight heparin)
- Inactivated influenza vaccinations
- Megestrol acetate administered as an appetite stimulant
- Mineralocorticoids (e.g., fludrocortisone)
- Corticosteroids administered for COPD or asthma
- Low-dose corticosteroids administered for orthostatic hypotension or adrenocortical insufficiency

Premedication with antihistamines, antipyretics, and/or analgesics may be administered for the second and subsequent infusions only, at the discretion of the investigator.

In general, investigators should manage a patient's care (including preexisting conditions) with supportive therapies other than those defined as cautionary or prohibited therapies as clinically indicated, per local standard practice. Patients who experience infusion-associated symptoms may be treated symptomatically with acetaminophen, ibuprofen, diphenhydramine, and/or H2-receptor antagonists (e.g., famotidine, cimetidine), or equivalent medications per local standard practice. Serious infusion-associated events manifested by dyspnea, hypotension, wheezing, bronchospasm, tachycardia, reduced oxygen saturation, or respiratory distress should be managed with supportive therapies as clinically indicated (e.g., supplemental oxygen and β2-adrenergic agonists).

5.4.2 Cautionary Therapy for Atezolizumab- and Tiragolumab-Treated Patients

5.4.2.1 Corticosteroids, Immunosuppressive Medications, and TNF-α Inhibitors

Systemic corticosteroids, immunosuppressive medications, and TNF-α inhibitors may attenuate potential beneficial immunologic effects of treatment with atezolizumab. Therefore, in situations in which systemic corticosteroids, immunosuppressive medications, or TNF-α inhibitors would be routinely administered, alternatives, including antihistamines, should be considered. If the alternatives are not feasible, systemic corticosteroids, immunosuppressive medications, and TNF-α inhibitors may be administered at the discretion of the investigator.

Systemic corticosteroids or immunosuppressive medications are recommended, at the discretion of the investigator, for the treatment of specific adverse events when associated with atezolizumab and/or tiragolumab therapy.

5.4.2.2 Herbal Therapies

Concomitant use of herbal therapies is not recommended because their pharmacokinetics, safety profiles, and potential drug-drug interactions are generally unknown. However, herbal therapies not intended for the treatment of cancer may be used during the study at the discretion of the investigator.

5.4.3 Prohibited Therapy

Use of the following concomitant therapies is prohibited as described below:

- Concomitant therapy intended for the treatment of cancer (including, but not limited to, chemotherapy, hormonal therapy, immunotherapy, radiotherapy, and herbal therapy), whether health authority-approved or experimental, is prohibited for various time periods prior to starting study treatment, depending on the agent, and during study treatment, until disease progression is documented and the patient has discontinued study treatment.
- Investigational therapy within 42 days prior to initiation of study treatment and during study treatment. Of note, COVID vaccines are not considered an investigational therapy.
- Live, attenuated vaccines (e.g., FluMist®) are prohibited within 4 weeks prior to initiation of study treatment, during study treatment, for 90 days after the final dose of tiragolumab, and for 5 months after the final dose of atezolizumab.
- Systemic immunostimulatory agents (including, but not limited to, interferons and IL 2) are prohibited within 4 weeks or 5 drug-elimination half-lives (whichever is longer) prior to initiation of study treatment and during study treatment because these agents could potentially increase the risk for autoimmune conditions when given in combination with atezolizumab and tiragolumab.

5.5 Women of Childbearing Potential

Women must remain abstinent or use contraceptive methods with a failure rate of < 1% per year during the treatment period and for 90 days after the final dose of tiragolumab and 5 months after the final dose of atezolizumab.

A woman is considered to be of childbearing potential if she is postmenarchal, has not reached a postmenopausal state (≥ 12 continuous months of amenorrhea with no identified cause other than menopause), and is not permanently infertile due to surgery (i.e., removal of ovaries, fallopian tubes, and/or uterus) or another cause as determined by the investigator

(e.g., Müllerian agenesis). The definition of childbearing potential may be adapted for alignment with local guidelines or regulations.

Examples of contraceptive methods with a failure rate of < 1% per year include bilateral tubal ligation, male sterilization, hormonal contraceptives that inhibit ovulation, hormone-releasing intrauterine devices, and copper intrauterine devices.

The reliability of sexual abstinence should be evaluated in relation to the duration of the clinical trial and the preferred and usual lifestyle of the patient. Periodic abstinence (e.g., calendar, ovulation, symptothermal, or postovulation methods) and withdrawal are not adequate methods of contraception.

With a female partner of childbearing potential, men who are not surgically sterile must remain abstinent or use a condom plus an additional contraceptive method that together result in a failure rate of < 1% per year during the treatment period, for 90 days after the final dose of tiragolumab. Men must refrain from donating sperm during this same period.

With a pregnant female partner, men must remain abstinent or use a condom during the treatment period for 90 days after the final dose of tiragolumab to avoid exposing the embryo.

The reliability of sexual abstinence should be evaluated in relation to the duration of the clinical trial and the preferred and usual lifestyle of the patient. Periodic abstinence (e.g., calendar, ovulation, symptothermal, or postovulation methods) and withdrawal are not adequate methods of contraception.

5.6 Duration of Therapy

If at any time the constraints of this protocol are considered to be detrimental to the patient's health and/or the patient no longer wishes to continue protocol therapy, the protocol therapy should be discontinued and the reason(s) for discontinuation documented in the case report forms.

In the absence of treatment delays due to adverse events, treatment may continue for up to 13 cycles or until one of the following criteria applies:

- Documented and confirmed disease progression
- Death
- Adverse event(s) that, in the judgment of the investigator, may cause severe or permanent harm or which rule out continuation of study drug
- General or specific changes in the patient's condition render the patient unable to receive further treatment in the judgment of the investigator
- Suspected pregnancy
- Serious noncompliance with the study protocol
- Lost to follow-up
- Patient withdraws consent

- Investigator removes the patient from study
- The Siteman Cancer Center decides to close the study

Patients who prematurely discontinue treatment for any reason will still be followed as indicated in the study calendar with the exception of discontinuation due to disease recurrence; patients who stop treatment due to recurrence will be considered off study and will no longer be followed after the 90-day follow-up for adverse events is completed.

5.7 Duration of Follow-up

Follow-up frequency is calculated from date of surgery. After surgery, patients will be followed every 12 weeks for 2 years and every 24 weeks for 1 additional year. Frequency and duration of follow-up after the end of treatment depends on when treatment is discontinued. Patients will be followed for 3 years from date of surgery regardless of when treatment is discontinued unless treatment is discontinued due to disease recurrence, in which case the patient will be considered off study after the 90-day follow-up for adverse events is completed. Patients removed from study for unacceptable adverse events will be followed until resolution or stabilization of the adverse event.

5.8 Lost to Follow-Up

A patient will be considered lost to follow-up if he or she fails to return for 2 scheduled visits and is unable to be contacted by the study team.

The following actions must be taken if the patient fails to return to clinic for a required study visit:

- The study team will attempt to contact the patient and reschedule the missed visit within 2 weeks and counsel the patient on the importance of maintaining the assigned visit schedule and ascertain if the patient wishes to and/or should continue in the study.
- Before a patient is deemed lost to follow-up, the investigator or designee will make every effort to regain contact with the patient (where possible, 3 telephone calls and, if necessary, a certified letter to the patient's last known mailing address). These contact attempts should be documented in the patient's medical record or study file.
- Should the patient continue to be unreachable, he or she will be considered to have withdrawn from the study with a primary reason of lost to follow-up.

6.0 DOSE DELAYS/DOSE MODIFICATIONS

Other holds and modifications beyond what is described below may be made at the discretion of the PI.

6.1 Dose Modifications

There will be no dose modifications, including dose reductions, for atezolizumab or tiragolumab in this study.

6.2 Treatment Interruptions

Study treatment may be temporarily suspended as appropriate for management of toxicity. On the basis of the available characterization of mechanism of action, tiragolumab may cause adverse events similar to but independent of atezolizumab, may exacerbate the frequency or severity of atezolizumab related adverse events, or may have non overlapping toxicities with atezolizumab. Because these scenarios may not be distinguished from one another in the clinical setting, immune mediated adverse events should generally be attributed to both study drugs, and dose interruptions or treatment discontinuation in response to immune mediated adverse events should be applied to both atezolizumab and tiragolumab.

In the adjuvant setting, atezolizumab and tiragolumab may be held for a maximum of 12 weeks. If tiragolumab is interrupted for > 12 weeks for any reason, the patient must permanently discontinue tiragolumab treatment, but may continue atezolizumab if there is no contraindication and after discussion with the PI to determine whether the toxicity is considered related to tiragolumab and/or to the combination. An exception can be made if in the judgment of the investigator, the patient is likely to derive clinical benefit from resuming tiragolumab after a hold of > 12 weeks. In this case, tiragolumab may be restarted with the approval of the PI. If atezolizumab is interrupted for > 12 weeks, the patient must permanently discontinue atezolizumab. However, if, in the judgment of the investigator, the patient is likely to derive clinical benefit from atezolizumab after a hold of > 12 weeks, atezolizumab may be restarted with the approval of the PI. Continued dosing of patients with single agent atezolizumab will require that all other study eligibility criteria continue to be met. Continued administration of single agent tiragolumab after permanent discontinuation of atezolizumab is not permitted.

If a patient must be tapered off steroids used to treat adverse events, atezolizumab and/or tiragolumab may be withheld for additional time beyond > 12 weeks from the last dose until steroids are discontinued, or until steroids are reduced to prednisone dose (or dose equivalent) ≤ 10 mg/day. The acceptable length of interruption will depend on an agreement with the PI.

Dose interruptions for reason(s) other than toxicity may be allowed with PI approval.

6.2.1 Study treatment discontinuation criteria specific to tiragolumab or atezolizumab-related AEs

- Any Grade ≥ 2 treatment-related uveitis, eye pain, or blurred vision that does not respond to topical therapy and does not improve to Grade 1 severity within 8 weeks or requires systemic treatment.

- Any Grade ≥ 2 treatment-related pneumonitis or interstitial lung disease that does not resolve following dose delay and systemic steroids.
- For Grade ≥ 3 adrenal insufficiency or Grade ≥ 4 hypophysitis, treatment needs to be discontinued regardless of control with hormone replacement (Note: Hospitalization for diagnostic workup of adrenal insufficiency without severe symptoms should not require discontinuation).
- Any Grade ≥ 3 non-skin, treatment-related AE lasting > 7 days or recurs, with the following exceptions for uveitis, pneumonitis, bronchospasm, diarrhea, colitis, neurologic toxicity, hypersensitivity reactions, infusion reactions, endocrinopathies, and laboratory abnormalities:
  - Grade 3 treatment-related uveitis, pneumonitis, bronchospasm, diarrhea, colitis, neurologic toxicity, hypersensitivity reaction, or infusion reaction of any duration requires discontinuation.
  - Grade 3 treatment-related endocrinopathies (excluding adrenal insufficiency and hypophysitis) adequately controlled with only physiologic hormone replacement do not require discontinuation.
  - Grade 3 treatment-related laboratory abnormalities do not require treatment discontinuation except:
    - Grade 3 treatment-related thrombocytopenia > 7 days or associated with clinically significant bleeding requires discontinuation.
- In most cases of Grade 3 AST or ALT elevation, study treatment will be permanently discontinued. If the Investigator determines a possible favorable benefit/risk ratio that warrants continuation of study treatment, a discussion between the Investigator and the Medical Monitor/designee must occur.
- Any treatment-related liver function test abnormality that meets the following criteria require discontinuation:
  - AST or ALT > 5 × ULN to 8 × ULN for > 2 weeks
  - AST or ALT > 8 × ULN
  - Total bilirubin > 5 × ULN
  - Concurrent AST or ALT > 3 × ULN and total bilirubin > 2 × ULN
- Grade 3 or 4 infusion reaction of any duration
- Any Grade 4 treatment -related AE or laboratory abnormality, except for the following events, which do not require discontinuation:
  - Grade 4 neutropenia ≤ 7 days.
  - Grade 4 lymphopenia or leukopenia ≤ 14 days in duration.
  - Isolated Grade 4 amylase or lipase abnormalities that are not associated with symptoms or clinical manifestations of pancreatitis and decrease to Grade < 4 or return to baseline within 7 days.
  - Isolated Grade 4 electrolyte imbalances/abnormalities that are not associated with clinical sequelae and are corrected with supplementation/appropriate management within 72 hours of their onset.
  - Grade 4 treatment -related endocrinopathy AEs (except adrenal insufficiency or hypophysitis), such as hyper- or hypothyroidism, or glucose intolerance, which resolve or are adequately controlled with physiologic hormone replacement (corticosteroids at ≤ 10 mg of prednisone or equivalent per day, thyroid hormones) or glucose-controlling agents,

respectively, may not require discontinuation after discussion with and approval from the Medical Monitor.

- Any AE, laboratory abnormality, or intercurrent illness that, in the judgment of the Investigator, presents a substantial clinical risk to the patient with continued treatment.
- Disease recurrence (local, regional, or distant), including new primary melanoma (except MMIS Stage 0) Note: Non-melanoma skin cancer does not require treatment discontinuation. In the absence of intolerable toxicity, patients with diagnosed MMIS Stage 0 melanoma will be eligible to continue study treatment using the same dose and schedule for their original melanoma of up to approximately 1 year of study treatment starting from first dose, at the discretion of the Investigator.

6.3 Management Guidelines for Atezolizumab- and Tiragolumab-Specific Adverse Events

6.3.1 Respiratory Disorders and Pulmonary Events

Immune-mediated pulmonary events are a potential risk with tiragolumab.

Pulmonary events may present as new or worsening cough, chest pain, fever, dyspnea, fatigue, hypoxia, pneumonitis, and pulmonary infiltrates. Patients will be assessed for pulmonary signs and symptoms throughout the study.

All pulmonary events should be thoroughly evaluated for other commonly reported etiologies such as pneumonia or other infection, lymphangitic carcinomatosis, pulmonary embolism, heart failure, chronic obstructive pulmonary disease, or pulmonary hypertension. COVID-19 evaluation should be performed per institutional guidelines where relevant. Management guidelines for pulmonary events are provided in the table below.

