novimmune

Clinical Study Protocol

A Phase 2/3 , Open-label, Single Arm, Multicentre Study to Assess Safety, Tolerability, Pharmacokinetics and Efficacy of Intravenous Multiple Administrations of NI-0501, an Anti-interferon Gamma (Anti-IFNγ) Monoclonal Antibody, in Paediatric Patients with Primary Haemophagocytic Lymphohistiocytosis (HLH)

Study number:	NI-0501-04
Protocol number:	NI-0501-04-EU-EudraCT#2012-003632-23
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[redacted]

INVESTIGATOR AGREEMENT

Protocol Number: NI-0501-04-EU-EudraCT#2012-003632-23

Protocol date and version: February 26, 2016 – Version 6.0

Study drug: NI-0501

Study title: A Phase 2/3, Open-label, Single Arm, Multicentre Study to Assess Safety, Tolerability, Pharmacokinetics and Efficacy of Intravenous Multiple Administrations of NI-0501, an Anti-interferon Gamma (Anti-IFNγ) Monoclonal Antibody, in Paediatric Patients with Primary Haemophagocytic Lymphohistiocytosis (HLH).

Investigator endorsement:

I, the undersigned, am responsible for the conduct of this study at this site and agree to conduct the study according to the protocol and any approved protocol amendments, ICH GCP and all applicable regulatory authority requirements.

I will not deviate from the protocol without prior permission from the Sponsor and prior review and written approval from the Institutional Review Board or Independent Ethics Committee, and where applicable, from the Competent Authorities, except where necessary to prevent any immediate danger to a patient.

I have read and understand fully the Investigator Brochure for NI-0501 and I am familiar with the investigational product and its use according to this protocol.

_____ _____

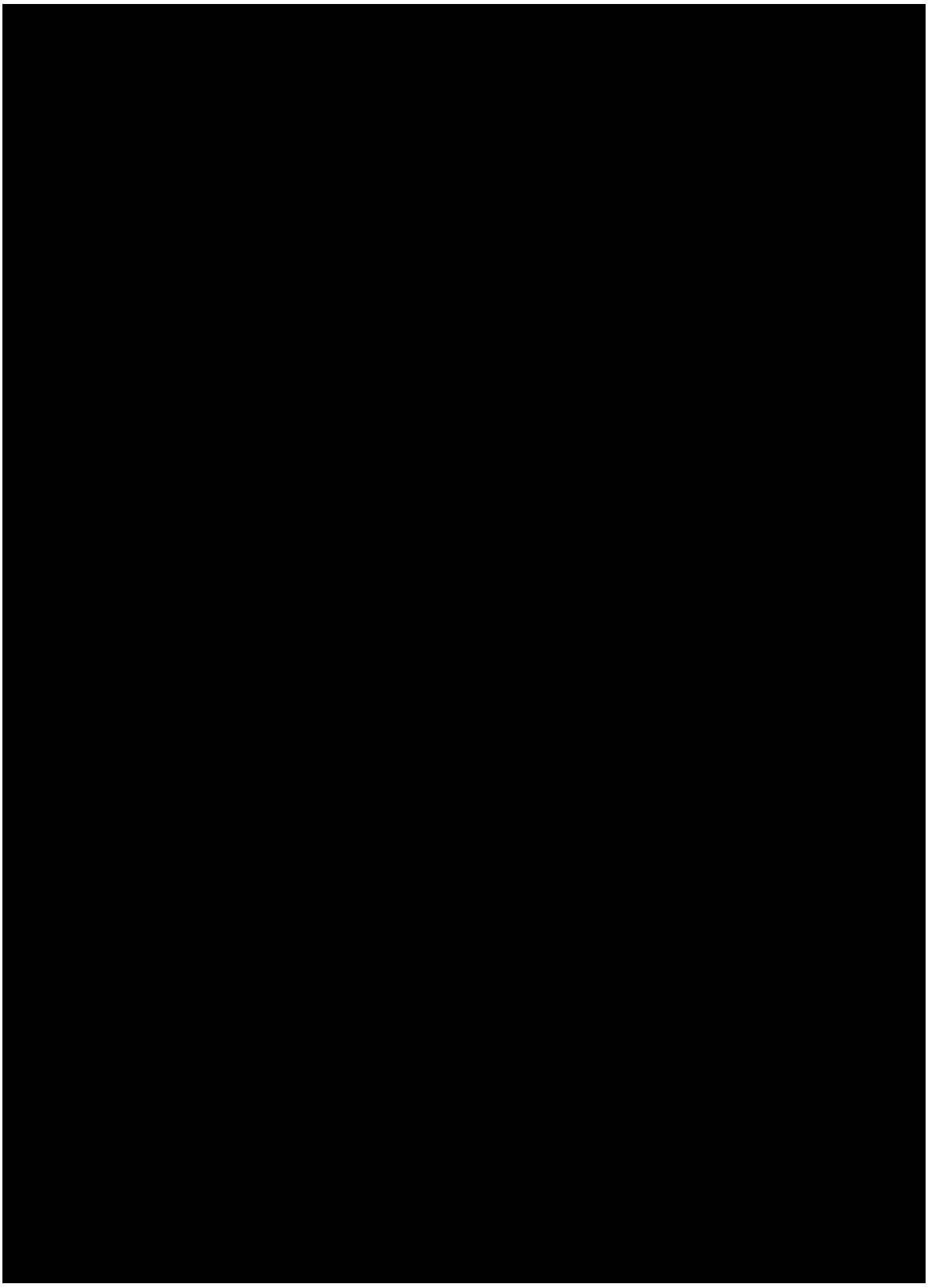
Site Investigator' Signature Date

_____

Site Investigator's Name

CONTACT LIST

Study Location: Multicentre



NI-0501-04 SYNOPSIS

Title:	A Phase 2/3, open-label, single arm, multicentre study to assess safety, tolerability, pharmacokinetics and efficacy of intravenous multiple administrations of NI-0501, an anti-interferon gamma (anti-IFNγ) monoclonal antibody, in paediatric patients with primary Haemophagocytic Lymphohistiocytosis (HLH)
Sponsor:	NovImmune SA, Switzerland
Study Type, Phase and Design:	• Interventional Phase 2/3 study • Open-label, single arm, international multicentre study • NI-0501-04 study is performed both in the US and in Europe according to twin protocols called NI-0501-04 (P-IND#111015) and NI-0501-04 (EudraCT#2012-003632-23), respectively
Study Objectives:	• To determine the safety and tolerability profile of multiple intravenous (IV) administrations of NI-0501. • To determine NI-0501 efficacy and benefit/risk profile in HLH patients. • To describe the PK profile of NI-0501 in HLH patients. • To define an appropriate NI-0501 therapeutic dose regimen for HLH. • To assess the immunogenicity of NI-0501.
Study Population:	• Primary HLH patients • Patients can be naïve to HLH treatment (first line patients), or may have already received conventional HLH therapy (second line patients) without having obtained a satisfactory response according to the treating physician or having shown signs of intolerance to it. • Patients who receive NI-0501 as second line treatment for HLH will represent the pivotal cohort of the study.
Main Inclusion Criteria:	• Gender: male and female. • Age: up to and including 18 years at diagnosis of HLH • Patient (if ≥ 18 years old), or patient's legally authorized representative(s) must have signed informed consent • Having accepted contraceptive measures whenever relevant
Exclusion Criteria:	• Diagnosis of secondary HLH consequent to a proven rheumatic or neoplastic disease. • Body weight < 3 kg. • Patients treated with: - any T-cell depleting agents (such as anti-thymocyte globulin [ATG], anti-CD52) during the previous 2 weeks prior to

	screening - any other biologic drug within 5 times their defined half-life period (except for rituximab in case of documented B-cell EBV infection). • Active mycobacteria, *Histoplasma Capsulatum, Shigella*, *Campylobacter, Leishmania* or *Salmonella* infections. • Evidence of past history of tuberculosis or latent tuberculosis. • Positive serology for HIV antibodies, hepatitis B surface antigen or hepatitis C antibodies. • Presence of malignancy. • Patients who have another concomitant disease or malformation severely affecting the cardiovascular, pulmonary, liver or renal function. • History of hypersensitivity or allergy to any component of the study regimen. • Receipt of a live or attenuated live (including BCG) vaccine within the previous 12 weeks from screening. • Pregnant or lactating female patients.
Study Drug:	• NI-0501 is a fully human IgG1 monoclonal antibody (mAb) directed against human IFNγ.
Dosing Regimen & Frequency of Administration:	• NI-0501 will be administered by IV infusion over a period of one hour at an initial dose of 1 mg/kg. • Infusions will be performed every 3 days until Study Day 15 (SD15) (infusion #6), and twice per week thereafter. • NI-0501 dose increase to 3 mg/kg will be possible according to pre-defined criteria guided by clinical and laboratory response in each patient (see Table 4, protocol section 5.2.2) at any time during the study. • After a minimum of two infusions at 3 mg/kg if, upon re-assessment, the same clinical and laboratory criteria qualifying the patient to receive 3 mg/kg of NI-0501 are found to still apply, the dose of NI-0501 may be increased to 6 mg/kg for up to four infusions, with a regular monitoring of the clinical and laboratory HLH parameters. • Based on the evolution of these parameters, the dose of NI-0501 may either *i*) be decreased back to 3 mg/kg, or *ii*) remain at 6 mg/kg for additional infusions (or be increased above 6 mg/kg), if PK and PD evidence indicates excessively high IFNγ production and, consequently, fast NI-0501 elimination (see Appendix B). • Dose increase may occur any time during the study, if the clinical and laboratory criteria in Table 4 are met.
Treatment Duration:	• NI-0501 administration is foreseen for 8 weeks. After this time period, pre-transplantation conditioning regimen can be initiated in preparation for Hematopoeitic Stem Cell Transplantation (HSCT). • The anticipated duration of treatment can be shortened, although not

	to less than 4 weeks, if the patient's condition and the donor availability allow earlier HSCT. • In the event that an appropriate donor has not been identified by Week 8 or in case of the need to delay the schedule of HSCT for reasons unrelated to the administration of NI-0501, NI-0501 treatment can be continued beyond this time, upon the request of the Investigator, in the context of an extension long-term follow-up study (NI-0501-05) to this proposed study, provided that a favourable benefit/risk has been established for the patient.
Background Therapy & Concomitant Medication:	• NI-0501 will be administered on a background of dexamethasone, which can be tapered depending on patient condition. • Patients will receive prophylactic treatment for *Pneumocystis jiroveci*, fungal and *Herpes Zoster* virus infection from the day before initiation of NI-0501 treatment until the end of the study. • Cyclosporine A can be continued if already being administered to the patient prior to screening. Cyclosporine can be withdrawn at any time, upon the judgement of the Investigator. Cyclosporine is not to be introduced *de novo* during the course of the study once NI-0501 administration has started. • If the patient is receiving intrathecal methotrexate and glucocorticoids at the time of NI-0501 treatment initiation, this treatment will be continued as required. • IV immunoglobulin (IVIG) use is allowed only as substitutive treatment in case of a documented immunoglobulin deficiency outside of screening and the 2 first weeks of NI-0501 administration. • Analgesic treatment, transfusion of blood products, electrolyte and glucose infusions, antibiotics, antifungal and anti-viral treatment and general supportive care (e.g. gastro-protective agents) are allowed. • Vaccination with a live or attenuated (including BCG) vaccine will be avoided during the whole study including the 4 weeks follow-up period. • Additional HLH treatments may be allowed in case of unsustained or limited HLH improvement once the maximum NI-0501 dose level is achieved. - Unsustained HLH Improvement: Patients who are unable to maintain at least 50% improvement from baseline for 3 HLH parameters (see Table 1). At least two consecutive measurements must document the loss of HLH improvement. - Limited HLH Improvement: Less than 50% change from baseline in a minimum of 3 HLH clinical and laboratory criteria. Etoposide should be administered as additional HLH treatment, unless clear evidence of lack of response or intolerance to the drug is derived from previous medical history. In this circumstance, the Investigator may propose an alternative agent which requires to be approved by the Data Monitoring Committee.

Sample Size:	• Sample size is estimated for the pivotal cohort of the study, i.e. patients receiving NI-0501 in second line. • A minimum of 28 evaluable second line patients will be enrolled in the study. Sample size calculation is based on the primary efficacy endpoint of "Overall Response Rate". Assuming an Overall Response Rate of 70%, the study will have 90% power to show a significant improvement above 40% using an exact binomial test at a one-sided significance level of 2.5%.
Number of Sites and Recruitment Duration:	• It is estimated that in EU approximately 14 sites will participate in this study. The time needed to complete enrolment of the required number of second line patients, in this rare population, is estimated to be approximately 1 year. • A twin protocol is actively recruiting in the US. The recruitment will be competitive across all US and European sites.
Study Duration and Study End Definition:	• After the treatment period, or, in any case, at treatment discontinuation, patients will enter a follow-up period of 4 weeks (short term follow-up). • End of the study is defined as last patient last visit. • After study end, a separate long term follow-up study (NI-0501-05) will allow: - long term safety surveillance, investigation of the impact of NI-0501 treatment on survival and post-HSCT outcome measures for all patients who have received at least one dose of NI-0501 - continuation of NI-0501 treatment for patients for whom an appropriate donor has not been identified by week 8 or in case of the need to delay HSCT for reasons unrelated to the administration of NI-0501.
Study Safety Monitoring and Stopping Rules:	• An independent Data Monitoring Committee (DMC) composed of relevant Experts (pediatric onco-hematologists, pediatric immune deficiency/infectious disease specialists, a bio-statistician and specialist in ethics) will oversee the study conduct, reviewing data generated both in the US and in Europe. • The main DMC responsibility is to review all safety and relevant efficacy data as they are generated on an on-going basis for the determination of the benefit/risk profile and to ensure that no patient is exposed to unnecessary risks. • The DMC can recommend treatment discontinuation for individual patients as well as to halt temporarily or permanently the entire study. Predefined stopping rules will guide the DMC review process. • Patients withdrawn from the study will receive rescue therapy, according to the standard of care at the site. • A patient's withdrawal can be decided at any time by the patient or his/her representative as well as by the Investigator. In any case this decision will have no impact on the patient's care.

Efficacy Endpoints:	Evolution of clinical signs (fever, splenomegaly, CNS symptoms) and laboratory parameters (CBC, fibrinogen, ferritin, sCD25 levels), which characterize the disease, will be used to assess the achievement of response and time to response. Primary efficacy endpoint: • Overall Response Rate, i.e. achievement of either Complete or Partial Response or HLH Improvement, at End of Treatment (EoT), as defined in Table 1. Secondary efficacy endpoints: • Time to Response any time during the study • Durability of Response, i.e. maintenance of response achieved any time during the study until EoT and beyond (including data collected in the long-term follow-up study NI-0501-05). • Number of patients able to reduce glucocorticoids by 50% or more of baseline dose. • Number of patients able to proceed to HSCT, when deemed indicated. • Survival at Week 8 (or EoT) and at the end of the study [Long-term survival (in particular D+30 and D+100 post-HSCT survival) will be assessed in the context of long-term study NI-0501-05]. • Serum concentration of NI-0501 to determine NI-0501 pharmacokinetic (PK) profile. • Determination of pharmacodynamic (PD) effects (levels of circulating total IFNγ and markers of its neutralization, namely CXCL9 and CXCL10). • Determination of other biomarkers, e.g. sCD25, IL-10.
Safety Endpoints:	• Safety parameters to be collected and assessed: - Incidence, severity, causality and outcomes of Adverse Events (serious and non-serious), with particular attention being paid to infections - Evolution of laboratory parameters such as complete blood cell count (CBC), with a focus on red cells (haemoglobin), neutrophils and platelets, liver tests, renal function tests and coagulation - Number of patients withdrawn for safety issues • Other parameters: - Level (if any) of circulating antibodies against NI-0501 to determine immunogenicity (ADA)
Statistical Analysis:	• The primary endpoint Overall Response Rate will be evaluated using the exact binomial test at the one-sided 0.025 level. • Time to Response, durability of Response and Survival time will be presented using Kaplan-Meier curves with medians calculated if available. 95% confidence intervals will be calculated for the median for each of these endpoints.

- Additional endpoints based on binary outcomes including number of patients who reduce glucocorticoids by 50% or more, and number of patients able to proceed to HSCT will be converted to proportions and associated 95% confidence intervals calculated.
- Statistical significance in terms of p-values will only be obtained for the primary endpoint. All other endpoints will be viewed as supportive for the primary endpoint and as a consequence no formal hierarchy of endpoints will be declared.

CONTENTS

LIST OF ABBREVIATIONS

Abbreviation	Term
ADA	Anti-drug-antibodies
ADCC	Antibody-dependent cell-mediated cytotoxicity
AE	Adverse event
ALP	Alkaline Phosphatase
ALT	Alanine aminotransferase
aPTT	Activated Prothrombin Time
AST	Aspartate aminotransferase
ATG	Anti-thymocyte globulin
AUC	Area Under the Curve
BCG	Bacillus Calmette-Guérin
BSA	Body Surface Area
CBC	Complete blood cell count
CDC	Complement Dependent Cytotoxicity
CL	Systemic drug clearance
C_{max}	Peak drug plasma concentration
CMV	Cytomegalovirus
CNS	Central nervous system
CRF	Case report form
CRP	C-reactive protein
CsA	Cyclosporin A
CSF	Cerebrospinal fluid
C_{trough}	Plasma drug concentration immediately prior next dosing
DMC	Data Monitoring Committee
eCRF	Electronic case report form
EBV	Epstein-Barr virus
EoT	End of treatment
γGT	Gamma Glutamyl Transferase
GM-CSF	Granulocyte-macrophage colony-stimulating factor
HBV	Hepatitis B virus
HCV	Hepatitis C virus
HIV	Human immunodeficiency virus
HLH	Haemophagocytic lymphohistiocytosis
HSCT	Haematopoietic stem cell transplantation
HSV	Herpes simplex virus
HZ	Herpes Zoster

HZV	Herpes zoster virus
ICMJE	International Committee of Medical Journal Editors
IFNγ	Interferon gamma
IFNγ-R1	Interferon gamma receptor chain 1
IFPMA	International Federation of Pharmaceutical Manufacturers & Associations
IgG1	Immunoglobulin G1
IL	Interleukin
IMP	Investigational medicinal product
IT	Intrathecal
ITT	Intention-to-treat
IVIG	IV immunoglobulin
KD	Dissociation constant
KO	Knock Out
LCMV	Lymphocytic choriomeningitis virus
LDH	Lactate dehydrogenase
mAb	Monoclonal antibody
MCSF	Macrophage colony stimulating factor
MRI	Magnetic resonance imaging
NK	Natural killer
NaCl	Sodium Chloride
PCR	Polymerase chain reaction
PD	Pharmacodynamic
PPD	Purified protein derivative
PK	Pharmacokinetic
PR	Partial Response
PT	Prothrombin Time
PVC	Polyvinyl chloride
SAE	Serious adverse event
SAP	Statistical analysis plan
SAD	Single ascending dose
sCD25	soluble CD25 (i.e. soluble IL-2 receptor)
SD(n)	Study Day number (e.g. Study day 1 = SD1)
SSC	Scientific steering committee
SUSAR	Suspected Unexpected Serious Adverse Reaction
TB	Tuberculosis
$t_{1/2}$	Elimination half-life
Tmax	Time when plasma concentration is at peak
TMDD	Target mediated drug disposition

TMF	Trial Master File
TNFα	Tumour necrosis factor alpha
US	Ultrasonography
Vss	Volume of distribution at steady state
WD	Withdrawal

Table 1: Definition of response

Overall Response Rate	
Complete Response	Complete Response is adjudicated if: - No fever = body temperature < 37.5°C - Normal spleen size as measured by 3D abdominal ultrasound - No cytopenia = Absolute Neutrophil Counts ≥ $1.0x10^9$/L and platelet count ≥ $100x10^9$/L [absence of G-CSF and transfusion support must be documented for at least 4 days to report no cytopenia] - No hyperferritinemia = serum level is < 2000 µg/L - No evidence of coagulopathy, i.e. normal D-Dimer and/or normal (> 150 mg/dL) fibrinogen levels - No neurological and CSF abnormalities attributed to HLH - No sustained worsening of sCD25 (as indicated by at least two consecutive measurements that are > 2-fold higher than baseline)
Partial Response	Partial Response is adjudicated if: - At least 3 of the HLH clinical and laboratory abnormalities (including CNS abnormalities) meet the above mentioned criteria for "Complete Response". In the case of "reactivated patients" who enter the study with 3 abnormal HLH features, at least 2 criteria should meet the definition given - There is no progression of other aspects of HLH disease pathology (e.g., jaundice, liver size, oedema, CNS clinical alterations)
HLH improvement	- Improvement (>50% change from baseline) of at least 3 HLH clinical and laboratory abnormalities (including CNS involvement). In the case of "reactivated patients" who enter the study with 2 abnormal HLH features, a change from baseline greater than 50% for both will define HLH as improved.
Limited Improvement/Lack of Improvement/No Response	
- Less than 50% change from baseline of 3 or more of the above mentioned HLH clinical and laboratory abnormalities [in the case of "reactivated patients" who enter the study with 2 abnormal HLH features, less than 50% change from baseline in both will be sufficient to define limited improvement] and - No apparent improvement in other aspects of disease pathology	
Reactivation	
- Deterioration of two or more HLH d clinical and laboratory criteria with the following specifications: 1. numerical laboratory values* must become abnormal and worsen by more than 30% compared to the previous evaluation, on two sequential assessments performed with an interval of minimum 1 day and maximum 1 week 2. deterioration of clinical criteria must be confirmed by consistent observations of worsening over three consecutive days - The development of new or recurrent CNS symptoms counts as a single criterion for reactivation. * *The following laboratory parameters are specifically considered for determination of reactivation:* *- platelets* *- neutrophils* *- fibrinogen* *- ferritin* *- soluble CD25 (sCD25; i.e. soluble IL-2 receptor).* *The assessment of NK function, red blood cells/hemoglobin and triglyceride levels cannot be considered for the determination of reactivation*.	

