A phase 1b/2a open label study to evaluate anti-tumor efficacy and safety of rhIL-7-hyFc (NT-I7) in combination with anti-PD-L1 (atezolizumab) in patients with anti-PD-1/PD-L1 naïve or relapsed/refractory high-risk skin cancers

NCT03901573

20 November 2023

Note: Signatures and audit trail have been redacted from this document.

Note: signatures and audit trail have been redacted from this document

Cytel

Statistical Analysis Plan Template

Sponsor:	NeoImmuneTech, Inc.		
Protocol:	NIT-106 (ION-02)		
Document Version No.:	1.0	**Document Date:**	20Nov2023

Protocol NIT-106 (ION-02) Amendment 2

Protocol Title: A phase 1b/2a, open label study to evaluate anti-tumor efficacy and safety of rhIL-7-hyFc (NT-I7) in combination with anti-PD-L1 (atezolizumab) in patients with anti-PD-1/PD-L1 naïve or relapsed/refractory high-risk skin cancers

Protocol Number: (Version Date)	NIT-106 (ION-02)_IL7+Atezo_AM 5_Ver 1.0 (19 Jan 2023))
Name of Test Drug:	Atezolizumab (MPDL3280A; NSC 783608) NT-I7 (rhIL-7-hyFc)
Phase:	Phase 1b/2a
Methodology:	Non-randomized, 2 arm, open label trial with initial NT-I7 dose escalation phase (phase 1b), followed by a dose expansion phase (phase 2a)
Sponsor:	NeoImmuneTech, Inc. 2400 Research Blvd, Suite 250 Rockville, MD 20850
Sponsor Representative, Principal Investigator:	[redacted]
Sponsor Representative, ION Clinical Trials Manager:	[redacted]

Sponsor:	NeoImmuneTech, Inc.		
Protocol:	NIT-106 (ION-02)		
Document Version No.:	1.0	**Document Date:**	20Nov2023

[redacted]

Document Date: 20Nov2023

Document Version: 1.0

Sponsor:	NeoImmuneTech, Inc.		
Protocol:	NIT-106 (ION-02)		
Document Version No.:	1.0	**Document Date:**	20Nov2023

SIGNATURE PAGE

Protocol Title:	A phase 1b/2a, open label study to evaluate anti-tumor efficacy and safety of rhIL-7-hyFc (NT-I7) in combination with anti-PD-L1 (atezolizumab) in patients with anti-PD-1/PD-L1 naïve or relapsed/refractory high-risk skin cancers
Sponsor:	NeoImmuneTech, Inc. 2400 Research Blvd, Suite 250 Rockville, MD 20850
Protocol Number:	NIT-106 (ION-02)
Document Date/Version:	20Nov2023/1.0

[redacted]

C t l, Inc.
675 Massachusetts Avenue
Cambridge, MA 02139

Date: Nov 21, 2023

Sponsor Approval

By signing this document, I acknowledge that I have read the document and approve of the planned statistical analyses described herein. I agree that the planned statistical analyses are appropriate for this study, are in accordance with the study objectives, and are consistent with the statistical methodology described in the protocol, clinical development plan, and all applicable regulatory guidance's and guidelines.

I have discussed any questions I have regarding the contents of this document with the biostatistical author.

I also understand that any subsequent changes to the planned statistical analyses, as described herein, may have a regulatory impact and/or result in timeline adjustments. All changes to the planned analyses will be described in the clinical study report (CSR).

Sponsor:	NeoImmuneTech, Inc.		
Protocol:	NIT-106 (ION-02)		
Document Version No.:	1.0	**Document Date:**	20Nov2023

[redacted]

NeoImmuneTech, Inc.
2400 Research Blvd, Suite 250
Rockville, MD 20850

Sponsor:	NeoImmuneTech, Inc.		
Protocol:	NIT-106 (ION-02)		
Document Version No.:	1.0	**Document Date:**	20Nov2023

TABLE OF CONTENTS

Sponsor:	NeoImmuneTech, Inc.		
Protocol:	NIT-106 (ION-02)		
Document Version No.:	1.0	**Document Date:**	20Nov2023

LIST OF IN-TEXT TABLES AND FIGURES

Table/Figure **Page**

Sponsor:	NeoImmuneTech, Inc.		
Protocol:	NIT-106 (ION-02)		
Document Version No.:	1.0	**Document Date:**	20Nov2023

ABBREVIATIONS

Abbreviation	Definition
ADA	Anti-drug antibody
AE	Adverse events
AESI	Adverse event of special interest
AUC_{0-24}	Area under the concentration versus time curve from time 0 to the end of the dosing interval 24 hours later, calculated using linear trapezoid rule
BOR	Best overall response
C1/D1	Cycle 1 Day 1
CBC	Complete blood count
C_{max}	Maximum plasma concentration
C_{min}	Trough plasma concentration, taken 24 hours after dose and prior to subsequent dose
CPI	Checkpoint inhibitors
CR	Complete response
cSCC	Cutaneous squamous cell carcinoma
CSR	Clinical study report
DCR	Disease control rate
DLT	Dose-limiting toxicity
DoSD	Duration of stable disease
ECG	Electrocardiogram
ECOG	Eastern Cooperative Oncology Group
FDA	Food and Drug Administration
ICF	Informed Consent Form
ICH	International Conference on Harmonisation
IDO	Indoleamine 2,3-dioxygenase
IFNγ	Interferon γ
IQR	Inter-quartile range
irRC	Immune-related response criteria
ITT	Intent-to-treat
MCC	Merkel cell carcinoma