Management Guidelines for Respiratory Disorders and Pulmonary Events, Including Pneumonitis

Event	Management
Respiratory disorder or other pulmonary event, Grade 1	● Continue atezolizumab and tiragolumab and monitor closely. ● Re-evaluate on serial imaging. ● Consider patient referral to pulmonary specialist.
Respiratory disorder or other pulmonary event, Grade 2	● Withhold atezolizumab and tiragolumab for up to 12 weeks after event onset. [a] ● Refer patient to pulmonary and infectious disease specialists and consider bronchoscopy or BAL with or without transbronchial biopsy. ● Initiate treatment with corticosteroids equivalent to 1−2 mg/kg/day oral prednisone. ● If event resolves to Grade 1 or better, resume atezolizumab and tiragolumab. [b]

Event	Management
	● If event does not resolve to Grade 1 or better while withholding atezolizumab and tiragolumab, permanently discontinue atezolizumab and tiragolumab and contact Medical Monitor. [c] ● For recurrent events, treat as a Grade 3 or 4 event.
Respiratory disorder or other pulmonary event, Grade 3 or 4	● Permanently discontinue atezolizumab and tiragolumab and contact Medical Monitor. [c] ● Oral or IV broad-spectrum antibiotics should be administered in parallel to the immunosuppressive treatment. ● Bronchoscopy or BAL with or without transbronchial biopsy is recommended. ● Initiate treatment with corticosteroids equivalent to 1−2 mg/kg/day oral prednisone. ● If event does not improve within 48 hours after initiating corticosteroids, consider adding an immunosuppressive agent. ● If event resolves to Grade 1 or better, taper corticosteroids over ≥ 1 month.

BAL = bronchoscopic alveolar lavage.

[a] Atezolizumab and tiragolumab may be withheld for a longer period of time (i.e., > 12 weeks after event onset) to allow for corticosteroids (if initiated) to be reduced to the equivalent of ≤ 10 mg/day oral prednisone. The acceptable length of the extended period of time must be based on the investigator's benefit risk assessment and in alignment with the protocol requirements for the duration of treatment and document by the investigator.

[b] If corticosteroids have been initiated, they must be tapered over ≥ 1 month to the equivalent of ≤ 10 mg/day oral prednisone before atezolizumab and tiragolumab can be resumed.

[c] Resumption of atezolizumab may be considered in patients who are deriving benefit and have fully recovered from the immune-mediated event. The decision to re challenge patients with atezolizumab should be based on the investigator's assessment of benefit risk assessment and documented by the investigator (or appropriate delegate)..

[d] In case of pneumonitis, atezolizumab should not be resumed after permanent discontinuation.

6.3.2 Hepatobiliary Disorders or Events

Eligible patients must have adequate liver function, as manifested by measurements of total bilirubin and hepatic transaminases, and liver function will be monitored throughout study treatment. Management guidelines for hepatic events are provided in the table below.

Patients with right upper-quadrant abdominal pain and/or unexplained nausea or vomiting should have liver function tests (LFTs) performed immediately and reviewed before administration of the next dose of study drug(s).

For patients with elevated LFTs, concurrent medication, viral hepatitis, and toxic or neoplastic etiologies should be considered and addressed, as appropriate.

Management Guidelines for Hepatobiliary Disorders or Events

Event	Management
Hepatobiliary disorder or event, Grade 1	● Continue atezolizumab and tiragolumab. ● Monitor LFTs until values resolve to within normal limits or to baseline values.
Hepatobiliary disorder or event, Grade 2	**All events:** ● Monitor LFTs more frequently until return to baseline values. **Events of > 5 days' duration:** ● Withhold atezolizumab and tiragolumab for up to 12 weeks after event onset. [a] ● Initiate treatment with corticosteroids equivalent to 1−2 mg/kg/day oral prednisone. ● If event resolves to Grade 1 or better, resume atezolizumab and tiragolumab. [b] ● If event does not resolve to Grade 1 or better while withholding atezolizumab and tiragolumab, permanently discontinue atezolizumab and tiragolumab. [c]
Hepatobiliary disorder or event, Grade 3 or 4	● Permanently discontinue atezolizumab and tiragolumab. [c] ● Consider patient referral to gastrointestinal specialist for evaluation and liver biopsy to establish etiology of hepatic injury. ● Initiate treatment with corticosteroids equivalent to 1−2 mg/kg/day oral prednisone. ● If event does not improve within 48 hours after initiating corticosteroids, consider adding an immunosuppressive agent. If event resolves to Grade 1 or better, taper corticosteroids over ≥ 1 month.

LFT = liver function test.

[a] Atezolizumab and tiragolumab may be withheld for a longer period of time (i.e., > 12 weeks after event onset) to allow for corticosteroids (if initiated) to be reduced to the equivalent of ≤ 10 mg/day oral prednisone. The acceptable length of the extended period of time must be based on the investigator's benefit-risk assessment and in alignment with the protocol requirements for the duration of treatment and documented by the investigator.

[b] If corticosteroids have been initiated, they must be tapered over ≥ 1 month to the equivalent of ≤ 10 mg/day oral prednisone before atezolizumab and tiragolumab can be resumed.

[c] Resumption of atezolizumab and tiragolumab may be considered in patients who are deriving benefit and have fully recovered from the immune-mediated event. The decision to re-challenge patients with atezolizumab should be based on the investigator's benefit-risk assessment and documented by the investigator.

6.3.3 Gastrointestinal Disorders or Events

Immune-mediated gastrointestinal events are a potential risk with tiragolumab.

Management guidelines for diarrhea or colitis are provided in the table below.

All events of diarrhea or colitis should be thoroughly evaluated for other more common etiologies. For events of significant duration or magnitude or associated with signs of systemic inflammation or acute-phase reactants (e.g., increased C-reactive protein, platelet count, or bandemia): Perform sigmoidoscopy (or colonoscopy, if appropriate) with colonic biopsy, with three to five specimens for

standard paraffin block to check for inflammation and lymphocytic infiltrates to confirm colitis diagnosis.

Management Guidelines for Gastrointestinal Disorders or Events (Diarrhea or Colitis)

Event	Management
Diarrhea or colitis, Grade 1	● Continue atezolizumab and tiragolumab. ● Initiate symptomatic treatment. ● Endoscopy is recommended if symptoms persist for > 7 days. ● Monitor closely.
Diarrhea or colitis, Grade 2	● Withhold atezolizumab and tiragolumab for up to 12 weeks after event onset. [a] ● Initiate symptomatic treatment. ● If strong clinical suspicion for immune-mediated colitis, start empiric IV steroids while waiting for definitive diagnosis. ● Patient referral to GI specialist is recommended. ● For recurrent events or events that persist > 5 days, initiate treatment with corticosteroids equivalent to 1−2 mg/kg/day oral prednisone. If the event does not improve within 48 hours after initiating corticosteroids, consider adding an immunosuppressive agent. ● If event resolves to Grade 1 or better, resume atezolizumab and tiragolumab. [b] ● If event does not resolve to Grade 1 or better while withholding atezolizumab and tiragolumab, permanently discontinue atezolizumab and tiragolumab and contact Medical Monitor. [c]
Diarrhea or colitis, Grade 3	● Withhold atezolizumab and tiragolumab for up to 12 weeks after event onset. [a] ● Refer patient to GI specialist for evaluation and confirmatory biopsy. ● Initiate treatment with corticosteroids equivalent to 1−2 mg/kg/day IV methylprednisolone and convert to 1−2 mg/kg/day oral prednisone or equivalent upon improvement. If event does not improve within 48 hours after initiating corticosteroids, consider adding an immunosuppressive agent. ● If event resolves to Grade 1 or better, resume atezolizumab and tiragolumab. [b] ● If event does not resolve to Grade 1 or better while withholding atezolizumab and tiragolumab, permanently discontinue atezolizumab and tiragolumab and contact Medical Monitor. [c]
Diarrhea or colitis, Grade 4	● Permanently discontinue atezolizumab and tiragolumab and contact Medical Monitor. [c] ● Refer patient to GI specialist for evaluation and confirmatory biopsy. ● Initiate treatment with corticosteroids equivalent to 1−2 mg/kg/day IV methylprednisolone and convert to 1−2 mg/kg/day oral prednisone or equivalent upon improvement. ● If event does not improve within 48 hours after initiating corticosteroids, consider adding an immunosuppressive agent. ● If event resolves to Grade 1 or better, taper corticosteroids over ≥ 1 month.

Event	Management

GI = gastrointestinal.

[a] Atezolizumab and tiragolumab may be withheld for a longer period of time (i.e., > 12 weeks after event onset) to allow for corticosteroids (if initiated) to be reduced to the equivalent of ≤ 10 mg/day oral prednisone. The acceptable length of the extended period of time must be based on the investigator's benefit-risk assessment and in alignment with the protocol requirements for the duration of treatment and documented by the investigator.

[b] If corticosteroids have been initiated, they must be tapered over ≥ 1 month to the equivalent of ≤ 10 mg/day oral prednisone before atezolizumab and tiragolumab can be resumed.

[c] Resumption of atezolizumab and tiragolumab may be considered in patients who are deriving benefit and have fully recovered from the immune-mediated event. Patients can be re-challenged with atezolizumab and tiragolumab only after approval has been documented by the investigator.

6.3.4 Endocrine Disorders or Events

Immune-related endocrine events are a potential risk with tiragolumab.

Management guidelines for endocrine events are provided in the table below.

Patients with unexplained symptoms such as headache, fatigue, myalgias, impotence, constipation, or mental status changes should be investigated for the presence of thyroid, pituitary, or adrenal endocrinopathies. The patient should be referred to an endocrinologist if an endocrinopathy is suspected. Thyroid-stimulating hormone (TSH) and free triiodothyronine and thyroxine levels should be measured to determine whether thyroid abnormalities are present. Pituitary hormone levels and function tests (e.g., TSH, growth hormone, luteinizing hormone, follicle-stimulating hormone, testosterone, prolactin, adrenocorticotropic hormone [ACTH] levels, and ACTH stimulation test) and magnetic resonance imaging (MRI) of the brain (with detailed pituitary sections) may help to differentiate primary pituitary insufficiency from primary adrenal insufficiency.

Management Guidelines for Endocrine Disorders or Events

Event	Management
Hypothyroidism, Grade 1	● Continue atezolizumab and tiragolumab. ● Initiate treatment with thyroid replacement hormone. ● Monitor TSH closely.
Hypothyroidism, Grade 2	● Consider withholding atezolizumab and tiragolumab. ● Initiate treatment with thyroid replacement hormone. ● Monitor TSH closely. ● Consider patient referral to endocrinologist. ● Resume atezolizumab and tiragolumab when symptoms are controlled and thyroid function is improving.
Hypothyroidism Grade 3 and 4	● Withhold atezolizumab and tiragolumab ● Initiate treatment with thyroid replacement hormone. ● Monitor TSH closely ● Refer to an endocrinologist ● Admit patient to the hospital for developing myxedema (bradycardia, hypothermia, and altered mental status) ● Resume atezolizumab when symptoms are controlled, and thyroid

Event	Management
	function is improving. ● Permanently discontinue atezolizumab.[c]
Hyperthyroidism, Grade 1	**TSH ≥ 0.1 mU/L and < 0.5 mU/L:** ● Continue atezolizumab and tiragolumab. ● Monitor TSH every 4 weeks. ● Consider patient referral to endocrinologist **TSH < 0.1 mU/L:** ● Follow guidelines for Grade 2 hyperthyroidism. ● Consider patient referral to endocrinologist
Hyperthyroidism, Grade 2	● Consider atezolizumab and tiragolumab. ● Initiate treatment with anti-thyroid drug such as methimazole or carbimazole as needed. ● Consider patient referral to endocrinologist. ● Resume atezolizumab and tiragolumab when symptoms are controlled and thyroid function is improving.
Grade 3 and 4 hyperthyroidism	● Withhold atezolizumab. ● Initiate treatment with anti-thyroid drug such as methimazole or carbimazole as needed. ● Refer to an endocrinologist. ● Resume atezolizumab and tiragolumab when symptoms are controlled, and thyroid function is improving. ● Permanently discontinue atezolizumab.[c]
Adrenal insufficiency, Grade 2−4	● Withhold atezolizumab and tiragolumab for up to 12 weeks after event onset. [a] ● Refer patient to endocrinologist. ● Perform appropriate imaging. ● Initiate treatment with corticosteroids equivalent to 1−2 mg/kg/day IV methylprednisolone and convert to 1−2 mg/kg/day oral prednisone or equivalent upon improvement. ● If event resolves to Grade 1 or better and patient is stable on replacement therapy, resume atezolizumab and tiragolumab. [b] ● If event does not resolve to Grade 1 or better or patient is not stable on replacement therapy while withholding atezolizumab and tiragolumab, permanently discontinue atezolizumab and tiragolumab and contact PI. [c]
Hypophysitis (panhypopituitarism), Grade 2 or 3	● Withhold atezolizumab and tiragolumab for up to 12 weeks after event onset. [a] ● Refer patient to endocrinologist. ● Perform brain MRI (pituitary protocol). ● Initiate treatment with corticosteroids equivalent to 1−2 mg/kg/day IV methylprednisolone and convert to 1−2 mg/kg/day oral prednisone or equivalent upon improvement. ● Initiate hormone replacement if clinically indicated. ● If event resolves to Grade 1 or better, resume atezolizumab and tiragolumab. [b]

Event	Management
	● If event does not resolve to Grade 1 or better while withholding atezolizumab and tiragolumab, permanently discontinue atezolizumab and tiragolumab and contact PI. [c] ● For recurrent hypophysitis, treat as a Grade 4 event.
Hypophysitis (panhypopituitarism), Grade 4	● Permanently discontinue atezolizumab and tiragolumab and contact Medical Monitor. [c] ● Refer patient to endocrinologist. ● Perform brain MRI (pituitary protocol). ● Initiate treatment with corticosteroids equivalent to 1−2 mg/kg/day IV methylprednisolone and convert to 1−2 mg/kg/day oral prednisone or equivalent upon improvement. ● Initiate hormone replacement if clinically indicated.

MRI = magnetic resonance imaging; TSH = thyroid-stimulating hormone.

[a] Atezolizumab and tiragolumab may be withheld for a longer period of time (i.e., > 12 weeks after event onset) to allow for corticosteroids (if initiated) to be reduced to the equivalent of ≤ 10 mg/day oral prednisone. The acceptable length of the extended period of time must bebased on the investigator's benefit-risk assessment and in alignment with the protocol requirements for the duration of treatment and documented by the investigator.

[b] If corticosteroids have been initiated, they must be tapered over ≥ 1 month to the equivalent of ≤ 10 mg/day oral prednisone before atezolizumab and tiragolumab can be resumed.