Table 2: Schedule of Assessments – Screening & Treatment Period 1 –SD0 to SD15 (Weeks 1 and 2)

Assessments		Screening		Treatment Period 1 - SD0 to SD15 (wk 1 and 2)											
		Up to one week prior to first infusion	SD-1	Inf. 1			Inf. 2		Inf. 3		Inf. 4		Inf. 5		Inf. 6
				SD0	SD1	SD2	SD3	SD5	SD6	SD8	SD9	SD11	SD12	SD14	SD15
Hospitalisation			Starting from SD-1												After this time-point patients may be discharged
Dexamethasone			Starting from SD-1												
Prophilactic treatment, as described in Section 6.2			Starting from SD-1												
Infusion				X			X		X		X		X		X
Patient Information		X													
Clinical Assessment	Vital signs [1]		X	X (Pre, during, post)	X	X	X (Pre, during, post)	X	X (Pre, during, post)	X	X (Pre, during, post)	X	X (Pre, during, post)	X	X (Pre, during, post)
	Continuous cardiac monitoring / pulse oxymetry			X (Pre, during, post)			X (Pre, during, post)		X (Pre, during, post)		X (Pre, during, post)		X (Pre, during, post)		X (Pre, during, post)
	Physical Examination [2]	X	X	X (Pre)	X	X	X (Pre)	X	X (Pre)	X	X (Pre)	X	X (Pre)	X	X (Pre)
Procedure	ECG	X		X (Post)	only if clinically indicated										
Search for Infections	TB [3]	X						X					X (Pre)		
	Adenoviruses, EBV, CMV (viral load)	X						X					X (Pre)		
	HSV, HZV, HIV, HBV, HCV	X		In case of suspicion of infection											
	Atypical mycobacteria, *Histoplasma Capsulatum* , *Shigella* , *Salmonella Campylobacter* , *Leishmania*	X		In case of suspicion of infection											
Laboratory	CBC	X		X (Pre)	X (morning)	X (morning)	X (Pre)	X	X (Pre)	X		X		X	
	Lymphocyte subsets	X					X (Pre)		X (Pre)						
	Coagulation (aPTT, PT, D.-dimers), fibrinogen	X		X (Pre)	X	X	X (Pre)	X	X (Pre)	X		X		X	
	Biochemistry[4], triglycerides	X		X (Pre)	X	X	X (Pre)	X	X (Pre)	X		X		X	
	IgG level	X													
	Pregnancy test (if applicable)	X													
	Urinalysis[5]	X		X (Pre)[5]	X	X	X (Pre)	X	X (Pre)	X		X		X	
Imaging	3D abdominal US (spleen and liver size)	X													X
	Chest X-ray[6]	X													
	Brain MRI	In case of CNS symptoms occurrence													
Histopathology	Cerebrospinal Fluid (CSF) analysis if coagulation allows	X		Only if clinically indicated (to monitor evolution or to confirm occurrence of new CNS symptoms)											
PD/Exploratory (sCD25, IL-10. CXCL9, CXCL10, CXCL11), IFNγ (free IFNγ at SD0 predose, total IFNγ for all other timepoints)				X (Pre)	X	X	X (Pre)	X	X (Pre)	X	X (Pre)		X (Pre)		X (Pre)
PK (NI-0501 circulating concentration)				X (Pre - post infusion)	X	X	X (Pre - post infusion)	X	X (Pre - post infusion)	X	X (Pre - post infusion)		X (Pre - post infusion)		X (Pre - post infusion)
Immunogenicity (ADA)		X													

1: Vital signs: Temperature, heart rate, blood pressure, respiratory rate. Oxygen saturation is also recorded at SD-1 and pre-, during and after infusion on infusion days

2: Physical examination: : includes as a minimum: weight (at screening, at SD-1, and prior to each infusion), height (at screening only), and in particular at each visit, occurrence of skin rashes, jaundice, purpura, bleeding, edema, ascites, search for tonsillitis, lymphadenopathies, dyspnea, cough, spleen and liver size, and neurological examination

3: TB: search for tuberculosis mycobacteria: At screening: IGRA/PPD and PCR; after screening by PCR

4: Biochemistry= glucose, electrolytes, ferritin, CRP, AST, ALT, ALP, gGT, LDH, bilirubin, albumin, creatinin, urea

5: Urinalysis= glucose, blood, protoein, leukocytes, ketone, pH, gravity. On SD0 urinalysis needs to be performed if not done at screening

6: Chest X-ray: every 4 weeks, except if required more frequently in case of clinical suspicion of a pulmonary infection

Table 3: Schedule of Assessments - Treatment Period 2 – SD 16 to EoT(3 days after last infusion) (Weeks 3 to 8) & Follow-up Period

Assessments		Treatment Period 2 - Week 3-8 SD16 until EoT (3 days after last NI-0501 infusion)			Follow-Up Period[7]			Wk 4/ Study completion visit or Withdrawal (WD) visit	Unscheduled Visit (UV)[9]
		Infusion visit	Efficacy/Safety visit[6]	End of treatment visit					
		Infusion X	Efficacy/Safety visit X	3 days post last infusion (± 1 day)	Week 2	Week 3	Pre-conditioning visit[8]		
	Infusion	X							
Clinical Assessment	Vital signs [1]	X (Pre, during, post)	X	X	X	X	X	X	X
	Continuous cardiac monitoring/ pulse oxymetry	X (Pre, during, post)							
	Physical Examination [2]	X (Pre)	X	X	X	X	X	X	X
Procedure	ECG	only if clinically indicated		X	only if clinically indicated			X	
Infections	TB [3]		X (every 2 weeks)	X	X			X	
	Adenoviruses, EBV, CMV (viral load)		X (every 2 weeks)		X			X	
	Atypical mycobacteria, *Histoplasma Capsulatum* , *Shigella, Salmonella Campylobacter, Leishmania*	In case of suspicion of infection							
Laboratory	CBC		X	X	X	X	X	X	
	Coagulation, fibrinogen		X	X	X	X	X	X	
	Biochemistry[4], triglycerides		X	X	X	X	X	X	
	Urinalysis (glucose, blood, protein, leukocytes, ketone, pH, gravity)		X	X	X	X	X	X	
Imaging	3D abdominal ultrasound (spleen and liver size)		X (every 2 weeks)	X			X		
	Chest X-ray[5]		X (every 4 weeks)	X				X	
	Brain MRI	In case of CNS symptoms							
Histopathology	CSF analysis	only if clinically indicated							
PD/Exploratory[10]		X (pre)		X	X	X	X	X	
PK (NI-0501 circulating concentration)[10]		X (pre and post)		X	X	X	X	X	
Immunogenicity (ADA)				X				X	

1: Vital signs: Temperature, heart rate, blood pressure, respiratory rate. Oxygen saturation is also recorded pre-, during and after infusion on infusion days

2: Physical examination: : includes: as a minimum weight prior to each infusion, at each follow-up visit and each unscheduled visit; weight and height at the end of treatment visit and at Week 4/Study completion visit or Withdrawal (WD) visit and in particular at each visit, occurrence of skin rashes, jaundice, purpura, bleeding, edema, ascites, search for tonsillitis, lymphadenopathies, dyspnea, cough, spleen and liver size, and neurological examination

3: TB: search for tuberculosis mycobacteria by PCR

4: Biochemistry= glucose, electrolytes, ferritin, CRP, AST, ALT, ALP, gGT, LDH, bilirubin, albumin, creatinin, urea

5: Chest X-ray: every 4 weeks, except if required more frequently in case of clinical suspicion of a pulmonary infection. At EoT and follow-up visits, chest X-ray will not be performed unnecessarily if a recent exam is available

6: Efficacy/Safety visits: should occur every 6 days, with a time-window of ± 48 hours in order to combine, whenever possible, with NI-0501 infusion visits.

7: Pre-HSCT visit: if applicable, i.e. if transplant takes place during the 4-week follow-up period, appropriate schedule will be applied to combine a weekly follow-up visit with the pre-HSCT visit at the site.

8: Pre-conditioning visit: if applicable, i.e. if the patient starts conditioning during the 4-week follow-up period, the closer weekly follow-up visit will be combined, in order to allow collection of clinical and laboratory HLH parameters before administration of the conditioning drugs.

9: Unscheduled Visit: These assessments should be performed at minimum, but additional assessments may be added according to the clinical judgment of the Investigator.

10: PK/PD: Additional PK/PD samples may be required to better characterize the PK/PD profile and/or for further safety assessments. Number of additional samples taken will be based on body weight, patient characteristics and clinical status of the patient.

PART I

1 BACKGROUND INFORMATION

1.1 NI-0501

1.1.1 Description and mode of action

NI-0501 is a fully human IgG1anti-interferon gamma (IFNγ) monoclonal antibody (mAb) which binds and neutralizes IFNγ. IFNγ is one of the most potent and pleiotropic cytokines of the immune system. It is critical for innate and adaptive immunity against viral and intracellular bacterial infections.

After binding to its receptor, IFNγ acts to produce a variety of physiological and cellular responses. Numerous studies over the last 20 years have associated IFNγ with the pathogenesis and the maintenance of inflammatory diseases[1-3].

IFNγ is produced predominantly by natural killer (NK) and natural killer T (NKT) cells, as part of the innate immune response, and by CD4 Th1 and CD8 cytotoxic T lymphocyte (CTL) effector T cells, once antigen-specific immunity develops.

1.1.2 Preclinical Data

1.1.2.1 Non-clinical Pharmacology

NI-0501 has shown similar binding affinity and blocking activity for IFNγ from non-human species, including *Rhesus* and *Cynomolgus* monkeys, but not from dogs, cats, pigs, rabbits, rats or mice.

NI-0501 binds to soluble and receptor (IFNγR1)-bound forms of IFNγ.

Since NI-0501 is a human IgG1, it retains the characteristics of this immunoglobulin isotype, including the capacity to engage Fcγ receptors and bind complement.

Due to NI-0501 capacity to bind free and IFNγR1-bound IFNγ, studies were performed to investigate the potential of NI-0501 to mediate ADCC and CDC activities, in the presence of target. A lack of ADCC activity was demonstrated and no induction of CDC activity was observed.

1.1.2.2 Toxicology

Binding and functional data demonstrated *Rhesus* or *Cynomolgus* monkeys to be relevant species to evaluate the safety of NI-0501. No off-target toxicity was attributed to the drug when administered to *Cynomolgus* monkeys in 13 weekly doses of up to 200 mg/kg. An enhanced susceptibility to infections due to an exaggerated pharmacological effect of the drug has been observed at all dose levels in animals originally harbouring gastrointestinal pathogens (*Shigella, Salmonella, Campylobacter*) prior to NI-0501 administration. In a second study, where *Cynomolgus* monkeys were not initially found to be harbouring gastrointestinal pathogens, weekly administrations of NI-0501 for 8 consecutive weeks at doses up to 30 mg/kg were well tolerated, without the need for antibiotic prophylaxis.

Results from a human tissue cross-reactivity study, involving a panel of 35 different human tissues, demonstrated that NI-0501 did not cross-react with any of the human samples tested.

1.1.2.3 Safety pharmacology

There were no abnormal findings in ECGs taken periodically during treatment and recovery periods in the 8 week and 13 week repeated dose toxicology studies in *Cynomologus* monkeys, where animals were exposed to doses up to 200 mg/kg of NI-0501 weekly. No abnormal findings were observed in the

histopathological investigations of the hearts and lungs in these animals compared to untreated animals. Histopathological analysis of kidneys from these animals revealed no abnormal findings and the periodic urinalysis readings were also normal, indicating no abnormal effects on renal function. There were no histopathological findings in brains in both studies. Furthermore, no abnormal behaviour of the animals was observed throughout the study periods, suggesting no effects on CNS.

1.1.3 Clinical Data

A Phase 1 randomized double-blinded placebo-controlled single ascending dose study in 20 healthy adult volunteers investigating the safety, tolerability and pharmacokinetic profiles of single intravenous (IV) administrations of NI-0501 took place between September 2011 and April 2012. During this study 6 subjects received placebo, while 3, 3, 4, and 4 subjects (in total 14 subjects) received NI-0501 doses of 0.01, 0.1, 1, and 3 mg/kg, respectively.

The pharmacokinetics (PK) analysis of NI-0501 revealed the expected profile for an IgG1 with a long half-life (around 22 days), a slow clearance (≤ 0.1 mL/kg/hr) and a low volume of distribution (< 65 mL/kg on average). The serum concentrations and the AUC measured at 1 and 3 mg/kg were in line with the prediction that NI-0501 induces at these doses a significant neutralization of IFNγ.

A total of 41 adverse events (AEs) were observed after start of drug infusion in 14 out of 20 subjects (70%), 10 of which were reported by 4 subjects having received placebo. Thirty-six (87.8%) AEs were of mild intensity and 5 (12.2%) were of moderate intensity. No severe or life-threatening AEs were reported. Twenty-three AEs (56.1%) in 10 of the 14 subjects who experienced an AE were reported as drug-related (at least with a reasonable possibility). Most AEs were singular occurrences and no trend in relation to increasing NI-0501dosage was observed.

All NI-0501 infusions were uneventful.

Among the AEs reported during the 24 hours post infusion, chills (1 in the 1 mg/kg cohort), myalgia (1 in the 3 mg/kg cohort) and pyrexia (3 in the 1 mg/kg cohort) were reported. These symptoms were assessed as mild, except one assessed as moderate (temperature increase up to 38.4°C, treated by paracetamol administration) and were concomitant to transient laboratory changes (CRP and neutrophil elevation) and some slight cytokine variations. These observations at the higher doses of NI-0501 administration suggest an attempt by the immune system to adjust to a new homeostasis upon IFNγ abrogation. They appear to be consistent with the mild flu-like symptoms reported with another IFNγ neutralizing antibody (Fontolizumab), after its first administration in Crohn's disease patients[4;5].

Regarding the occurrence of infections, only common infections such as upper respiratory tract infections were observed after administration of NI-0501, with a similar incidence reported in placebo subjects.
Herpes zoster (HZ) was also reported in one subject (male, aged 26) 14 days after the infusion of 3 mg/kg of NI-0501. This event was assessed as related to the NI-0501 infusion and considered as serious (medically significant) in the context of a Phase I study in healthy volunteers (HVs). Its intensity was moderate and its course normal under antiviral therapy and the subject recovered with no sequelae.
The occurrence of an HZ infection in a subject who has received a dose of NI-0501 intended to fully neutralize IFNγ can be attributed to the expected pharmacological effect of the drug. An increased susceptibility to HZ infections in patients having developed auto-antibodies against IFNγ[6] or having received ustekinumab (a mAb which decreases IFNγ production by inhibiting the p40 subunit of IL-12) has been described in the literature. HZ infection is reported in the ustekinumab USPI with a frequency rate less than 1% in controlled clinical studies. However, the subject who suffered from the HZ infection also exhibited unexplained elevated serum IgE levels prior to inclusion in, and during, the study. A non-detectable subtle defect in the immune system linked to the elevated IgE levels may have increased the susceptibility of this subject to the pharmacological effect of NI-0501.

A blood sample was taken pre-infusion and at Week 8 for anti-drug antibody (ADA) detection. All samples were reported as negative for the presence of anti-NI-0501 antibodies. As in a few subjects NI-0501 levels were still detectable at Week 8, additional samples at follow-up monthly visits were taken. The presence of ADA was found in one subject (having received 1 mg/kg) at Week 28, not affecting PK.

In conclusion, the infusion of NI-0501 was well tolerated and the effects observed during the 8 week monitoring after drug infusion did not reveal any serious or unexpected off-target safety or immunogenicity concerns. Therefore, neither the clinical features and laboratory results, nor the PK profile and pharmacodynamic (PD) effects observed after administration of NI-0501 in HVs prevent moving NI-0501 into the next development phase.

At the cut-off date of January 31st 2016, seventeen patients with confirmed or suspected primary HLH have been enrolled in the present NI-0501-04 study (8 in US, 9 in EU). Two patients received NI-0501 as a first-line treatment and 15 patients received NI-0501 as a second-line treatment because of an inadequate response to previous HLH treatments. For interim outcome data please refer to the latest Investigator's Brochure (presently version 5.0, dated 20 January 2016). A favourable benefit risk profile of NI-0501 in primary HLH patients has been assessed based on data so far obtained in the study.

Please refer to section 9.5.2 – General Benefit/Risk Considerations, for the current benefit/risk assessment on the use of NI-0501 in HLH patients.

1.2 HAEMOPHAGOCYTIC LYMPHOHISTIOCYTOSIS (HLH)

HLH is a syndrome characterized by a severe impairment or absence of cytotoxic function by NK and CD8+ T cells with striking activation of the immune system.

HLH comprises primary (genetic/familial) HLH and secondary HLH, both clinically described by a dysregulation of the immune system leading to a profound hypercytokinaemia with deleterious consequences on various tissues and organs[7].

Primary HLH is a heterogeneous autosomal recessive disorder found to be more prevalent with parental consanguinity. Primary HLH is mostly seen in infancy and early childhood with an estimated prevalence in Europe of 1/50,000 live births[8]. The disease is invariably fatal with a median survival of less than 2 months after diagnosis, if untreated[9;10].

The impaired cytotoxic function present in HLH leads to hypercytokinemia and heamophagocytosis. These in turn cause all the typical symptoms of HLH[11-13]:

- Prolonged fever
- Splenomegaly
- Cytopenia
- Hyperferritinemia
- Hypertriglyceridemia
- Hypofibrinogenemia
- Lymphohistiocytic infiltrate, bone marrow hypoplasia, meningeal infiltrate.

Among the cytokines dramatically elevated in HLH patients are: IFNγ, interleukin (IL)-6, IL-10, tumour necrosis factor (TNF) α, IL-8, macrophage colony stimulating factor (MCSF) and granulocyte-macrophage colony-stimulating factor (GM-CSF).

HLH can also occur during the course of an infection, a rheumatic or a neoplastic disease and in this case it is referred to as secondary HLH. Secondary HLH presents with the same signs and symptoms of primary forms and can be equally severe. The current treatment of secondary HLH is aimed at addressing the cause of the underlying disease. This is certainly the case for HLH caused by infections such as Leishmaniasis. Of note, the presence of certain infections, in particular viral infections such as those due

to CMV or EBV, is very often the trigger for the manifestation of primary forms of HLH. This observation is also supported by the evidence that in animal models of primary HLH[14-17], infection with lymphocytic choriomeningitis virus (LCMV) is required for the development of the disease.

When HLH manifests during a neoplastic disease, in particular an hematological malignancy, often the severity of the patient condition requires the immediate treatment of HLH, prior to specifically addressing the underlying disease.

The presence of signs and symptoms of HLH in patients suffering from a rheumatic disease, such as systemic Juvenile Idiopathic Arthritis (sJIA) and Systemic Lupus Erythematosus (SLE), is often referred to by rheumatologists as Macrophage Activation Syndrome (MAS) and can precede the appearance of the rheumatic disease itself. The majority of patients with MAS have impaired NK and perforin functional tests and a significant number of patients show polymorphisms or heterozygous mutations in PRF1 and UNC13D. Although it is an extremely severe and life threatening condition, usually it resolves when an adequate treatment is initiated, consisting in most cases of corticosteroids and cyclosporine. However, in approximately 15%[18] of patients developing MAS, the disease can be difficult to control and the use of etoposide may be considered.

While primary HLH is recognized as predominantly a childhood disease, HLH is a condition that can be found in adults, and increased awareness indicates this may happen more often than recognized in the past. In the majority of adult patients the disease develops during malignancies (mainly non-Hodgkin lymphomas), infections, auto-inflammatory or autoimmune diseases and iatrogenic immune deficiencies.

There are currently no approved drugs for the treatment of HLH. However, experts in the field have established guidelines for the management of HLH patients[19-21].

The management of primary HLH patients currently comprises of the following steps[19]:

- Induction therapy of 8 weeks with a combination of corticosteroids and immunosuppressive drugs (e.g. VP-16, cyclosporin, alemtuzumab, ATG);
- Maintenance therapy up to haematopoietic stem cell transplantation (HSCT);
- HSCT for all patients with an identified genetic deficiency and eventually in very severe HLH cases with no disease-associated mutations.

The main goal of induction therapy is to suppress the life-threatening inflammatory process that characterizes HLH, enabling HSCT in those patients who require it[22]. HSCT is the only curative treatment for HLH associated with high penetrance genetic mutations[20].

Despite the adoption of such guidelines the overall mortality rate for primary HLH remains around 40 to 50%[20;23].

The need to use, during the induction period, drugs associated with severe short and long term-safety issues further contributes to the already high mortality. This constitutes a strong argument for the development of a targeted treatment ensuring efficacy with less toxicity.

1.3 STUDY RATIONALE

During the last years, growing evidence of the pivotal role of IFNγ in the development of HLH has been demonstrated[7;14;15;24-26].

The mutations of genes which characterize primary forms of HLH all affect proteins involved in the same process, ultimately impairing cytotoxic activity. Perforin mutations were the first identified in HLH patients.

Perforin knocked out (KO) mice are considered a relevant model for the human disease. In fact, these mice, once infected with LCMV, develop all the diagnostic and many of the clinical and laboratory characteristic features of the human disease, and they die if untreated. For these reasons, perforin KO mice have been used to study the pathophysiology of HLH. The HLH-like pathology that they develop is dependent on CD8+ T cells and IFNγ produced in response to antigen stimulation.

It was demonstrated that when the high circulating levels of IFNγ are neutralized, with the administration of an anti-IFNγ antibody, not only are the clinical and laboratory abnormalities reverted, but also survival rate is dramatically improved. On the contrary, the ablation of any other cytokine had no impact on survival[14;15].

Two models of secondary HLH have been investigated in the context of the NI-0501 development program. In one model, repeated administration of CpG (causing TLR9 stimulation) has been used to mimic a chronic severe hyperstimulation in healthy mice (i.e. with normal genetics of the cytotoxic pathway) as a model of HLH secondary to infection. Although these mice do not necessarily die, they develop typical clinical and laboratory features of HLH. When IFNγ is neutralized, with the administration of an anti-IFNγ antibody, clinical and laboratory features of the disease are reverted. Interestingly, in this model it has been demonstrated that administration of the anti-IFNγ antibody leads to full neutralization of IFNγ effects also in relevant target tissues, such as liver and spleen (manuscript in preparation).

To study the physiopathology of secondary HLH occurring in the context of rheumatic diseases, an animal model has been generated using IL-6 transgenic mice expressing high levels of IL-6, similarly to what occurs in patients with sJIA, the rheumatic disease most frequently associated with secondary forms of HLH. When triggered with Toll Like Receptor (TLR) ligands, these mice die with many of the features of the human disease[27]. In these mice, when IFNγ is neutralized with the administration of an anti-IFNγ antibody, survival is markedly improved and laboratory parameters reverted (Prencipe G et al, manuscript in preparation).

Further strengthening the importance of IFNγ in HLH are the high concentrations of circulating IFNγ levels in primary HLH patients[7;25]. In a series of 71 patients monitored from HLH diagnosis to treatment and follow-up (including 2 with leukaemia and 3 with lymphoma), IFNγ levels were above upper limit of normal (17.3 pg/mL) in all patients, and in particular 53.5 % had levels above 1000 pg/mL. It was also reported that IFNγ levels rise early and quickly, and can fall from > 5000 pg/mL to normal in 48 hours upon effective treatment of HLH.

More recently, in an observational study in patients with secondary forms of HLH, high levels of IFNγ were demonstrated both in patients with HLH secondary to infections and in patients with HLH occurring in the context of sJIA. The levels of CXCL9, CXCL10 and CXCL11, three chemokines that are known to be induced by IFNγ, were also significantly elevated. Noteworthy, levels of IFNγ, and of the three IFNγ chemokines, were found to be significantly correlated with laboratory parameters of disease severity, such as ferritin, platelet count and transaminases (Bracaglia et al., manuscript submitted).

As hypercytokinemia and organ infiltration by activated lymphocytes and hystiocytes are responsible for all symptoms present in HLH patients and are dependent on CD8+ T cells hyperactivity and high IFNγ levels, the neutralization of IFNγ represents a rational approach, as no agents specifically targeting CD8+ T cells are available at the moment, and targeting individual cytokines downstream of IFNγ would not necessarily be feasible.

Therefore, based on the data from animal models of primary and secondary HLH and from the observation made in patients with both primary and secondary HLH, confirming the critical role played by IFNγ in the pathogenesis of this disease, the neutralization of IFNγ offers a robust rationale to develop a targeted therapy for HLH, which must be effective with no or limited toxicity.

2 OBJECTIVES

- To determine the safety and tolerability profile of multiple IV administrations of NI-0501
- To determine the efficacy and benefit/risk profile of NI-0501 in HLH patients
- To describe the PK profile of NI-0501 in HLH patients
- To define an appropriate NI-0501 therapeutic dose regimen for HLH
- To determine the PD effects (levels of circulating Total IFNγ and biomarkers of its neutralization, namely CXCL9 and CXCL10)
- To determine other biomarkers, e.g. sCD25, IL-10
- To assess the immunogenicity of NI-0501

3 STUDY DESIGN

3.1 OVERALL DESIGN

This is an open-label, single arm, international multicentre Phase 2/3 study.

The study, initially designed as a pilot Phase 2 study, with the target of enrolling 10 evaluable patients with primary HLH, based on the positive benefit risk profile observed in the patients enrolled in the study so far and in consideration of:

i) the rare nature of the disease;
ii) the lack of valuable therapeutic options especially for patients having failed previous HLH therapies or being unable to continue due to toxicity;
iii) the significant number of requests for compassionate use of NI-0501 for HLH patients;

has been amended to become a Phase 2/3 continuing enrolment of both first and second line patients.

Patients who receive NI-0501 after having failed conventional HLH therapy or having shown intolerance to it represent the pivotal cohort of the study, to demonstrate the efficacy of NI-0501 as second line treatment of primary HLH.

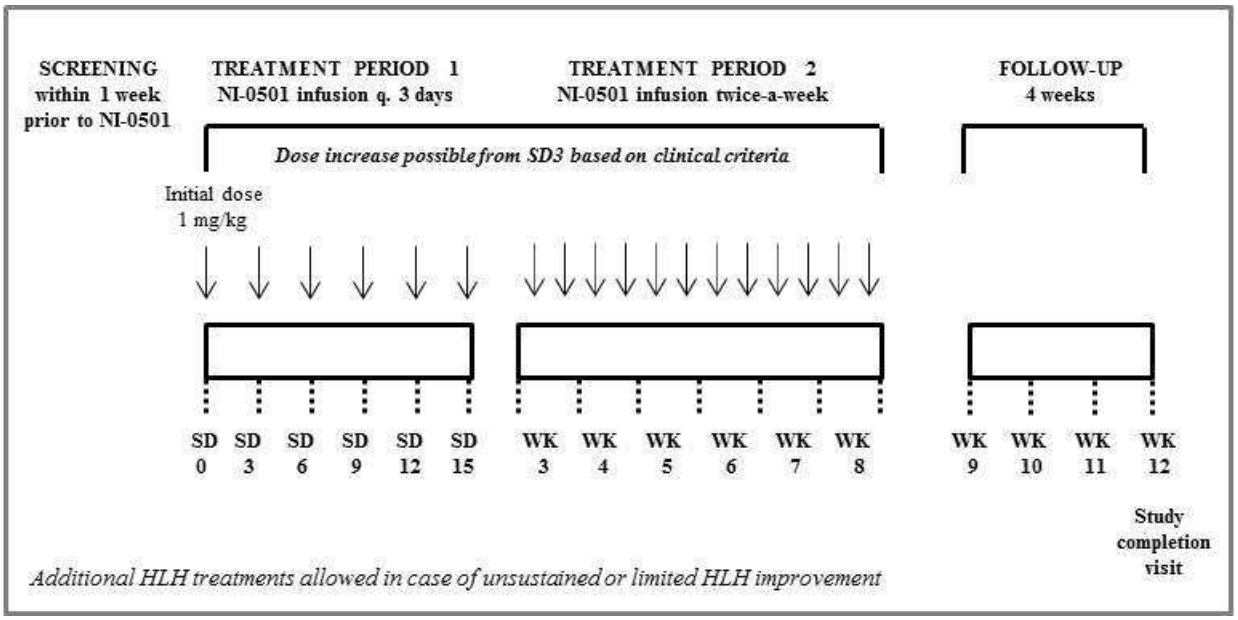
Treatment-naïve patients will be enrolled for collection of efficacy and safety data in the first line setting.