Sponsor:	NeoImmuneTech, Inc.		
Protocol:	NIT-106 (ION-02)		
Document Version No.:	1.0	**Document Date:**	20Nov2023

Abbreviation	Definition
MED	Maximum effective dose
MedDRA	Medical Dictionary for Regulatory Activities
MTD	Maximum tolerated dose
NE	Not evaluable
NIT	NeoImmuneTech
NTL	Non-target lesion
OR	Objective response
ORR	Objective response rate
OS	Overall survival
PD (evaluating response)	Progressive disease
PD	Pharmacodynamics
PD-1	Programmed cell death protein 1
PD-L1	Programmed death-ligand 1
PFS	Progression Free Survival
PK	Pharmacokinetic
PPP	Per-Protocol Population
PR	Partial response
PT	Preferred term
RP2D	Recommended Phase 2 dose
SAP	Statistical Analysis Plan
SD	Stable disease
SOC	System organ class
T_{max}	Time to maximum plasma concentration
TGF-β	Transforming growth factor β
TME	Tumor microenvironment
WHO	World Health Organization

Sponsor:	NeoImmuneTech, Inc.		
Protocol:	NIT-106 (ION-02)		
Document Version No.:	1.0	**Document Date:**	20Nov2023

1. INTRODUCTION AND OBJECTIVES OF ANALYSIS

1.1. Introduction

Very few preclinical studies have combined interleukin 7 (IL-7) with programmed cell death protein 1 (PD-1), programmed death-ligand 1 (PD-L1), or CTLA-4 checkpoint blockade. One murine study showed that the therapeutic efficacy of concomitant blockade of CTLA-4 and PD-1 relies on interdependence of IL-7 and interferon-gamma (IFN-γ) signaling in T-cells, predicting that exogenous IL-7 will boost the therapeutic efficacy 1. Another study of murine sepsis showed that IL-7 reverses immune suppression by increasing lymphocyte proliferation, expression of lymphocyte adhesion molecules, IFN-γ production, and CD28 expression on splenic CD8+ T-cells. Combined treatment with IL-7 and anti-PD-1 produced additive effects on CD28 expression, lymphocyte proliferation, and splenic secretion of IFN-γ 2. A Science paper 3 using the mouse model of chronic lymphocytic choriomeningitis virus (LCMV) infection explored the combined effect of IL-7 and anti-PD-L1 on the development and functionality of exhausted T-cells. Anti-PD-L1 treatment moderately improved IL-7 signaling by increasing CD127 (IL-7Rα) expression on exhausted T-cells and exploited potential pathways to improve checkpoint blockade efficacy by combining with IL-7.

Anti-PD-1 and anti-PD-L1 including atezolizumab, can induce remarkable responses in a subset of patients with cancer including skin cancers, yet very few are cured. The mechanisms of immune escape, including (1) the lack of strong, tumor-specific antigens or epitopes recognized by T-cells, (2) impaired or suppressed antigen presentation machinery including downregulation of major histocompatibility complex on cancer cells, (3) impaired or suppressed cytotoxic T-cell activation, (4) poor infiltration of T-cells into the tumor microenvironment (TME), and (5) increased immunosuppressive cytokines and cells in the TME, prevent a large proportion of cancer patients from deriving clinical benefit from anti-PD-1/PD-L1 therapies 4. Thus, additional therapies to increase the frequency and depth of responses to anti-PD-1/anti-PD-L1 such as atezolizumab are needed.

The current trial will determine whether:

1. Administration of NT-I7 at a dose known to expand T-cell number and repertoire will: (a) increase the number of tumor-infiltrating T-cells; and (b) increase the efficacy of atezolizumab therapy.
2. Administration of NT-I7 with atezolizumab will result in clinically meaningful increases in objective response rate (ORR), disease control rate (DCR), duration of objective response (DOR), progression-free survival (PFS) and overall survival (OS).
3. Administration of NT-I7 with atezolizumab exerts certain effects on the TME, e.g., favors T cell and myeloid infiltration, or overexpression of indoleamine 2,3-dioxygenase (IDO), arginase and transforming growth factor β (TGF-β).

1.2. Objectives of Statistical Analysis

This study will evaluate the safety and anti-tumor activity of NT-I7 in combination with atezolizumab, including estimation of the Maximum Tolerated Dose (MTD) and/or the Recommended Phase 2 Dose (RP2D). The trial is designed to determine whether NT-I7, at doses known to increase the peripheral blood T-cell level, will also increase the level of tumor infiltrating T-cells and thereby increase the efficacy of atezolizumab.

1.2.1. Primary Study Objective

- To evaluate the safety and tolerability of NT-I7 in combination with atezolizumab (Phase 1b), including estimation of the MTD and/or the RP2D

1.2.2. Secondary Objectives

- To evaluate immunogenicity of NT-I7
- To make a preliminary assessment of the anti-tumor activity of NT-I7 in combination with atezolizumab

1.2.3. Exploratory Objectives

- To make a preliminary assessment of pharmacokinetic (PK) parameters of NT-I7 in combination with atezolizumab
- To make a preliminary assessment of biomarkers that might act as pharmacodynamic indicators of activity of NT-I7 in combination with atezolizumab
- To make a preliminary assessment of biomarkers that might act as predictors of anti-tumor activity of NT-I7 in combination with atezolizumab

This statistical analysis plan (SAP) describes the populations for analysis, data handling rules including handling of missing data and imputation, statistical methods, and table/listing/figure formats for data presentation ("TLF shells"). The statistical analyses and summary tabulations prepared according to this SAP will provide the basis for the results sections of the CSR for this trial.

This SAP also will outline any differences in the currently planned analyses relative to those planned in the study protocol.