[c] Resumption of atezolizumab and tiragolumab may be considered in patients who are deriving benefit and have fully recovered from the immune-mediated event. The decision to re-challenge patients with atezolizumab should be based on the investigator's benefit-risk assessment and documented by the investigator.

6.3.5 Metabolic Disorders or Events

Event	Management
Hyperglycemia, Grade 1 or 2	● Continue atezolizumab and tiragolumab. ● Investigate for diabetes. If patient has Type 1 diabetes, treat as grade 3 event. If patient does not have Type 1 diabetes, treat as per institutional guidelines. ● Monitor for glucose control.
Hyperglycemia, Grade 3 or 4	● Withhold atezolizumab and tiragolumab. ● Initiate treatment with insulin. ● Monitor for glucose control. ● Resume atezolizumab and tiragolumab when symptoms resolve and glucose levels are stable.

6.3.6 Eye Disorders or Events

An ophthalmologist should evaluate visual complaints (e.g., uveitis, retinal events). Management guidelines for ocular events are provided in the table below.

Management Guidelines for Eye Disorders or Events

Event	Management
Eye disorders, Grade 1	● Continue atezolizumab and tiragolumab. ● Patient referral to ophthalmologist is strongly recommended.

Event	Management
	● Initiate treatment with topical corticosteroid eye drops and topical immunosuppressive therapy. ● If symptoms persist, treat as a Grade 2 event.
Eye disorders, Grade 2	● Withhold atezolizumab and tiragolumab for up to 12 weeks after event onset. [a] ● Patient referral to ophthalmologist is strongly recommended. ● Initiate treatment with topical corticosteroid eye drops and topical immunosuppressive therapy. ● If event resolves to Grade 1 or better, resume atezolizumab and tiragolumab. [b] ● If event does not resolve to Grade 1 or better while withholding atezolizumab and tiragolumab, permanently discontinue atezolizumab and tiragolumab and contact PI. [c]
Eye disorders, Grade 3 or 4	● Permanently discontinue atezolizumab and tiragolumab and contact PI. [c] ● Refer patient to ophthalmologist. ● Initiate treatment with corticosteroids equivalent to 1−2 mg/kg/day oral prednisone. ● If event resolves to Grade 1 or better, taper corticosteroids over ≥ 1 month.

[a] Atezolizumab and tiragolumab may be withheld for a longer period of time (i.e., ≥ 12 weeks after event onset) to allow for corticosteroids (if initiated) to be reduced to the equivalent of ≤ 10 mg/day oral prednisone. The acceptable length of the extended period of time must be based on the investigator's benefit-risk assessment and in alignment with the protocol requirements for the duration of treatment and documented by the investigator.

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[b] If corticosteroids have been initiated, they must be tapered over ≥ 1 month to the equivalent of ≤ 10 mg/day oral prednisone before atezolizumab and tiragolumab can be resumed.

[c] Resumption of atezolizumab and tiragolumab may be considered in patients who are deriving benefit and have fully recovered from the immune-mediated event. The decision to re-challenge patients with atezolizumab should be based on the investigator's benefit-risk assessment and documented by the investigator.

6.3.7 Immune-Mediated Cardiac Events

IMMUNE-MEDIATED MYOCARDITIS

Immune-mediated myocarditis should be suspected in any patient presenting with signs or symptoms suggestive of myocarditis, including, but not limited to, laboratory (e.g., B-type natriuretic peptide) or cardiac imaging abnormalities, dyspnea, chest pain, palpitations, fatigue, decreased exercise tolerance, or syncope. Myocarditis may also be a clinical manifestation of myositis or associated with pericarditis (see section on pericardial disorders below) and should be managed accordingly. Immune-mediated myocarditis needs to be distinguished from myocarditis resulting from infection (commonly viral, e.g., in a patient who reports a recent history of gastrointestinal illness), ischemic events, underlying arrhythmias, exacerbation of preexisting cardiac conditions, or progression of malignancy.

All patients with possible myocarditis should be urgently evaluated by performing cardiac enzyme assessment, an ECG, a chest X-ray, an echocardiogram, and a

cardiac MRI as appropriate per institutional guidelines. A cardiologist should be consulted. An endomyocardial biopsy may be considered to enable a definitive diagnosis and appropriate treatment, if clinically indicated.

Patients with signs and symptoms of myocarditis, in the absence of an identified alternate etiology, should be treated according to the guidelines in the table below.

IMMUNE-MEDIATED PERICARDIAL DISORDERS

Immune-mediated pericarditis should be suspected in any patient presenting with chest pain and may be associated with immune-mediated myocarditis (see section on myocarditis above).
Immune-mediated pericardial effusion and cardiac tamponade should be suspected in any patient presenting with chest pain associated with dyspnea or hemodynamic instability.
Patients should be evaluated for other causes of pericardial disorders such as infection (commonly viral), cancer related (metastatic disease or chest radiotherapy), cardiac injury related (post myocardial infarction or iatrogenic), and autoimmune disorders, and should be managed accordingly.
All patients with suspected pericardial disorders should be urgently evaluated by performing an ECG, chest X-ray, transthoracic echocardiogram, and cardiac MRI as appropriate per institutional guidelines. A cardiologist should be consulted. Pericardiocentesis should be considered for diagnostic or therapeutic purposes, if clinically indicated.
Patients with signs and symptoms of pericarditis, pericardial effusion, or cardiac tamponade, in the absence of an identified alternate etiology, should be treated according to the guidelines in the table below. Withhold treatment with atezolizumab for Grade 1 pericarditis and conduct a detailed cardiac evaluation to determine the etiology and manage accordingly.

Management Guidelines for Immune-Mediated Cardiac Events

Event	Management
Immune-mediated myocarditis, Grades 2-4 Immune-mediated pericardial disorders, Grades 2-4	● Withhold atezolizumab and tiragolumab for up to 12 weeks after event onset and contact PI. ● Refer patient to cardiologist. ● Initiate treatment as per institutional guidelines and consider antiarrhythmic drugs, temporary pacemaker, ECMO, VAD, or pericardiocentesis as appropriate. ● Initiate treatment with corticosteroids equivalent to 1−2 mg/kg/day IV methylprednisolone and convert to 1−2 mg/kg/day oral prednisone or equivalent upon improvement. ● If event does not improve within 48 hours after initiating corticosteroids, consider adding an immunosuppressive agent. ● If event resolves to Grade 1 or better, taper corticosteroids over≥1 month.

ECMO = extracorporeal membrane oxygenation; VAD = ventricular assist device.

6.3.8 Infusion-Related Reactions

No premedication is indicated for the administration of Cycle 1 of atezolizumab or tiragolumab. However, patients who experience an infusion-related reaction (IRR) or (CRS) with atezolizumab may receive premedication with antihistamines, antipyretic medications, and/or analgesics (e.g., acetaminophen) for subsequent infusions. Metamizole (dipyrone) is prohibited in treating atezolizumab-associated IRRs because of its potential for causing agranulocytosis.

IRRs are known to occur with the administration of monoclonal antibodies and have been reported with tiragolumab and atezolizumab. These reactions, which are thought to be due to release of cytokines and/or other chemical mediators, occur within 24 hours of atezolizumab or tiragolumab administration and are generally mild to moderate in severity.

Guidelines for medical management of IRRs during Cycle 1 are provided in the table below. For subsequent cycles, IRRs should be managed according to institutional guidelines.

Management Guidelines for Infusion-Related Reactions

Event	Management
IRR, Grade 1	● Reduce infusion rate to half the rate being given at the time of event onset. ● After the event has resolved, the investigator should wait for 30 minutes while delivering the infusion at the reduced rate. ● If the infusion is tolerated at the reduced rate for 30 minutes after symptoms have resolved, the infusion rate may be increased to the original rate.
IRR, Grade 2	● Interrupt infusion. ● Administer aggressive symptomatic treatment (e.g., oral or IV antihistamine, antipyretic medication, glucocorticoids, epinephrine, bronchodilators, oxygen, IV fluids). ● After symptoms have resolved to baseline, resume infusion at half the rate being given at the time of event onset. ● For subsequent infusions, consider administration of oral premedication with antihistamines, antipyretic medications, and/or analgesics and monitor closely for IRRs.
IRR, Grade 3 or 4	● Stop infusion. ● Administer aggressive symptomatic treatment (e.g., oral or IV antihistamine, antipyretic medication, glucocorticoids, epinephrine, bronchodilators, oxygen, IV fluids). ● Permanently discontinue atezolizumab or tiragolumab and contact PI. [a]

IRR = infusion-related reaction.

[a] Resumption of atezolizumab or tiragolumab may be considered in patients who are deriving benefit and have fully recovered from the event. Patients can be re-challenged with atezolizumab or tiragolumab only after approval has been documented by the investigator.

6.3.9 Cytokine-Release Syndrome

No premedication is indicated for the administration of Cycle 1 of atezolizumab or tiragolumab. However, patients who experience CRS with atezolizumab or tiragolumab may receive premedication with antihistamines, anti-pyretics, and/or analgesics (e.g., acetaminophen) for subsequent infusions.

CRS is defined as a supraphysiologic response following administration of any immune therapy that results in activation or engagement of endogenous or infused T cells and/or other immune effector cells. Symptoms can be progressive, always include fever at the onset, and may include hypotension, capillary leak (hypoxia), and end-organ dysfunction (Lee et al. 2019). CRS has been well documented with chimeric antigen receptor T-cell therapies and bispecific T-cell engager antibody therapies but has also been reported with immunotherapies that target PD-1 or PD-L1 (Rotz et al. 2017; Adashek and Feldman 2019), including atezolizumab.

Severe SARS CoV 2 infection is associated with a CRS involving the inflammatory cytokines interleukin (IL)-6, IL-10, IL-2, and IFN-© (Merad and Martin 2020). If a patient develops suspected CRS during the study, a differential diagnosis should include SARS CoV 2 infection, which should be confirmed or refuted through assessment of exposure history, appropriate laboratory testing, and clinical or radiologic evaluations per investigator's judgment. If a diagnosis of SARS CoV 2 infection is confirmed, the disease should be managed as per local or institutional guidelines.

Guidelines for medical management of CRS are provided in the table below.

Management Guidelines for Infusion-Related Reactions and Cytokine-Release Syndrome

Event	Management
Grade 1 [a] fever [b] with or without constitutional symptoms	● Immediately interrupt infusion. ● Upon symptom resolution, wait for 30 minutes and then restart infusion at half the rate being given at the time of event onset. ● If the infusion is tolerated at the reduced rate for 30 minutes, the infusion rate may be increased to the original rate. ● If symptoms recur, discontinue infusion of this dose. ● Administer symptomatic treatment, [c] including maintenance of IV fluids for hydration. ● In case of rapid decline or prolonged CRS (> 2 days) or in patients with significant symptoms and/or comorbidities, consider managing as per Grade 2. ● For subsequent infusions, consider administration of oral premedication with antihistamines, antipyretic medications, and/or analgesics, and monitor closely for CRS.
Grade 2 [a] fever [b] with at least one of the	● Immediately interrupt infusion. ● Upon symptom resolution, wait for 30 minutes and then restart infusion at half the rate being given at the time of event onset.

Event	Management
following: Hypotension not requiring vasopressors Hypoxia requiring low-flow oxygen [d] by nasal cannula or blow-by	● If symptoms recur, discontinue infusion of this dose. ● Administer symptomatic treatment. [c] ● For hypotension, administer IV fluid bolus as needed. ● Monitor cardiopulmonary and other organ function closely (in the ICU, if appropriate). Administer IV fluids as clinically indicated, and manage constitutional symptoms and organ toxicities as per institutional practice. ● Rule out other inflammatory conditions that can mimic CRS (e.g., sepsis). If no improvement within 24 hours, initiate workup and assess for signs and symptoms of HLH or MAS as described in this appendix. ● Consider IV corticosteroids (e.g., methylprednisolone 2 mg/kg/day or dexamethasone 10 mg every 6 hours). ● Consider anti-cytokine therapy. ● Consider hospitalization until complete resolution of symptoms. If no improvement within 24 hours, manage as per Grade 3, that is, hospitalize patient (monitoring in the ICU is recommended), permanently discontinue atezolizumab or tiragolumab, and contact Medical Monitor. ● If symptoms resolve to Grade 1 or better for 3 consecutive days, the next dose of atezolizumab or tiragolumab may be administered. For subsequent infusions, consider administration of oral premedication with antihistamines, antipyretic medications, and/or analgesics and monitor closely for CRS. ● If symptoms do not resolve to Grade 1 or better for 3 consecutive days, contact PI.
<u>Grade 3</u> [a] Fever [b] with at least one of the following: ● Hypotension requiring a vasopressor (with or without vasopressin) ● Hypoxia requiring high-flow oxygen [d] by nasal cannula, face mask, non-rebreather mask, or Venturi mask	● Permanently discontinue atezolizumab or tiragolumab and contact Medical Monitor. [e] ● Administer symptomatic treatment. [c] ● For hypotension, administer IV fluid bolus and vasopressor as needed. ● Monitor cardiopulmonary and other organ function closely; monitoring in the ICU is recommended. Administer IV fluids as clinically indicated, and manage constitutional symptoms and organ toxicities as per institutional practice. ● Rule out other inflammatory conditions that can mimic CRS (e.g., sepsis). If no improvement within 24 hours, initiate workup and assess for signs and symptoms of HLH or MAS as described in this appendix. ● Administer IV corticosteroids (e.g., methylprednisolone 2 mg/kg/day or dexamethasone 10 mg every 6 hours). ● Consider anti-cytokine therapy. ● Hospitalize patient until complete resolution of symptoms. If no improvement within 24 hours, manage as per Grade 4, that is, admit patient to ICU and initiate hemodynamic monitoring, mechanical ventilation, and/or IV fluids and vasopressors as needed; for patients who are refractory to anti-cytokine therapy, experimental treatments may be considered at the discretion of the investigator and in consultation with the PI.
<u>Grade 4</u> [a]	● Permanently discontinue atezolizumab or tiragolumab and contact PI. [f]

Event	Management
Fever [b] with at least one of the following: ● Hypotension requiring multiple vasopressors (excluding vasopressin) ● Hypoxia requiring oxygen by positive pressure (e.g., CPAP, BiPAP, intubation and mechanical ventilation)	● Administer symptomatic treatment. [c] ● Admit patient to ICU and initiate hemodynamic monitoring, mechanical ventilation, and/or IV fluids and vasopressors as needed. Monitor other organ function closely. Manage constitutional symptoms and organ toxicities as per institutional practice. ● Rule out other inflammatory conditions that can mimic CRS (e.g., sepsis). If no improvement within 24 hours, initiate workup and assess for signs and symptoms of HLH or MAS as described in this appendix. ● Administer IV corticosteroids (e.g., methylprednisolone 2 mg/kg/day or dexamethasone 10 mg every 6 hours). ● Consider anti-cytokine therapy. For patients who are refractory to anti-cytokine therapy, experimental treatments [f] may be considered at the discretion of the investigator and in consultation with the PI. ● Hospitalize patient until complete resolution of symptoms.