The study foresees a screening, a treatment and a follow-up period. The study design is summarised below (Figure 1).

If not already hospitalised, patients will be admitted to the unit the day before the first administration of the study drug (study day minus one, SD-1). Discharge from the hospital cannot occur before SD15. After SD15, in case the patient condition allows, the Investigator at the site can discharge a patient from the hospital provided the following condition has been met:

- no active infections requiring IV antimicrobial therapy are present.

Figure 1: NI-0501-04 Study Design



3.2 SCREENING PERIOD

Screening will be carried out within 1 week prior to first administration of NI-0501 (SD0) to enable confirmation of patient eligibility and following the signature of the Informed Consent Form.

In the event that a patient's medical condition warrants rapid treatment initiation, and to avoid unnecessary repetition of recent interventions, results of tests which have been performed as part of the normal patient's care at the site not more than 12 days prior to first NI-0501 infusion, can be considered for screening purposes (inclusion/exclusion criteria checks), with the agreement of both the Sponsor and the Investigator.

Samples for infection screening need to be collected for analysis according to the protocol requirements; however availability of the results is not required prior to initiation of NI-0501 treatment provided that there are no clinical findings suggestive for the presence of any of the infections which represent exclusion criteria.

3.3 TREATMENT PERIOD

NI-0501 will be administered for 8 weeks as induction treatment of HLH.

The treatment period will be divided in 2 separate periods: Treatment Period 1 and 2 (please refer to Figure 1).

NI-0501 treatment can be shortened, although not less than 4 weeks, if patient's condition and donor availability allow an earlier HSCT.

Based on the current knowledge, no wash-out period is required between the last administration of NI-0501 and the start of conditioning.

In the event that an appropriate donor has not been identified by Week 8 or in case of the need to delay the schedule of HSCT for reasons unrelated to the administration of NI-0501, NI-0501 treatment can be continued beyond this time upon the request of the Investigator, provided a favourable benefit/risk has been established for that patient.

3.4 FOLLOW-UP PERIOD

All patients who have received at least one dose of NI-0501 will be monitored for 4 weeks after the last administration of NI-0501 within the context of the NI-0501-04 protocol, independently of the duration of treatment with NI-0501.

In the event that the NI-0501 concentration is still measurable after the 4 week follow-up period (i.e. short-term follow-up), NI-0501 monitoring will continue until a measurable concentration of NI-0501 is no longer detectable. This monitoring should occur, whenever possible, in the context of the long-term follow-up study, NI-0501-05.

Patients for whom an Investigator has requested a prolongation of NI-0501 treatment beyond Week 8 will directly enter the long-term follow-up study NI-0501-05, without having to complete the 4 week short-term follow-up.

All patients having completed the follow-up period will also be asked to enter the open label safety extension study (NI-0501-05).

3.5 STUDY END

The end of the study is defined as the last visit of the last patient.

The last visit of a patient should occur, at the latest, 4 weeks after the last administration of NI-0501, except for patients continuing NI-0501 treatment beyond Week 8 (see paragraph below).

In case of an ongoing serious adverse event (SAE), the patient will continue to be monitored until resolution or until the outcome of the event is known and stable, beyond the defined study end as necessary. In the event that additional NI-0501 concentration monitoring is not performed in the context of the NI-0501-05 study, this monitoring will continue beyond the 4 week follow-up period, until a measurable concentration of NI-0501 is no longer detectable. This measurement should occur not less than every two weeks.

In this study, no further treatment beyond Week 8 is planned. However, in the event that an appropriate donor has not been identified by Week 8 or in case of the need to delay the schedule of HSCT for reasons unrelated to the administration of NI-0501, NI-0501 treatment can be continued beyond this time upon the request of the Investigator, in the context of an open label safety extension long term follow-up study, providing a favourable benefit/risk has been established. For these patients, end of study NI-0501-04 is defined as their last visit (i.e. three days after the last infusion administered in the context of NI-0501-04 study).

3.6 LONG-TERM FOLLOW-UP STUDY (NI-0501-05)

All patients having received at least one dose of NI-0501 in the pilot study will be asked to be part of the long-term follow-up study (NI-0501-05), to allow long-term safety surveillance, and, when relevant, investigation of the impact of NI-0501 treatment on survival and post-HSCT outcome measures.

This study will allow monitoring of patients who have been considered for a treatment extension.

4 TARGET POPULATION

The study population comprises patients of both genders, up to and including 18 years[i] at diagnosis, suffering from confirmed or suspected primary HLH.

Patients can be naïve to HLH treatment (first line patients), or may have already received conventional HLH therapy[ii] (second line patients), without obtaining a satisfactory response according to the treating physician or having shown signs of intolerance to it.

4.1 ELIGIBILITY CRITERIA

Patients included in the study must be compliant with the following inclusion/exclusion criteria:

4.1.1 Inclusion Criteria

1. Primary HLH patients of both genders, up to and including 18 years[i] at diagnosis of HLH. The diagnosis of HLH must be made on the basis of the following criteria (as per HLH-2004 protocol):
   a. A molecular diagnosis or familial history consistent with primary HLH
      OR
   b. Five out of the eight criteria below are fulfilled:
      - Fever
      - Splenomegaly
      - Cytopenias affecting 2 of 3 lineages in the peripheral blood (hemoglobin < 90 g/L; platelets < 100 x 10^9/L; neutrophils < 1 x 10^9/L)
      - Hypertriglyceridemia (fasting triglycerides ≥ 3 mmol/L or ≥ 265 mg/dL) and/or hypofibrinogenemia (≤ 1.5 g/L)
      - Hemophagocytosis in bone marrow, spleen or lymph nodes, with no evidence of malignancy
      - Low or absent natural killer (NK)-cell activity
      - Ferritin ≥ 500 µg/L
      - Soluble CD25 (sCD25; i.e. soluble IL-2 receptor) ≥ 2400 U/mL.
2. Presence of active disease in patients as assessed by the treating physician.
3. Patients having already received HLH conventional therapy must fulfill one of the following criteria as assessed by the treating physician:
   - Having not responded
   - Having not achieved a satisfactory response
   - Having not maintained a satisfactory response
   - Showing intolerance to conventional treatment of HLH

   At the time of enrollment, eligible patients might still be receiving treatment (induction or maintenance) or might have already discontinued it.

[i] Or an age appropriate to be treated in the Investigator's practice
[ii] Conventional HLH therapy as per site standard of care, e.g. any of the following alone or in combination (Etoposide, ATG, Alemtuzumab and Cyclosporine A) or glucocorticoids, namely Dexamethasone at 10 mg/m2 for at least 7 days or methylprednisolone pulses for 3 consecutive days

4. Informed consent signed by the patient (if ≥ 18 years old), or by the patient's legally authorized representative(s) with the assent of patients who are legally capable of providing it.
5. Having received guidance on contraception for both male and female patients sexually active and having reached puberty:

   Females of child-bearing potential, having a negative pregnancy test at screening, and unless true abstinence is in line with the preferred and usual lifestyle of the patient, must agree to use adequate method(s) of birth control from screening until 6 months after receiving last dose of the study drug. Males with partners(s) of child-bearing potential must agree to take appropriate precautions to avoid fathering a child from screening until 6 months after receiving last dose of the study drug.

4.1.2 Exclusion Criteria

1. Diagnosis of secondary HLH consequent to a proven rheumatic or neoplastic disease.
2. Body weight < 3 kg.
3. Patients treated with:
   - any T-cell depleting agents (such as anti-thymocyte globulin [ATG], anti-CD52) during the previous 2 weeks prior to screening
   - any other biologic drug within 5 times their defined half-life period, except for rituximab in case of documented B-cell EBV infection (a list of some of the most commonly used biologic half-lives is included in the Study Specific Risk Management Plan)
4. Active mycobacteria*, Histoplasma Capsulatum, Shigella, Campylobacter, Leishmania* or *Salmonella* infections.
5. Evidence of past history of tuberculosis or of latent tuberculosis.
6. Positive serology for HIV antibodies, hepatitis B surface antigen or hepatitis C antibodies.
7. Presence of malignancy.
8. Patients who have another concomitant disease or malformation severely affecting cardiovascular, pulmonary, liver or renal function.
9. History of hypersensitivity or allergy to any component of the study regimen.
10. Vaccination with a live or attenuated live (including BCG) vaccine within the previous 12 weeks prior to screening.
11. Pregnant or lactating female patients.

5 INVESTIGATIONAL MEDICINAL PRODUCT (IMP)

5.1 DESCRIPTION OF IMP

NI-0501 is a fully human anti-IFNγ monoclonal antibody which binds and neutralises IFNγ.

NI-0501 is manufactured by a third party manufacturing facility duly qualified by Novimmune and is supplied to study sites in either 2 mL and/or 10 mL filled single-use glass vials at a concentration of 5 mg/mL, for dilution prior to administration.

The nominal composition of the NI-0501 sterile concentrate for infusion (per mL) is as follows:

Ingredient	Quantity (per mL)
NI-0501	5 mg
L-Histidine	1.55 mg
L-Histidine monohydrochloride, monohydrate	3.14 mg
Sodium chloride (NaCl)	7.31 mg
Polysorbate 80	0.05 mg
pH	6.0 ± 0.2

The solution contains no antimicrobial preservative, and therefore each vial must be used only once.

5.2 RATIONALE FOR DOSE SELECTION

5.2.1 Initial dose (see Appendix A)

Data from *in vitro* experiments investigating the binding kinetics of NI-0501 to human IFNγ and the functional inhibition of human IFNγ by NI-0501 have been used for predicting the concentrations of NI-0501 expected to inhibit (e.g. 99%) the effect of circulating IFNγ concentrations.

Based on:

- the calculated neutralizing concentrations of NI-0501
- the PK parameters of NI-0501 in healthy volunteers
- the PK information from recombinant IFNγ in humans,

simulations were performed regarding the dose that would inhibit the effect of circulating and newly formed IFNγ by up to 99% over a period of 3 days in HLH patients.

Based on these simulations, the starting dose in HLH patients is 1 mg/kg. This dose is predicted to inhibit for 3 days at least 99% of IFNγ effect in patients with baseline IFNγ concentrations lower or equal to 3400 pg/mL. This dose is mainly driven by the estimated production of IFNγ which is expected to impact the clearance of NI-0501 (due to target-mediated drug disposition) and varies considerably between patients as already indicated by the wide range of baseline IFNγ concentrations observed in these patients.

5.2.2 Subsequent doses (see Appendices A and B)

Due to the expected target-mediated drug disposition (TMDD) effect and to the high inter-individual variability of IFNγ concentrations in HLH patients, doses subsequent to the initial one can be increased to 3 mg/kg, if required, based on clinical and laboratory criteria (see Table 4).

After a minimum of two infusions at 3 mg/kg, if upon re-assessmen, the same clinical and laboratory criteria are found to still apply, the dose of NI-0501 may be increased to 6 mg/kg for up to four infusions, with a regular monitoring of the clinical and laboratory HLH parameters.

If clinical and laboratory response criteria are no longer applicable, the dose of NI-0501 will be decreased back to 3 mg/kg,

On the other hand, if criteria still apply, based on careful benefit/risk assessment, the Investigator may propose either

i) to continue treatment at 6 mg/kg for additional infusions

or

ii) to increase NI-0501 dose above 6 mg/kg if PK and PD evidence indicates extremely high IFNγ levels and, consequently, fast NI-0501 elimination.

However, the Investigator's proposal of either continuing 6 mg/kg infusions or increasing the dose above 6 mg/kg has to be discussed and approved by the DMC, after thorough assessment of all available data, including PK and PD.

Appendix A "NI-0501 dose justification in HLH patients" provides a detailed descriptions of the rationale supporting the initial and subsequent NI-0501 doses in HLH patients.

Appendix B "Decision making process on dose selection" includes a detailed description of the clinical criteria and the process to guide decision on dosing increase by the Investigator.

Table 4 : Clinical and laboratory criteria to guide dose increase

Study day (SD)	NI-0501 dose	
On SD0	Starting dose of 1 mg/kg	
On SD3	Increase to 3 mg/kg	*Criteria to be met*: - if fever persists or reoccurs (when present at baseline) or - if significant worsening of clinical conditions
From SD6 onwards[a]	Increase to 3 mg/kg[b]	*Criteria to be met*: - if no satisfactory improvement in clinical conditions (as assessed by the Investigators) and - at least 1 of the followings: *Platelet counts* (x10^3/mcl) If bsl. counts < 50 → no improvement to > 50 bsl. counts 50-100 → less than 30% improvement bsl. counts > 100 → any decrease to < 100 *ANC* (count/mcl) If bsl. counts < 500 → no improvement to > 500 bsl. counts 500-1000 → any decrease to < 500 bsl. counts > 1000 → any decrease to < 1000 *Ferritin* (ng/ml) If bsl. levels ≥ 3000 → no improvement (<20% decrease) bsl. levels < 3000 → any increase to > 3000 *Splenomegaly* → worsening (at clinical or US examination) *Coagulopathy* (both D-Dimer and Fibrinogen have to apply) D-Dimer If abnormal at bsl. → no improvement Fibrinogen (mg/dL) If bsl. levels ≤ 100 → no improvement bsl. levels > 100 → any decrease to < 100
From SD9 or SD12 onwards[c]	Increase to 6 mg/kg[d]	*Criteria to be met*: - In case, after a minimum of two infusions at 3 mg/kg, the criteria above reported have been reassessed and found to be still met

[a] NI-0501 dose has to be increased from 1 to 3 mg/kg if these criteria apply after SD6.

[b] If NI-0501 dose has been already increased on SD3, at least two infusions at the dose of 3 mg/kg have to be performed before criteria re-assessment.

[c] Depending on whether dose increase to 3 mg/kg has occurred on SD3 or SD6.

[d] For a maximum of four infusions.

Abbreviations: bsl. = baseline; ANC = absolute neutrophil count; US = ultrasound

5.3 DOSING REGIMEN

NI-0501 will be administered by IV infusion over a period of one hour, at a starting dose of 1 mg/kg.

Infusions will be performed every 3 days for the first NI-0501 six infusions (until SD15). Dose increases are possible at any time during the study (see 5.2.2 above).

From SD15 during Treatment Period 2, the administration of NI-0501 will occur on a twice-a-week schedule. Elongation of the dosing interval to 1 week can occur after 4 weeks of treatment, if the patient has achieved Complete Response and maintained it for at least one week.

5.4 IMP HANDLING

5.4.1 Packaging and Labelling

NI-0501 will be supplied to study sites in glass vials containing a either 2 and/or 10 ml solution at a concentration of 5mg/ml. Labelling and packaging will be prepared to meet local regulatory requirements.

5.4.2 IMP Supply

NI-0501 will be supplied to the study sites as open-label supplies.

5.4.3 IMP Receipt and Storage

The NI-0501 vials will be transported with temperature deviation alarms (TempTale 4 or equivalent device), in order to ensure consistent temperatures during transit. When the study drug is received at the site, the Investigator or Pharmacist will check for accurate delivery and absence of temperature deviation alarms.

The study drug should be stored between 2-8°C (36 - 46°F). All vials must be stored in a secure locked location in a temperature-controlled refrigerator or cold room. Any deviations from the recommended storage conditions should be immediately reported to the Sponsor and responsible study monitor or contract research organisation (CRO). Affected vials should not be used and should be quarantined until the Sponsor has authorised their use, return or destruction.

Documentation of the storage conditions of the study drug must be maintained for the duration of the time the study drug is stored at the site, until such time as it is used, disposed of, or returned to NovImmune or designee.

Regular inspections of the NI-0501 vials are required, as detailed in the IMP manual's directions for the Preparation and Administration of Individual Doses of Study Drug NI-0501.

5.4.4 IMP Preparation, Administration, Accountability and Destruction

5.4.4.1 Preparation

The study drug must be prepared only by a Pharmacist or other appropriately qualified staff member, specifically authorised by the Investigator/Pharmacist.

The specific dose to be administered for an individual infusion is determined as detailed in Section 5.3. As NI-0501 is dosed in mg/kg, the weight of the patient must be taken within 24 hours of the preparation of the study drug for administration.

Full instructions for the preparation, including dilution steps, and method for administration of NI-0501 are available in the IMP manual's directions for the Preparation and Administration of Individual Doses of Study Drug NI-0501.

5.4.4.2 Administration

The patient should receive the designated volume through an infusion pump over 1 hour (or more depending on the volume to infuse). A 0.2 μm filter has to be added to all infusion lines.

It is recommended that an intravenous central line remains in place to ensure venous access during the treatment period. Since no data is available on the compatibility of NI-0501 with other intravenous substances or additives, other medications/substances should not be added to the infusion material or infused simultaneously through the same intravenous line. If the same intravenous line is used for subsequent infusions of other drugs, the line should be flushed with saline before and after infusion of NI-0501.

The infusion of NI-0501 will be administered under the direct supervision of the Investigator (or delegate). It must be performed in the morning, preferably always at the same time, with a maximal tolerated difference in onset of infusion of not more than 3 hours compared to the initial infusion. Details of the infusion administered must be recorded in the patient's Medical Notes (source documents) and CRF with:

- The date of administration
- The time (start and finish) of administration
- The volume administered
- Any incidence of adverse effects or general illness experienced by the patient.
- Any other event(s) judged relevant by the site personnel.

5.4.4.3 Accountability

When the study drug is received at the site, the Investigator or Pharmacist (or appropriate designee) should acknowledge its receipt by signing (or initialling) and dating the documentation. Documentation should be returned to NovImmune (or its designee) and a copy retained in the Investigator's file.

The dispensing of the study drug shall be carefully recorded on Drug Accountability Forms and an accurate accounting must be available for verification by the Monitor at each monitoring visit.

The used (or unused) infusion material should be sent back to the Pharmacist at the end of the infusion, if possible, for later inventory. If this is not possible, accountability should be made at the bed-side before discarding the material.

Drug accountability records shall include:

- Confirmation of the study drug's delivery to the study site
- The inventory at the study site
- The use of study drug by each patient
- The return to the Sponsor or alternative disposition of unused products.

The records should include dates, quantities, expiration dates, if applicable, and batch number and patient number.

Unused study drug must not be discarded or used for any purpose other than the present study. Study drug that has been dispensed to a patient and returned unused must not be re-dispensed to a different patient.

5.4.4.4 Destruction, Return and Disposal

Periodically during the study and at the conclusion of participation of the study by the site, the clinical research associate (CRA) will monitor and collect the Drug Accountability Forms, before making arrangements for study drug return or authorisation of destruction by the study site.

6 PATIENT BACKGROUND TREATMENT AND CARE

6.1 BACKGROUND THERAPY WITH DEXAMETHASONE

In treatment-naïve patients, NI-0501 will be administered on a background of 10 mg/m^2 of dexamethasone. In patient receiving NI-0501 as second line HLH treatment, dexamethasone has to be administered at the dose of at least 5mg/m^2, or at the same dose administered prior to screening if higher. Patients are required to have received dexamethasone from at least SD-1.

Dexamethasone can be tapered depending on patient condition, according to the judgment of the treating physician. The tapering scheme can be selected by the treating physician, provided that the dexamethasone dose, at each step, is not more than halved and frequency of change is not more than weekly.

In the event of disease worsening after tapering of dexamethasone, the dose of dexamethasone can be increased and maintained until a satisfactory response is achieved according to the treating physician.

6.2 PROPHYLACTIC TREATMENT

As recommended in HLH treatment guidelines, patients will receive prophylactic treatment for *Pneumocystis jiroveci* and fungal infections. In addition, prophylaxis for *HZ* virus infection will be performed to mitigate the potential risk associated to NI-0501 administration (see Benefit/Risk Management, Section 9.5). In the unlikely event that a patient, previously vaccinated for TB, shows a Purified Protein Derivative (PPD) test result ≥ 5mm and a negative IFNγ-release assay, the patients will receive prophylaxis.

Patients will therefore receive prophylactic treatments starting from the day prior to initiation of NI-0501 treatment (i.e. SD-1) until the end of the study:

- For *Pneumocystis jiroveci* prevention, according to Institution Guidelines/ Recommendations (e.g. 750 mg/m2/day sulfamethoxazole with 150 mg/m2/day trimethoprim given orally in equally divided doses twice a day, on 3 consecutive days per week).
- For fungal infection prevention, according to Institution Guidelines/Recommendations (e.g. Fluconazole 12 mg/kg daily with a maximum of 400 mg daily dose).
- For Varicella Zoster virus prevention, according to Institution Guidelines/Recommendations (e.g. Acyclovir 200 mg four times daily for children over two years, for children under two years 100 mg four times daily).
- For TB, if required (see above), according to Institution Guidelines/Recommendations (e.g. Isoniazid).

These treatments will be given orally, whenever possible, otherwise intravenously.

In the event that NI-0501 concentrations remain at therapeutic levels after the end of the study, it is strongly recommended that the above mentioned prophylaxis be maintained, regardless of the participation of the patient in the NI-0501-05 study.

6.3 CONCOMITANT THERAPY

6.3.1 Cyclosporin A

CsA can be continued, if already administered prior to screening. CsA can be withdrawn at any time, upon the judgement of the Investigator. CsA is not to be introduced *de novo* during the course of the study, once NI-0501 administration has started. As it is recommended in other HLH protocols, CsA concentrations should be monitored at least weekly to maintain trough levels of 150-200 ng/ml.

6.3.2 Intrathecal Methotrexate and Glucocorticoids

For patients receiving intrathecal methotrexate and glucocorticoids at the time of NI-0501 treatment initiation, this treatment should be continued as required. If the appearance of CNS symptoms occurs before the initiation of NI-0501 treatment, therapy with intrathecal methotrexate and glucocorticoids must be initiated prior to the first administration of NI-0501.

6.3.3 Other possible concomitant therapies

Intravenous immunoglobulin (IVIG) will not be administered at the dose expected to produce an immunomodulator effect (e.g. 2 g/kg). However, if deemed justified by the treating physician, in case of a documented immunoglobulin deficiency justifying replacement, IVIG can be given at a dose of 0.5 g/kg, every 4 weeks or more frequently in order to maintain adequate IgG levels. Any infusion within the previous 4 weeks prior to screening, as well as any infusion during NI-0501 treatment, should be documented in the CRF (dose, date of administration).

Analgesic treatment, transfusion of blood products, electrolyte and glucose infusions, antibiotics, anti-fungal and anti-viral treatment and general supportive care (e.g. gastro-protective agents) are permitted within the study.

Use of any additional prescription drugs or over-the-counter medication (including herbal and homeopathic preparations), with the exception of multi-vitamins, requires approval from the Investigator.

Contraception guidance: See Inclusion Criteria Section 4.1.1, point 6.

6.3.4 Not allowed concomitant therapies

During treatment with NI-0501, concomitant use of etoposide, T-cell depleting agents, or any other biologic drug is generally not allowed except for the followings:

- G-CSF, in case of prolonged neutropenia
- Rituximab, in case of documented B-cell EBV infection
- additional HLH treatments, in case of unsustained or limited HLH improvement (as defined below) at the maximum NI-0501 dose level. Etoposide should be administered, unless a clear evidence of lack of response or intolerance to the drug is derived from previous medical history.
  In this circumstance, the Investigator may propose an alternative agent which requires to be approved by the Data Monitoring Committee.

  The definitions below apply:

  - Unsustained HLH Improvement:
    Patients who are unable to maintain at least 50% improvement from baseline in 3 HLH parameters (see Table 1). At least two consecutive measurements must document the loss of HLH improvement.
  - Limited HLH Improvement:

Less than 50% change from baseline in a minimum of 3 HLH clinical and laboratory criteria.

Vaccination with a live or attenuated-live (including BCG) vaccine will be avoided during the whole study including the 4-week follow-up period. In the event that NI-0501 concentrations remain at therapeutic levels after the end of the study, the period with no vaccinations should be extended until measurable concentration of NI-0501 are no longer detectable.

6.4 EMERGENCY TREATMENT

Severe allergic reactions such as anaphylactic shock require prompt IV treatment with adrenaline and antihistamines. Oxygen shall be supplied through a face mask. Patients must have an appropriately sized IV line that allows rapid infusion of colloid volume substitution. In case of an anaphylactic reaction patients shall be transferred as soon as possible to the intensive care unit of the hospital.

Following the first administration of NI-0501 and before leaving their reference centre, each patient (and/or patient's legal representative) will be given a card to carry at all times in case of any emergency. The card gives details of the name of the drug, name of the responsible physician, and the address and telephone number of the study site; this card will be collected by the Investigator from the patient after the end of the study.

6.5 RESCUE THERAPY

Patients who are withdrawn from the study due to a safety issue or for lack of efficacy (i.e. worsening of HLH or no response to NI-0501; see Section 10.1.3) will be treated with rescue therapy, according to the standard of care at the site.