2. STUDY DESIGN

2.1. Synopsis of Study Design

This is a Phase 1b/2a, open-label, multicenter study to evaluate the safety, tolerability and anti-tumor effect of NT-I7 (recombinant human interleukin 7 [rhIL-7]-hybrid Fc [hyFc]) in combination with atezolizumab (MPDL3280A) in patients with anti-PD-1/PD-L1 naïve or relapsed/refractory high-risk skin cancers including cSCC, MCC and melanoma.

This study has been designed to allow for an investigation of MTD or RP2D of NT-I7 in combination with atezolizumab. There are two phases to this study:

- Phase 1b, a NT-I7 dose escalation phase to determine the MTD or RP2D
- Phase 2a, a non-randomized parallel dose expansion phase to confirm the MTD or RP2D in both arms

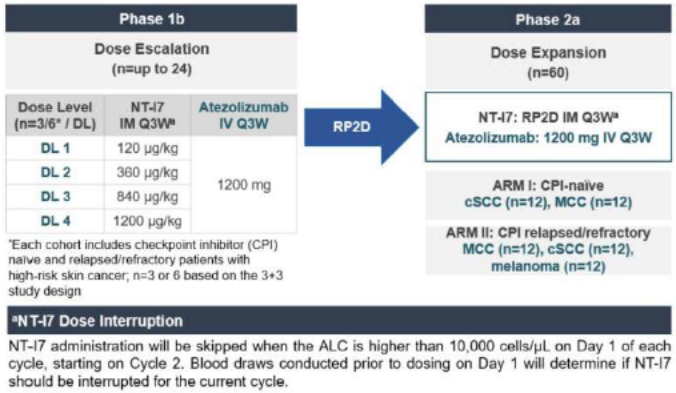
  Arm I: Anti-PD-1/PD-L1 (checkpoint inhibitors [CPI]) naïve patients with cSCC and MCC

  Arm II: Anti-PD-1/PD-L1 relapsed/refractory patients with MCC, cSCC and melanoma

Number of Patients

Approximately 84 patients will be enrolled; up to 24 patients will be enrolled in the Phase 1b (3 + 3 dose escalation design will be used), and approximately 60 patients will be enrolled in the Phase 2a (24 patients in Arm I, i.e., 12 patients for each indication, and 36 patients in Arm II, i.e., 12 patients for each indication).

Figure 1 Phase 1b\2a Schema



Each patient will participate in the trial ("enrolled") from the time the Informed Consent Form (ICF) is signed through final protocol-specified contact. The active study will end when the last patient completes the 90-day safety follow up visit, approximately 27 months after enrollment. Trial is expected to be completed approximately 36 months after the last patient is enrolled.

2.2. Randomization Methodology

Not applicable to the present study.

2.3. Stopping Rules and Unblinding

Not applicable to the present study.

Sponsor:	NeoImmuneTech, Inc.		
Protocol:	NIT-106 (ION-02)		
Document Version No.:	1.0	**Document Date:**	20Nov2023

2.4. Study Procedures

Baseline evaluations (screening visit) are to be conducted within 1 week before the initiation of study treatment. Scans and x-rays must be done ≤ 4 weeks (28 days) before the initiation of study treatment. In the event that the patient's condition is deteriorating, laboratory evaluations should be repeated within 48 hours before the initiation of the next cycle of therapy. Patients will be assessed for pulmonary signs and symptoms throughout the study.

Figure 2 Schedule of Assessments

ARM I AND ARM II															
Treatment Cycle/Visit	**Screening**	**Cycle 1**			**Cycle 2**		**Cycle 3**		**Cycle 4**	**Cycle 5**	**Subsequent Cycle** (Repeat up to 2 year)	**End of Tx**	**Post-Treatment Follow-up**		
												30 days ± 5 days post Tx	Safety FU	Disease Assessment[8]	Survival FU
Scheduling Window	Within 7 days	Day 1	Day 2	Day 8± 2 days	Day 1± 2 days	Day 8± 2 days	Day 1± 2 days	Day 8± 2 days	Day 1± 2 days	Day 1± 2 days	Day 1± 2 days		60 and 90 days ± 7 days post Tx	Every 12 wks ± 1 wk	Every 12 wks ± 1 wk
NT-I7*		A					A			A	A				
Atezolizumab		B			B		B		B	B	B				
Administrative procedures															
Informed consent	X														
Demographics	X														
Medical history	X														
Concurrent meds	X	X----------X										X	X	X	X

Sponsor:	NeoImmuneTech, Inc.		
Protocol:	NIT-106 (ION-02)		
Document Version No.:	1.0	**Document Date:**	20Nov2023

ARM I AND ARM II															
Treatment Cycle/Visit	**Screening**	**Cycle 1**			**Cycle 2**		**Cycle 3**		**Cycle 4**	**Cycle 5**	**Subsequent Cycle** (Repeat up to 2 year)	**End of Tx**	**Post-Treatment Follow-up**		
												30 days ± 5 days post Tx	Safety FU	Disease Assessment[g]	Survival FU
Scheduling Window	Within 7 days	Day 1	Day 2	Day 8± 2 days	Day 1± 2 days	Day 8± 2 days	Day 1± 2 days	Day 8± 2 days	Day 1± 2 days	Day 1± 2 days	Day 1± 2 days		60 and 90 days ± 7 days post Tx	Every 12 wks ± 1 wk	Every 12 wks ± 1 wk
Clinical Procedures/Assessments															
Physical exam	X				X		X		X	X	X	X	X		
Vital signs[a]	X	X		X	X		X	X	X	X	X	X	X		
Height	X														
Weight	X	X			X		X		X	X	X	X	X		
AEs evaluation		X----------									X	X	X	X	
Immune-related AEs evaluation		X----------									X	X	X	X	
Performance status	X	X		X	X		X	X	X	X	X	X	X		
EKG	X	X[l] (as indicated) ----------									X[l]				
Laboratory Assessments (Safety Labs)															
CBC w/diff, platelets	X	X		X	X		X	X	X	X	X	X	X		
Serum chemistry[b]	X	X		X	X		X	X	X	X	X	X	X		
Pregnancy Test (serum)	X[c]												X[c]		