ASTCT = American Society for Transplantation and Cellular Therapy; BiPAP = bi-level positive airway pressure; CAR = chimeric antigen receptor; CPAP = continuous positive airway pressure; CRS = cytokine-release syndrome; CTCAE = Common Terminology Criteria for Adverse Events; eCRF = electronic Case Report Form; HLH = hemophagocytic lymphohistiocytosis; ICU = intensive care unit; MAS = macrophage activation syndrome; NCCN = National Cancer Comprehensive Network; NCI = National Cancer Institute.

The management guidelines have been adapted from the NCCN guidelines for the management of CAR T-cell−related toxicities (Version 2.2019).

[a] Grading system for management guidelines is based on the ASTCT CRS Consensus Grading Scale. NCI CTCAE v5.0 and the ASTCT CRS Consensus Grading Scale should be used when reporting severity of CRS on the Adverse Event eCRF. NCI CTCAE v5.0 should be used when reporting severity of organ toxicities associated with CRS on the dedicated Cytokine Release Syndrome eCRF. Organ toxicities associated with CRS should not influence overall CRS grading.

[b] Fever is defined as temperature ≥ 38°C not attributable to any other cause. In patients who develop CRS and then receive anti-pyretic, anti-cytokine, or corticosteroid therapy, fever is no longer required when subsequently determining event severity (grade). In this case, the grade is driven by the presence of hypotension and/or hypoxia.

[c] Symptomatic treatment may include oral or IV antihistamines, antipyretic medications, analgesics, bronchodilators, and/or oxygen. For bronchospasm, urticaria, or dyspnea, additional treatment may be administered as per institutional practice.

[d] Low flow is defined as oxygen delivered at ≤ 6 L/min, and high flow is defined as oxygen delivered at > 6 L/min.

[e] Resumption of atezolizumab or tiragolumab may be considered in patients who are deriving benefit and have fully recovered from the event. The decision to re-challenge patients with atezolizumab or tiragolumab should be based on the investigator's benefit-risk assessment and documented by the investigator. The Medical Monitor is available to advise is needed. For subsequent infusions, administer oral premedication with antihistamines, antipyretics medications, and/or analgesics, and monitor closely for CRS. Premedication with corticosteroids and extending the infusion time may also be considered after consulting the PI and considering the benefit−risk ratio.

f Refer to Riegler et al. (2019)

6.3.10 Pancreatic Events

The differential diagnosis of acute abdominal pain should include pancreatitis. Appropriate workup should include an evaluation for ductal obstruction, as well as

serum amylase and lipase tests. Management guidelines for pancreatic events, including pancreatitis, are provided in the table below.

Management Guidelines for Pancreatic Events, Including Pancreatitis

Event	Management
Amylase and/or lipase elevation, Grade 2	**Amylase and/or lipase > 1.5-2.0 · ULN:** ● Continue atezolizumab and tiragolumab. ● Monitor amylase and lipase weekly. ● For prolonged elevation (e.g., > 3 weeks), consider treatment with corticosteroids equivalent to 10 mg/day oral prednisone. **Asymptomatic with amylase and/or lipase > 2.0-5.0 · ULN:** ● Treat as a Grade 3 event.
Amylase and/or lipase elevation, Grade 3 or 4	● Withhold atezolizumab and tiragolumab for up to 12 weeks after event onset. [a] ● Refer patient to GI specialist. ● Monitor amylase and lipase every other day. ● If no improvement, consider treatment with corticosteroids equivalent to 1-2 mg/kg/day oral prednisone. ● If event resolves to Grade 1 or better, resume atezolizumab and tiragolumab. [b] ● If event does not resolve to Grade 1 or better while withholding atezolizumab and tiragolumab, permanently discontinue atezolizumab and tiragolumab and contact PI. [c] ● For recurrent events, permanently discontinue atezolizumab and tiragolumab and contact PI. [c]
Immune-mediated pancreatitis, Grade 2 or 3	● Withhold atezolizumab and tiragolumab for up to 12 weeks after event onset. [a] ● Refer patient to GI specialist. ● Initiate treatment with corticosteroids equivalent to 1-2 mg/kg/day IV methylprednisolone and convert to 1-2 mg/kg/day oral prednisone or equivalent upon improvement. ● If event resolves to Grade 1 or better, resume atezolizumab and tiragolumab. [b] ● If event does not resolve to Grade 1 or better while withholding atezolizumab and tiragolumab, permanently discontinue atezolizumab and tiragolumab and contact PI. [c] ● For recurrent events, permanently discontinue atezolizumab and tiragolumab and contact PI. [c]
Immune-mediated pancreatitis, Grade 4	● Permanently discontinue atezolizumab and tiragolumab and contact PI. [c] ● Refer patient to GI specialist. ● Initiate treatment with corticosteroids equivalent to 1-2 mg/kg/day IV methylprednisolone and convert to 1-2 mg/kg/day oral prednisone or equivalent upon improvement. ● If event does not improve within 48 hours after initiating corticosteroids, consider adding an immunosuppressive agent. ● If event resolves to Grade 1 or better, taper corticosteroids over ≥ 1

Event	Management
	month.

GI = gastrointestinal; ULN=upper limit of normal.

[a] Atezolizumab and tiragolumab may be withheld for a longer period of time (i.e., > 12 weeks after event onset) to allow for corticosteroids (if initiated) to be reduced to the equivalent of ≤ 10 mg/day oral prednisone. The acceptable length of the extended period of time must be based on the investigator's benefit-risk assessment and in alignment with the protocol requirements for the duration of treatment and documented by the investigator.

[b] If corticosteroids have been initiated, they must be tapered over ≥ 1 month to the equivalent of ≤ 10 mg/day oral prednisone before atezolizumab and tiragolumab can be resumed.

[c] Resumption of atezolizumab and tiragolumab may be considered in patients who are deriving benefit and have fully recovered from the immune-mediated event. The decision to re-challenge patients with atezolizumab should be based on the investigator's benefit-risk assessment and documented by the investigator.

6.3.11 Skin and subcutaneous tissue disorders

Immune-related dermatologic events are a potential risk with tiragolumab. The majority of cases of rash reported with the use of atezolizumab were mild in severity and self limiting, with or without pruritus. Although uncommon, cases of severe cutaneous adverse reactions such as Stevens-Johnson syndrome and toxic epidermal necrolysis have been reported with atezolizumab. A dermatologist should evaluate persistent and/or severe rash or pruritus. A biopsy should be considered unless contraindicated. Management guidelines for dermatologic events are provided in the table below.

Management Guidelines for Dermatologic Events

Event	Management
Dermatologic event, Grade 1	● Continue atezolizumab and tiragolumab. ● Consider treatment with topical corticosteroids and/or other symptomatic therapy (e.g., antihistamines).
Dermatologic event, Grade 2	● Continue atezolizumab and tiragolumab. ● Consider patient referral to dermatologist for evaluation and, if indicated, biopsy. ● Initiate treatment with topical corticosteroids. ● Consider treatment with higher-potency topical corticosteroids if event does not improve.
Dermatologic event, Grade 3	● Withhold atezolizumab and tiragolumab for up to 12 weeks after event onset. [a] ● Refer patient to dermatologist for evaluation and, if indicated, biopsy. ● Initiate treatment with corticosteroids equivalent to 10 mg/day oral prednisone, increasing dose to 1-2 mg/kg/day if event does not improve within 48−72 hours. ● If event resolves to Grade 1 or better, resume atezolizumab and tiragolumab. [b] ● If event does not resolve to Grade 1 or better while withholding atezolizumab and tiragolumab, permanently discontinue atezolizumab and tiragolumab and contact PI. [c]
Dermatologic event, Grade 4	● Permanently discontinue atezolizumab and tiragolumab and contact PI. [c]

Event	Management
Stevens Johnson syndrome or toxic epidermal necrolysis (any grade)	Additional guidance for Stevens Johnson syndrome or toxic epidermal necrolysis: ● Withhold tiragolumab and atezolizumab for suspected Stevens Johnson syndrome or toxic epidermal necrolysis. ● Confirm diagnosis by referring patient to a specialist (dermatologist, ophthalmologist or urologist as relevant) for evaluation and, if indicated, biopsy. ● Follow the applicable treatment and management guidelines above. ● If Stevens Johnson syndrome or toxic epidermal necrolysis, permanently discontinue tiragolumab and atezolizumab.

[a] Atezolizumab and tiragolumab may be withheld for a longer period of time (i.e., > 12 weeks after event onset) to allow for corticosteroids (if initiated) to be reduced to the equivalent of ≤ 10 mg/day oral prednisone. The acceptable length of the extended period of time must be based on the investigator's benefit-risk assessment and in alignment with the protocol requirements for the duration of treatment and documented by the investigator. [b]

If corticosteroids have been initiated, they must be tapered over ≥ 1 month to the equivalent of ≤ 10 mg/day oral prednisone before atezolizumab and tiragolumab can be resumed.

[c] Resumption of atezolizumab and tiragolumab may be considered in patients who are deriving benefit and have fully recovered from the immune-mediated event. The decision to re-challenge patients with atezolizumab should be based on the investigator's benefit-risk assessment and documented by the investigator.

6.3.12 Nervous System Disorders

Immune-related neurologic events are a potential risk with tiragolumab.

Patients may present with signs and symptoms of sensory and/or motor neuropathy. Diagnostic workup is essential for an accurate characterization to differentiate between alternative etiologies. Management guidelines for neurologic disorders are provided in the table below with specific guidelines for myelitis provided in a separate table.

Management Guidelines for Neurologic Disorders

Event	Management
Immune-mediated neuropathy, Grade 1	● Continue atezolizumab and tiragolumab. ● Investigate etiology. ● Any cranial nerve disorder (including facial paresis) should be managed as per Grade 2 management guidelines below.
Immune-mediated neuropathy, including facial paresis, Grade 2	● Withhold atezolizumab and tiragolumab for up to 12 weeks after event onset. [a] ● Investigate etiology. ● Initiate treatment as per institutional guidelines. ● For general immune-mediated neuropathy: o If event resolves to Grade 1 or better, resume atezolizumab and tiragolumab. [b] o If event does not resolve to Grade 1 or better while withholding atezolizumab and tiragolumab, permanently discontinue atezolizumab and tiragolumab and contact PI. [c] • For facial paresis:

Event	Management
	○ If event resolves fully, resume atezolizumab b ○ If event does not resolve fully while withholding atezolizumab, permanently discontinue atezolizumab. c
Immune-mediated neuropathy, including facial paresis, Grade 3 or 4	● Permanently discontinue atezolizumab and tiragolumab and contact PI. [c] ● Refer patient to neurologist ● Initiate treatment as per institutional guidelines.
Myasthenia gravis and Guillain-Barré syndrome (any grade)	● Permanently discontinue atezolizumab and tiragolumab and contact PI. [c] ● Refer patient to neurologist. ● Initiate treatment as per institutional guidelines. ● Consider initiation of corticosteroids equivalent to 1-2 mg/kg/day oral or IV prednisone.

[a] Atezolizumab and tiragolumab may be withheld for a longer period of time (i.e., > 12 weeks after event onset) to allow for corticosteroids (if initiated) to be reduced to the equivalent of ≤ 10 mg/day oral prednisone. The acceptable length of the extended period of time must be based on the investigator's benefit-risk and in alignment with the protocol requirements for the duration and treatment and documented by the investigator.

[b] If corticosteroids have been initiated, they must be tapered over ≥ 1 month to the equivalent of ≤ 10 mg/day oral prednisone before atezolizumab and tiragolumab can be resumed.

[c] Resumption of atezolizumab and tiragolumab may be considered in patients who are deriving benefit and have fully recovered from the immune-mediated event. The decision to re-challenge patients with atezolizumab should be based on the investigator's benefit-risk assessment and documented by the investigator.

Management Guidelines for Immune-Mediated Myelitis

Event	Management
Immune-mediated myelitis, Grade 1	• Continue atezolizumab unless symptoms worsen or do not improve. • Investigate etiology and refer patient to a neurologist.
Immune-mediated myelitis, Grade 2	• Permanently discontinue atezolizumab. • Investigate etiology and refer patient to a neurologist. • Rule out infection. • Initiate treatment with corticosteroids equivalent to 1-2 mg/kg/day oral prednisone.
Immune-mediated myelitis, Grade 3 or 4	• Permanently discontinue atezolizumab. • Refer patient to a neurologist. • Initiate treatment as per institutional guidelines.

6.3.13 Immune-Mediated Meningoencephalitis

Immune-mediated meningoencephalitis should be suspected in any patient presenting with signs or symptoms suggestive of meningitis or encephalitis, including, but not limited to, headache, neck pain, confusion, seizure, motor or

sensory dysfunction, and altered or depressed level of consciousness. Encephalopathy from metabolic or electrolyte imbalances needs to be distinguished from potential meningoencephalitis resulting from infection (bacterial, viral, or fungal) or progression of malignancy, or secondary to a paraneoplastic process.

All patients being considered for meningoencephalitis should be urgently evaluated with a CT scan and/or MRI scan of the brain to evaluate for metastasis, inflammation, or edema. If deemed safe by the treating physician, a lumbar puncture should be performed and a neurologist should be consulted.

Patients with signs and symptoms of meningoencephalitis, in the absence of an identified alternate etiology, should be treated according to the guidelines in the table below.

Management Guidelines for Immune-Mediated Meningoencephalitis

Event	Management
Immune-mediated meningoencephalitis, all grades	● Permanently discontinue atezolizumab and tiragolumab and contact PI. ● Refer patient to neurologist. ● Initiate treatment with corticosteroids equivalent to 1-2 mg/kg/day IV methylprednisolone and convert to 1-2 mg/kg/day oral prednisone or equivalent upon improvement. ● If event does not improve within 48 hours after initiating corticosteroids, consider adding an immunosuppressive agent. ● If event resolves to Grade 1 or better, taper corticosteroids over ≥ 1 month.