7 ENDPOINTS

7.1 SAFETY ENDPOINTS

Safety and tolerability of multiple IV infusions of NI-0501 will be assessed as follows:

- Incidence, severity, causality and outcomes of AEs (serious and non-serious), with particular attention being paid to infections
- Evolution of laboratory parameters such as complete blood cell count (CBC), with focus on red cells (haemoglobin), neutrophils and platelets, liver tests, renal function tests and coagulation
- Number of patients withdrawn for safety issues
- Level (if any) of circulating antibodies against NI-0501 to determine immunogenicity; i.e. the development of anti-drug antibodies (ADAs).
  The detection of ADA (Anti-Drug Antibodies) in human serum will be assessed with a GYROLAB-based assay. This homogeneous assay utilises a bridging format composed of a biotinylated NI-0501 antibody, streptavidin coated beads and an Alexa-Fluor labelled NI-0501 antibody as the detection antibody. The fluorescent signal produced is proportional to the amount of ADA present in the human serum sample.

7.2 EFFICACY ENDPOINTS

Primary efficacy endpoint

- Overall Response Rate, i.e. achievement of either Complete or Partial Response or HLH Improvement, at End of Treatment (EoT).

Criteria for the definition of Overall Response Rate are reported in Table 1.

Secondary efficacy endpoints:

- Time to Response any time during the study
- Durability of Response, i.e. maintenance of response achieved any time during the study until EoT and beyond (including data collected in the long-term follow-up study NI-0501-05)
- Number of patients who reduce glucocorticoids by 50% or more of the baseline dose
- Number of patients able to proceed to HSCT, when deemed indicated
- Survival at Week 8 (or EoT) and at the end of the study [Long-term survival (in particular D+30 and D+100 post-HSCT survival) will be assessed in the context of long-term study NI-0501-05]

7.3 PHARMACOKINETIC ENDPOINTS

All PK data will be summarised using appropriate graphical and tabular presentations. Descriptive non-compartmental pharmacokinetic analysis (NCA) will be applied: C_{max} (concentration corresponding to T_{max}), T_{max} (time of maximum observed concentration), C_{EOI} (concentration at the end of infusion), C_{trough} (concentration just before administration), AUCτ (area under curve of a dosing interval), AUC_{last} (area under curve from the time of dosing to the last measurable concentration), λz (first order rate constant associated with the terminal (log-linear) portion of the curve, estimated via linear regression of time versus log concentration), $t_{½}$ (plasma half-life), CL (systemic drug clearance), Vss (volume of distribution at steady state). Individual and mean PK parameters will be tabulated. Exploratory compartmental pharmacokinetic analysis and population pharmacokinetic analysis will be undertaken to investigate linear and non-linear (TMDD) kinetics. Pharmacokinetic-pharmacodynamic analysis will also be undertaken.

7.4 PHARMACODYNAMIC ENDPOINTS

Determination of PD parameters will include, but will not be limited to, the following:

- Levels of circulating free IFNγ at predose, and of total IFNγ (free + bound) at any subsequent time-point
- Markers of IFNγ neutralization, namely CXCL-9, CXCL-10, CXCL-11
- Other biomarkers (e.g. sCD25, IL-10).

8 OUTLINE OF STUDY PROCEDURES

Patients will be recruited from specialised study sites, with an intensive care unit.

For a detailed description of the schedule of visits and assessments, please refer to the Schedule of Assessment **Error! Reference source not found.** (Screening and Treatment Period 1) and

Table 3 (Treatment Period 2 and Follow-up Period).

The informed consent form must be signed by the patient or his/her legally authorized representative prior to any study-related procedures, with the assent of patients who are legally capable of providing it.

Some procedures are not to be done systematically but only if clinically relevant.

For example:

- ECG is only mandatory at screening, after first infusion, after last infusion and at the end of the study, but can also be done at any other time point, if relevant,
- Brain MRI should be done in case of neurological symptoms occurrence,
- Lumbar puncture for CSF analysis is done (providing that coagulation allows) only at screening but should be repeated during the study course if the initial analysis at screening was abnormal or in case of occurrence of neurological symptoms,
- Search for pathogens during the study should be done if there is any suspicion of infection,
- Chest X-ray during the study should be done more frequently than indicated in case of clinical suspicion of a pulmonary infection.

Analysis done on blood samples will favour as much as possible the use of micro-sampling techniques.
In case of a need for prioritization of blood analysis, laboratory safety parameters (which would have been done as normal disease monitoring) will be prioritized. For details on blood sampling, please refer to Appendix C.
For safety laboratory and search for pathogens, the planned schedule of assessment is in accordance with recommendations for the monitoring of disease evolution and potential for infection in these severely sick patients (HLH-2004 protocol of the Histiocyte Society[iii]). This has been agreed by the study's Scientific Steering Committee (SSC).
The additional amounts of blood which will be drawn for study mandatory specific assessments represent only 24.5 mL for PK assessments and 1.5 mL for immunogenicity monitoring. A maximum of 29 ml of blood are required for PD assessments, and will be only taken if the amount of blood required is acceptable in the context of the EMA guideline[iv].

The following situations will not be considered as protocol deviations:

- Missing data if not occurring at 2 consecutive time-points,
- Missing urinalysis, except for the test to be performed at Screening (or SD0), EoT and Study Completion,
- Vital signs measured within no more than 1 minute before or after the planned time-point when measured every 5, 10 or 15 minutes, within no more than 10 minutes before or after the planned time-

[iii] Treatment protocol of the second international HLH study 2004 available online at http://www.uni-ulm.de/expane/docs/HLH%202004%20Study%20Protocol.pdf

[iv] ETHICAL CONSIDERATIONS FOR CLINICAL TRIALS ON MEDICINAL PRODUCTS CONDUCTED WITH THE PAEDIATRIC POPULATION (2008) - Recommendations of the ad hoc group for the development of implementing guidelines for Directive 2001/20/EC relating to good clinical practice in the conduct of clinical trials on medicinal products for human use

point when measured every hour or 2 hours, and within no more than 15 minutes before or after the planned time-point when measured every 4 or 8 hours.

- Infusion start delayed by no more than 3 hours,
- Physical examination prior to each infusion performed in the late afternoon of the previous day instead of the morning of the infusion (the data will be captured on the infusion day of the eCRF),
- Assessments performed within no more than 48h before or after the planned time-point during the Follow-up Period.

8.1 SCREENING

Patients will be screened for eligibility prior to enrolment into the study. The Investigator must keep a log of the patients screened for the study and reasons for non-eligibility, if applicable.

Screening evaluations should be completed within 1 week prior to the first administration of study drug (SD0) as detailed below. In the event that a patient's medical condition warrants rapid treatment initiation, and to avoid unnecessary repetition of recent interventions, results of tests which have been performed as part of the normal patient's care at the site not more than 12 days prior to first NI-0501 infusion, may be considered for screening purposes (inclusion/exclusion criteria checks) with the agreement of both the Sponsor and Investigator.

The following information must be collected and the following procedures must be performed:

Patient information:
- Demographic and medical history
- Medications at screening
- HLH induction treatment received
- Date of HLH diagnosis
- Molecular diagnosis and perforin expression granule release assay and other functional tests performed for the diagnosis of HLH if available
- Date and criteria of eligibility

Clinical Assessment:
- Physical examination, including liver and spleen size (in cm from costal grill) as well as height (in cm) and weight (in kg) to measure Body Surface Area (BSA)

Procedure:
- Electrocardiogram (ECG)

Imaging:
- 3D abdominal ultrasound with measurements of spleen and liver size
- Chest X-ray
- Brain magnetic resonance imaging (MRI): only if reactivation due to CNS symptoms

Search for infections:
- Tuberculosis (via IFNγ-release assay or PPD test. In a patient having received BCG vaccination a PPD test must be performed and combined with IFNγ-release assay if the PPD result is ≥5mm. In addition, search for Tuberculosis via polymerase chain reaction [PCR] in any relevant specimen should be performed to have a baseline, as this test will be used during the course of the study to

perform regular TB monitoring.

- *Adenoviruses*, *EBV, CMV* by quantitative PCR
- *Herpes Simplex Virus* (*HSV*), *Herpes Zoster Virus* (*HZV*), *Hepatitis B Virus* (*HBV*), *Hepatitis C Virus* (*HCV*), *Human Immune Deficiency Virus* (*HIV*) by PCR monitoring or serology
- *Atypical mycobacteria*, *Histoplasma Capsulatum, Shigella*, *Salmonella*, *Campylobacter* and *Leishmania*, as appropriate[v]. The presence of *Leishmania* can also be ascertained by direct bone marrow observation

Laboratory:

- Complete blood count (CBC) with differential count in order to define an absolute lymphocyte count, and a dedicated lymphocyte subsets count
- Triglycerides (fasting)
- Coagulation tests: activated partial thromboplastin, prothrombin time, D-dimers and fibrinogen
- Biochemistry: glucose and electrolytes, ferritin, C-Reactive Protein (CRP), Aspartate Aminotransferase (AST), Alanine Aminotransferase (ALT), gamma Glutamyl Transferase (γGT), Alkaline Phosphatase (ALP), Lactate Dehydrogenase (LDH), bilirubin, albumin, creatinine and urea
- IgG level
- Urinalysis: glucose, blood, protein, leucocytes, ketones, pH and specific gravity
- Pregnancy test (blood or urine), if applicable

Histopathology:

- Lumbar puncture for cerebrospinal fluid (CSF), if the coagulation function allows

Immunogenicity:

- Baseline assessment for anti-drug antibodies (ADAs)

8.2 STUDY DAY-1 (SD-1)

If not already hospitalized, the patient will enter the hospital on the day prior to the first NI-0501 infusion (i.e. on SD-1).

On this day, the following treatments will be administered:

- Dexamethasone will be administered daily, as described in Section 6.1
- Prophylactic treatment, as described in Section 6.2

[v] A patient with a clinical assessment (including chest X-ray) not indicative of the presence of an active infection, provided that a usable specimen has been taken, and the microbiological analysis is ongoing, can be enrolled prior to the availability of the results.

Clinical assessments:
- Vital signs: temperature, heart and respiratory rate, blood pressure and oxygen saturation
- Physical examination, including spleen and liver size (in cm from costal grill) as well as the weight

8.3 STUDY DAY 0 (SD0, Day of first infusion of NI-0501)

8.3.1 Pre-NI-0501 infusion

The following baseline assessments are conducted on SD0, before NI-0501 is administered:

Clinical assessments:
- Vital signs: temperature, heart and respiratory rate, blood pressure and oxygen saturation
- Physical examination, including spleen and liver size (in cm from costal grill) as well as the weight
- Initiation of continuous cardiac monitoring and pulse oxymetry

Laboratory:
- Complete blood count (CBC) with differential count in order to define an absolute lymphocyte count
- Triglycerides (fasting)
- Coagulation tests: aPTT, PT, D-dimers and fibrinogen
- Biochemistry: glucose and electrolytes, ferritin, CRP, AST, ALT, γGT, ALP, LDH, bilirubin, albumin, creatinine and urea
- Urinalysis: glucose, blood, protein, leucocytes, ketones, pH and specific gravity (if not done at screening)

Pharmacokinetics:
- NI-0501 serum concentration

Pharmacodynamics/ Exploratory parameters:
- sCD25, IL-10, CXCL-9, CXCL-10 and CXCL-11
- Free IFNγ

8.3.2 During NI-0501 infusion

Immediately after the infusion of NI-0501 has started, the following assessments will be conducted:

Clinical assessment:
- Vital signs (blood pressure, heart and respiratory rate and oxygen saturation) and skin aspect (rash, coloration, sweating):
  - every 5 minutes during the first 15 minutes of the infusion and then
  - every 10 minutes until completion of infusion
- Continuous cardiac and pulse oxymetry monitoring.

8.3.3 At the end of NI-0501 infusion

At the end of the infusion of NI-0501, the following assessments will be conducted:

Pharmacokinetics:
- NI-0501 serum concentration

8.3.4 During the 24 hours following NI-0501 infusion

During the 24 hours following NI-0501 infusion, the following procedures will be carried out:

Clinical assessments:

- Vital signs (blood pressure, heart and respiratory rate and oxygen saturation), temperature, and skin aspect (rash, coloration and sweating):
  - every hour for the first 4 hours after infusion and then
  - every 2 hours for the next 12 hours after infusion and then
  - every 4 hours until completion of 24-hour monitoring
- Continuous cardiac and pulse oxymetry monitoring

Procedure:

- ECG (in the afternoon)

8.4 STUDY DAY 1 (SD1)

The following assessments will be carried out 24 hours after the first NI-0501 infusion on SD1 (the day after the first infusion of NI-0501):

Clinical assessments:

- Vital signs: temperature, heart rate, blood pressure and respiratory rate every 8 hours
- Physical examination, including spleen and liver size (in cm from costal grill)

Laboratory :

- CBC with differential count in order to define an absolute lymphocyte count will be performed. In case of a 5-fold increase in lymphocyte count compared to baseline (SD0 pre infusion) or of a total lymphocyte count greater than the upper limit of normal for age, additional investigations will be required, e.g. blood flow cytometry
- Triglycerides (fasting)
- Coagulation tests: aPTT, PT, D-dimers and fibrinogen
- Biochemistry: glucose and electrolytes, ferritin, CRP, AST, ALT, γGT, ALP, LDH, bilirubin, albumin, creatinine and urea
- Urinalysis: glucose, blood, protein, leucocytes, ketones, pH and specific gravity

Pharmacokinetics:

- NI-0501 serum concentration from blood sample taken around 24 h after start of NI-0501 infusion

Pharmacodynamics/ Exploratory Parameters:

- sCD25, IL-10, CXCL-9, CXCL-10 and CXCL-11
- Total IFNγ

8.5 STUDY DAY 2 (SD2)

48 hours after the first administration of NI-0501, the same procedure performed on SD1 (see Section 8.4) must be repeated, with PK sample taken around 48h after start of NI-0501 infusion.

8.6 FROM SD3 TO SD15 (REMAINDER OF TREATMENT PERIOD 1)

8.6.1 Assessments to be performed pre-NI-0501 infusion on SD3, SD6, SD9, SD12 and SD15

The following procedures will be performed prior to the start of the NI-0501 infusion. Please note some assessments will only be performed at selected visits as indicated below:

Clinical assessments: Mandatory at each visit (namely SD3, SD6, SD9 , SD12 and SD15)	▪ Vital signs (blood pressure, oxygen saturation, heart and respiratory rate), temperature, and skin aspect (rash, coloration and sweating) ▪ Physical examination, including spleen and liver size (in cm from costal grill) as well as weight ▪ Initiation of continuous cardiac monitoring and pulse oxymetry
Laboratory: Mandatory only on SD3 and SD6	▪ CBC with differential count in order to define an absolute lymphocyte count and a dedicated lymphocyte subsets count. In case of a 5-fold increase in lymphocyte count compared to baseline (SD0 pre infusion) or of a total lymphocyte count greater than the upper limit of normal for age, additional investigations will be required, e.g. blood flow cytometry. ▪ Triglycerides (fasting) ▪ Coagulation tests: aPTT, PT, D-dimers and fibrinogen ▪ Biochemistry: glucose and electrolytes, ferritin, CRP, AST, ALT, γGT, ALP, LDH, bilirubin, albumin, creatinine and urea ▪ Urinalysis: glucose, blood, protein, leucocytes, ketones, pH and specific gravity
Search for infections: Mandatory only on SD12	▪ Tuberculosis via polymerase chain reaction [PCR] in any relevant specimen ▪ *Adenoviruses, EBV, CMV* by quantitative PCR
Imaging: Mandatory only on SD15	▪ 3D abdominal ultrasound with measurements of spleen and liver size
Pharmacokinetics: Mandatory at each visit	▪ NI-0501 serum concentration
Pharmacodynamics/ Exploratory parameters: Mandatory at each visit	▪ sCD25, IL-10, CXCL-9, CXCL-10 and CXCL-11 ▪ Total IFNγ

8.6.2 During and immediately after NI-0501 infusion

Following completion of all pre-infusion assessments as listed above, on SD3, SD6, SD9, SD12 and SD15, patients will receive NI-0501 by IV infusion over a period of 1 hour (or more depending on the volume to infuse).

The following procedures will be carried out (or continued) during or immediately after the infusion of NI-0501 at each visit:

Clinical assessments:	▪ Vital signs (blood pressure, oxygen saturation, heart and respiratory rate), temperature, and skin aspect (rash, coloration and sweating): - about every 15 minutes until completion of infusion ▪ Continuous cardiac and pulse oxymetry monitoring
Pharmacokinetics: Mandatory at each visit after end of NI-0501 infusion	▪ NI-0501: serum concentration

8.6.3 During the 24 hours following NI-0501 infusion

The following assessments will also be performed during the 24 hours after the infusion of NI-0501 at each visit:

Clinical assessments:	▪ Vital signs (blood pressure, oxygen saturation, heart and respiratory rate), temperature, and skin aspect (rash, coloration and sweating): - about every 4 hours until completion of 24-hour monitoring ▪ Continuous cardiac and pulse oxymetry monitoring

8.6.4 Assessments to be performed on the day before the infusion

Assessments will also be performed in-between infusion days between SD3 and SD15. Please note that some assessments will only be performed at selected time-points:

Clinical assessments: Mandatory at each visit	▪ Vital signs (blood pressure, heart and respiratory rate), temperature, and skin aspect (rash, coloration and sweating): - every 8 hours ▪ Physical examination, including spleen and liver size (in cm from costal grill)
Laboratory: Mandatory at each visit	▪ CBC with differential count ▪ Triglycerides (fasting) ▪ Coagulation tests: aPTT, PT, D-dimers and fibrinogen ▪ Biochemistry: glucose and electrolytes, ferritin, CRP, AST, ALT, γGT, ALP, LDH, bilirubin, albumin, creatinine and urea ▪ Urinalysis: glucose, blood, protein, leucocytes, ketones, pH and specific gravity
Search for infections: Mandatory only on SD5	▪ Tuberculosis through PCR in any relevant specimen ▪ *Adenoviruses, EBV, CMV* by quantitative PCR
Pharmacokinetics: Mandatory only on SD5 and SD8	▪ NI-0501 serum concentrations from blood taken around 48 h after start of previous NI-0501 infusion

Pharmacodynamics/ Exploratory parameters: Mandatory only on SD5 and SD8	▪ sCD25, IL-10, CXCL-9, CXCL-10 and CXCL-11 ▪ Total IFNγ

8.7 TREATMENT PERIOD 2 (Weeks 3 to 8)

Treatment period 2 includes the next infusions that will be performed twice a week, no closer together than 3 days.

The safety and efficacy monitoring/assessment visits should occur every 6 days in Treatment Period 2, with an allowed time-window of ± 48 hours in order to be able to combine these visits with NI-0501 infusions. In case a dose modification is deemed necessary, the required clinical and laboratory data must be recorded/reported.

If patients are no longer hospitalized, they must return to the sites to receive their NI-0501 infusions and to perform the planned assessments as per protocol. Patients in out-patient status must remain at the site for 8 hours following each NI-0501 infusion.

8.7.1 Assessments to be performed on all Infusion Days

8.7.1.1 Prior to Infusion

The following procedures will be performed on infusion days prior to the start of NI-0501 infusion:

Clinical assessments: Mandatory prior to each infusion	▪ Vital signs (blood pressure, oxygen saturation, heart and respiratory rate), temperature, and skin aspect (rash, coloration and sweating) ▪ Physical examination, including spleen and liver size (measure in cm from costal grill) as well as weight ▪ Initiation of continuous cardiac monitoring and pulse oxymetry
Pharmacokinetics: At maximum, every infusion	▪ NI-0501 serum concentration will be measured if required, depending on patient weight, condition, previous drug concentration and potential change in dosing regimen. Sampling schedule will be proposed by the Sponsor and discussed with the site.
Pharmacodynamics/ Exploratory parameters: At maximum, every infusion	▪ sCD25, IL-10, CXCL-9, CXCL-10 and CXCL-11 ▪ Total IFNγ These parameters will be measured if required, depending on patient weight, condition, previous drug concentration and potential change in dosing regimen. Sampling schedule will be proposed by the Sponsor and discussed with the site.

8.7.1.2 During Infusion and at the end of infusion

Following completion of all pre-infusion assessments as listed above, patients will receive NI-0501 by IV infusion over a period of 1 hour (or more depending on the volume to infuse).

The following procedures will be carried out (or continued) during infusion of NI-0501 or just after its end:

Clinical assessments:

- Vital signs (blood pressure, oxygen saturation, heart and respiratory rate), temperature, and skin aspect (rash, coloration and sweating):
  - every 15 minutes until completion of infusion
- Continuous cardiac and pulse oxymetry monitoring

Pharmacokinetics at the end of infusion:
At maximum, every infusion

- NI-0501 serum concentration will be measured if required, depending on patient weight, condition, previous drug concentration and potential change in dosing regimen. Sampling schedule will be proposed by the Sponsor and discussed with the site.

8.7.1.3 Post Infusion

Until the patient is discharged, the assessments detailed in Section 8.3.4 above (for SD0 post-infusion) can be performed over 24 hours or 8 hours, depending on the Investigator's judgment and patient's condition, but in any case with the time intervals reduced to 4-hourly monitoring.

From patient's discharge until last infusion, the assessments detailed in Section 8.3.4 above (for SD0 post-infusion) will be performed over the 8 hours post NI-0501 infusion with the time intervals reduced to 4-hourly monitoring.

Clinical assessments:

- Vital signs (blood pressure, heart and respiratory rate and oxygen saturation), temperature, and skin aspect (rash, coloration and sweating):
  - every 4 hours until completion of either 8 or 24-hour monitoring
- Continuous cardiac and pulse oxymetry monitoring

8.7.2 Efficacy and safety assessments

These assessments will be carried out systematically as described in section 8.7.

In case of an early transplant (after 4 weeks of treatment), the follow-up visit schedule should be commenced after the last infusion.

Some assessments will only be performed at selected visits as indicated:

Clinical assessments:
Mandatory at each visit

- Vital signs (blood pressure, heart and respiratory), temperature and skin aspect (rash, coloration and sweating)
- Physical examination, including spleen and liver size (in cm from costal grill)

Laboratory:
Mandatory at each visit

- CBC with differential count
- Triglycerides (fasting)
- Coagulation tests: aPTT, PT, D-dimers and fibrinogen
- Biochemistry: glucose and electrolytes, ferritin, CRP, AST, ALT, γGT, ALP, LDH, bilirubin, albumin, creatinine and urea
- Urinalysis: glucose, blood, protein, leucocytes, ketones, pH and specific gravity

Search for infections: Mandatory every 2 weeks (unless required more frequently by the presence of a documented or suspected infection)	▪ Tuberculosis via PCR in any relevant specimen ▪ *Adenoviruses, EBV, CMV* by quantitative PCR
Imaging:	▪ Chest X-Ray (every 4 weeks) ▪ 3D abdominal ultrasound with measurements of spleen and liver size (every 2 weeks)

8.7.3 End of Treatment visit: three days after the last NI-0501 infusion

The end of treatment visit should always be carried out 3 days (±1 day) after last NI-0501 infusion.

This visit will include the following:

Clinical assessment:	▪ Vital signs (blood pressure, oxygen saturation, heart and respiratory rate) and temperature ▪ Physical examination, including spleen and liver size (measure in cm from costal grill) as well as height and weight
Procedure:	▪ ECG
Laboratory:	▪ CBC with differential count ▪ Triglycerides (fasting) ▪ Coagulation tests: aPTT, PT, D-dimers and fibrinogen ▪ Biochemistry: glucose and electrolytes, ferritin, CRP, AST, ALT, γGT, ALP, LDH, bilirubin, albumin, creatinine and urea ▪ Urinalysis: glucose, blood, protein, leucocytes, ketones, pH and specific gravity
Search for infections	▪ Tuberculosis via PCR
Imaging:	▪ Chest X-ray ▪ 3D abdominal ultrasound with measurements of spleen and liver size
Pharmacokinetics:	▪ NI-0501 serum concentration
Pharmacodynamics/ Exploratory parameters:	▪ sCD25, IL-10, CXCL-9, CXCL-10 and CXCL-11 ▪ Total IFNγ
Immunogenicity:	▪ Presence of anti-NI-0501 antibodies (ADA)

8.8 FOLLOW-UP VISITS (WEEKLY POST LAST NI-0501 INFUSION)

The follow-up period consists of weekly visits, to be performed approximately two (2) and three (3) weeks after the last NI-0501 infusion.

In case the patient starts conditioning during the 4-week follow-up, the closer weekly follow-up visit will be combined with the pre-conditioning visit scheduled at the site, so that clinical and laboratory parameters can be recorded before the administration of the conditioning drugs.

Combining the pre-transplant visit with the weekly follow-up visit should also be attempted if transplant takes place during the follow-up period.