Sponsor:	NeoImmuneTech, Inc.		
Protocol:	NIT-106 (ION-02)		
Document Version No.:	1.0	**Document Date:**	20Nov2023

ARM I AND ARM II															
Treatment Cycle/Visit	**Screening**	**Cycle 1**			**Cycle 2**		**Cycle 3**		**Cycle 4**	**Cycle 5**	**Subsequent Cycle** (Repeat up to 2 year)	**End of Tx**	**Post-Treatment Follow-up**		
												30 days ± 5 days post Tx	Safety FU	Disease Assessment[g]	Survival FU
Scheduling Window	Within 7 days	Day 1	Day 2	Day 8± 2 days	Day 1± 2 days	Day 8± 2 days	Day 1± 2 days	Day 8± 2 days	Day 1± 2 days	Day 1± 2 days	Day 1± 2 days		60 and 90 days ± 7 days post Tx	Every 12 wks ± 1 wk	Every 12 wks ± 1 wk
Hepatitis B testing	X														
Efficacy Measurements															
Tumor measurements	Within 28 days	Tumor measurements are performed at *9 weeks ± 1 week for the first two timepoints, then every 12 weeks ± 1 week*. Documentation (radiologic) must be provided for patients removed from study for progressive disease.										X[i]		X	
Radiologic evaluation	Within 28 days	Radiologic measurements should be performed at *9 weeks ± 1 week for the first two timepoints, then every 12 weeks ± 1 week*										X[i]		X	
Tumor Biopsies/Archival Tissue Collection															
Tumor Biopsy	X[d]					X[d]									
Correlative Studies Blood Draws[e]															
T-cell count (CD4+ and CD8+)		X[e]			X[e]		X[e]		X[e]	X[e]	X[e]				
Pharmacokinetics[h]		X	X**	X			X			X		X			
Immunophenotyping		X		X[k]	X		X	X	X	X		X			
Multiplex Cytokines		X			X		X		X	X		X			
TCR sequencing		X			X		X		X	X					

Sponsor:	NeoImmuneTech, Inc.		
Protocol:	NIT-106 (ION-02)		
Document Version No.:	1.0	**Document Date:**	20Nov2023

ARM I AND ARM II

Treatment Cycle/Visit	Screening	Cycle 1			Cycle 2		Cycle 3		Cycle 4	Cycle 5	Subsequent Cycle (Repeat up to 2 year)	End of Tx	Post-Treatment Follow-up		
												30 days ± 5 days post Tx	Safety FU	Disease Assessment[g]	Survival FU
Scheduling Window	Within 7 days	Day 1	Day 2	Day 8± 2 days	Day 1± 2 days	Day 8± 2 days	Day 1± 2 days	Day 8± 2 days	Day 1± 2 days	Day 1± 2 days	Day 1± 2 days		60 and 90 days ± 7 days post Tx	Every 12 wks ± 1 wk	Every 12 wks ± 1 wk
ELISPOT		X			X		X		X	X					
Kyn/Trp and Arginine		X			X		X		X	X		X			
Immunogenicity Testing[j] (NT-I7)		X			X		X		X	X	X	X	X[j]		

A NT-I7: Dose as assigned; starting with DL4 (1200 µg/kg), NT-I7 dosing is changed from Q3W to Q6W (i.e., Cycle 1, Day 1, Cycle 3, Day 1, Cycle 5, Day 1, etc.). NT-I7 will be skipped when the ALC is higher than 10,000 cells/µL on Day 1 of each cycle, starting on Cycle 3 (protocol Section 5.2).

* NT-I7 must be administered 60 (± 10) minutes before atezolizumab.

B Atezolizumab: Dose as assigned; once every 3 weeks, starting Cycle 1, Day 1.

a Vital signs assessment before and after study treatment is required (Section 6.1.1 and 6.1.2).

b Albumin, alkaline phosphatase, total bilirubin, bicarbonate, BUN, calcium, chloride, creatinine, glucose, LDH, phosphorus, potassium, total protein, SGOT [AST], SGPT [ALT], lipase, amylase, TSH and sodium.

c Pregnancy test (women of childbearing potential) must be performed within 72 hours prior to initiation of study treatment per eligibility criteria and at the 90-day Safety Follow up visit.

d Tumor biopsy/tissue collection (fresh) must be obtained within 7 days prior to Cycle 1, Day 1. If an archival tumor sample is available, the pre-treatment biopsy is not required. Post-treatment tumor biopsy must be obtained between Cycle 2, Day 8 and Cycle 2, Day 21 (Section 5.4)

e To be performed **prior** dosing (NT-I7 and/or atezolizumab)

g Disease assessment per standard of care

h To be collected on all patients (dose escalation and dose expansion); refer to the Pharmacokinetic timepoints tables for details (12.3.2); **Day 2 on Dose Escalation (Phase 1b) patients only, and is

Sponsor:	NeoImmuneTech, Inc.		
Protocol:	NIT-106 (ION-02)		
Document Version No.:	1.0	**Document Date:**	20Nov2023