6.3.14 Renal and Urinary Disorders

Eligible patients must have adequate renal function. Renal function, including serum creatinine, should be monitored throughout study treatment. Patients with abnormal renal function should be evaluated and treated for other more common etiologies (including prerenal and postrenal causes, and concomitant medications such as nonsteroidal anti-inflammatory drugs). Refer the patient to a renal specialist if clinically indicated. A renal biopsy may be required to enable a definitive diagnosis and appropriate treatment.

Patients with signs and symptoms of nephritis, in the absence of an identified alternate etiology, should be treated according to the guidelines in the table below.

Management Guidelines for Renal Events

Event	Management
Renal event, Grade 1	● Continue atezolizumab and tiragolumab. ● Monitor kidney function, including creatinine, closely until values resolve to within normal limits or to baseline values.

Event	Management
Renal event, Grade 2	● Withhold atezolizumab and tiragolumab for up to 12 weeks after event onset. [a] ● Refer patient to renal specialist. ● Initiate treatment with corticosteroids equivalent to 1-2 mg/kg/day oral prednisone. ● If event resolves to Grade 1 or better, resume atezolizumab and tiragolumab. [b] ● If event does not resolve to Grade 1 or better while withholding atezolizumab and tiragolumab, permanently discontinue atezolizumab and tiragolumab and contact PI. [c]
Renal event, Grade 3 or 4	● Permanently discontinue atezolizumab and tiragolumab and contact PI. ● Refer patient to renal specialist and consider renal biopsy. ● Initiate treatment with corticosteroids equivalent to 1-2 mg/kg/day oral prednisone. ● If event does not improve within 48 hours after initiating corticosteroids, consider adding an immunosuppressive agent. ● If event resolves to Grade 1 or better, taper corticosteroids over ≥ 1 month.

[a] Atezolizumab and tiragolumab may be withheld for a longer period of time (i.e., > 12 weeks after event onset) to allow for corticosteroids (if initiated) to be reduced to the equivalent of ≤ 10 mg/day oral prednisone. The acceptable length of the extended period of time must be based on the investigator's benefit-risk and in alignment with the protocol requirements for the duration and treatment and documented by the investigator.

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[b] If corticosteroids have been initiated, they must be tapered over ≥ 1 month to the equivalent of ≤ 10 mg/day oral prednisone before atezolizumab and tiragolumab can be resumed.

[c] Resumption of atezolizumab and tiragolumab may be considered in patients who are deriving benefit and have fully recovered from the immune-mediated event. The decision to re-challenge patients with atezolizumab should be based on the investigator's benefit-risk assessment and documented by the investigator.

6.3.15 Immune-Mediated Myositis

Myositis or inflammatory myopathies are a group of disorders sharing the common feature of inflammatory muscle injury; dermatomyositis and polymyositis are among the most common disorders. Initial diagnosis is based on clinical (muscle weakness, muscle pain, skin rash in dermatomyositis), biochemical (serum creatine kinase increase), and imaging (electromyography/MRI) features, and is confirmed with a muscle biopsy.

Patients with signs and symptoms of myositis, in the absence of an identified alternate etiology, should be treated according to the guidelines in the table below.

Management Guidelines for Immune-Mediated Myositis

Event	Management
Immune-mediated myositis, Grade 1	● Continue atezolizumab and tiragolumab. ● Refer patient to rheumatologist or neurologist. ● Initiate treatment as per institutional guidelines.

Event	Management
Immune-mediated myositis, Grade 2	● Withhold atezolizumab and tiragolumab for up to 12 weeks after event onset [a] and contact PI. ● Refer patient to rheumatologist or neurologist. ● Initiate treatment as per institutional guidelines. ● Consider treatment with corticosteroids equivalent to 1-2 mg/kg/day IV methylprednisolone and convert to 1-2 mg/kg/day oral prednisone or equivalent upon improvement. ● If corticosteroids are initiated and event does not improve within 48 hours after initiating corticosteroids, consider adding an immunosuppressive agent. ● If event resolves to Grade 1 or better, resume atezolizumab and tiragolumab. [b] ● If event does not resolve to Grade 1 or better while withholding atezolizumab and tiragolumab, permanently discontinue atezolizumab and tiragolumab and contact PI. [c]
Immune-mediated myositis, Grade 3	● Withhold atezolizumab and tiragolumab for up to 12 weeks after event onset [a] and contact PI. ● Refer patient to rheumatologist or neurologist. ● Initiate treatment as per institutional guidelines. ● Respiratory support may be required in more severe cases. ● Initiate treatment with corticosteroids equivalent to 1-2 mg/kg/day IV methylprednisolone, or higher-dose bolus if patient is severely compromised (e.g., cardiac or respiratory symptoms, dysphagia, or weakness that severely limits mobility); convert to 1-2 mg/kg/day oral prednisone or equivalent upon improvement. ● If event does not improve within 48 hours after initiating corticosteroids, consider adding an immunosuppressive agent. ● If event resolves to Grade 1 or better, resume atezolizumab and tiragolumab. [b] ● If event does not resolve to Grade 1 or better while withholding atezolizumab and tiragolumab, permanently discontinue atezolizumab and tiragolumab and contact PI. [c] ● For recurrent events, treat as a Grade 4 event. Permanently discontinue atezolizumab and tiragolumab.[c]
Immune-mediated myositis, Grade 4	● Permanently discontinue atezolizumab and tiragolumab and contact PI. [c] ● Refer patient to rheumatologist or neurologist. ● Initiate treatment as per institutional guidelines. ● Respiratory support may be required in more severe cases. ● Initiate treatment with corticosteroids equivalent to 1-2 mg/kg/day IV methylprednisolone, or higher-dose bolus if patient is severely compromised (e.g., cardiac or respiratory symptoms, dysphagia, or weakness that severely limits mobility); convert to 1-2 mg/kg/day oral prednisone or equivalent upon improvement. ● If event does not improve within 48 hours after initiating corticosteroids, consider adding an immunosuppressive agent.

Event	Management
	● If event resolves to Grade 1 or better, taper corticosteroids over ≥ 1 month.

[a] Atezolizumab and tiragolumab may be withheld for a longer period of time (i.e., > 12 weeks after event onset) to allow for corticosteroids (if initiated) to be reduced to the equivalent of ≤ 10 mg/day oral prednisone. The acceptable length of the extended period of time must be based on the investigator's benefit-risk and in alignment with the protocol requirements for the duration and treatment and documented by the investigator. agreed upon by the investigator.

[b] If corticosteroids have been initiated, they must be tapered over ≥ 1 month to the equivalent of ≤ 10 mg/day oral prednisone before atezolizumab and tiragolumab can be resumed.

[c] Resumption of atezolizumab and tiragolumab may be considered in patients who are deriving benefit and have fully recovered from the immune-mediated event. The decision to re-challenge patients with atezolizumab should be based on the investigator's benefit-risk assessment and documented by the investigatorPatients can be rechallenged with atezolizumab and tiragolumab- only after approval has been documented by the investigator..

6.3.16 Hemophagocytic lymphohistiocytosis and Macrophage Activation Syndrome

Immune-mediated reactions may involve any organ system and may lead to hemophagocytic lymphohistiocytosis (HLH) and macrophage activation syndrome (MAS).

Clinical and laboratory features of severe CRS overlap with HLH, and HLH should be considered when CRS presentation is atypical or prolonged.

Patients with suspected HLH should be diagnosed according to published criteria by McClain and Eckstein (2014). A patient should be classified as having HLH if five of the following eight criteria are met:

- Fever ≥ 38.5°C
- Splenomegaly
- Peripheral blood cytopenia consisting of at least two of the following:
  - Hemoglobin < 90 g/L (9 g/dL) (< 100 g/L [10 g/dL] for infants < 4 weeks old)
  - Platelet count < $100 \cdot 10^9$/L (100,000/⌈L)
  - ANC < $1.0 \cdot 10^9$/L (1000/⌈L)
- Fasting triglycerides > 2.992 mmol/L (265 mg/dL) and/or fibrinogen < 1.5 g/L (150 mg/dL)
- Hemophagocytosis in bone marrow, spleen, lymph node, or liver
- Low or absent natural killer cell activity
- Ferritin > 500 mg/L (500 ng/mL)
- Soluble IL-2 receptor (soluble CD25) elevated ≥ 2 standard deviations above age-adjusted laboratory-specific norms

Patients with suspected MAS should be diagnosed according to published criteria for systemic juvenile idiopathic arthritis by Ravelli et al. (2016). A febrile patient should be classified as having MAS if the following criteria are met:

- Ferritin > 684 mg/L (684 ng/mL)

- At least two of the following:
  - Platelet count ≤ 181 · 10^9/L (181,000/⌈L)
  - AST ≥ 48 U/L
  - Triglycerides > 1.761 mmol/L (156 mg/dL)
  - Fibrinogen ≤ 3.6 g/L (360 mg/dL)

Patients with suspected HLH or MAS should be treated according to the guidelines in the table below.

Management Guidelines for Suspected Hemophagocytic Lymphohistiocytosis or Macrophage Activation Syndrome

Event	Management
Suspected HLH or MAS	● Permanently discontinue atezolizumab and tiragolumab and contact PI. ● Consider patient referral to hematologist. ● Initiate supportive care, including intensive care monitoring if indicated per institutional guidelines. ● Consider initiation of IV corticosteroids, an immunosuppressive agent, and/or anti-cytokine therapy. ● If event does not respond to treatment within 24 hours, contact PI and initiate treatment as appropriate according to published guidelines (La Rosée 2015; Schram and Berliner 2015; La Rosée et al. 2019). ● If event resolves to Grade 1 or better, taper corticosteroids over ≥ 1 month.

HLH = hemophagocytic lymphohistiocytosis; MAS = macrophage activation syndrome.

7.0 REGULATORY AND REPORTING REQUIREMENTS

The entities providing oversight of safety and compliance with the protocol require reporting as outlined below. Please refer to Appendix B for definitions and Appendix C for a grid of reporting timelines.

Adverse events will be tracked from start of treatment through 90 days after the last day of treatment with atezolizumab and tiragolumab. All adverse events must be recorded on the toxicity tracking case report form (CRF) with the exception of:

- Baseline adverse events, which shall be recorded on the medical history CRF

Refer to the data submission schedule in Section 11 for instructions on the collection of AEs in the EDC.

Reporting requirements for Washington University study team may be found in Section 7.1. Reporting requirements for secondary site study teams participating in Washington University-coordinated research may be found in Section 7.2.

7.1 Sponsor-Investigator Reporting Requirements

7.1.1 Reporting to the Human Research Protection Office (HRPO) at Washington University

Reporting will be conducted in accordance with Washington University IRB Policies.

Pre-approval of all protocol exceptions must be obtained prior to implementing the change.

7.1.2 Reporting to the Quality Assurance and Safety Monitoring Committee (QASMC) at Washington University

The Washington University Sponsor Investigator (or designee) is required to notify the QASMC of any unanticipated problems involving risks to participants or others occurring at WU or any BJH or SLCH institution that has been reported to and acknowledged by HRPO. (Unanticipated problems reported to HRPO and withdrawn during the review process need not be reported to QASMC.)

QASMC must be notified within **10 days** of receipt of IRB acknowledgment via email to qasmc@wustl.edu. Submission to QASMC must include the myIRB form and any supporting documentation sent with the form.

For events that occur at secondary sites, the Washington University Sponsor Investigator (or designee) is required to notify the QASMC within 10 days of Washington University notification via email to qasmc@wustl.edu. Submission to QASMC must include either the myIRB form and supporting documentation or (if not submitted to myIRB) the date of occurrence, description of the event, whether the event is described in the currently IRB approved materials, the event outcome, determination of relatedness, whether currently enrolled participants will be notified, and whether the informed consent document and/or any study procedures will be modified as a result of this event.

7.1.3 Reporting to Genentech

Certain events require immediate reporting. The investigator must report such events to Genentech immediately; under no circumstances should reporting take place more than 24 hours after the investigator learns of the event. The following is a list of events that the investigator must report to Genentech within 24 hours after learning of the event, regardless of relationship to study treatment:

- Serious adverse events
- Adverse events of special interest (defined in Section 7.1.3.1)
- Pregnancies (within 90 days after the last dose of tiragolumab or within 5 months after the last dose of atezolizumab)
- Data related to the product usage during breastfeeding

- Data related to overdose, abuse, misuse, or medication error

In addition, reasonable attempts should be made to obtain and submit the age or age group of the patient, in order to be able to identify potential safety signals specific to a particular population.

For serious adverse events and adverse events of special interest, the investigator must report new significant follow-up information to Genentech immediately (i.e., no more than 24 hours after becoming aware of the information). New significant information includes the following:

- New signs or symptoms or a change in the diagnosis
- Significant new diagnostic test results
- Change in causality based on new information
- Change in the event's outcome, including recovery
- Additional narrative information on the clinical course of the event

The completed MedWatch form should be sent to the Genentech contact specified below. Transmission of these reports will be either electronically via email or by vax and within the timelines specified below.

All written IND Safety Reports submitted to the FDA by the Investigator must also be faxed to Genentech Drug Safety:
Fax: (650) 225-4682 or (650) 225-4630
Email: usds_aereporting-d@gene.com

All product complaints (refer to Appendix B) without an AE should call via:

PC hotline number: 800-334-0290 (M-F 5am-5pm PST)

Investigators must report all of the above-mentioned single case reports adequately to Genentech on a MedWatch within one (1) business day of the awareness date.

The Investigator will email Genentech a quarterly line-listing documenting single case reports sent by the Investigator to Genentech in the preceding time period. These will be sent to ctvist_drugsafety@gene.com. Following Case Transmission Verification, single case reports which have not been received by Genentech shall be forwarded by the Investigator to Genentech within five (5) calendar days from request by Genentech.

The investigator will forward listings of non-serious AEs originating from the study to Genentech/Roche as needed on an ad-hoc basis.

For questions related to safety reporting, please contact Genentech Drug Safety:
Tel: 888-835-2555
Fax: 650-225-4682 or 650-225-4630

Additionally, all IND annual reports submitted to the FDA by the Sponsor-Investigator should be copied to Genentech.. The Investigator will forward a copy of the Final Study Report to Genentech upon completion of the study. Finally, any literature articles that are a result of the study should be sent to Genentech. Copies of such reports should be sent to the Genentech Drug Safety CTV oversight mailbox at: ctvist_drugsafety@gene.com and to the assigned Clinical Operations contact for the study:
Erica Winsch (imCORE-USMA-OPS-d@gene.com)

Post-Study Adverse Events

The investigator should expeditiously report any SAE occurring after a subject has completed or discontinued study participation if attributed to prior to atezolizumab or tiragolumab exposure. If the investigator should become aware of the development of cancer or a congenital anomaly in a subsequently conceived offspring of a female subject who participated in the study, this should be reported as an SAE adequately to Genentech drug Safety during follow up period.