Some assessments will only be performed at selected visits as indicated below:

Clinical assessments:	▪ Vital signs (blood pressure, oxygen saturation, heart and respiratory rate) and temperature ▪ Physical examination, including spleen and liver size (measure in cm from costal grill) as well as weight
Laboratory:	▪ CBC with differential count ▪ Triglycerides (fasting) ▪ Coagulation tests: aPTT, PT, D-dimers and fibrinogen ▪ Biochemistry: glucose and electrolytes, ferritin, CRP, AST, ALT, γGT, ALP, LDH, bilirubin, albumin, creatinine and urea ▪ Urinalysis: glucose, blood, protein, leucocytes, ketones, pH and specific gravity
Search for infection: Mandatory only on week 2 post last infusion	▪ Tuberculosis via PCR ▪ *Adenoviruses, EBV, CMV* by quantitative PCR
Imaging: Mandatory only at pre-conditioning visit	▪ 3D Abdominal ultrasound with measurements of spleen and liver size
Pharmacokinetics:	▪ NI-0501 serum concentration
Pharmacodynamics/ Exploratory parameters:	▪ sCD25, IL-10, CXCL-9, CXCL-10 and CXCL-11 ▪ Total IFNγ

8.9 STUDY COMPLETION VISIT (4TH WEEK AFTER THE LAST NI-0501 INFUSION) OR WITHDRAWAL VISIT

Patients will attend a final study completion visit, four weeks after the last NI-0501 administration. In case a patient enters the NI-0501-05 study while still receiving NI-0501, the study completion visit is replaced by the EoT visit under the NI-0501-04 study.

The same assessment procedures should also be followed by the Investigator for any patient who is withdrawn prematurely from the study as soon as possible after the decision to withdraw is made. For patients who withdraw from the study as a result of their own decision or the decision of their parent/guardian, the Investigator should contact the patient (or parent/guardian) and ask them to attend a withdrawal visit as soon as possible. Withdrawal (WD) visits should be scheduled within 30 days of termination whenever possible.

Patients who are withdrawn due to a serious adverse event (SAE) should be followed-up until the resolution of the event or until the outcome of the event is known and stable.

The following assessments will be performed at the Study Completion Visit or WD visit:

Clinical assessments:	▪ Vital signs (blood pressure, oxygen saturation, heart and respiratory rate) and temperature ▪ Physical examination, including spleen and liver size (measure in cm from costal grill) as well as height and weight
Procedure:	▪ ECG
Laboratory:	▪ CBC with differential count ▪ Triglycerides (fasting) ▪ Coagulation tests: aPTT, PT, D-dimers and fibrinogen ▪ Biochemistry: glucose and electrolytes, ferritin, CRP, AST, ALT, γGT, ALP, LDH, bilirubin, albumin, creatinine and urea ▪ Urinalysis: glucose, blood, protein, leucocytes, ketones, pH and specific gravity
Imaging:	▪ Chest X-ray
Search for infection:	▪ Tuberculosis via PCR*Adenoviruses, EBV, CMV* by quantitative PCR
Pharmacokinetics:	▪ NI-0501 serum concentration
Pharmacodynamics/ Exploratory parameters:	▪ sCD25, IL-10, CXCL-9, CXCL-10 and CXCL-11 ▪ Total IFNγ
Immunogenicity:	▪ Presence of anti-NI-0501 antibodies (ADA)

8.10 ASSESSMENTS IN CASE OF UNPLANNED (UNSCHEDULED) VISITS

Unplanned visits may occur should the patient need to be assessed or treated for any clinical condition that arises during the study. This may include the evaluation and follow-up of AEs, SAEs or laboratory tests. The following assessments (as detailed in the Schedule of Assessments) should always be performed *at minimum*, but additional assessments may be added according to the clinical judgment of the Investigator.

Clinical assessments:	▪ Vital signs (blood pressure, heart and respiratory rate and oxygen saturation) and temperature ▪ Physical examination, including spleen and liver size (in cm from costal grill) as well as weight

8.11 UNPLANNED ASSESSMENTS

Additional PK/PD samples may be required to better characterize the PK/PD profile and/or for safety reasons.

The number of additional samples taken will depend on the weight and health status of the patient. Sampling schedule will be proposed by the Sponsor and discussed with the site.

9 SAFETY MONITORING

9.1 STUDY SCIENTIFIC OVERSIGHT

A Scientific Steering Committee (SSC) composed of international experts in primary and secondary HLH has been involved in the preparation of study design and protocol writing.

This SSC has also been consulted for the composition of the DMC, the selection of its members and the elaboration of the DMC Charter (see Section 9.5.1).

The SSC will continue to play an advisory role throughout the course of the study and will perform evaluations of the results for the Sponsor as well as for the DMC. Please refer to Appendix D for full details of membership of the SSC.

The DMC, composed of relevant experts (2 pediatric onco-hematologists, 2 pediatric immunodeficiency/infectious disease specialists, a bio-statistician and a specialist in ethics), will oversee the study conduct and evaluate safety and relevant efficacy parameters. See Section 9.5.1 for further details of the DMC.

9.2 DESCRIPTION OF SAFETY PARAMETERS

Evaluation of NI-0501 tolerability and safety will be based on the following parameters:

- Adverse events (AEs), with special attention being paid to events potentially related to the infusion of NI-0501 (events occurring during the infusions and within 24 hours post infusion) and to the occurrence of infections
- Laboratory parameters:
  - Complete blood count (CBC),
  - Coagulation tests (activated partial thromboplastin, prothrombin time), d-Dimer and fibrinogen
  - Biochemistry: glucose and electrolytes, ferritin, C-reactive protein (CRP), aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), γGT, LDH, triglycerides, bilirubin, albumin, creatinine and urea.
  - Urinalysis
- Vital signs: temperature, heart and respiratory rate, blood pressure, oxygen saturation
- Physical examination with particular attention paid to:
  - weight evolution, occurrence of oedema or ascite
  - occurrence of skin rashes, jaundice, purpura, bleeding, edema
  - signs of infections (e.g tonsillitis, lymphadenopathies, cough and/or dyspnea),
  - neurological examination
  - liver and spleen size
- Immunogenicity: development of anti-NI-0501antibodies

9.3 RECORDING AND REPORTING SAFETY PARAMETERS

9.3.1 Adverse events

Adverse events (AEs) are defined as any undesirable experience occurring to a subject during the study, whether or not considered related to the investigational product. All adverse events (AEs) reported spontaneously by the patients or his/her relatives or observed by the Investigator or his staff during the

clinical study from the signature of the ICF up to and including the end-of-study visit will be reported on the AE data collection form.

Medical conditions present at screening should be recorded in the medical history section of the eCRF.

An AE which occurs between start of screening visit and start of first IMP administration will be considered as a pre-treatment AE.

Any AE that occurs after the start of the first IMP administration will be considered as a Treatment Emergent Adverse Event (TEAE).

However, if a pre-existing medical condition recorded in the medical history worsens (clinically significant change in intensity or frequency), it must be recorded as an AE in the eCRF and, depending on the time of its occurrence, will be considered as a pre-treatment AE or a TEAE. If a medical condition recorded as a pre-treatment AE worsens post IMP administration, it will be recorded in the eCRF as a separate TEAE.

For all AEs, the following will be assessed and recorded: intensity, relationship to IMP, action taken regarding IMP, any treatment received and outcome to date.

Intensity of adverse events will be graded on a three-points scale (mild, moderate, severe) using the modified WHO (World Health Organization) toxicity scale (Grade 3 and 4 are considered to be the severe grade). If AE severity is not mentioned in the scale, assessment will be made using the following definitions:

- Mild: Discomfort noticed but no disruption of normal activity
- Moderate: Discomfort sufficient to reduce or affect normal daily activity
- Severe: Inability to work or perform normal daily activity.

For a given AE, the assessment of its intensity should reflect the highest grade (on the 3 points scale mentioned above) reported during its course (except when the intensity of a pre-treatment AE increases after treatment initiation, as indicated above).

AEs characterized as intermittent require documentation of onset and duration of each episode.

The relationship of adverse events to the Investigational Medicinal Product (IMP) will be assessed by the Investigator using a “Yes/No” classification. A “Yes” relationship infers that there is a reasonable suspected causal relationship to the trial medication. The expression “reasonable causal relationship” is meant to convey that there are facts, evidence or arguments to suggest a causal relationship. In this study NI-0501 is the only IMP.

9.3.2 Serious Adverse Events

A serious adverse event (SAE) is any untoward medical occurrence that:

- results in death (note: death is an outcome, not an event);
- is life-threatening; (note: the term "life-threatening" refers to an event in which the patient was at immediate risk of death at the time of the event; it does not refer to an event which could hypothetically have caused a death had it been more severe);
- requires in-patient hospitalization or prolongs an existing hospitalization;
- results in persistent or significant disability/incapacity;
- is a congenital anomaly/birth defect;
- is medically significant or requires intervention to prevent one or other of the outcomes listed above (note: examples are intensive treatment in an emergency room or at home for allergic bronchospasm; blood dyscrasias or convulsions that do not result in hospitalization).

SAEs are to be reported to NovImmune immediately.

The term severe is a measure of intensity/severity: thus a severe adverse event is not necessarily serious. For example, nausea of several hours' duration may be rated as severe, but may not be clinically serious.

For the purposes of this study, the following will not be considered as serious adverse events:

- Elective hospitalizations or surgical procedures that are a result of a patient's pre-existing condition(s) which have not worsened since receiving IMP. Such events should still be recorded as adverse events in the eCRF;
- Hospitalization as requested per protocol for NI-0501infusion and study visits.

Any serious clinical adverse event or clinically significant abnormal laboratory test value that occurs during the course of the study, irrespective of the treatment received by the subject, must be communicated by the Investigator to NovImmune, by fax or electronic transmission, within 24 hours of awareness.

For the initial SAE report, the Investigator should report all available case details concerning the patient and the event, using the NovImmune SAE reporting standard form (see Appendix F).

NovImmune contact information for SAE reporting:

Relevant follow-up information on SAEs should be forwarded to NovImmune as soon as it becomes available. In addition, the Investigator must be available to answer without delay any request for follow-up information or questions NovImmune may have regarding the SAE.

All SAEs will be recorded on the appropriate page of the eCRF. They will be reviewed, evaluated and followed through to resolution by a study physician-

Relationship to the study drug will be established by the Investigator in order to define the need for expedited reporting.

9.3.3 SUSAR reporting

Unexpected adverse reactions are all untoward and unintended responses to an investigational product related to any dose administered

These reactions are SUSARs if the following two conditions are met:
1) the event must be serious (see section 9.3.2);
2) there must be a certain degree of probability that the event is a harmful and an undesirable reaction to the medicinal product under investigation, regardless of the administered dose;

NovImmune will report directly or through delegation to a third party all SUSARs to the investigators and the relevant Ethics Committees, in writing as soon as practical, but at least within 15 days. Fatal and life-threatening suspected SUSARs will be reported within 7 calendar days, with another 8 days for completion of the report.

NovImmune will report all SUSARs to the EMA's EudraVigilance database within 15 days, as well as to the relevant National Competent Authorities when required. Fatal and life-threatening suspected SUSARs will be reported within 7 calendar days, with another 8 days for completion of the report.

9.3.4 Managing Abnormal Laboratory Test Values

All safety laboratory tests (haematology and blood biochemistry), for each visit time-point, should be captured in the database from the local laboratory and should not be reported as AEs unless specific treatment is given for the abnormality (e.g. a blood transfusion is given for a low haemoglobin) or a laboratory abnormality leads to further investigation and the diagnosis of a new clinical event (e.g. a high white cell count is found to be due to incidental leukaemia). In this event the clinical diagnosis should be reported on the AE form, not the laboratory abnormality leading to the diagnosis.

9.4 FOLLOW-UP OF SAFETY PARAMETERS

9.4.1 Treatment and Follow-up of Adverse Events

Adverse events, especially those for which the relationship to the study drug has been assessed as 'Yes', should be followed-up until the event has returned to baseline status or has stabilised. If a clear explanation is established, it should be recorded on the CRF.

All SAEs must be followed-up until the event has either resolved or reached a stable clinical outcome.

9.4.2 Follow-up of Abnormal Laboratory Test Values

In the event of unexplained clinically relevant abnormal laboratory test values, the tests should be repeated immediately and followed-up until the values have returned to within normal range and/or an adequate explanation of the abnormality is found. If a clear explanation is established, it should be recorded on the eCRF.

9.4.3 Pregnancy

In the event that a pregnancy occurs during the trial course, it must be reported to NovImmune within 24 hours of awareness. This includes pregnancies occurring in partners of male enrolled patients. All information pertaining to the pregnancy should be reported using the NovImmune Pregnancy form (see Appendix F). Pregnancies should be followed until conclusion to obtain outcome information.

Occurrence of a pregnancy in a study participant will preclude any further IMP administration and the patient will be withdrawn.

On withdrawal the assessments presented in the Schedule of Assessments are to be performed (see Section 8.9).

9.5 BENEFIT/RISK MANAGEMENT

9.5.1 Safety Surveillance Management

The main responsibility of the DMC is to review all safety and efficacy data as they are generated to ensure that no patient is exposed to unnecessary risk and to continuously assess the benefit/risk profile of NI-0501.

The DMC can recommend treatment discontinuation for individual patients as well as to halt the entire study temporarily or permanently. Predefined stopping rules will guide the DMC review process. For more details, see stopping rules in Section 10.

9.5.2 General Benefit/Risk Considerations

9.5.2.1 Potential benefits

On the basis of the data available to date[28], NI-0501 administration has shown the potential to improve or resolve relevant clinical and laboratory abnormalities of HLH, including CNS signs and symptoms when present, allowing most of the patients to proceed to HSCT (9 out of 15 patients who have completed the

study with 3 additional patient ready for HSCT pending donor availability). Three patients underwent HSCT only after 4 weeks of NI-0501 treatment. The response to NI-0501 seems independent of:
- the presence and type of causative mutations
- the presence and type of an infectious trigger
- the line of treatment, although data obtained in patients in first line are still limited.

For more details refer to the latest Investigator's Brochure (presently version 5.0, dated 20 January 2016).

9.5.2.2 Risks analysis

- Risks related to NI-0501

NI-0501 is a monoclonal antibody of IgG1.
Upon the administration of mAbs, which are proteins, acute infusion reactions can occur. These may happen during the infusion or in the subsequent hours (usually within the first 24 hours)[29].

These reactions are either IgE-mediated type I hypersensitivity reactions (anaphylactic reactions), or anaphylactoid reactions not mediated by IgE. True anaphylactic reactions usually do not occur upon initial infusion and require a certain sensitization. In contrast, the pathophysiology of anaphylactoid reactions appears to be secondary to the release of cytokines consequent to a mAb binding to circulating antigen-expressing cells. However, the clinical manifestations of anaphylactic and anaphylactoid reactions overlap, and both may lead to life-threatening conditions, involving cardiovascular, respiratory, central nervous, gastro-intestinal, and cutaneous systems. The management of anaphylactic and anaphylactoid reactions involves immediate administration of oxygen, epinephrine, vasopressors, bronchodilators, corticosteroids, and/or antihistamines.

After 14 single infusions to HVs up to and including the dose of 3 mg/kg and more than 380 infusions administered to HLH patients (either in the context of the NI-0501-04 and NI-0501-05 studies or in patients who received NI-0501 in compassionate use) up to and including the dose of 10 mg/kg, no significant infusion related reaction has been observed. Only transient erythematous rashes localized to the extremities (feet and/or hands) resolving spontaneously have been observed in a few patients during the first infusions of NI-0501 in the NI-0501 study. On a few occasions, administration of NI-0501 has been performed through a peripheral venous access and all infusions were uneventful.

- When administered to humans, most mAb therapeutics elicit some level of antibody response (anti-drug antibodies or ADAs) against the therapeutic product, as early as after the first exposure. No sign of immunogenicity has been reported in the NI-0501 study in healthy volunteers. The presence of ADAs will be measured throughout this study as per regulatory recommendations. The full analysis is planned to be performed at the end of the NI-0501-04 study. Data accumulated so far (in particular PK profiles and a negative ADA search performed in the only patient who developed hemolytic anemia during conditioning) have not led to suspect the presence of ADA. Risks related to the target

The impact on the immune defense caused by the neutralization of IFNγ is known from patients with inborn errors of the IL-12/23-IFN-γ circuit, particularly patients with complete or partial IFNγ receptor (R) deficiency, and subjects developing neutralizing auto anti-IFNγ antibodies.

Patients with IFNγ R deficiency are prone to developing mycobacterial infections and, although to a lesser extent, *Salmonella* infections[30,31].
The mean age of the first environmental mycobacterial infection is 3.1 and 13.4 years in patients with complete and partial deficiency, respectively[32]. No systematic prophylaxis has been recommended in these patients.

If an infection occurs, appropriate antibiotherapy based on sensitivity of isolated species is prescribed. Individuals with anti-IFNγ auto-antibodies are also susceptible to develop mycobacterial infections (for the vast majority atypical mycobacterial infections), but also opportunistic infections (e.g. by *Histoplasma Capsulatum, Salmonella*, *Herpes Zoster* virus infections)[6].
Toxicological studies carried out with NI-0501 have shown an increased susceptibility of the monkeys having received NI-0501 to enteral pathogen infections when the pathogen is present into the intestinal tract prior to NI-0501 administration. Presence of infections due to *Shigella, Salmonella* and *Campylobacter* is part of the exclusion criteria.

NI-0501 has been administered to 14 healthy volunteers in a single ascending dose study, which confirmed the absence of off-target toxicity. However a reactivation of herpes zoster virus, at a dose of 3 mg/kg, observed in one healthy volunteer, may have been due to the pharmacodynamics effect of the drug (see clinical information section 1.1.3). It is difficult to draw a firm conclusion from one case, but it may be prudent, in the context of this pilot study, to include herpes zoster virus (HZV) prophylaxis for all patients.

Preliminary data on infections reported in 17 patients enrolled in the NI-0501-04 study allow the following conclusions to be drawn:

- Active infections, in particular EBV or CMV infections, which are often the trigger of the HLH initial episode or reactivation/worsening, were present at the first administration of NI-0501. During NI-0501 administration these infections resolved with appropriate antimicrobial treatment and while achieving control of HLH.

- Patients developed infections during the course of NI-0501 treatment. However in the presence of a satisfactory control of HLH, and upon appropriate anti-microbial treatment, infections resolved.

- None of the bacterial, viral or fungal infections reported were due to pathogens known to be favored by IFNγ neutralization.

- The severity and duration of neutropenia, a hallmark of HLH as well as a potential consequence of previous HLH treatments, seemed to contribute significantly to the development of infections.
A long and profound generalized immune suppression caused by HLH treatments administered prior to the initiation of NI-0501 constitutes a higher risk for infection development.

- The patients who have received NI-0501 treatment as first line HLH therapy, no infection was reported during the NI-0501-04 study.

- Systematic search for tuberculosis was negative and no atypical mycobacteria were detected in any of the patients. Stool/blood cultures were negative for *Salmonella*, *Shigella* or *Campylobacter* in all patients.

For more details refer to the latest Investigator's Brochure (presently version 5.0, dated 20 January 2016).

- Risk related to the study population

Most of the patients are expected to be children of a young age affected by a life-threatening disease and are expected to have already received HLH treatments; therefore they carry variable degree of toxicities caused by those treatments. The data collected to date show that administration of NI-0501 does not aggravate these toxicities while showing, in general, a favorable impact on disease activity.

All information collected with regards to disease stage and previous treatments will be taken into account for the analysis of adverse events.

Toxicities of concomitant treatments, authorized or recommended during the administration of NI-0501, may also potentially expose the patients to adverse events; however their benefits may outweigh their risks. No safety concern related to the concomitant administration of NI-0501 with other treatments (e.g.,

antimicrobial agents, anti-hypertensive drugs) has been reported so far. Corticosteroids have already been administered with IFNγ therapy in Crohn's Disease without any particular safety concerns[4]. Of interest, tapering of glucocorticoids had no impact on safety and tolerability of NI-0501 infusions and has shown benefit for patients with steroids-related hypertension and generalized immunosuppression.

Treatment-naïve patients are not exposed to risks associated with the drugs used conventionally to treat HLH (e.g., generalized immune suppression).

As the risk that the NI-0501 treatment will not be able to control the disease may exist, thepossibility to receive additional HLH therapy to NI-0501 or to receive rescue therapy upon discontinuation of NI-0501 is foreseen. Risks associated with the administration of other therapies after having received NI-0501 seems to be very low, since no particular safety concerns related to the administration of NI-0501 were observed in patients receiving NI-0501 as second line therapy. Furthermore, two primary HLH patients have received other HLH therapies (e.g., etoposide and alemtuzumab) after NI-0501 discontinuation with no particular safety concern. For one of them, NI-0501 was re-initiated in Compassionate Use as the patient continued to worsen after the reintroduction of etoposide and the independent DMC judged the benefit risk profile of NI-0501 positive for this particular patient. Both these patients underwent HSCT.

9.5.2.3 Risk minimization measures

In view of the expected benefits the above listed risks are considered to be manageable in this patient population, if adequate minimization measures are put in place. An overview of specific measures to minimize the subject's risk is provided below:

- Study designed with HLH experts forming the SSC
- Patients are hospitalized in specialized centers for the treatment of HLH, and therefore with all necessary emergency assistance equipment
- Inclusion/exclusion criteria: patients with malformations or severely altered functions (either due to the disease stage or to a concomitant disease), as well as patients with evidence of patent or latent TB infections or active mycobacteria*, Shigella, Salmonella, Campylobacter* or *Leishmania* infections, will not be included in the study (for details see Section 4.1)
- Infusion Related Reaction (IRR) monitoring: patients will be very closely monitored during the study drug infusions and for 24 hours following them to immediately identify if the subject is experiencing any IRRs. Each of the specialized centers will have physicians adequately trained in IRR management (please see NI-0501-04 Study Specific Risk Management Plan for further details).
- Recommendations on prophylaxis for *Pneumocystis jiroveci*, fungal infections, Herpes Zoster virus for all patients and Tuberculosis for a defined subpopulation (see section 6.2) in the protocol to avoid occurrence of these infections
- Close monitoring of potential infections through careful physical examination, laboratory parameters, active search for EBV, CMV, Adenoviruses, detection of tuberculosis
- Study safety surveillance by a Data Monitoring Committee

The Development Risk Management Plan addresses risks, identify signals for early detection of safety concerns and propose mitigating actions. It will be part of the study documentation shared with Investigators and any relevant third party involved in the study.

Stopping rules have been also developed to ensure individual patient safety and determine whether the study should be put on hold or terminated prematurely.

10 STOPPING RULES

10.1 AT PATIENT LEVEL

10.1.1 Decision to slow down or stop NI-0501 infusion due to systemic reaction

During the infusion of NI-0501, any significant change compared to pre-infusion values in vital signs, such as those listed below, should trigger appropriate immediate care:

- Sudden and sustained increase or paradoxical decrease of heart rate (duration of more than 1 minute) compared to pre-infusion value
- Respiratory rate abnormality (significant change compared to measure prior to infusion)
- Blood pressure drop or peak compared to the value prior to infusion for at least 2 consecutive measurements
- Significant skin modifications (change in color or abundant sweating)
- Sustained (an episode of more than 3 minutes duration or more than 3 episodes of shorter duration i.e. 1 minute) oxygen desaturation (below 90%)

The decision to slow down the infusion will be taken by the physician in the event of any of the above mentioned occurrences.

The decision to stop the infusion will be based on the evolution of patient status after appropriate symptomatic measures, e.g. oxygenation, and upon physician's own medical judgment.

All changes in infusion rate will be recorded in the eCRF: each time with a rate modification as well as end of the premature or delayed termination of the infusion.

10.1.2 Local reaction to NI-0501 infusion

Unless related to a hypersensitivity reaction, a local infusion issue such as catheter displacement, obstruction or product extravasation, will trigger the infusion of the remaining quantity through a new venous access as soon as possible. All information related to the incident will be recorded accurately. This includes reasons, volume of IMP potentially lost (in order to calculate the quantity of drug infused), time at which the infusion stopped, time at which the infusion was resumed and time of end of the infusion.

To avoid this type of incident, it is preferable for a central venous access to be used: this will improve patient's comfort and ensure a reliable drug administration in particular in infants and toddlers or in case of foreseen difficulties with peripheral venous access.

10.1.3 Decision to Discontinue Treatment

An Investigator can decide at any time during the study to discontinue the treatment for an individual patient based on his/her own medical judgment, taking into account the individual benefit risk ratio for his/her patient. In addition, the patient (or their legal representative) can decide at any time to withdraw from the study.

In any case the decision to withdraw or be withdrawn will have no impact on the patient's care and further treatments administered to him/her after withdrawal. The management of these patients is described in Section 10.3.

Nevertheless some situations should trigger an immediate decision to permanently discontinue treatment:

10.1.3.1 Treatment Discontinuation for a Safety Reason

A patient should be discontinued from study treatment if a SAE occurring after NI-0501 administration is:

1. Considered by the Investigator to be related to NI-0501 (with guidance from the DMC if needed) **AND**
2. is a life-threatening event.

All other AEs will be judged by the DMC on a case-by-case basis taking into account the disease evolution (such as signs of improvement in HLH) and the possibility of managing the AE and ensuring that no patient is exposed to unnecessary risks.

10.1.3.2 Treatment Discontinuation for Lack of Efficacy

A patient should be withdrawn from the NI-0501-04 study if, after having added an additional therapy concomitantly to NI-0501, no response or lack of improvement is observed.