ARM I AND ARM II

Treatment Cycle/Visit	Screening	Cycle 1			Cycle 2		Cycle 3		Cycle 4	Cycle 5	Subsequent Cycle (Repeat up to 2 year)	End of Tx	Post-Treatment Follow-up		
												30 days ± 5 days post Tx	Safety FU	Disease Assessment[g]	Survival FU
Scheduling Window	Within 7 days	Day 1	Day 2	Day 8± 2 days	Day 1± 2 days	Day 8± 2 days	Day 1± 2 days	Day 8± 2 days	Day 1± 2 days	Day 1± 2 days	Day 1± 2 days		60 and 90 days ± 7 days post Tx	Every 12 wks ± 1 wk	Every 12 wks ± 1 wk

optional.

i Only required if not performed within the past 9 or 12 ± 1 weeks – Section 6.6

j Refer to Sections 11.3.3 and 12.3.1 for all details. Testing to be repeated at 90-day Safety Follow up visit *only if* positive at End of Treatment visit.

k To be collected on all patients (dose escalation and dose expansion).

l EKG must be obtained at Screening. As clinically indicated, EKGs are to be obtained to evaluate for cardiac toxicity during subsequent cycles of therapy and follow-up visits.

2.5. Efficacy, Pharmacokinetic, and Safety Variables

2.5.1. Efficacy Variables

- Objective Response Rate (ORR) defined as the percentage of patients who have at least one confirmed partial response (PR) or complete response (CR) according to Response Evaluation Criteria in Solid Tumors (RECIST) v.1.1
- Disease Control Rate (DCR) defined as the percentage of patients with a response of CR, PR or stable disease (SD) according to RECIST v.1.1
- Progression Free Survival (PFS) defined as the time from the first study treatment to the first occurrence of progression, as measured by RECIST v1.1 or death from any cause, whichever occurs first
- Overall survival (OS) defined as the time from first study treatment to death from any cause.
- Duration of stable disease (DOSD) defined as the time from the first occurrence of stable disease or better (PR or CR or SD) to the time of the first documented disease progression or death from any cause, whichever occurs first per RECIST v1.1

2.5.2. Pharmacokinetics and Pharmacodynamics Variables

To evaluate the pharmacokinetics of NT-I7 administered in combination with atezolizumab:

- Serum concentration of NT-I7 will be used to estimate the following parameters: Area under the concentration time-curve (AUC), Maximum serum concentration (C_{max}), Minimum serum concentration (C_{min}), Clearance (CL) to be completed by Third Party Vendor.

To evaluate the immunogenicity of NT-I7, the following endpoint will be analyzed:

- Incidence of anti-drug antibody (ADA) to NT-I7 during the study relative to prevalence of ADA at baseline, defined as ADA titer relative to baseline to Day 1 of each cycle.

To evaluate the effect of the investigational treatment combination on the tumor microenvironment, based upon baseline and post-baseline tumor biopsy, the following endpoints will be assessed, but are outside of the scope of this plan:

- Number, distribution, and phenotype of tumor-infiltrating cells
- Expression of Interferon γ (IFNγ) and associated inflammatory gene expression in the tumor microenvironment

- Changes in tumor microenvironment that correlate with response or provide information on potential actionable causes for lack of clinical benefit

Potential correlation with PK, pharmacodynamics (PD), safety, and efficacy parameters can be explored but are outside of the scope of this plan.

2.5.3. Safety Variables

To confirm the safety and tolerability of the following safety variables:

- Adverse Events (AE) incidence, nature, and severity of AEs graded according to NCI CTCAE v5.0
- Clinical Laboratory Assessments (CBC and serum chemistry)
- Physical Examinations
- Vital signs
- Electrocardiograms (ECG)
- Eastern Cooperative Oncology Group (ECOG) Performance Status

3. PATIENT POPULATIONS

3.1. Population Definitions

The following patient populations will be evaluated and used for presentation and analysis of the data:

3.1.1. Safety Population

The Safety Population will include all patients who receive at least 1 dose of the investigational regimen. The Safety Population will be the primary set for the analysis of safety and non-efficacy data.

3.1.2. Efficacy Population

The efficacy analysis will be conducted on all patients who received at least one cycle of study treatment and have at least one evaluable post baseline tumor assessment.

The efficacy population will be used for all efficacy analysis.

3.1.3. Pharmacokinetic (PK) Population

The PK Population will include all patients who receive at least 1 dose of the investigational regimen and have at least 1 non-missing PK sample available for analysis.

The PK Population will be used for the PK analysis.

3.2. Protocol Violations

Violations of the protocol will be collected and reported. The sponsor, or designee, will be responsible for producing the final protocol violation file (formatted as a Microsoft Excel file), in collaboration with Cytel.

All protocol violations will be presented in the data listings.

4. STATISTICAL METHODS

4.1. Sample Size Justification

During the dose escalation phase (Phase 1b), a 3+3 design will be used for identifying the MTD and/or the RP2D. The following design will be used:

Number of Patients with Dose-Limiting Toxicity (DLT) at a Given Dose Level	Escalation Decision Rule
0 out of 3	Enroll 3 patients at the next dose level.
1 out of 3	Enroll at least 3 more patients at this dose level. - If 0 of these 3 additional patients experience a DLT (totaling 1/6), enroll at the next dose level. - If ≥1 of these 3 additional patients experiences DLT (totaling ≥2/6 at the dose level), the dose escalation is stopped, and the next lower dose level will be declared the MTD. In the case this happens at the 120 µg/kg dose level, a lower dose of 60 µg/kg will be tested, and the same 3+ 3 design followed.
≥2 out of 3	Dose escalation will be stopped. The next lower dose level will be declared the MTD. Three (3) additional patients will be entered at the next lower dose level if only 3 patients were treated previously at that dose. In the case this happens at the 120 µg/kg dose level, a lower dose of 60 µg/kg will be tested, and the same 3+3 design followed.