Investigators must report all the above mentioned single case reports adequately to Genentech on a MedWatch within one (1) business day of the awareness date.

Batch ID/lot ID for biologics associated with AE/SSR/PC/AESI must be included when submitting the case reports to Genentech.

For blinded studies, batch ID/lot ID for biologics associated with AE/SSR/PC/AESI must be submitted at the end of the study to Genentech.

7.1.3.1 Adverse Events of Special Interest

Adverse events of special interest are required to be reported by the investigator to Genentech immediately (i.e., no more than 24 hours after learning of the event; see Section 5.4.2 for reporting instructions). Adverse events of special interest for this study are as follows:

- Cases of potential drug-induced liver injury that include an elevated ALT or AST in combination with either an elevated bilirubin or clinical jaundice, as defined by Hy's Law (see Section 5.3.5.7)
- Suspected transmission of an infectious agent by the study treatment, as defined as: Any organism, virus, or infectious particle (e.g., prion protein transmitting transmissible spongiform encephalopathy), pathogenic or non-pathogenic, is considered an infectious agent. A transmission of an infectious agent may be suspected from clinical symptoms or laboratory findings that indicate an infection in a patient exposed to a medicinal product. This term applies only when a contamination of study treatment is suspected.

- Pneumonitis
- Colitis
- Endocrinopathies: diabetes mellitus, pancreatitis, adrenal insufficiency, hyperthyroidism, and hypophysitis
- Hepatitis, including AST or ALT > 10 x ULN
- Systemic lupus erythematosus
- Neurological disorders: Guillain Barré syndrome, myasthenic syndrome or myasthenia gravis, and meningoencephalitis, facial paresis, myelitis
- Events suggestive of hypersensitivity, infusion-related reactions, cytokine release syndrome, and hemophagocytic lymphohistiocytosis,
- Pericardial disorders
- Nephritis
- Ocular toxicities (e.g., uveitis, retinitis, optic neuritis)
- Myositis
- Myopathies, including rhabdomyolysis
- Grade ≥ 2 cardiac disorders
- Vasculitis
- Autoimmune hemolytic anemia
- Severe cutaneous reactions (e.g., Stevens-Johnson syndrome, dermatitis bullous, toxic epidermal necrolysis)

7.1.4 Reporting to the FDA

The conduct of the study will comply with all FDA safety reporting requirements. **PLEASE NOTE THAT REPORTING REQUIREMENTS FOR THE FDA DIFFER FROM REPORTING REQUIREMENTS FOR HRPO/QASMC.** It is the responsibility of the Washington University Sponsor-Investigator to report to the FDA as follows:

- Report any unexpected fatal or life-threatening suspected adverse reaction (refer to Appendix B for definitions) no later than **7 calendar days** after initial receipt of the information.
- Report a suspected adverse reaction that is both serious and unexpected (SUSAR, refer to Appendix B) no later than **15 calendar days** after it is determined that the information qualifies for reporting. Report an adverse event (refer to Appendix B) as a suspected adverse reaction only if there is evidence to suggest a causal relationship between the drug and the adverse event, such as:
  - A single occurrence of an event that is uncommon and known to be strongly associated with drug exposure
  - One or more occurrences of an event that is not commonly associated with drug exposure but is otherwise uncommon in the population exposed to the drug
  - An aggregate analysis of specific events observed in a clinical trial that

indicates those events occur more frequently in the drug treatment group than in a concurrent or historical control group

- Report any findings from epidemiological studies, pooled analysis of multiple studies, or clinical studies that suggest a significant risk in humans exposed to the drug no later than **15 calendar days** after it is determined that the information qualifies for reporting.
- Report any findings from animal or in vitro testing that suggest significant risk in humans exposed to the drug no later than **15 calendar days** after it is determined that the information qualifies for reporting.
- Report any clinically important increase in the rate of a serious suspected adverse reaction of that listed in the protocol or IB within **15 calendar days** after it is determined that the information qualifies for reporting.

Submit each report as an IND safety report in a narrative format or on FDA Form 3500A or in an electronic format that FDA can process, review, and archive. Study teams must notify the Siteman Cancer Center Protocol Development team of each potentially reportable event within 1 business day after initial receipt of the information, and must bring the signed 1571 and FDA Form 3500A to the Siteman Cancer Center Protocol Development team no later than 1 business day prior to the due date for reporting to the FDA.

Each notification to FDA must bear prominent identification of its contents ("IND Safety Report") and must be transmitted to the review division in the Center for Drug Evaluation and Research (CDER) or in the Center for Biologics Evaluation and Research (CBER) that has responsibility for review of the IND. Relevant follow-up information to an IND safety report must be submitted as soon as the information is available and must be identified as such ("Follow-up IND Safety Report").

7.1.5 Reporting to Secondary Sites

The Washington University Sponsor-Investigator (or designee) will notify the research team at each secondary site of all unanticipated problems involving risks to participants or others that have occurred at other sites within **10 working days** of the occurrence of the event or notification of the Sponsor-Investigator (or designee) of the event. This includes events that take place both at Washington University and at other secondary sites, if applicable. Refer to Section 16.0 (Multicenter Management) for more information.

7.2 Secondary Site Reporting Requirements

The research team at each secondary site is required to promptly notify the Washington University Sponsor-Investigator and designee of all serious adverse events (refer to Appendix B, Section D) within **1 working day** of the occurrence of the event or notification of the secondary site's PI of the event. This notification may take place via email if there is not yet enough information for a formal written report (using FDA Form 3500a

(MedWatch) and Washington University's cover sheet (Appendix D)). A formal written report must be sent to the Washington University Sponsor-Investigator and designee within **4 calendar days** (for fatal or life-threatening suspected adverse reactions) or **11 calendar** days (for serious unexpected adverse reactions) of the occurrence of the event or notification of the secondary site's PI of the event.

The research team at a secondary site is responsible for following its site's guidelines for reporting applicable events to its site's IRB according to its own institutional guidelines. The research team at Washington University is responsible for reporting all applicable events to the FDA and Genentech as needed.

Washington University pre-approval of all protocol exceptions must be obtained prior to implementing the change. Local IRB approval must be obtained as per local guidelines. Washington University IRB approval is not required for protocol exceptions occurring at secondary sites.

7.3 Exceptions to Expedited Reporting

Events that do not require expedited reporting as described in Section 7.1 include:

- planned hospitalizations
- hospitalizations < 24 hours
- respite care
- events related to disease progression

Events that do not require expedited reporting must still be captured in the EDC.

8.0 PHARMACEUTICAL INFORMATION

8.1 Atezolizumab (Tecentriq)

8.1.1 Atezolizumab Description

Atezolizumab is a humanized IgG1 monoclonal antibody consisting of two heavy chains (448 amino acids) and two light chains (214 amino acids). Atezolizumab targets human PD-L1 and inhibits its interaction with its receptor PD-1. Atezolizumab also blocks the binding of PD-L1 to B7.1, an interaction that is reported to provide additional inhibitory signals to T cells (Butte et al. 2007).

8.1.2 Clinical Pharmacology

PD-L1 may be expressed on tumor cells and/or tumor infiltrating immune cells and can contribute to the inhibition of the anti-tumor immune response in the tumor microenvironment. Binding of PD-L1 to the PD-1 and B7.1 receptors found on T cells and antigen presenting cells suppresses cytotoxic T-cell activity, T-cell proliferation and cytokine production.

Atezolizumab is a monoclonal antibody that binds to PD-L1 and blocks its interactions with both PD-1 and B7.1 receptors. This releases the PD-L1/PD-1 mediated inhibition of the immune response, including activation of the anti-tumor immune response without inducing antibody dependent cellular cytotoxicity. In syngeneic mouse tumor models, blocking PD-L1 activity resulted in decreased tumor growth.

8.1.3 Pharmacokinetics and Drug Metabolism

Patients' exposure to atezolizumab increased dose proportionally over the dose range of 1 mg/kg to 20 mg/kg, including a dose of 1200 mg administered every 3 weeks. The clearance (CV%) was 0.20 L/day (29%), the volume of distribution at steady state was 6.9 L, and the terminal half-life was 27 days. Steady state was achieved after 6 to 9 weeks following multiple doses. The systemic accumulation ratio for every 2 weeks administration and every 3 weeks administration was 3.3- and 1.9-fold, respectively. Atezolizumab clearance was found to decrease over time, with a mean maximal reduction (CV%) from baseline value of approximately 17% (41%); however, the decrease in clearance was not considered clinically relevant.

8.1.4 Supplier

Atezolizumab is provided by Genentech.

8.1.5 Dosage Form and Preparation

The atezolizumab 840 mg drug product will be supplied in a single-use, 15-mL USP/Ph. Eur. Type 1 glass vial as a colorless to slightly yellow, sterile, preservative-free clear liquid solution intended for IV administration. The vial is designed to deliver 14 mL (840 mg) of atezolizumab solution, but may contain more than the stated volume to enable delivery of the entire 14-mL volume. Patients will receive 1680 mg of atezolizumab, so 2 vials will be used for each dose.

The prescribed dose of atezolizumab should be diluted in 250 mL 0.9% NaCl and infused through a 0.2 micrometer in-line filter. The IV bag may be constructed of PVC or PO; the IV infusion line may be constructed of PVC or PE; and the 0.2 micrometer in-line filter may be constructed of PES. The prepared solution may be stored at 2°C-8°C or room temperature for up to 8 hours.

8.1.6 Storage and Stability

2°C-8°C (36°F-46°F) Vial contents should not be frozen or shaken and should be protected from direct sunlight.

Stability studies are ongoing.

CAUTION: No preservative is used in atezolizumab; therefore, the vial is intended for single use only. Discard any unused portion of drug remaining in a vial.

8.1.7 Administration

Atezolizumab is administered as an intravenous infusion over 60 minutes. If the first infusion is tolerated, all subsequent infusions may be delivered over 30 minutes. Do not administer atezolizumab as an intravenous push or bolus. No premedication is indicated for administration of Cycle 1 of atezolizumab. Patients who experience an infusion related reaction with Cycle 1 of atezolizumab may receive premedication with antihistamines or antipyretics/analgesics (e.g. acetaminophen) for subsequent infusions.

8.2 Tiragolumab

8.2.1 Tiragolumab Description

Tiragolumab is a fully human IgG1/kappa mAb that binds TIGIT and prevents its interaction with PVR. The blockade of TIGIT by tiragolumab represents a novel strategy for cancer therapy because its mechanism of action is complementary to that of other checkpoint inhibitors, such as anti-PD-L1/PD-1. Consequently, tiragolumab as a single-agent or in combination with anti-cancer therapies, may enhance the magnitude and quality of tumor-specific T-cell responses, which may result in meaningful anti-tumor activity in solid tumors and in hematological malignancies.

8.2.2 Clinical Pharmacology

The PK of tiragolumab was evaluated as a single-agent in the Phase Ia portion and in combination with atezolizumab in the Phase Ib portion of Study GO30103. As of 2 December 2019, preliminary PK data at tiragolumab doses ranging from 2 to 1200 mg every 21 days (Q3W) show that tiragolumab exposures increased approximately doseproportionally with increasing dose levels. Similarly, tiragolumab exposures increased approximately in a dose-proportional manner from 2 to 1200 mg in combination with 1200 mg atezolizumab Q3W. Further, the PK of tiragolumab in combination with atezolizumab appeared to be consistent with the PK of tiragolumab administered as a single-agent. The geometric mean Cycle 1 Cmax of tiragolumab ranged from 0.771 to 268 µg/mL and geometric mean Cycle 1 AUC0-21 ranged from 2.61 to 2880 day · µg/mL for the dose levels 2 to 1200 mg in the Phase Ia portion of the study. Similarly, the geometric mean Cycle 1 Cmax ranged from 0.582 to 339 µg/mL and geometric mean AUC0-21 ranged from 2.60 to 3000 day · µg/mL for tiragolumab dose levels 2 to 1200 mg in combination with atezolizumab. Preliminary population-PK analysis estimated tiragolumab clearance at 0.28 L/day and linear drug elimination half-life at approximately 15 days.

8.2.3 Pharmacokinetics and Drug Metabolism in Animals

Tiragolumab binds to recombinant human and cynomolgus monkey TIGIT at comparable affinity (0.81 and 4.42 nM, respectively) and blocks binding to its high-affinity receptor PVR at comparable 50% inhibitor concentration reduction (IC50) (± standard deviation [SD]) values of 2.15± 0.38 nM and 0.44± 0.06 nM respectively. Tiragolumab does not bind mouse or rat TIGIT. The PK of tiragolumab following a single IV injection to cynomolgus monkeys was evaluated. In summary, all animals developed anti-drug antibodies (ADAs) from Day 10 onward. Up to Day 7 post-dose, prior to the onset of the ADA response, tiragolumab demonstrated biphasic disposition, and PK parameters such as observed maximum observed serum concentration (Cmax) and area under the concentration-time curve (AUC) from time 0 to 7 days post-dose (AUC0-7) were similar to other IgG1 mAbs against low-expressing targets (Deng et al. 2011). The toxicokinetics (TK) of tiragolumab were evaluated as part of the repeat-dose Good Laboratory Practice (GLP) toxicity studies in cynomolgus monkeys (Study 15-1334 and Study 17-2910). After repeated administration of MTIG7192A in the 1-month study (Study 15-1334), all animals in the 10 and 30 mg/kg dose groups developed a detectable ADA response that led to the loss of exposure in most animals. However, only 1 of 10 animals in the 100 mg/kg dose group developed a detectable ADA response. In the 26-week study (Study 17-2910), 5 of 6 animals in the 30 mg/kg and 5 of 6 animals in the 100 mg/kg dose groups were ADA-positive and had slightly lower MTIG7192A serum concentrations when compared to animals with negative ADA results. In both studies, systemic exposure was maintained in the ADA-negative animals, and the TK profile was consistent with that expected based on the single-dose PK study. The serum concentration□time profiles of all dose groups exhibited apparent biphasic disposition in which a rapid initial distribution phase was followed by a slower elimination phase.

8.2.4 Supplier

Tiragolumab is provided by Genentech.