However, at the request of the investigator, the DMC can agree to maintain the patient in the study if, after having determined a favorable benefit/risk for NI-0501 in this patient through a thorough review of patient's data, it is considered that loss of neutralization of IFNγ could expose patient to the risk of HLH worsening. The DMC may propose continuing the administration of NI-0501 under certain conditions which have to be accepted and implemented by the Investigator at the site.

10.2 AT STUDY LEVEL

10.2.1 Recruitment Suspension

Recruitment may be suspended in the following situations:

- Any occurrence of death or life-threatening SAE related to the drug
- At the DMC's own request as an outcome of their regular study review

Patients already enrolled in the study should continue receiving NI-0501 as per protocol unless decided otherwise by the Investigator.

The suspension will allow the DMC to analyse the data already generated and consider a recommendation.

After re-evaluation of benefit/risk profile, the DMC may recommend any of the following:

- To resume recruitment without any change
- To implement minimisation measures that may require protocol amendment
- To implement conditions for study termination: e.g. next occurrence of a particular serious drug reaction

10.2.2 Study Termination

Occurrence of two deaths suggesting a reasonably possible relationship with continuous exposure to NI-0501 and occurring in similar conditions will trigger the decision to terminate the study.

This process will involve both the DMC and the Investigators. The management of patients already enrolled in the study will also be part of the DMC recommendations.

10.3 MANAGEMENT OF TREATMENT DISCONTINUATION

All patients who are withdrawn from the study will be treated, according to the standard care at the site. They should be assessed as indicated in Section 8.9.

11 STATISTICAL CONSIDERATIONS AND ANALYTICAL PLAN

Full details of all statistical issues and planned statistical analyses will be specified in a separate Statistical Analysis Plan (SAP), which will be finalised prior to the locking of the study database. This section contains an overview of the planned methods of analysis.

11.1 SAMPLE SIZE

Sample size estimation has been done for the pivotal cohort of the study, i.e. patients receiving NI-0501 in second line.

A minimum of 28 patients treated with NI-0501 in second line will be enrolled in the study. Sample size calculation is based on the primary efficacy endpoint of "Overall Response Rate". Assuming an Overall Response Rate of 70%, the study will have 90% power to show a significant improvement above 40% using an exact binomial test[33] at a one-sided significance level of 2.5%.

Due to the rarity of primary HLH, the recruitment is competitive across all EU and US sites in order to gather data in a reasonable timeframe.

11.2 ANALYSIS SETS

All analysis sets will be defined prior to final database closure. In addition to the analysis sets listed below, further exploratory analyses may be performed using other subgroups of patients.

11.2.1 Safety Analysis Set

The safety analysis set will include all patients who receive any part of an infusion of study drug.

11.2.2 Intent-to-Treat Analysis Set

The intent-to-treat (ITT) analysis set will include all patients who receive any part of an infusion of study drug.

11.2.3 Per-Protocol Analysis Set

The per-protocol analysis set will consist of all ITT patients who complete the study without violations of the study protocol. Details of the analysis will be defined in the SAP prior to locking of the final database.

11.3 STATISTICAL AND ANALYTICAL METHODS

For measurements of continuous endpoints, summary statistics will include n, mean, median, standard deviation, minimum and maximum values. For binary data (proportions of patients showing a defined response for example) numbers and percentages will be tabulated. For time to event data, Kaplan-Meier plots will be provided together with the median should this be available. Finally 95% confidence intervals will be calculated for suitable summary statistics associated with endpoints of interest.

All efficacy analyses will be undertaken on both the ITT and PP analysis sets although the primary efficacy analyses will focus on the subgroup of second line patients. All safety analyses will be conducted on the safety set.

11.3.1 Efficacy Data

The analysis of the primary endpoint, Overall Response Rate will utilize an exact binomial test to evaluate the null hypothesis that the response rate is at most 40%. This test will be undertaken at the one-sided 0.025 level.

Time to Response, Durability of Response, and Survival time will be presented using Kaplan-Meier curves with medians calculated if available. Ninety-five percent confidence intervals will be calculated for the median for each of these endpoints.
For maintenance of response achieved any time during study until EoT and beyond (including data collected in the long-term follow-up study NI-0501-05) different follow-up periods will be considered, firstly censoring at *i*) EoT and *ii*) the day prior to starting conditioning; and secondly by taking the complete follow-up period beyond HSCT.

Additional endpoints based on binary outcomes including number of patients who reduce glucocorticoids by 50% or more, and number of patients able to proceed to HSCT will be converted to proportions and associated 95% confidence intervals calculated.

Statistical significance in terms of p-values will only be obtained for the primary endpoint in both the ITT and PP analysis sets. All other endpoints will be viewed as supportive for the primary endpoint and as a consequence no formal hierarchy of endpoints will be declared.

11.3.2 Safety Data

All data relating to safety will be listed and summarised using descriptive statistics.

AEs will be coded and tabulated by body system, and by individual events within each body system. AEs will also be tabulated by severity and relationship to the study medication. Summaries will also be produced of SAEs, and AEs leading to withdrawal from the study.

For each clinical laboratory test, individual patient values will be listed and summarised and change from pre-treatment baseline values calculated and summarised. Any values outside the standard reference range will be flagged. Summaries of marked abnormalities and shift tables will be tabulated for each laboratory test.

In addition, other exploratory analyses of safety data, including summaries for different subsets of patients, may be conducted.

11.3.3 Pharmacodynamic Data

All PD data will be summarised using appropriate graphical and tabular presentations.

Exploratory statistical models will be fitted, and correlation analyses undertaken, to investigate the relationships between PD data and other biomarkers and the clinical measures of response. ROC curves may be used to summarise any relationships that are found.

In addition, other exploratory analyses of pharmacodynamic endpoints, including summaries for different subsets of patients, may be conducted.

11.3.4 Immunogenicity Data

The numbers of patients with anti-drug antibodies present at each assessment point will be summarised.

11.3.5 Missing Data

No imputations of missing data will be performed. However, the following rules will be applied to ensure that all patients can be included in the final analysis:

- Patients who are withdrawn from the study prior to Week 8 because of safety concerns or poor efficacy will be classified as non-responders from the time of their withdrawal in all analyses of response status, and their data will be censored at time of withdrawal in all time-to-event analyses. For continuous endpoints in such patients, all analyses for time points beyond the point of withdrawal will exclude missing data for these patients.

- Patients who do not reach Week 8 because of early transplant will be classified as responders beyond their time of withdrawal in all analyses of response status, and their data will be censored at time of withdrawal.

11.4 REPLACEMENT POLICY

11.4.1 For Patients

Any patient withdrawn from the study for reasons other than safety or efficacy concerns will be replaced.

11.4.2 For Centres

A centre may be replaced for the following administrative reasons: excessively slow recruitment, poor protocol adherence.

PART II

12 ETHICAL AND LEGAL ASPECTS

12.1 GOOD CLINICAL PRACTICE

The procedures set out in this study protocol pertaining to the conduct, evaluation, and documentation of this study are designed to ensure that NovImmune, its authorised representative, and Investigator abide by Good Clinical Practice (GCP), as described in International Conference on Harmonization (ICH) Guideline E6 and in accordance with the general ethical principles outlined in the Declaration of Helsinki. The study will receive approval from an Independent Ethics Committee (IEC) prior to commencement and where applicable by law also from National Competent Authorities. The Investigator will conduct all aspects of this study in accordance with applicable national, state, and local laws of the pertinent regulatory authorities.

12.2 RESPONSIBILITIES

Investigator

The Investigator should ensure that all persons assisting with the trial are appropriately qualified and adequately informed about the protocol, amendments, study treatments, as well as study-related duties and functions. The Investigator should maintain a list of sub-investigators and other appropriately qualified persons to whom he or she has delegated significant study-related duties.

The Investigator is responsible for keeping a record of all patients (or their legally authorised representative) who sign an informed consent document and are screened for entry into the study. Patients who fail screening must have the reason(s) recorded in their source documents and the study-screening log.

The Investigator, or a designated member of the Investigators' staff, must be available during monitoring visits, audits and inspections to review data, resolve queries and allow direct access to subjects' records (e.g. medical/hospital records, office charts, hospital charts, and study related charts) for source data and other type of verification. The Investigator must ensure timely and accurate completion of CRFs and queries.

12.3 CONSENT

Before being admitted to the clinical study, the patient must consent to participate after the nature, scope, and possible consequences of the clinical study have been explained in a form understandable to him or her. An informed consent document that includes both information about the study and the consent form will be prepared and given to the patient. This document will contain all ICH, GCP, and locally required regulatory elements (whichever is more stringent). The document must be in a language understandable to the patient and must specify who informed the patient, and when the informed consent was obtained.

Information to patients will be split into a Patient Information Sheet that provides detailed information about the trial and its benefits and risks, and the Informed Consent Form that summarises the content of the Patient Information Sheet and is used to obtain the dated signature from the patient as evidence of the patient's agreement to partake in the study.

If applicable, since minors are involved in the trial, assent must be obtained from the minor and informed consent from at least one of the parents or as mandated by local rules (individual or judicial or other body authorised under applicable law to consent on behalf of a prospective patient to the patient's participation in the procedures involved in the research). The language used in the Assent Form is adapted to the

maturity level of the minor involved in the trial. Since minors of different age groups are likely to be entered into the trial different versions of the Assent Form will be provided. The modalities for obtaining informed consent from the parents and Assent from the minor will be defined at the site initiation visit and documented in the clinical trial centre Trial Master File (TMF).

After reading and understanding the informed consent document, the patient (or their legally authorised representative) must give consent in writing. The written informed consent will be obtained prior to conducting any study-related procedures or tests. The patient's consent (or the consent of the patient's legally authorised representative) must be confirmed at the time of consent by the personally dated signature of the person conducting the informed consent discussions. A copy of the signed consent document must be given to the patient or their legally authorised representative. The Investigator will retain the original signed consent document. The Investigator will not undertake any measures specifically required only for the clinical study until valid consent has been obtained.

If an amended protocol impacts the content of the informed consent document, the consent document must be revised. Patients already participating in the study when the amended protocol is implemented must be re-consented with the revised version of the informed consent document. A copy of the revised informed consent document must be given to the patient or their legally authorised representative. The Investigator will retain the original signed updated consent document in the study files.

12.4 CONFIDENTIALITY AND DATA PRIVACY

NovImmune affirms the patient's right to protection against invasion of privacy and to be in compliance with ICH and other local regulations (whichever is more stringent). NovImmune requires the Investigator to permit NovImmune representatives and when necessary, representatives from regulatory authorities, to review and/or copy any medical records relevant to the study in accordance with local laws (any copies of patients' records must be duly anonymised to protect patients' confidentiality).

Should direct access to medical records require a waiver or authorisation separate from the patient's statement of informed consent, it is the responsibility of the Investigator to obtain such permission in writing from the appropriate individual.

12.5 PROTOCOL AMENDMENTS

Substantial amendments will be submitted to the IEC for written approval and where applicable to National Competent Authorities. Written approval must be obtained before implementation of the amended version occurs. The written signed approval from the IEC should specifically reference the Principal Investigator's name, protocol number, study title and amendment number(s) that is/are applicable.

12.6 APPROVAL OF THE CLINICAL STUDY PROTOCOL AND AMENDMENTS

Before the start of the study, the study protocol, informed consent document, and any other appropriate documents will be submitted to the IEC with a cover letter or a form listing the documents submitted, their dates of issue, and the site (or region or area of jurisdiction, as applicable) for which approval is sought. If applicable, the documents will also be submitted to the authorities in accordance with local legal requirements.

NovImmune can only supply study drug to an Investigator after NovImmune or their authorised representative, an international CRO, has received documentation on all ethical and legal requirements for starting the study. This documentation must also include a list of the members of the IEC and their occupation and qualifications. If the IEC will not disclose the names of the committee members, it should be asked to issue a statement confirming that the composition of the committee is in accordance with GCP. Formal approval by the IEC should preferably mention the study title, study code, study site, and

any other documents reviewed. It must mention the date on which the decision was made and must be officially signed by a committee member (chairman or secretary of the IEC. Before the first patient is enrolled in the study, all ethical and legal requirements must be met.

The IEC and, if applicable, the authorities, must be informed of all subsequent protocol amendments in accordance with local legal requirements. Amendments must be evaluated to determine whether formal approval must be sought and whether the informed consent document should also be revised.

The Investigator must keep a record of all communication with the IEC and, if applicable, between a coordinating Investigator and the IEC. This statement also applies to any communication between the Investigator (or coordinating Investigator, if applicable) and regulatory authorities.

All documents handed over to patients or their legal representative prior to use must first be reviewed and approved by NovImmune, and upon approval by NovImmune submitted to and reviewed and approved by, the competent IEC. This includes but is not limited to the informed consent form, patient information sheet, assent form, advertisements, training materials, etc.

12.7 ONGOING INFORMATION FOR INDEPENDENT ETHICS COMMITTEE

If required by legislation or the IEC, the investigator must submit to the IEC:

- Information on SAEs or SUSARs as per local applicable rules and timelines;
- Periodic reports on the progress of the study;
- Deviations from the protocol or anything that may involve added risk to subjects.

12.8 CLOSURE OF THE STUDY

NovImmune reserves the right to terminate this study at any time for reasonable medical or administrative reasons. Any premature discontinuation will be appropriately documented according to local requirements (e.g., IRB/IEC, regulatory authorities).

In addition, the Investigator or NovImmune has the right to discontinue a single site at any time during the study for medical or administrative reasons such as:

- Significant non-compliance with contractual enrolment timelines and targets
- Serious or continued GCP non-compliance
- Inaccurate, incomplete or delayed data collection
- Failure to adhere to the study protocol
- Failure to provide requested follow-up information for data queries

12.9 RECORD RETENTION

The investigator will ensure that essential records are kept in a secure archiving facility for the retention period stipulated in the study contract. Essential documents include, but are not limited to, the following:

- Signed informed consent documents for all subjects
- Subject identification code list, screening log (if applicable), and enrolment log
- Record of all communications between the investigator and the IEC
- Composition of the IEC
- Record of all communications between the investigator, NovImmune and their authorised representative
- List of sub-investigators and other appropriately qualified persons to whom the investigator has delegated significant trial-related duties, together with their roles in the study, curricula vitae and their signatures
- Copies of CRFs and of documentation of corrections for all subjects

- "Drug accountability" records
- Record of any body fluids or tissue samples retained
- All other source documents (subject records, hospital records, laboratory records, etc.)
- All other documents as listed in Section 8 of the ICH consolidated guideline on GCP (Essential Documents for the Conduct of a Clinical Trial)

Normally, these records will be held in the Investigator's archives. If the Investigator is unable to meet this obligation, the Investigator must ask NovImmune for permission to make alternative arrangements. Details of these arrangements should be documented in the clinical trial centre TMF.

12.10 LIABILITY AND INSURANCE

Liability and insurance provisions for this study are provided in the investigator contract.

12.11 FINANCIAL DISCLOSURE

Investigators are required to provide financial disclosure information to allow NovImmune to submit complete and accurate certification or disclosure statements in accordance with applicable national and local regulations. In addition, investigators must provide NovImmune with a commitment to promptly update this information if any relevant changes occur during the course of the investigation and for 1 year following the completion of the study.

12.12 DISCLOSURE OF PROTOCOL AND STUDY RESULTS AND PUBLICATION POLICY

Information about this trial will be posted following the principles of the International Committee of Medical Journal Editors (ICMJE), the International Federation of Pharmaceutical Manufacturers & Associations (IFPMA) Industry Position Paper and applicable national or regional regulations and laws.

The results of this study may be published or presented at scientific meetings. If this is foreseen, the Investigator agrees to submit all manuscripts or abstracts to NovImmune prior to submission. This allows NovImmune to protect proprietary information and to provide comments based on information from other studies that may not yet be available to the investigator.

NovImmune will comply with the requirements for publication of study results. In accordance with standard editorial and ethical practice, the NovImmune will support publication of multicentre trials only in their entirety and not as individual centre data. In this case, a coordinating investigator will be designated by mutual agreement prior to the start of the trial.

Authorship will be determined by mutual agreement and in line with ICMJE authorship requirements. Any formal publication of the study in which contribution of NovImmune personnel exceeded that of conventional monitoring will be considered as a joint publication by the investigator and the appropriate NovImmune personnel.

So-called 'ghost writing' is not permitted. All contributors who do not meet the criteria for authorship should be listed in an acknowledgments section. Examples of those who might be acknowledged include a person who provided purely technical help, writing assistance, or a department chairperson who provided only general support.

Any inventions and resulting patents, improvements, and/or know-how originating from the use of data from this study will become and remain the exclusive and unburdened property of NovImmune, except where agreed otherwise.

13 MONITORING AND AUDITING

All aspects of the study will be monitored by NovImmune or its representative for this study (NovImmune authorised representative), for compliance with applicable government regulations with respect to current GCP and standard operating procedures. Direct access to the on-site study documentation and medical records must be ensured.

13.1 STUDY MONITORING AND SOURCE DATA VERIFICATION

As part of the responsibilities commensurate with participating in the study, the investigator agrees to maintain and have available for monitoring, adequate case records (accurate source documents and CRFs) for the patients treated under this protocol. In addition, the investigator agrees to maintain all administrative documents (e.g. IEC correspondence, investigational product and supplies shipment manifests, monitoring logs, or correspondence with NovImmune and with any of its representative for this study).

13.2 ON-SITE AUDITS

Investigators and institutions involved in the study will permit trial-related monitoring, audits, IEC review, and domestic or foreign regulatory inspection(s) by providing direct access to source documents, CRFs, and all other study documentation.

The Investigator should promptly notify NovImmune of any inspections scheduled by any regulatory authorities and promptly forward to NovImmune copies of any audit reports received.

13.3 SERIOUS GCP BREACHES

NovImmune is required to report a serious GCP Breach within 7 days to applicable health authorities. Therefore, should an Investigator become aware of a possible serious GCP breach, e.g. a protocol violation, or non-reporting of critical safety information that has the potential of jeopardising patients' safety, NovImmune must be notified within 24 hours.

14 DOCUMENTATION AND USE OF STUDY FINDINGS

14.1 DOCUMENTATION OF STUDY RESULTS

An electronic CRF is used in this study and a specific electronic CRF will correspond to each subject.

All required information must be entered on the CRFs. If an item is not available or is not applicable, this fact should be indicated and no blank spaces must be left. The data collected on the CRF will be entered into the study database. If the investigator authorises other personnel to enter data into the CRF, the names, positions, signatures, and initials of these persons must be supplied to NovImmune or their authorised representative before these individuals start completing CRF information.

The CRF pages must be reviewed and signed by the Investigator named in the study protocol or by a designated sub-investigator. NovImmune will ensure that the CRF copy left with the Investigator (print-outs and/or CD-ROM) has never been under the direct or indirect control of NovImmune.

14.2 USE OF COMPUTERISED SYSTEMS AT THE CLINICAL TRIAL CENTRE

When clinical observations are entered directly into an investigational site's computerised medical record system (i.e. in lieu of original hardcopy records), the electronic record can serve as the source document if the system has been validated in accordance with health authority requirements pertaining to computerised systems used in clinical research. An acceptable computerised data collection system allows preservation of the original entry of data. If original data are modified, the system should maintain

a viewable audit trail that shows the original data as well as the reason for the change, name of the person making the change, and date of the change.

The system must allow the clinical research associate, auditors or inspectors to verify source data without infringing privacy rights of other patients, e.g. access must be restricted to records pertaining to the study patients and access to other patients must not be possible.

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APPENDICES

Appendix A - NI-0501 dose justification in HLH patients

Appendix B - Decision making process on dose increase

Appendix C – Detail of estimated blood volumes to be drawn during the study

Appendix D –Membership of the Study Scientific Committee (SSC)

Appendix E – NovImmune SAE Reporting Form

Appendix F – NovImmune Pregnancy Form

APPENDIX A: NI-0501 DOSE JUSTIFICATION IN HLH PATIENTS

Based on the evidence accumulated during the past years, it can be concluded that the over-production of IFNγ in HLH patients has a pivotal pathogenic role in this disease. Based on the key role played by IFNγ in the HLH, the investigation of the use of an anti-IFNγ mAb in HLH is deemed justified.

NI-0501 is a fully human anti-IFNγ monoclonal antibody (mAb) which binds and neutralizes IFNγ. The dose of NI-0501 required for reaching and maintaining over time a certain percentage of inhibition of IFNγ depends on the amount of IFNγ produced, circulating as well as present in tissues. The NI-0501 concentrations that inhibit the effect of IFNγ and the doses that reach and maintain these NI-0501concentrations have been predicted.

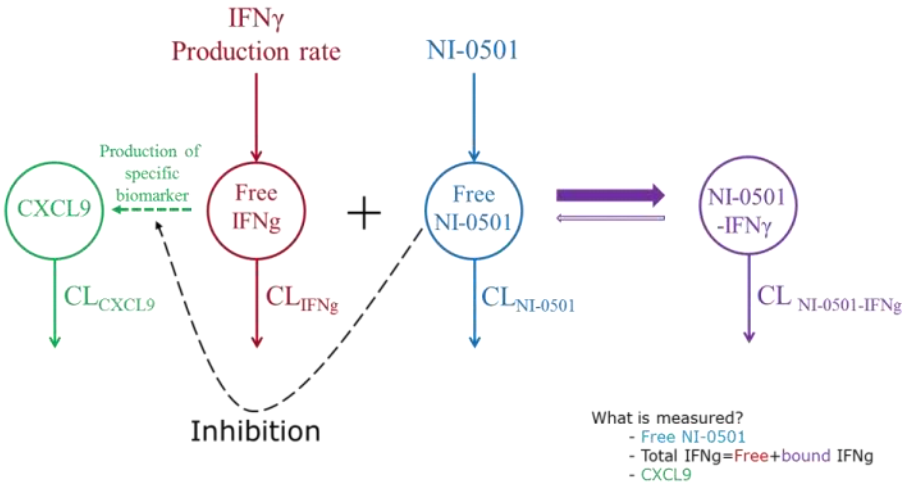


Figure 1: Schematic representation of the interaction between IFNγ, NI-0501 and CXCL9

Figure 1 schematically represents the intended interaction between IFNγ and NI-0501. In HLH patients, the production of IFNγ (red) is exacerbated and leads to high circulating concentrations of free IFNγ. NI-0501 (blue) can be administered to these patients with the goal to bind to free IFNγ and reduce its concentration and consequently its damaging effects. The binding of NI-0501 to IFNγ leads to the production of an IFNγ-NI-0501 complex (violet) which is in equilibrium with the free moieties. To confirm the neutralization of IFNγ by NI-0501, the decrease in serum concentration of CXCL-9 (green), one of the cytokines specifically produced through the activation of the IFNγ receptor, can be monitored. All the components described above (i.e. free IFNγ, free NI-0501, IFNγ-NI-0501 complex and CXCL-9) have their own intrinsic clearance and their concentrations correspond to an equilibrium between production and elimination. Furthermore, for NI-0501, the formation of the complex corresponds to an additional target mediated clearance proportional to the IFNγ production. In HLH patients receiving NI-0501, serum concentrations of free NI-0501, total IFNγ (i.e. free IFNγ + IFNγ-NI-0501 complex) and CXCL-9 have been measured.

Data from *in vitro* experiments investigating the binding of NI-0501 to human IFNγ and the functional inhibition of human IFNγ by NI-0501 have been used for predicting the concentrations of NI-0501 that inhibit (e.g., 99%) the effect of circulating IFNγ concentrations. The results indicate that these NI-0501 concentrations highly depend on the IFNγ concentrations themselves.

Based on the calculated neutralizing concentrations of NI-0501 (e.g., for 99% inhibition of the effect of a baseline IFNγ concentration of 0.1 nM, 3400 pg/mL), on the PK parameters of NI-0501 in Healthy

Volunteers and on the PK information from recombinant IFNγ in human, predictions were performed regarding the dose that would inhibit in the majority of patients the effect of circulating and newly formed IFNγ over a period of 3 days (i.e. dosing interval). Based on these predictions, the starting dose in HLH patients was determined to be 1 mg/kg. This dose, predicted to inhibit for 3 days at least 99% of IFNγ effect in patients with baseline IFNγ concentrations lower or equal to 3400 pg/mL, was mainly driven by the estimated production of IFNγ which impacts the clearance of NI-0501 and varies considerably between patients as already indicated by the wide range of baseline IFNγ concentrations observed in HLH patients.

The originally planned dosing strategy foresaw that, after the initial administration of NI-0501 at 1 mg/kg, the dose could have been adapted depending on PK and clinical response (fever and thrombocytopenia), with clinical response overriding PK, in case of a favorable response to treatment.

The rationale for choosing the initial interval of administration of 3 days for the pilot Phase 2 study, at least until steady state has been achieved, was based on the expected need to adjust in a short period of time and in an informed manner the dose of NI-0501.

The selection of an interval of administration of 3 days has allowed:

- Having a fast initial assessment of the patient's PK (concentration measured at the end of NI-0501 infusion, 24 h and 48 h post infusion), confirming, after the first dose, whether the assumptions used in the mathematical Models were correct. The PK samples mentioned above have been analyzed so far in a timely manner, so that the results were available for an informed decision on the selection of the subsequent dose to be administered (provided no safety concerns and no worsening were observed);
- The possibility to rapidly adjust the dose of NI-0501 depending on PK and clinical response;
- Overcoming the expected effect of TMDD of IFNγ turnover on the bioavailability of NI-0501 in HLH patients;
- To avoid in patient with high clearance due to TMDD to have high peak and trough fluctuations which would have been the case with longer dosing intervals.
- To satisfy the request of the Scientific Steering Committee of the study to ensure a fast and safe dose finding process in this fragile and severe patient population, in parallel to the close laboratory and clinical monitoring of patients.