Based on the available data (Section 2.2 of the protocol), the average historical ORR for the patients in this study would be 45.25% in Arm I and less than 5% in Arm II. With a total of 24 patients treated at the RP2D in Phase 2a of the study in Arm I, the 95% confidence interval for an observed ORR of 17/24 or higher would exclude 45.25%. With a total of 36 patients treated at the RP2D in Phase 2a of the study in Arm II, the 95% confidence interval for an observed ORR of 6/36 or higher would exclude 5%.

Approximately 84 patients will be enrolled in this study; up to approximately 24 patients in the Phase 1b and approximately 60 patients in the Phase 2a of the study. The expected accrual is 4 patients per month total at approximately 8 ION sites.

4.2. General Statistical Methods and Data Handling

4.2.1. General Methods

All output will be incorporated into RTF or PDFs, sorted and labeled according to the International Conference on Harmonisation (ICH) recommendations, and formatted to the appropriate page size(s).

Phase 1b/dose escalation will be presented by dose levels 120 µg/kg, 360 µg/kg, 840 µg/kg, 1200 µg/kg and Total. Phase 2a/dose expansion will be summarized under the 1200 µg/kg dose. Efficacy analysis will analyze two groups of pooled data. The first group will pool 120 µg/kg, 360 µg/kg, and 840 µg/kg doses levels of Phase 1b; and the second group will pool the 1200 µg/kg dose level from both phases.

For categorical variables, summary tabulations of the number and percentage within each category (with a category for missing data) of the parameter will be presented.

Continuous variables will be summarized using mean, standard deviation, median, inter-quartile range (IQR) and range; 95% confidence intervals will be presented where appropriate.

4.2.2. Computing Environment

All descriptive statistical analyses will be performed using SAS statistical software (Version 9.4), unless otherwise noted. Medical history and adverse events will be coding using MedDRA version 'MedDRA v22.0 - Mar 2019'. Concomitant medications will be coded using World Health Organization (WHO) Drug version 'B3 WHO Drug Global – Mar 2020'.

4.2.3. Methods of Pooling Data

Not applicable to the present study.

4.2.4. Adjustments for Covariates

No formal statistical analysis that adjusts for possible covariate effects is planned.

4.2.5. Multiple Comparisons/Multiplicity

Multiplicity is not of concern for this study since no p-values will be reported.

4.2.6. Subpopulations

No analyses of subpopulations of patients are planned.

4.2.7. Withdrawals, Dropouts, Loss to Follow-up

Not applicable to this analysis.

4.2.8. Missing, Unused, and Spurious Data

In general, there will be no imputation of missing data points. All data recorded on the CRF will be included in data listings that will accompany the CSR.

4.2.9. Definition of Baseline

The baseline will be the last measurement obtained prior to the drug delivery on Cycle 1 Day 1 (C1/D1). When no measurements are available on C1/D1, data obtained at screening will be regarded as the baseline measurement.

4.2.10. Study Day

Study day is defined as (study date – C1/D1 date + 1) for post-baseline dates, (study date – C1/D1 date) otherwise. C1/D1 is the actual start date for the first dose of any study treatment.

4.2.11. Years and Months

For any calculations, Years will be 365.25 days and Months will be 30.4375 (or 365.25/12).

4.2.12. Imputation of Partial Dates

Partial Diagnosis dates will be imputed to the earliest Day of the month or year.

Stop date imputation will not be applied to ongoing AEs or concomitant medications.

Missing or partial dates for AEs and prior/concomitant medications will be imputed as follows:

- Start Date
  - If only the day is missing and the month and year are the same as the month and year of the first dose date of any treatment, then the day will be imputed with the day of the first dose date. Otherwise, the day will be imputed with the first day of the event month.
  - If both the day and month are missing and the year is the same as the year of the first dose date of any treatment, then they will be imputed with the month and day of the first dose date. Otherwise, they will be imputed with the date of January 1st of event year.
  - If the start date is completely missing, the date will be imputed with the first dose date of any treatment.
  - If the stop date is complete and the imputed start date is after the stop date, then the imputed start date will be set to the stop date.
- Stop Date

- If only the day is missing and the month and year are the same as the month and year of the study discontinuation date, then the day will be imputed with the day of the study discontinuation date. Otherwise, the day will be imputed with the last day of the event month.
- If both the day and month are missing and the year is the same as the year of the study discontinuation, then they will be imputed with the month and day of the study discontinuation date. Otherwise, they will be imputed with the date of December 31st.
- If the stop date is completely missing, then it will be imputed with the study discontinuation date.
- If the imputed stop date is greater than the last contact date, then the imputed stop date will be set to last contact date.

4.2.13. Dose-Limiting Toxicity

Dose-limiting toxicity is defined as any AE occurring within the first 21 days (i.e., C1/D1 through Day 21), that is considered to be at least possibly related to the combination study treatment (atezolizumab and NT-I7), and that meets at least one of the non-hematologic or hematologic criteria (Appendix A).

4.3. Interim Analyses

An interim analysis is not planned for this study.

4.4. Patient Disposition

Patient disposition will be tabulated, including the number of patients who: were screened, enrolled, received study treatment, discontinued treatment and the primary reason for discontinuation, withdrew from the study and the primary reasons for withdrawal, and completed 60- and 90-day follow-up visits.