8.2.5 Dosage Form and Preparation

Tiragolumab (also known as MTIG7192A) is a fully human mAb that binds to TIGIT. The recombinant antibody is produced in Chinese hamster ovary (CHO) cells, and consists of two heavy chains (456 amino acid residues each) and two light chains (220 amino acid residues each). There are two N-linked glycosylation sites (Asn306) in the fragment crystallizable (Fc) domain. Tiragolumab will be supplied as a sterile liquid in a single-use, 15-mL glass vial. The vial contains approximately 10 mL (600 mg) of tiragolumab solution.

Tiragolumab is to be administered by IV infusion either neat or after dilution in 0.9% NaCl. The concentration of tiragolumab is 60 mg/mL. Using this

concentration, calculate the appropriate volume of undiluted study drug according to patient dose and product concentration.

Tiragolumab dose	Tiragolumab (60 mg/mL, 600 mg)
120 mg	2.0 mL*
300 mg	5.0 mL
420 mg	7.0 mL
600 mg	10 mL
840 mg	14 mL
1200 mg	20 mL

* doses must be administered to the gradation on syringes (e.g. 1 mL increments)

The final concentration of drug in the IV bag should be between 0.2 and 12.0 mg/mL. It may be necessary to remove an amount of saline from the IV bag equivalent to the amount of study drug volume to be added.

A 0.2 µm in-line filter must be used with the infusion set during administration. The compatibility of tiragolumab with diluents other than described is unknown.

8.2.6 Storage and Stability

Tiragolumab must be prepared for dosing under appropriate aseptic conditions as it does not contain antimicrobial preservatives. The dose solution should be used immediately. If not used immediately, the total storage time of the dose solution prior to administration should not exceed 24 hours to limit the risk of microbial growth in case of accidental contamination. The recommended storage condition for the drug product and dose solution is 2°C–8°C, but dose solutions may be stored at room temperature for up to a maximum of 4 hours. The compatibility of tiragolumab with diluents other than described is unknown.

8.2.7 Administration

Tiragolumab must not be infused into the same line or cannula concomitantly with other drug infusions, including parenteral nutrition. Infusions of blood products and any electrolyte supplementation must not occur simultaneously with infusion of tiragolumab. A 0.2 micron in line filter must be used with the infusion set during administration.

9.0 CORRELATIVE STUDIES

9.1 Tissue for Whole Exome Sequencing

At least 11 unstained positively charged 5µm slides from resection or shave biopsyplus one contiguous H&E stained slide will be shipped to Natera using the address in Section

9.4. A de-identified copy of the associated pathology report should accompany the tissue specimen.

9.2 Blood for Whole Exome Sequencing

Six mL of blood will be collected into a K2-EDTA tube for whole exome sequencing for development of the Signatera assay during Step 1 screening. Immediately after the draw, thoroughly mix the blood with the anticoagulant by gently inverting the tube 8x. Within 2 hours of draw, the blood will be shipped at ambient temperature with a refrigerated gel pack overnight to Natera using the address in Section 9.4.

9.3 Blood for ctDNA

Twenty mL of peripheral blood will be drawn into 2 pre-labeled circulating tumor DNA BCT tubes (Streck 218962, NAT-800632) at the following time points:

- Step 2 screening (4 to 12 weeks after date of surgery)
- C3D1
- C6D1
- C9D1
- C12D1
- End of treatment (OPTIONAL)

We will follow ctDNA titers longitudinally and correlate level changes with treatment response and/or relapse based on clinical and/or radiographical assessments. Note that ctDNA testing will be discontinued after two consecutive negative tests.

Immediately after blood is drawn, thoroughly mix the blood with the anticoagulant by gently inverting it 8 times. Do not shake vigorously.

Tubes should be shipped to Natera at ambient temperature within 24 hours of collection.

9.4 Shipping Instructions

All specimens should be shipped to the following address:

ATTN: Accessioning – Signatera
Natera Inc.
13011A McCallen Pass
Austin, TX 78753
(877) 869-3052

10.0 DATA SUBMISSION SCHEDULE

Case report forms with appropriate source documentation will be completed according to the schedule listed in this section. Data may be up to 1 cycle behind at any given time.

Case Report Form	Submission Schedule
Original Consent Form	Prior to registration
On-Study Form Medical History Form Treatment Assignment Form	Prior to starting treatment
ctDNA Form	Baseline, C3D1, C6D1, C9D1, C12D1, end of treatment
Treatment Form	Every cycle
Toxicity Form	Continuous
Treatment Summary Form	Completion of treatment
Follow Up Form	Frequency per study calendar
Imaging Form	Baseline, every 12 weeks for the first 2 years after surgery, every 24 weeks for the third year after surgery
Recurrence Form	Time of disease recurrence
Death Form	Time of death
MedWatch Form	See Section 7.0 for reporting requirements

Any queries generated by Washington University must be responded to within 28 days of receipt by the participating site. The Washington University research team will conduct a regular review of data status at all secondary sites, with appropriate corrective action to be requested as needed.

10.1 Adverse Event Collection in the Case Report Forms

All adverse events that occur beginning with start of treatment (minus exceptions defined in Section 7.0) must be captured in the Toxicity Form. Baseline AEs should be captured on the Medical History Form.

Participant death due to disease progression should be reported on the Toxicity Form as grade 5 disease progression. If death is due to an AE (e.g. cardiac disorders: cardiac arrest), report as a grade 5 event under that AE. Participant death must also be recorded on the Death Form.

11.0 MEASUREMENT OF EFFECT

Tumor imaging is strongly preferred to be acquired by CT. For the abdomen and pelvis, contrast-enhanced MRI may be used when CT with iodinated contrast is contraindicated, or when mandated by local practice. The same imaging technique regarding modality, ideally the same scanner, and the use of contrast should be used in a participant throughout the study to optimize the reproducibility of the assessment of existing and new tumor burden and improve the accuracy of the assessment of response or progression based on imaging.

For the purposes of this study, patients should be re-evaluated every 12 weeks (+/- 2 weeks) for the first two years then every 24 weeks (+/- 2weeks) x2 for the third year, in addition to a baseline scan.

Relapse-free survival is defined as the time from randomization to any of the following events:

recurrence of melanoma at any site (local, in-transit or regional lymph nodes or distant recurrence) or death due to any cause. New incident cases of melanoma and second cancer diagnoses are not counted as events for recurrence-free survival.

12.0 DATA AND SAFETY MONITORING

In compliance with the Washington University Institutional Data and Safety Monitoring Plan, an independent Data and Safety Monitoring Board (DSMB) will be specifically convened for this trial to review toxicity data. A DSMB will consist of no fewer than 3 members including 2 clinical investigators and a biostatistician. DSMB members must be employed by Washington University, Barnes-Jewish Hospital, or St. Louis Children's Hospital. Like investigators, DSMB members are subject to the Washington University School of Medicine policies regarding standards of conduct. Individuals invited to serve on the DSMB will disclose any potential conflicts of interest to the trial principal investigator and/or appropriate university officials, in accordance with institution policies. Potential conflicts that develop during a trial or a member's tenure on a DSMB must also be disclosed.

Until such a time as the first secondary site enrolls its first patient, a semi-annual DSM report to be prepared by the study team will be submitted to the Quality Assurance and Safety Monitoring Committee (QASMC) semi-annually beginning six months after study activation at Washington University (if at least one patient has been enrolled) or one year after study activation (if no patients have been enrolled at the six-month mark).

The DSM report for the DSMB will be prepared by the study team with assistance from the study statistician, will be reviewed by the DSMB, and will be submitted to the QASM Committee. The DSMB must meet at least every six months beginning six months after enrollment of the first patient at a secondary site, no more than one month prior to the due date of the DSM report to QASMC. This report will include:

- HRPO protocol number, protocol title, Principal Investigator name, data coordinator name, regulatory coordinator name, and statistician
- Date of initial HRPO approval, date of most recent consent HRPO approval/revision, date of HRPO expiration, date of most recent QA audit, study status, and phase of study
- History of study including summary of substantive amendments; summary of accrual suspensions including start/stop dates and reason; and summary of protocol exceptions, error, or breach of confidentiality including start/stop dates and reason
- Study-wide target accrual and study-wide actual accrual including numbers from participating sites
- Protocol activation date at each participating site
- Average rate of accrual observed in year 1, year 2, and subsequent years at each participating site
- Expected accrual end date, accrual by site, and accrual by cohort
- Objectives of protocol with supporting data and list the number of participants who have met each objective
- Measures of efficacy

- Early stopping rules with supporting data and list the number of participants who have met the early stopping rules
- Summary of toxicities at all participating sites and separated by cohorts
- Abstract submissions/publications
- Summary of any recent literature that may affect the safety or ethics of the study

Further DSMB responsibilities are described in the DSMB charter.

The study principal investigator and coordinator will monitor for serious toxicities on an ongoing basis. Once the principal investigator or coordinator becomes aware of an adverse event, the AE will be reported to the HRPO and QASMC according to institutional guidelines (please refer to Section 7.0).

Refer to the Washington University Quality Assurance and Data Safety Monitoring Committee Policies and Procedures for full details on the responsibilities of the DSMB. This is located on the QASMC website at https://siteman.wustl.edu/research/clinical-research-resources/protocol-office-prmcqasmc/.

13.0 AUDITING

As coordinating center of this trial, Washington University (via the Quality Assurance and Safety Monitoring Committee (QASMC) will monitor each participating site to ensure that all protocol requirements are being met; that applicable federal regulations are being followed; and that best practices for patient safety and data collection are being followed per protocol. Participating sites will be asked to send copies of all audit materials, including source documentation. The audit notification will be sent to the Washington University Research Patient Coordinator, who will obtain the audit materials from the participating institution.

Notification of an upcoming audit will be sent to the research team one month ahead of the audit. Once accrual numbers are confirmed, and approximately 30 days prior to the audit, a list of the cases selected for review (up to 10 for each site) will be sent to the research team. However, if during the audit the need arises to review cases not initially selected, the research team will be asked to provide the additional charts within two working days.

Items to be evaluated include:
- Subject screening and enrollment
- Reporting of adverse events
- Maintenance of HIPAA compliance
- Completeness of regulatory documentation
- Completeness of participant documentation
- Acquisition of informed consent
- IRB documentation
- Issues of protocol adherence

Additional details regarding the auditing policies and procedures can be found at https://siteman.wustl.edu/research/clinical-research-resources/protocol-office-prmcqasmc/.

14.0 STATISTICAL CONSIDERATIONS

14.1 Study Design

This is an open-label, multi-center, Simon two-stage phase II study consisting of random allocation of a portion of patients to a small reference arm to offer some protection against the uncertainties and potential biases of a one-armed study while retaining some of the statistical efficiency. The reference arm is not a control group in the traditional sense and there will not be a formal efficacy comparison between the treatment and reference groups (Herson and Carter, 1986; Rubinstein, et. al., 2009).

The purpose of this study is to evaluate the efficacy and safety of atezolizumab and tiragolumab in stage II melanoma patients who had undergone complete resection and been tested positive for circulating tumor DNA (ctDNA). In this study, the treatment arm (i.e., atezolizumab and tiragolumab) will not be compared with the reference arm (i.e., atezolizumab only); it will be analyzed against historical control as a one-armed study. The reference arm is intended to only act as a check on the similarity of the current patients to the historical control for a descriptive estimation of ctDNA clearance.

14.2 Primary Endpoint

ctDNA clearance rate, defined as the proportion of ctDNA+ patients having a ctDNA negative test at C3D1.

14.3 Secondary Endpoints

- Confirmed ctDNA negative rate, defined as the proportion of ctDNA-positive patients having two consecutive negative ctDNA tests.

- Relapse free survival (RFS), defined as the duration of time from the date of positive ctDNA being confirmed to the date of earliest disease relapse or death, whichever occurs first. Patients who neither relapse nor die by the data cutoff date will be censored at the last follow up date.

- Distant metastasis free survival (DMFS), defined as the duration of time from the date of positive ctDNA being confirmed to the date of appearance of a distant metastasis or death, whichever occurs first. Patients who neither develop distant metastasis nor die by the data cutoff date will be censored at the last follow up date.

- Overall survival (OS), defined as the duration of time from the date of positive ctDNA being confirmed to death from any cause. Patients who are alive by the data cutoff date will be censored at the last follow up date.

- Safety and tolerability will be defined as the number of treatment (i.e., atezolizumab and tiragolumab) related grade 3 or greater adverse events (AEs). Adverse events will be assessed using CTCAE v5.0 criteria. Safety will be monitored from the time of initiation of study treatment to 30 days after discontinuation of therapy.

14.4 Statistical Hypotheses

We hypothesize that for ctDNA+ stage II melanoma patients, using atezolizumab and tiragolumab will lead to at least a 25% ctDNA clearance at C3D1, while a null ctDNA clearance rate is 10%. [22]

14.5 Sample Size Determination

Under Simon's two-stage optimal design (Simon, 1989), with one-sided alpha = 0.05 and power = 0.8, assuming using atezolizumab and tiragolumab will have a ctDNA clearance rate of 25% and a null rate is 10%, in stage 1, 18 patients will be accrued for the treatment arm (atezolizumab + tiragolumab). If 3 or more patients are observed to be ctDNA-negative at C3D1 for the treatment arm, we will continue the treatment of atezolizumab and tiragolumab to stage 2 with an additional 25 patients being accrued. Otherwise the study will be stopped. If 8 or more ctDNA clearances are observed at the study completion, the combination of atezolizumab and tiragolumab will be considered promising for ctDNA-positive stage II melanoma patients.

Randomization will only happen in the stage 1. In total, 36 patients (18 per arm) will be randomly assigned in a 1:1 ratio to the treatment group (atezolizumab and tiragolumab) or reference group (atezolizumab monotherapy). There is no randomization in stage 2. Stage 2 is single-arm (i.e., the treatment arm only), with a sample size of 25.

According to PI's personal communication with the Natera team, we assume the ctDNA positive rate will be 25%. In stage 1, it is expected that up to 144 patients will be screened in order to randomize 36 patients (18 to each arm). If we continue the study to stage 2 with the treatment arm only, an additional 100 patients will be screened (in order to enroll 25 patients). In total, we expect to accrue 244 patients for ctDNA testing with 61 patients going on to treatment.

14.6 Study Population

If the study stops after stage 1, we will screen 144 melanoma patients who had undergone complete resection. Among them, 18 ctDNA-positive patients being treated with the combination of atezolizumab and tiragolumab will be compared to a historical control and 18 ctDNA-positive patients on atezolizumab monotherapy will be analyzed as a descriptive comparison.