Data gathered so far in the Phase 2 study have shown that an initial dose regimen of 1 mg/kg every 3 days was appropriate to obtain a rapid onset of NI-0501 effects on HLH parameters in the majority of the patients treated so far. However, it has also been observed that in patients having a high production of IFNγ, evidenced by high circulating total IFNγ concentrations, a higher dose of NI-0501 was required, demonstrating the presence of a target mediated drug disposition (TMDD) with NI-0501. This phenomenon has shown to induce a pronounced increased clearance of NI-0501 due to the high production of IFNγ. The presence of TMDD, while requiring the administration of NI-0501 doses higher than 1 mg/kg, prevents NI-0501 accumulation to occur.

Based on experience gained from the first 16 evaluable primary HLH patients treated with NI-0501, and the excellent safety and tolerability profile of NI-0501, the possibility that a dose increase could be required at different times during treatment has been confirmed and therefore a standardized approach to guide dose changes has been introduced through this amendment.

From the available data it has also been possible to estimate the total production of IFNγ in HLH patients. In fact, it is not possible to estimate the total amount of a given cytokine produced in the body simply based on its free circulating concentrations. However, once an anti-IFNγ antibody is administered, it moves from the circulation to tissues and organs where it binds to IFNγ, and then returns into circulation. It is at this point that the amount of IFNγ bound to the antibody and present in blood reflects the total production of IFNγ. This is what we measure as "Total IFNγ" (Finkelman and Morris, 1999). Applying this principle, from a preliminary analysis of the data from the Phase 2 study, it can be concluded that the production rate of IFNγ is extremely high in patients with HLH, as demonstrated by the elevated concentrations of total IFNγ detected in the vast majority of the HLH patients receiving NI-0501

(reaching the concentration of several hundred thousand pg/mL) and that inflamed tissues are certainly a main source of the produced IFNγ.

A significant INFγ production was present in all patients in whom a dose increase was applied during the ongoing Phase 2 study. Importantly, it was also demonstrated that the production of IFNγ can vary during the course of the disease, particularly influenced by the presence of infections, as proven by the high inter- and intra-patient variability of IFNγ levels (from 1 to 10,000 pM) shown in the patients treated so far with NI-0501 (Figure 2).

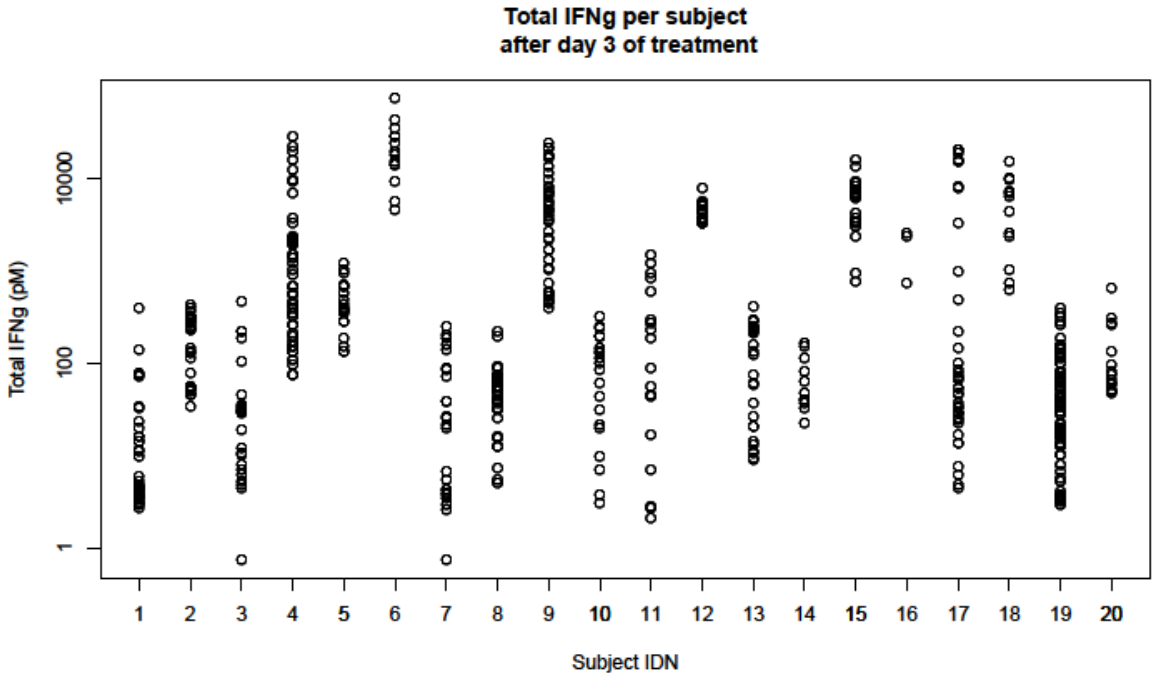


Figure 2: Total IFNγ concentrations (log scale) in HLH patients treated with NI-0501. Concentrations before day 3 are omitted because there are not yet at steady state.

All this taken together, in protocol Version 5 of study NI-0501-04, after the initial administration of 1 mg/kg of NI-0501, dose adjustments can be applied. In particular, a dose increase to 3 mg/kg (and, if needed, to 6 mg/kg) will be possible according to pre-defined criteria guided by clinical and laboratory response in each patient (see Table 4 and Appendix B).

Infusions will be performed every 3 days until SD15 (infusion #6), and twice per week thereafter. Elongation of the dosing interval to 1 week can occur only after the first 4 weeks of treatment, if the patient has achieved and maintained Complete Response for at least 1 week. Dose increases to 3 and 6 mg/kg are possible at any time during the study. More than 4 administrations at 6 mg/kg, or the introduction of a dose higher than 6 mg/kg may be proposed by the Investigator based on a positive benefit/risk assessment. However, the implementation of the above has to be discussed and approved by the Data Monitoring Committee after thorough assessment of available data, including PK and PD.

Predictions to guide the initial and subsequent NI-0501 doses in HLH patients

The original calculation of the first NI-0501 dose in HLH patients was based on:

- The affinity of NI-0501 for human IFNγ (Report NI-0501-MIF-01)
- Inhibition of human IFNγ-induced STAT-1 induction by NI-0501 (Report NI-0501-PHARMACO-01)
- NI-0501 population pharmacokinetic parameters estimated in Healthy Volunteers (SAD study NI-0501-03) (Modeling and Simulation support to NI-0501: PK analysis of study NI-0501-003) (Draft Report available)
- Literature information on the in vivo PK of recombinant IFNγ in human (http://www.drugs.com/pro/actimmune.html)

The data from the *in vitro* experiments have been used for predicting the NI-0501 concentrations that inhibit up to 90 or 99% the effect of high IFNγ concentrations (i.e. 0.1 and 0.01 nM) (Fig. 3, Table 1).

Figure 3: Inhibition by NI-0501 of the effect induced by different concentrations of IFNγ. The parameters used in the simulations are given in Table 4 assuming 1 binding site per antibody.

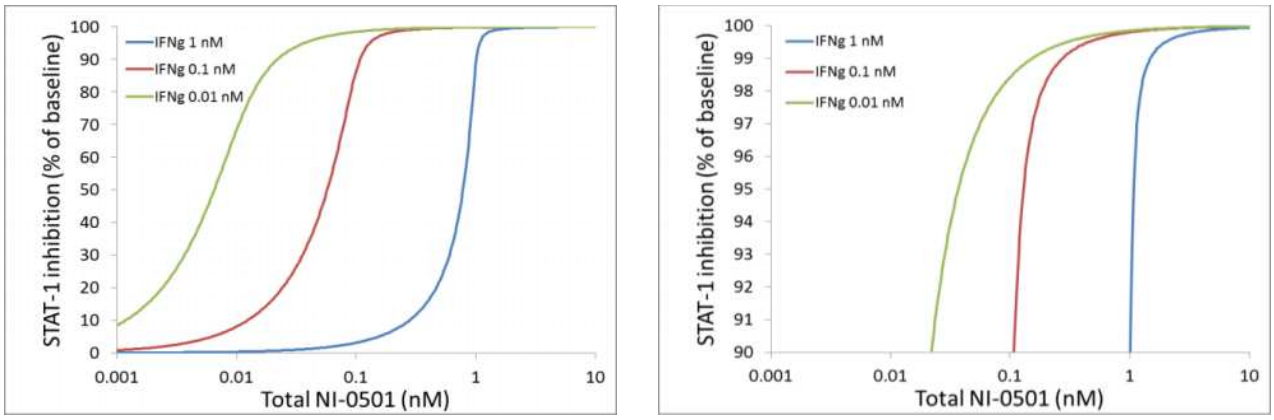


Table 5: Simulations of inhibition of IFNγ-induced STAT-1 induction by NI-0501

	Simulation based on KD and IC50 assuming one binding site per Ab (Model 1)		Simulation based on estimated parameters (Model 2)	
KD (nM)	0.0014	0.0014	0.0017	0.0017
IC_{50} (nM)	0.4	0.4	0.523	0.523
GA	1	1	0.737	0.737
BS Binding sites per NI-0501 molecule	1	1	1.58	1.58
IFNγc concentration (nM)	0.1	0.01	0.1	0.01
IFNγ effect (fraction of maximum effect)	0.200	0.0244	0.228	0.0514
EFF1F (fraction of IFNγ effect)	0.01	0.01	0.01	0.01
EFF1 (fraction of maximum effect)	0.00200	0.00024	0.00228	0.00051
EFF2F (fraction of IFNγ effect)	0.1	0.1	0.1	0.1
EFF2 (fraction of maximum effect)	0.02000	0.00244	0.02281	0.00514
Free IFNγ for EFF1	0.00080	0.00010	0.00014	0.00002
Free IFNγ for EFF2	0.00816	0.00098	0.00319	0.00041
Bound IFNγ for EFF1	0.09920	0.00990	0.09986	0.00998
Bound IFNγ for EFF2	0.09184	0.00902	0.09681	0.00959
Total NI-0501 (nM) for EFF1 (1%)	**0.272**	**0.152**	**0.850**	**0.603**
Total NI-0501 (nM) for EFF2 (10%)	0.108	0.0219	0.0939	0.0311
Free NI-0501 (nM) for EFF1 (1%)	0.173	0.142	0.787	0.596
Free NI-0501 (nM) for EFF2 (10%)	0.0158	0.0129	0.0326	0.0250

Parameters are derived from the IB or estimated by modeling of the experiment.

- Two levels of IFNγ (0.1 and 0.01 nM) are simulated with two levels of inhibition each (99%, i.e. EFF1F = 0.01 and 90%, i.e. EFF2F = 0.1).
- IFNγ effect = IFNγc^GA / (IC_{50}^GA +IFNγc^GA).
- EFFx = IFNγ effect* EFFxF.
- Free IFNγ = exp(ln(EFFx*(IC_{50}^GA)/(1-EFFx))/GA).
- Bound IFNγ = IFNγc-free IFNγ.
- Total NI-0501 = bound IFNγ*(KD+free IFNγ)/free IFNγ/BS.
- Free NI-0501 = Total NI-0501-(bound IFNγ /BS).

Note: the residual effect after inhibition is a percentage of the baseline effect (not an absolute residual effect).

Based on the predicted IFNγ-neutralizing concentrations of NI-0501 (Table 1), on PK parameters of NI-0501 in healthy volunteers and on PK parameters from recombinant IFNγ in human, calculations were performed to predict the doses that inhibit the effect of circulating and newly formed IFNγ up to 99% over a period of 3 days (dosing interval) in HLH patients (Table 6**Error! Reference source not found.**).

Table 6: Predictions of the first dose of NI-0501 in HLH patients for 'neutralizing' circulating and produced IFNγ.

	Simulation based on KD and IC_{50} assuming one binding site per Ab (Model 1)		Simulation based on estimated parameters (Model 2)	
IFNγ (nM)	0.1	0.01	0.1	0.01
MW of IFNγ	34000	34000	34000	34000
IFNγ (pg/mL)	3400	340	3400	340
Volume of distribution of IFNγ (L/kg)	1.10	1.10	1.10	1.10
Clearance of IFNγ (L/h/kg)	1.2	1.2	1.2	1.2
Total NI-0501 conc. for 99% inhibition of IFNγ effect (nM)	0.272	0.152	0.850	0.603
MW of NI-0501 (Dalton)	147987	147987	147987	147987
Total NI-0501 conc. for 99% inhibition of IFNγ effect (ng/mL or ug/L)	40	22	126	89
Volume of distribution of NI-0501 (L/kg)	0.079	0.079	0.079	0.079
Neutralizing dose of NI-0501 (mg/kg)	0.00318	0.00178	0.0099	0.00705
Neutralizing dose of NI-0501 corrected for difference in volume of distribution between IFNγ and NI-0501 (mg/kg)	0.0441	0.0247	0.138	0.0978
Maintenance dose of NI-0501 (mg/kg/h)	0.018	0.0018	0.011	0.0011
Maintenance dose of NI-0501 (mg/kg for 3 days)	1.28	0.128	0.809	0.0809
Loading dose (mg/kg)	1.32	0.153	0.947	0.179

- IFNγc = anticipated plasma concentration in HLH patients.
- MW = molecular weight.
- Volume of distribution of IFNγ = 1.4 L/min /(0.693/38 min)/70 kg.
- Clearance of IFNγ = 1.4 L/min*60 min/70 kg.
- Total NI-0501 conc. for 99% inhibition (nM) of IFNγ effect is obtained from Table 4.
- Volume of distribution of NI-0501 is from preliminary POP PK analysis of SAD data in healthy volunteers.
- Neutralizing dose of NI-0501 = total NI-0501 conc * Volume of distribution of NI-0501.
- Correcting factor for the difference in volume of distribution = volume of IFNγ / volume of NI-0501.
- Maintenance dose of NI-0501 = IFNγc (nM) * Clearance of IFNγ * MW of NI-0501 / 10^6/number of binding sites per molecule of NI-0501 (BS from Table 4).
- The loading dose is the sum of the neutralizing dose of NI-0501 corrected for difference in volume of distribution between IFNγ and NI-0501 and the maintenance dose of NI-0501 for 3 days.
- Note: the amount of NI-0501 eliminated over a period of 3 days after the first dose and assuming no TMDD is predicted to be lower than 36 % of the administered dose (e.g. for a body weight of 5 kg and a dose of 1 mg/kg the approximate calculation based on C_{max} (end infusion) is: 22.9 mg/L x 0.00102 L/h x 24h x 3 = 1.7 mg x 100 / 5 mg = 34%).

Based on the predicted 'IFNγ-neutralizing' doses of NI-0501 presented in Table 6, the proposed starting dose in HLH patients was 1 mg/kg. During the Phase 2 study (Study NI-0501-04 Protocol Version 4), due to the uncertainty in the predictions, especially from the target mediated drug disposition effect of IFNγ turnover on the clearance of NI-0501 in HLH patients, subsequent doses could be adapted based on NI-0501 concentrations measured at the end of the infusion ($C_{end\ infusion}$) and at subsequent time-points (e.g., 24h (C_{24h})/48h (C_{48h}) after administration and on clinical response. The observed concentrations and their ratios were compared to predicted concentrations and ratios obtained from the population PK model of NI-0501 in Healthy Volunteers after allometric scaling and assuming linear (i.e. without TMDD see Figures **Error! Reference source not found.**4 and 5 top graphs) or non-linear (i.e. with TMDD see Figures 4 and 5 bottom graphs) kinetics. If the observed concentrations and ratios were markedly low and below predicted reference values (see Figures 4, and 5, and Table 3), marked TMDD could be assumed and a dose increase could be recommended. Figure 6 shows the predictions of NI-0501 concentration-time profiles after the first administration of 1 mg/kg NI-0501 in HLH patients.

Prior to the initiation of the NI-0501-04 study, it was established that during treatment with NI-0501, the minimal NI-0501 concentration to be achieved should have been the one required to neutralize 0.1 nmol of INFγ for 3 days, while the initial maximal concentration ("ceiling") should have not exceeded an arbitrary concentration corresponding to the median maximal concentration reached in Healthy Volunteers following the single administration of the NI-0501 at the highest dose tested (3 mg/kg). The experience gathered during the conduct of the Phase 2 study has allowed to determine that, in the presence of a high INFγ production, there is fast clearance of NI-0501 (TMDD) and NI-0501 dose increases greater than 3 mg/kg are required.

So far, during the conduct of the study, as well as in patients receiving NI-0501 in Compassionate Use, the safety and tolerability profile of NI-0501 was very good. In particular, to date, in no circumstances an increase in the NI-0501 dose was associated with the occurrence of safety concerns. No SAEs related to NI-0501 and no increased severity or frequency of non-serious AEs was reported. All infusions were uneventful.

Taking all these data together, it can be concluded that, applying the dosing strategy proposed for the continuation of the Phase 2 study as a Phase 2/3 study, HLH patients will be exposed to NI-0501 concentrations already achieved during the Phase 2 study, at which no safety concern has emerged to date and well within the exposure achieved during the toxicology studies, with a safety margin of approx. 7.5 as to Cmax. In particular, as the administration of more than 4 NI-0501 doses at 6 mg/kg can only occur after a review of the PK and PD data, the possibility that an accumulation of NI-0501 occurs is to be excluded.

Simulations to help in characterizing the PK in HLH patients and recommending dose adjustment

Method

The PK model used in the simulations below is a two compartment model with linear elimination assuming allometric scaling based on body weight (BW) to which an additional non-linear (TMDD) elimination pathway characterized by a VMAX and a KM has been added.

Parameters used in the simulations are from a population pharmacokinetic analysis of study NI-0501-03 and assuming allometric scaling. VMAX is the IFNγ concentration (0.1 nM) multiplied by the recombinant IFNγ clearance (1.2 L/h/kg) divided by the number of binding sites (1 for model 1 or 1.58 for model 2, Table 4) per antibody. KM is assumed to be equal to KD.

CL = $0.00737*(BW/70)^{+0.75}$ L/h
V1 = $3.03*((BW/70)^{+1})$ L
Q = $0.0218*(BW/70)^{+0.75}$ L/h
V2 = $2.98*(BW/70)^{+1}$ L
VMAX = 0.12*BW nmol/h (model 1) or 0.076*BW nmol/h (model 2)
KM = 0.0014 nM (considered to be approximately the same for both models 1 and 2 and other VMAX values)

The VMAX value of 0.076 nmol/h/kg from model 2 is considered to be pivotal for the dose of 1 mg/kg NI-0501. At higher VMAX, the dose of 1 mg/kg NI-0501 is predicted not to optimally neutralize the effect of IFNγ 0.1 nM.

On the graphs:

The pink lines represent the median NI-0501 maximum concentration observed in study NI-0501-03 (SAD study) for the highest dose tested 3 mg/kg (78508 ng/mL or 533.85 nM).The red lines represent total NI-0501 concentration (40 ng/mL or 0.272 nM) that inhibits 99% of IFNγ (free concentration of 3400 pg/mL or 0.1 nM) in blood. The orange lines indicate the reference values on day 2 for model 2 with VMAX=0.076 nmol/h/kg.

Results

Figure 4: Predicted NI-0501 plasma concentration-time profiles after administration (1-hour infusion) of 1mg/kg NI-0501 every 3 days in a HLH patient of 7 kg assuming VMAX values equal to 0, (no TMDD, top graphs: semi-log scales on the left, linear scales on the right), 0.12 (model 1, bottom left graph) or 0.076 (model 2, bottom right graph) nmol/h/kg.

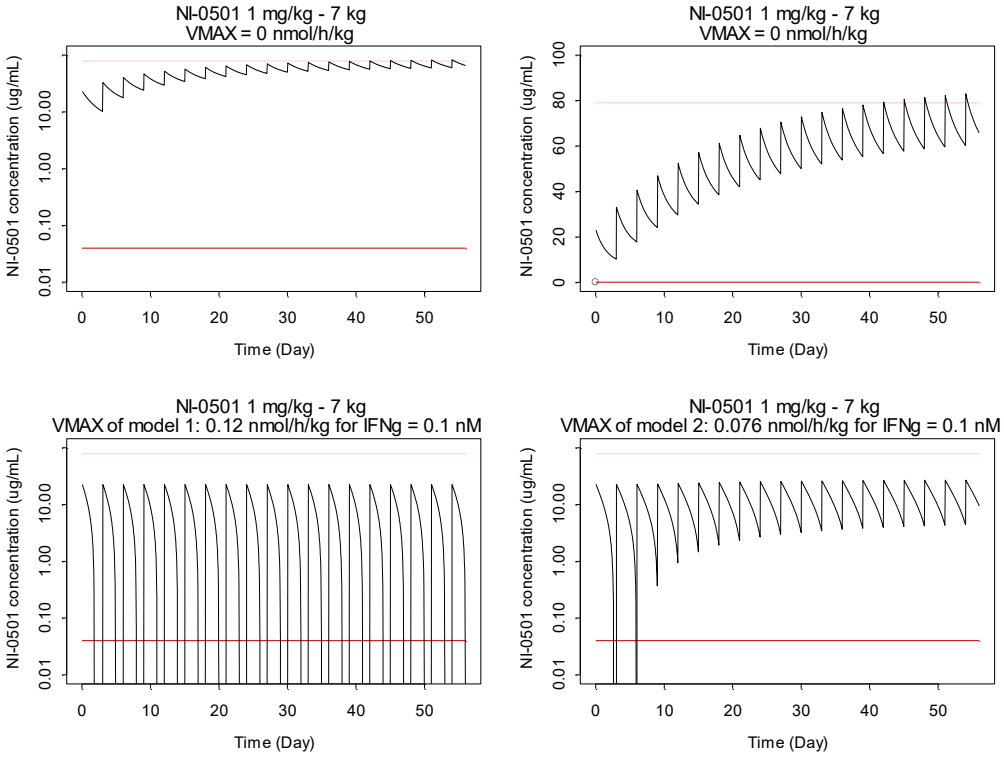


Figure 5: Predicted NI-0501 plasma concentration-time profiles after administration (1-hour infusion) of 1mg/kg NI-0501 every 3 days in a HLH patient of 23 kg assuming VMAX values equal to 0 (no TMDD, top graphs: semi-log scales on the left, linear scales on the right), 0.12 (model 1, bottom left graph) or 0.076 (model 2, bottom right graph) nmol/h/kg.

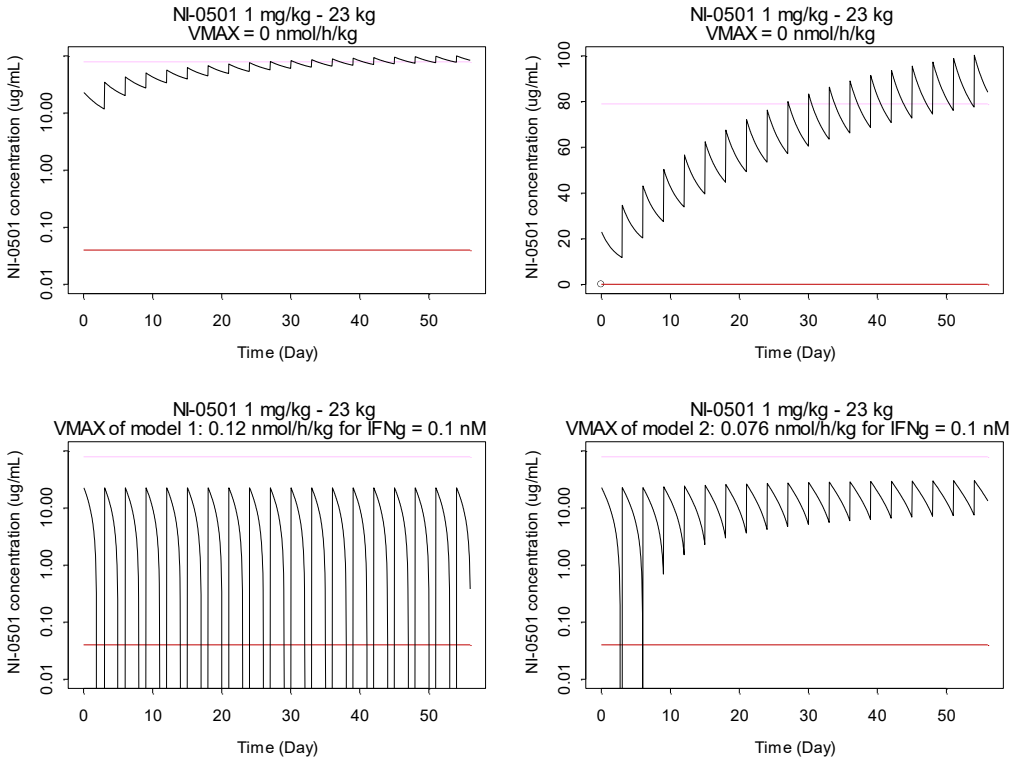


Figure 6: Predictions of NI-0501 concentration-time profiles after the first administration of 1 mg/kg NI-0501 in HLH patients of 23 kg with various levels of VMAX. From bottom to top VMAX=0.12 (model 1), 0.076 (model 2), 0.06, 0.03, 0.015, 0.0075, 0.00375, 0.001875 and 0 (no TMDD) nmol/h/kg.