A by-patient listing of study completion information, including the reason for study withdrawal, if applicable, will be presented.

4.5. Demographic and Baseline Characteristics

A summary of demographic characteristics will be provided by dose group, including summaries of age, sex, race, ethnicity, weight, diagnosis of disease, duration of diagnosis, metastatic site, and ECOG performance status. This summary will be based on the Safety Population by phase 1b and 2a and dose level.

A by-patient listing of demographic and baseline data will also be provided.

4.6. Medical History

Medical history will be included in a by-patient listing.

4.7. Study Treatment

Dose intensities will be summarized based on the Safety Population, with phases 1b and 2a presented separately by dose level/arm. Patients who have completed each cycle will be summarized.

The dose intensity of NT-I7 and atezolizumab is defined as:

> Total actual dose received /duration of treatment(months); where treatment duration is last dose date – first dose date + 1

Dose intensity will be summarized overall for each product.

Study treatment data will be included in a by-patient listing.

4.8. Efficacy Evaluation

All Efficacy analysis will be assessed using the Efficacy Population, unless specified otherwise.

Time to event data will be summarized using Kaplan-Meier Methodology using 25th, 50th (median), and 75th percentiles with associated 2-sided 95% confidence intervals, as well as percent of censored observations. The survival curves will be generated using the Kaplan-Meier method. The rate at 3, 6, 9, 12, 18 and 24 months will be calculated for PFS and OS.

4.8.1. Objective Response Rate and Disease Control Rate

ORR is defined as the percentage of patients who have at least one confirmed partial response (PR) or complete response (CR) according to RECIST v1.1 as determined by the investigator.

DCR is defined as percentage of patients with a response of CR, PR or Stable Disease (SD) according to RECIST v1.1.

The best overall response (BOR) is the best response recorded from the start of the treatment until disease progression/recurrence. Patients without baseline or post-baseline assessments will be considered Not Evaluable (NE).

The number and percentage of patients in each BOR category (CR, PR, SD, PD, NE), and ORR and DCR will be presented.

Listings of tumor assessments and responses assessment will be provided.

4.8.2. Progression Free Survival

PFS is defined as the time from C1/D1 to the first occurrence of progression or death from any cause, whichever occurs first, per RECIST v1.1 and irRC as determined by the investigator. PFS (in months) is calculated as (death or censor date – date of C1/D1 + 1) / 30.4375.

Rules for progression or censoring are described in Table 4-1. Patients who are not known to have progressed prior to analysis cut-off date will be censored at the date of last adequate tumor assessment or clinical cut-off, whichever comes first.

Table 1 PFS analyses and censoring rules.

Situation	Date of Progression or Censoring	Outcome
Incomplete or no baseline tumor assessment	C1/D1 date	Censored
Documented progression (per RECIST 1)	Earliest date on which radiological progression is documented	Progressed
Death	Date of death	Progressed
No progression	Date of last adequate tumor assessment with no documented progression	Censored
Treatment discontinuation	Date of Treatment discontinuation	Progressed

A summary of the number and percentage of patients experiencing a PFS event, and the type of event (progression or death) will be provided along with median PFS. A Kaplan-Meier plot of PFS will be presented by pool dose.

4.8.3. Overall survival

OS is defined as the time from C1/D1 to death from any cause, for patients who do not die before the end of the study or were lost to follow up, their OS will be censored on the last study follow-up date the patient is reported to be alive. OS (in months) is calculated as (death or censor date – date of C1/D1 + 1) / 30.4375.

Rules for selecting date of last contact are described in Table 4-2. Patients who are not known to have died prior to data cut-off will be censored at the date of last contact or clinical cut-off, whichever comes first.

Table 2 Rules for the last contact date for OS analysis

Source Data	Conditions
Date of C1/D1	No condition

Off study date	Only if patient status is reported to be alive
Start/end dates of concomitant medications	Non-missing verbatim term
Start/end dates from drug administration	Non-missing dose
Off treatment date	No condition
Tumor assessment (RECIST) date	Evaluation is marked as done.
Lab assessment date	Non-missing parameter value
Vital signs date	Non-missing parameter value
ECOG performance status date	Non-missing ECOG performance status
Start/end dates of adverse events	Non-missing verbatim term
Physical examination	At least 1 non-missing parameter value

A summary of the number and percentage of patients experiencing a death event will be provided along with median OS.

A Kaplan-Meier plot of OS will be presented by pool dose.

4.8.4. Duration of Stable Disease (DoSD)

The duration of stable disease is measured from the time measurement criteria are met for CR or PR or SD (whichever is first recorded) until the first date that recurrent or progressive disease is objectively documented or death, per RECIST v1.1 as determined by the investigator. Patients will be censored as PFS. DoSD (in months) is calculated as (event or censor date – first stable disease date + 1) / 30.4375.

DoSD analysis will be summarized based on Efficacy Population patients who had stable disease (CR or PR or SD). A summary of the number and percentage of patients experiencing an event, and the type of event (progression or death) will be provided along with median DoSD and Kaplan-Meier estimates.

A Kaplan-Meier plot of DoSD will be presented by pooled dose.

4.9. Pharmacokinetic Evaluations (NT-I7 serum concentrations)

Pharmacokinetic analyses will be conducted on participants in the PK Analysis Set.

Individual plasma concentration summary statistics will be computed using all available PK data. Individual concentration data will be listed for each participant and summarized by nominal sampling time point and visit as applicable with descriptive statistics (Mean, Std Dev, %CV, Min, Median, Max, and Geo Mean).

4.10. ADA/Immunogenicity Evaluations

Analysis of ADA data will be based on the Safety Analysis Set.