If we continue the study after stage 1, the study will screen 244 patients in total. Among them, 43 ctDNA-positive patients being treated with the combination of atezolizumab and

tiragolumab will be compared to a historical control and 18 ctDNA-positive patients on atezolizumab monotherapy will be analyzed as a descriptive comparison.

14.7 Statistical Analyses

Descriptive statistics will be used to summarize the trial results. Statistics for continuous variables may include means, medians, ranges and appropriate measures of variability. Qualitative variables will be summarized by counts and percentages. The uncertainty of estimates will be assessed by confidence intervals (CIs). All analyses will be considered as exploratory, even if statistical tests will be used.

ctDNA clearance and confirmed negative rate, as well as safety data will be summarized by the number and proportions. RFS, DMFS, and OS will be presented via the Kaplan-Meier method. Median survival and corresponding two-sided 95% CIs will be computed. Survival rates with 95% CIs will be estimated. The group survival difference (i.e., treatment vs. reference) will be examined by the log-rank test. Further exploratory correlative analyses will be performed via Fisher exact test, t-test, or nonparametric Wilcoxon–Mann–Whitney test, if needed.

14.7.1 Safety Early Stopping Rule

Early stopping of this trial is calculated based upon the report of study treatment (i.e., the combination of atezolizumab and tiragolumab, or atezolizumab monotherapy) related grade 3 or greater adverse events (AEs) (i.e., toxicities). A Bayesian safety-monitoring plan (Thall PF, Simon RM, Estey EH., 1996)[21] will be implemented once the first 12 evaluable patients are available. A Bayesian sequential safety monitoring will be defined as $Pr(\theta_T > \theta_0 \mid data) > 0.80$, where θ_T denotes the proportion of toxicity and θ_0 (θ_0=20%) represents the rate under null hypothesis (maximum probability of toxicity allowed). Enrollment will be stopped early whenever there is >80% chance that the event rate is larger than the null. The trial will be recommended to stop for excessive toxicities according to the toxicity stopping boundaries listed in the table below:

Stop for excessive toxicities (grade 3 or greater) if:	
# patients who experience toxicity is ≥:	In first # patients being determined ctDNA positive
4	12
7	24
10	36
12	48
15	61

The above stopping boundaries were obtained using Bayesian Toxicity Monitoring online application version 2.2.1.0 (available at http://ibl.mdanderson.org/BTM/) developed by M.D. Anderson Cancer Center. We assume that the parameter θ_0 is a constant and that θ_T follows a non-

informative prior of beta (0.5, 0.5). Accrual will be stopped and the event will be reviewed by the Data Safety and Monitoring Board if a study treatment-related death is observed.

15.0 MULTICENTER REGULATORY REQUIREMENTS

Washington University requires that each participating site sends its informed consent document to be reviewed and approved by the Washington University Regulatory Coordinator (or designee) prior to IRB/IEC submission.

Site activation is defined as when the secondary site has received official written documentation from the coordinating center that the site has been approved to begin enrollment. At a minimum, each participating institution must have the following documents on file at Washington University prior to study activation:

- Documentation of IRB approval of the study in the form of a letter or other official document from the participating institution's IRB. This documentation must show which version of the protocol was approved by the IRB.
- Documentation of IRB approval of an informed consent form. The consent must include a statement that data will be shared with Washington University, including the Quality Assurance and Safety Monitoring Committee (QASMC), the DSMC (if applicable), and the Washington University study team.
- Documentation of FWA, signed FDA Form 1572 (if applicable), and the CVs of all participating investigators.
- Protocol signature page signed and dated by the investigator at each participating site.

The coordinating center Principal Investigator (or designee) is responsible for disseminating to the participating sites all study updates, amendments, reportable adverse events, etc. Protocol/consent modifications and IB updates will be forwarded electronically to the secondary sites within 4 weeks of obtaining Washington University IRB approval. Activated secondary sites are expected to submit protocol/consent/IB modifications to their local IRBs within 4 weeks of receipt unless otherwise noted. Upon the secondary sites obtaining local IRB approval, documentation of such shall be sent to the Washington University study team within 2 weeks of receipt of approval.

Documentation of participating sites' IRB approval of annual continuing reviews, protocol amendments or revisions, all SAE reports, and all protocol violations/deviations/exceptions must be kept on file at Washington University.

The investigator or a designee from each institution must participate in a regular conference call to update and inform regarding the progress of the trial.

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APPENDIX A: ECOG Performance Status Scale

Grade	Description
0	Normal activity. Fully active, able to carry on all pre-disease performance without restriction.
1	Symptoms, but ambulatory. Restricted in physically strenuous activity, but ambulatory and able to carry out work of a light or sedentary nature (e.g., light housework, office work).
2	In bed <50% of the time. Ambulatory and capable of all self-care, but unable to carry out any work activities. Up and about more than 50% of waking hours.
3	In bed >50% of the time. Capable of only limited self-care, confined to bed or chair more than 50% of waking hours.
4	100% bedridden. Completely disabled. Cannot carry on any self-care. Totally confined to bed or chair.
5	Dead.

APPENDIX B: Definitions for Adverse Event Reporting

A. Adverse Events (AEs)

As defined in 21 CFR 312.32:

Definition: any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of an investigational medicinal product (IMP) or other protocol-imposed intervention, regardless of attribution.

This includes the following:

- AEs not previously observed in the subject that emerge during the protocol-specified
- AE reporting period, including signs or symptoms associated with
- {insert condition being studied} that were not present prior to the AE reporting period.
- Complications that occur as a result of protocol-mandated interventions (e.g., invasive procedures such as cardiac catheterizations)
- If applicable, AEs that occur prior to assignment of study treatment associated with medication washout, no treatment run-in, or other protocol-mandated intervention.
- Preexisting medical conditions (other than the condition being studied) judged by the investigator to have worsened in severity or frequency or changed in character during the protocol-specified AE reporting period.

Grading: the descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 5.0 will be utilized for all toxicity reporting. A copy of the CTCAE version 5.0 can be downloaded from the CTEP website. The American Society for Transplantation and Cellular Therapy (ASTCT) CRS Consensus Grading Scale should be used in addition to NCI CTCAE v5.0 when reporting the severity of CRS.

ASTCT CRS Consensus Grading

Grade	Symptoms
1	● Fever [a] with or without constitutional symptoms ● No hypotension ● No hypoxia
2	● Fever [a] combined with at least one of the following: – Hypotension not requiring vasopressors – Hypoxia requiring low-flow oxygen [b] by nasal cannula or blow-by
3	● Fever [a] combined with at least one of the following: – Hypotension requiring a vasopressor (with or without vasopressin) – Hypoxia requiring high-flow oxygen [b] by nasal cannula, face mask, non-rebreather mask, or Venturi mask
4	● Fever [a] combined with at least one of the following: ● Hypotension requiring multiple vasopressors (excluding vasopressin) ● Hypoxia requiring oxygen by positive pressure (e.g., CPAP, BiPAP, intubation and mechanical ventilation)
5	● Death due to CRS in which another cause is not the principal factor leading to this outcome

ASTCT = American Society for Transplantation and Cellular Therapy; BiPAP = bi-level positive airway pressure; CPAP = continuous positive airway pressure; CRS = cytokine-release syndrome.

[a] Fever is defined as temperature ε 38°C not attributable to any other cause. In patients who develop CRS and then receive anti-pyretic, anti-cytokine, or corticosteroid therapy, fever is no longer required when subsequently determining CRS severity (grade). In this case, the CRS grade is driven by the presence of hypotension and/or hypoxia.

[b] Low flow is defined as oxygen delivered at δ 6 L/min, and high flow is defined as oxygen delivered at > 6 L/min.

Attribution (relatedness), Expectedness, and Seriousness: the definitions for the terms listed that should be used are those provided by the Department of Health and Human Services' Office for Human Research Protections (OHRP). A copy of this guidance can be found on OHRP's website:
http://www.hhs.gov/ohrp/policy/advevntguid.html

B. Suspected Adverse Reaction (SAR)

As defined in 21 CFR 312.32:

Definition: any adverse event for which there is a reasonable possibility that the drug caused the adverse event. "Reasonable possibility" means there is evidence to suggest a causal relationship between the drug and the adverse event. "Suspected adverse reaction" implies a lesser degree of certainty about causality than adverse reaction, which means any adverse event caused by a drug.

C. Life-Threatening Adverse Event / Life Threatening Suspected Adverse Reaction 16.1.1

As defined in 21 CFR 312.32:

Definition: any adverse drug event or suspected adverse reaction is considered "life-threatening" if, in the view of the investigator, its occurrence places the patient at immediate risk of death. It does not include an adverse event or suspected adverse reaction that, had it occurred in a more severe form, might have caused death.

D. Serious Adverse Event (SAE) or Serious Suspected Adverse Reaction

As defined in 21 CFR 312.32:

Definition: an adverse event or suspected adverse reaction is considered "serious" if, in the view of the investigator, it results in any of the following outcomes:

- Death
- A life-threatening adverse event
- Inpatient hospitalization or prolongation of existing hospitalization
- A persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
- A congenital anomaly/birth defect
- Any other important medical event that does not fit the criteria above but, based upon appropriate medical judgment, may jeopardize the subject and may require medical or

surgical intervention to prevent one of the outcomes listed above

E. Protocol Exceptions

Definition: A planned change in the conduct of the research for one participant.

F. Deviation

Definition: Any alteration or modification to the IRB-approved research without prospective IRB approval. The term "research" encompasses all IRB-approved materials and documents including the detailed protocol, IRB application, consent form, recruitment materials, questionnaires/data collection forms, and any other information relating to the research study.

A minor or administrative deviation is one that does not have the potential to negatively impact the rights, safety, or welfare of participants or others or the scientific validity of the study.

A major deviation is one that does have the potential to negatively impact the rights, safety, or welfare of participants or others or the scientific validity of the study.

G. Product Complaints

Definition: Any written or oral information received from a complainant that alleges deficiencies related to identity, quality, safety, strength, purity, reliability, durability, effectiveness, or performance of a product after it has bene released and distributed to the commercial market or clinical trial.

APPENDIX C: Reporting Timelines

Expedited Reporting Timelines				
Event	**HRPO**	**QASMC**	**FDA**	**Genentech**
Serious AND unexpected suspected adverse reaction			Report no later than 15 calendar days after it is determined that the information qualifies for reporting	
Unexpected fatal or life-threatening suspected adverse reaction			Report no later than 7 calendar days after initial receipt of the information	
Serious adverse event				Report within 24 hours of learning of the event
Unanticipated problem involving risk to participants or others	Report within 10 working days. If the event results in the death of a participant enrolled at WU/BJH/SLCH, report within 1 working day.	Report via email after IRB acknowledgment		
Adverse event of special interest				Report within 24 hours of learning of the event
Abnormal liver function tests defined as Hy's law				Report within 24 hours of learning of the event
Accidental overdose or medication error (if fulfilling the seriousness criteria)				Report within 24 hours of learning of the event
Pregnancy of female patient or female partner of male patient, spontaneous abortion, therapeutic abortion, or congenital anomaly/birth defect				Report within 24 hours of learning of the event
Major deviation	Report within 10 working days. If the event results in the death of a participant enrolled at			

Expedited Reporting Timelines				
Event	**HRPO**	**QASMC**	**FDA**	**Genentech**
	WU/BJH/SLCH, report within 1 working day.			
A series of minor deviations that are being reported as a continuing noncompliance	Report within 10 working days.			
Protocol exception	Approval must be obtained prior to implementing the change			
Clinically important increase in the rate of a serious suspected adverse reaction of that list in the protocol or IB			Report no later than 15 calendar days after it is determined that the information qualifies for reporting	
Complaints	If the complaint reveals an unanticipated problem involving risks to participants or others OR noncompliance, report within 10 working days. If the event results in the death of a participant enrolled at WU/BJH/SLCH, report within 1 working day. Otherwise, report at the time of continuing review.			
Breach of confidentiality	Within 10 working days.			
Incarceration	If withdrawing the participant poses a safety issue, report within 10 working days. If withdrawing the participant does not represent a safety issue and the patient will be withdrawn, report at continuing review.			

Routine Reporting Timelines				
Event	**HRPO**	**QASMC**	**FDA**	**Genentech**
Adverse event or SAE that does not require expedited reporting	If they do not meet the definition of an unanticipated problem involving risks to participants or others, report summary information at the time of continuing review	Adverse events will be reported in the toxicity table in the DSM report which is typically due every 6 months.	The most current toxicity table from the DSM report is provided to the FDA with the IND's annual report.	
Minor deviation	Report summary information at the time of continuing review.			
Complaints	If the complaint reveals an unanticipated problem involving risks to participants or others OR noncompliance, report within 10 working days. If the event results in the death of a participant enrolled at WU/BJH/SLCH, report within 1 working day. Otherwise, report at the time of continuing review.			
Incarceration	If withdrawing the participant poses a safety issue, report within 10 working days. If withdrawing the participant does not represent a safety issue and the patient will be withdrawn, report at continuing review.			

Expedited Reporting Timelines for Secondary Sites				
Event	**WU (Coordinating Center)**	**Local IRB**	**FDA**	**Genentech**
Serious AND unexpected suspected adverse reaction	Report no later than 11 calendar days after it is determined that the information qualifies for reporting.	Report all applicable events to local IRB according to local institutional guidelines.	The research team at Washington University is responsible for reporting all applicable events to the FDA as needed.	The research team at Washington University is responsible for reporting all applicable events to Genentech as needed.
Unexpected fatal or life-threatening suspected adverse reaction	Report no later than 4 calendar days after initial receipt of the information.			
Unanticipated problem involving risk to participants or others	Report no later than 4 calendar days after initial receipt of the information.			

Expedited Reporting Timelines for Secondary Sites				
Event	**WU (Coordinating Center)**	**Local IRB**	**FDA**	**Genentech**
Adverse event or SAE that does not require expedited reporting	As per routine data entry expectations			
Protocol exception	Approval must be obtained prior to implementing the change.			

APPENDIX D: Washington University Unanticipated Problem Reporting Cover Sheet

SAE COVER SHEET- Secondary Site Assessment

Washington University HRPO#:	Sponsor-Investigator:
Subject Initials:	Subject ID:
Treating MD:	Treating Site:
EVENT TERM:	Admission Date:
EVENT GRADE:	Date of site's first notification:

Treating MD Event Assessment:

Is this event **possibly, probably, or definitely** related study treatment?

☐ yes ☐ no

If yes, please list which drug (if more than one)_____

Explain _____

_____

_____ _____ _____

Physician's Name **Physician's Signature** **Date**