The ADA status will be defined using the baseline and postbaseline results. Anti-drug antibody negative is defined as negative results at all time points. Anti-drug antibody positive is defined as a positive result at any time point, including baseline.

The number and percentage of subjects with negative and positive ADA results will be tabulated by dose and phase.

All ADA results will be presented in a data listing, by date for unscheduled or repeated values.

NAb (neutralizing anti-drug antibody) data will be summarized and reported as number and percentage of subjects with NAb positive results per dose level by visit.

All NAb data will be listed by subject, visit and dose level.

4.11. Safety Analyses

Safety analyses will be conducted using the Safety Population.

4.11.1. Adverse Events

Adverse events will be coded using the Medical Dictionary for Regulatory Activities (MedDRA) and displayed in tables and listings using System Organ Class (SOC) and Preferred Term (PT). AE severity will be graded according to the CTCAE v5.0.

Analyses of adverse events will be performed for those events that are considered treatment emergent, where a treatment emergent adverse event (TEAE) is defined as any adverse event with an onset date on or after the administration of study treatment through the end of the study (defined as 30 days after the last dose of study treatment) or any event that was present at baseline but worsened in intensity or was subsequently considered drug-related by the investigator through the end of the study.

The number and percentage of patients with any TEAE will be reported, TEAEs with CTCAE Grade 3 or Higher, TEAEs assessed by the Investigator as related to treatment (definite, probable, or possible relationship), Treatment-related TEAE with Grade 3 or Higher, TEAEs leading to NT-I7/atezo Interrupted (also Dose Reduced and Withdrawn), Adverse Events of Special Interest (AESI; as designated by the CRF), Serious TEAEs, Serious Treatment-related TEAE, and TEAEs leading to Death; summarized by SOC and PT. In these tabulations, each patient will contribute only once in each category regardless of the number of events.

No formal hypothesis-testing analysis of adverse events incidence rates will be performed.

All adverse events occurring on study will be listed in by-patient listings.

By-patient listings also will be provided for the following: patient deaths and serious adverse events.

4.11.2. Laboratory Data

A listing of all laboratory data will be generated; normal ranges and CTCAE grades will be included. A subset listing will be presented for all notable abnormal laboratory values.

Complete Blood Count (CBC) parameters include: White Blood Cells (WBC), Red Blood Cells (RBC), Hemoglobin, Hematocrit, Platelets, Absolute Neutrophil Count (ANC), Absolute Lymphocyte Count (ALC), Absolute Eosinophil Count, Absolute Basophil Count, Absolute Monocyte Count, Neutrophils, Lymphocytes, Eosinophils, Basophils and Monocytes.

Serum Chemistry parameters include: Albumin, Alkaline Phosphatase, Bilirubin, Bicarbonate, Blood Urea Nitrogen (BUN), Calcium, Chloride, Creatinine, Glucose (Non Fasting), Lactate Dehydrogenase (LDH), Phosphorus, Potassium, Total Protein, Aspartate Aminotransferase (AST or SGOT), Alanine Aminotransferase (ALT or SGPT), Sodium, Lipase, Amylase, TSH.

4.11.3. Vital Signs and Physical Examinations

The value and change from baseline at each time point will be summarized for vital signs, including systolic and diastolic blood pressure, heart rate, respiratory rate, and temperature.

By-patient listings of vital sign measurements will be presented in data listings.

All physical examination findings will be presented in a data listing.

4.11.4. Electrocardiogram

All ECG data for each patient will be provided in data listings.

4.11.5. ECOG Performance Status

ECOG data will be listed.

4.11.6. Prior and Concomitant Medications

Prior medications will be defined as all medications taken before the administration of study treatment.

Concomitant medications will be defined as all medications taken on or after the administration of study treatment or within 30 days after the last administration date.

Concomitant medications will be coded using the WHO Drug dictionary. Results will be tabulated by Anatomic Therapeutic Class (ATC) and preferred term.

The use of concomitant medications will be included in by-patient data listing.

5. CHANGES TO PLANNED ANALYSES

Due to the premature closure of the study the following changes are made in comparison to the protocol:

- AEs leading to withdrawal and DLTs will not be listed separately.
- Laboratory values will only be listed.
- irRC (iRECIST) will not be summarized.
- Efficacy summaries will be performed on Efficacy Population only.
- All phases will be summarized within the same table.
- Duration of Stable Disease will replace Duration of Response analysis, due to limited patient responses.

6. REFERENCES

1. Shi LZ, Fu T, Guan B, et al. Interdependent IL-7 and IFN-gamma signalling in T-cell controls tumour eradication by combined alpha-CTLA-4+alpha-PD-1 therapy. Nat Commun. 2016;7:12335.
2. Shindo Y, Unsinger J, Burnham CA, Green JM, Hotchkiss RS. Interleukin-7 and anti-programmed cell death 1 antibody have differing effects to reverse sepsis-induced immunosuppression. Shock. 2015;43(4):334-343.
3. Pauken KE, Sammons MA, Odorizzi PM, et al. Epigenetic stability of exhausted T cells limits durability of reinvigoration by PD-1 blockade. Science. 2016;354(6316):1160-1165.
4. Kim JM, Chen DS. Immune escape to PD-L1/PD-1 blockade: seven steps to success (or failure). Annals of oncology : official journal of the European Society for Medical Oncology. 2016;27(8):1492-1504.
5. Nghiem PT, Bhatia S, Lipson EJ, et al. PD-1 Blockade with Pembrolizumab in Advanced Merkel-Cell Carcinoma. The New England journal of medicine. 2016;374(26):2542-2552.