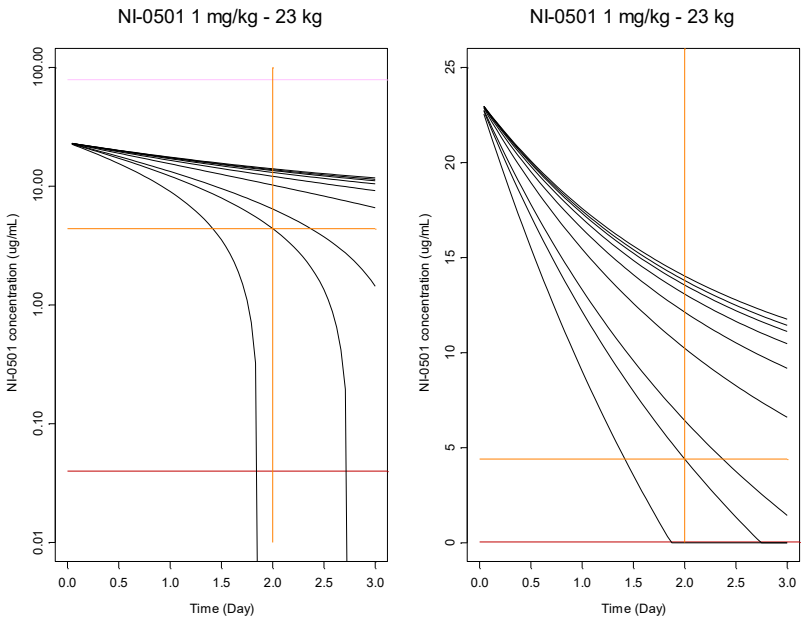


Table 7: NI-0501 concentrations at 48h (C48h) and C48h/Cend infusion ratios below which a dose higher than 1 mg/kg might be required. These values are predicted after the first administration of 1 mg/kg NI-0501 in HLH patients assuming a VMAX of 0.076 nmol/h/kg for a baseline IFNγ concentration of 0.1 nM (3400 pg/mL).

BW (kg)	C_{48h} (ug/mL)	C_{48h} / $C_{end\ infusion}$
5	3.14	0.14
10	3.69	0.16
15	4.03	0.18
20	4.28	0.19
25	4.48	0.20
30	4.63	0.20
35	4.77	0.21
40	4.88	0.21
45	4.99	0.22
50	5.08	0.22
55	5.16	0.23
60	5.24	0.23
65	5.30	0.23
70	5.37	0.24

Interim (up to December 2, 2015) PK-PD data from studies NI-0501-04 and NI-0501-05 in HLH patients and in Compassionate Use patients receiving NI-0501 have been analysed in order to explore the structural and quantitative relationships between NI-0501, total IFNγ (free + complex with NI-0501) and CXCL-9 a chemonokine specifically induced by IFNγ.

The results of this PK-PD exploratory analysis indicated that:

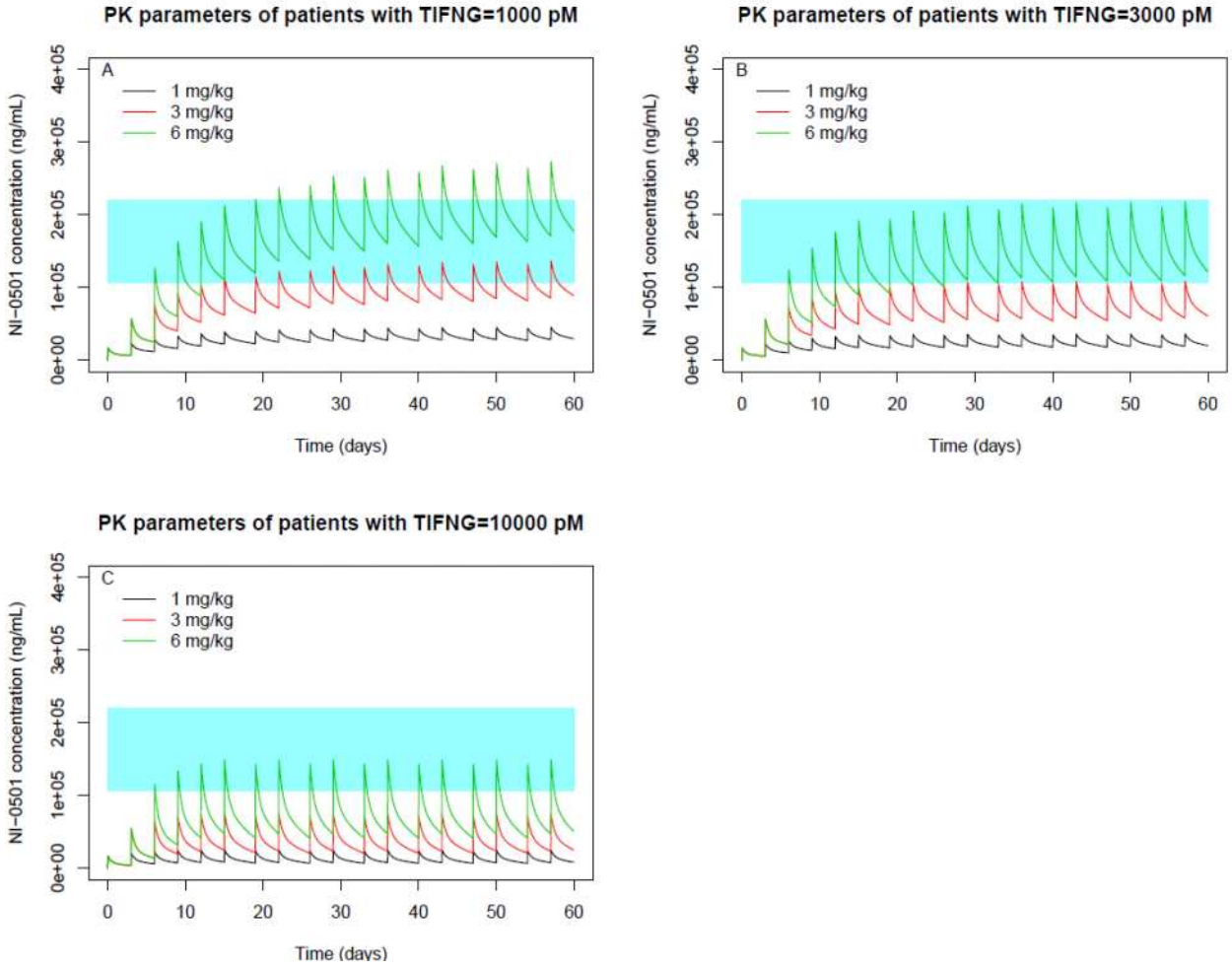
- CXCL-9 concentration is a good surrogate of IFNγ activity. In patients treated with NI-0501, CXCL-9 concentrations depend on the total IFNγ concentration (free + complex with NI-0501) and free concentration of NI-0501.
- Total IFNγ concentration at steady-state is an expression of IFNγ production: the higher the total concentration of IFNγ in serum, the higher the IFNγ production.
- Total IFNγ concentration/production in HLH patients shows a high inter- and intra-individual variability.
- The kinetics of NI-0501 in HLH patients show target mediated disposition:
  - At total IFNγ concentration lower than 300 pM the kinetics of NI-0501 is rather linear.
  - At total IFNγ concentration higher than 300 pM the clearance of NI-0501 increases to become proportional to the total IFNγ concentration/ production.
- The higher the total concentration of IFNγ in serum:
  - The higher the concentration of NI-0501 to neutralize IFNγ (evidenced by a higher CXCL9 concentration to inhibit).
  - The higher the dose of NI-0501 to reach the neutralizing concentration of IFNγ (evidenced by a higher target mediated clearance of NI-0501).

Although the PK/PD analysis performed so far was based on interim data, and for this reason it should be considered exploratory, the derived information is deemed enough explanatory to guide the selection of a safe and efficacious dosing algorithm of NI-0501 in HLH, allowing to perform explorative simulations to anticipate the exposure to be expected if the proposed new dosing strategy would be applied.

These quantitative findings have been used to evaluate an amended dosing strategy of NI-0501 in HLH patients. The goal of the adapted dosing strategy is to allow dose increases based on clinical judgment only in patients who deteriorate or have no clinical improvement after the first administration(s) of NI-0501 at 1 mg/kg/3days.

To evaluate the impact of these dose increases on concentration levels, simulations have been performed. In these simulations, a time aggressive scheme consisting of increasing the dose from 1 to 3 mg/kg on day 3 and, if necessary, to 6 mg/kg on day 6 have been simulated. The doses were assumed to be given every 3 days till day 15 and afterwards bi-weekly (intervals of 4 and 3 days). Since patients with insufficient response are potentially patients with significant levels of total IFNγ productions, simulations have been performed with different clearance levels based on the estimated correlation that exists between NI-0501 clearance and total IFNγ and for various plausible levels of total IFNγ concentrations as observed in study NI-0501-04 and NI-0501-05 (see Figure 7 below).

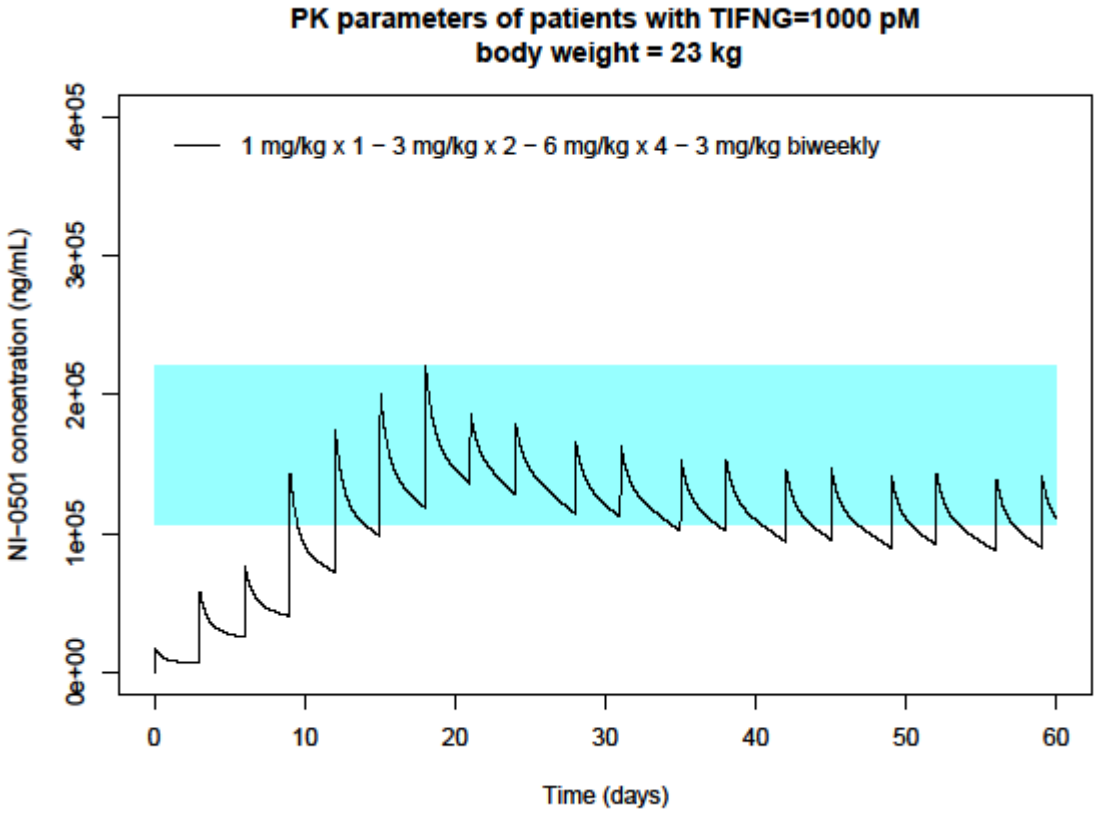
Figure 7: Simulated NI-0501 serum concentrations as a function of time for different dosing and total IFNγ concentration scenarios. Doses are administered every 3 days till day 15 and bi-weekly afterwards. Dose regimens are: 1 mg/kg from day 0 (1 mg/kg); 1 mg/kg on day 0 followed by 3 mg/kg from day 3 on (3 mg/kg); 1 mg/kg on day 0 followed by 3 mg/kg on day 3 and followed by 6 mg/kg from day 6 on (6 mg/kg). Simulations are performed using PK parameters from patients assuming total IFNγ concentrations of 1000 pM (A), 3000 pM (B) and 10000 pM (C). The blue shaded area represents the interval between the highest (i.e. mean of the 3 highest values in the database) peak and trough NI-0501 concentrations observed so far in studies NI-0501-04 and NI-0501-05 in HLH patients and in Compassionate Use patients.



Although patients receiving 3 and 6 mg/kg will have significantly higher concentrations, as observed in study NI-0501-04 and NI-0501-05, patients who usually require higher doses are patients with increased clearance due to high circulating IFNγ. Patients who will probably be subject to a dose increase to 3 mg/kg are most likely patients with a total IFNγ concentration around 1000 pM and patients who will require a dose increase to 6 mg/kg are most likely patients with total IFNγ concentration higher than 3000 pM.

Predictions for the proposed NI-0501 dose increase to 6 mg/kg allowed in Version 5 of the NI-0501 study protocol are represented in Figure 8 below.

Figure 8: Simulated NI-0501 serum concentrations as a function of time for the proposed NI-0501 dose increase to 6 mg/kg allowed in Version 5 of the NI-0501-04 study protocol and for a constant total IFNγ concentration of 1000 pM. The simulated dosing regimen is: 1 mg/kg on day 0; 3 mg/kg on days 3 and 6; 6 mg/kg on days 9, 12, 15 and 18, followed by 3 mg/kg on day 21 and then bi-weekly (e.g., days 24, 28, 31, 35). The blue shaded area represents the interval between the highest (i.e. mean of the 3 highest values in the database) peak and trough NI-0501 concentrations observed so far in studies NI-0501-04 and NI-0501-05 in HLH patients and in Compassionate Use patients.



In conclusion, since dose increases will most probably occur in patients with increased clearance due to a high production of IFNγ, the exposure will be less than dose proportional and is predicted to remain within (or close to) the concentration range that has already been observed in study NI-0501-04 and NI-0501-05.

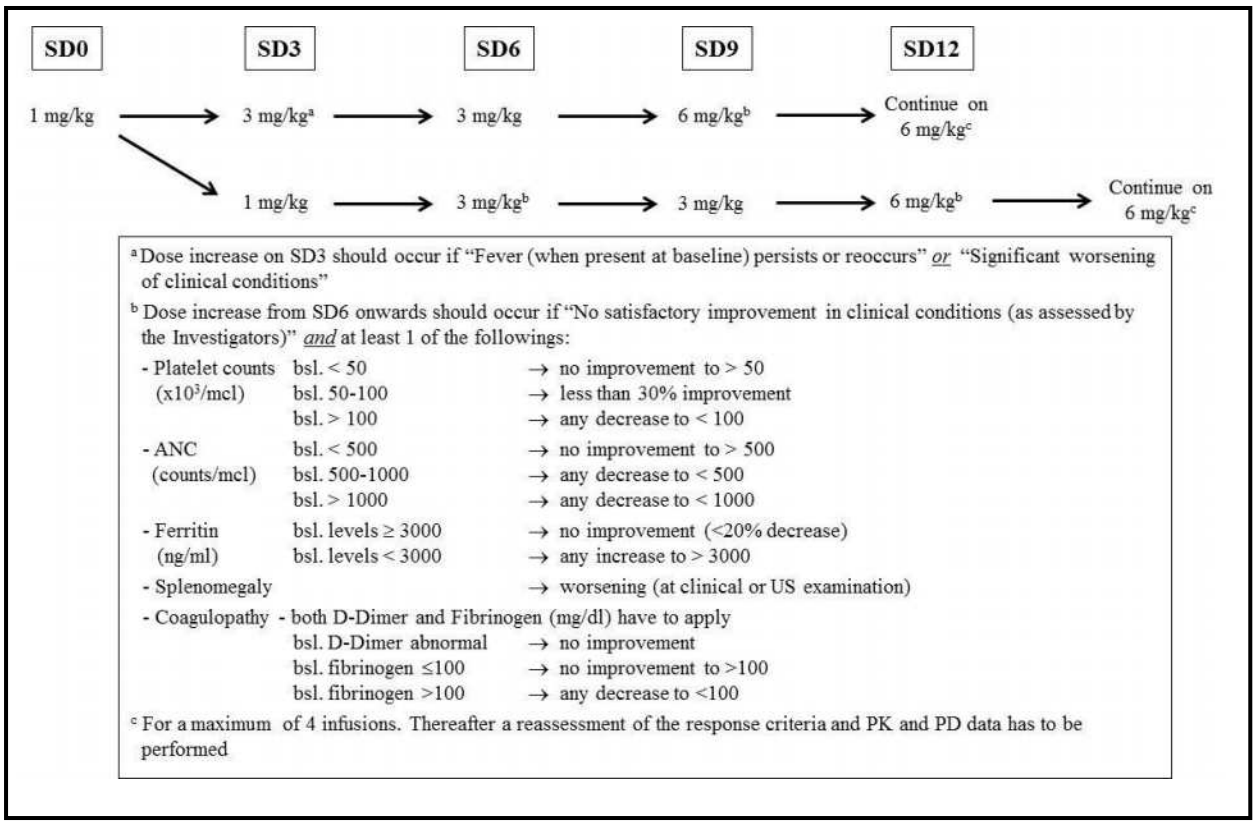
APPENDIX B: DECISION MAKING PROCESS ON DOSE INCREASE

This document has the objective of:

1. guiding the Investigator on the clinical and laboratory parameters for the assessment on whether NI-0501 dose has to be increased
2. giving a clear overview of the decision making process for dose increase during the study.

Figure 1 gives an example of the flow for assessing a potential NI-0501 dose increase occurring at the start of the study.

Figure 1 – Flow-chart of the decision making process on dose increase



Please note:

- Dose increase may occur any time during the study, if the clinical and laboratory criteria reported on SD6 are met.
- When NI-0501 dose is increased to 6 mg/kg, a maximum of four infusions at this dose levels will be administered. Thereafter, reassessment needs to occur by the Investigator and the DMC, in order to establish the appropriate regimen for continuation of NI-0501 treatment (see below)

Study Day 0 _____

The parameters collected at this time point constitute the baseline.

The initial dose of NI-0501 to be administered is **1 mg/kg**.

Study Day 3 _____

Parameters to consider:

- body temperature
- patient's clinical conditions

Decision to be taken by the Investigator:*

Is there a need to increase NI-0501 dose to **3 mg/kg**?

- if fever, present at baseline, persists or reoccurs, NI-0501 dose needs to be increased
- if there is a significant worsening of the patient's clinical conditions, NI-0501 dose needs to be increased

**Note:* the Investigator can decide at any time during the study to discontinue NI-0501 treatment based on the individual benefit/risk assessment.

Study Day 6 _____

Parameters to consider:

- patient's clinical conditions
- clinical and laboratory response parameters (as presented in Table 4)

Decisions to be taken by the Investigator:

a) If the patient is receiving NI-0501 at the dose of 1 mg/kg, should the dose be increased to **3 mg/kg**?

- if no satisfactory improvement in clinical conditions is assessed by the Investigator
  and
- if any of the clinical and laboratory criteria presented in Table 4 and Figure 1 is met

  NI-0501 dose needs to be increased

  If the above criteria do not apply, then the patient should continue with NI-0501 dose of 1 mg/kg.

b) If NI-0501 dose has been increased on SD3, the dose of 3 mg/kg should be maintained

Study Day 9 _____

Parameters to consider:

- patient's clinical conditions
- clinical and laboratory response parameters (as presented in Table 4 and Figure 1)

Decisions to be taken by the Investigator:

a) If the patient is receiving NI-0501 at the dose of 1 mg/kg, should the dose be increased to **3 mg/kg**?

- if no satisfactory improvement in clinical conditions is assessed by the Investigator

and

- if any of the clinical and laboratory criteria presented in Table 4 is met

NI-0501 dose needs to be increased

If the above criteria do not apply, then the patient should continue with NI-0501 dose of 1 mg/kg.

b) If NI-0501 dose has been increased on SD3, should the dose be increased to **6 mg/kg**?

- if no satisfactory improvement in clinical conditions is assessed by the Investigator
  and
- if any of the clinical and laboratory criteria presented in Table 4 and Figure 1 is met

NI-0501 dose needs to be increased

If the above criteria do not apply, then the patient should continue with NI-0501 dose of 3 mg/kg.

Study Day 12 _____

Parameters to consider:

- patient's clinical conditions
- clinical and laboratory response parameters (as presented in Table 4)

Decisions to be taken by the Investigator:

c) If the patient is receiving NI-0501 at the dose of 1 mg/kg, should the dose be increased to **3 mg/kg**?

- if no satisfactory improvement in clinical conditions is assessed by the Investigator
  and
- if any of the clinical and laboratory criteria presented in Table 4 is met

NI-0501 dose needs to be increased

If the above criteria do not apply, then the patient should continue with NI-0501 dose of 1 mg/kg.

d) If NI-0501 dose has been increased on SD6, should the dose be increased to **6 mg/kg**?

- if no satisfactory improvement in clinical conditions is assessed by the Investigator
  and
- if any of the clinical and laboratory criteria presented in Table 4 is met

NI-0501 dose needs to be increased

If the above criteria do not apply, then the patient should continue with NI-0501 dose of 3 mg/kg.

MANAGEMENT OF THE PATIENTS AFTER NI-0501 DOSE INCREASE TO 6 mg/kg

After the patient has received four infusions at the dose of 6 mg/kg with a regular monitoring of the clinical and laboratory HLH parameters, a thorough assessment has to be made by the Investigator with regard to:

- clinical and laboratory response criteria presented in Table 4 and Figure 1
- PK and PD data available

If clinical and laboratory response criteria are no longer applicable, the dose of NI-0501 will be decreased back to 3 mg/kg.

On the other hand, if criteria still apply, based on careful benefit/risk assessment, the Investigator may propose either

i) to continue treatment at 6 mg/kg for additional infusions

or

ii) to increase NI-0501 dose above 6 mg/kg, if PK and PD evidence indicates extremely high IFNγ levels and, consequently, fast NI-0501 elimination.

However, the Investigator's proposal of either continuing 6 mg/kg infusions or increasing the dose above 6 mg/kg has to be discussed and approved by the DMC, after thorough assessment of all available data, including PK and PD.

APPENDIX C: DETAIL OF ESTIMATED BLOOD VOLUMES TO BE DRAWN DURING THE STUDY

Volume of blood per visit (in mL)

Assessment		Screening (over a week)	Treatment Period 1												Treatment Period 2			Follow-Up Period			Wk4/ Study Compl. Visit or WD	Total max
			SD0	SD1	SD2	SD3	SD5	SD6	SD8	SD9	SD11	SD12	SD14	SD15	INF visit[3]	Efficacy/ Safety Visits (every 6-7 days)	Efficacy/ Safety Visits (every 2 weeks)	End of treat. visit	Wk2	Wk3	Pre-conditioning		
Safety Laboratory[1]	CBC	1	1	1	1	1	1	1	1		1		1			1		1	1	1	1	1	21
	Coagulation, fibrinogen	1	1	1	1	1	1	1	1		1		1			1		1	1	1	1	1	21
	Biochemistry, Triglycerides	2	2	2	2	2	2	2	2		2		2			2		2	2	2	2	2	42
	IgG level	0.5																					0.5
Infections	Search for CMV, EBV, Adenoviruses, TB	0.5					0.5					0.5					0.5	0.5	0.5			0.5	4.5
	Search for HIV, HepB, HepC, HZV, HSV	0.5																					0.5
	Search for other pathogens	1																					1
Subtotal		6.5	4	4	4	4	4.5	4	4	0	4	0.5	4	0	0	4	0.5	4.5	4.5	4	4	4.5	90.5
Subtotal per Month																	**46**	**22.5**				**17**	
PK			1	0.5	0.5	1	0.5	1	0.5	1		1		1	1			0.5	0.5	0.5	0.5	0.5	**24.5**
PD[2]			1	1	1	1	1	1	1	1		1		1	1			1	1	1	1	1	**29**
Immunogenicity		0.5																0.5				0.5	**1.5**
Total per visit		7	6	5.5	5.5	6	6	6	5.5	2	4	2.5	4	2	2	4	0.5	6.5	6	5.5	5.5	6.5	
Total per month																	68	31.5				23.5	
TOTAL OVERALL STUDY (maximum)																						**130**	

1 = calculation made without specific micro sampling techniques

2 = if possible - can be reduced

3 = maximum 14 infusions visits, which would mean maximum 14 mL for PK and maximum 14 mL for PD

APPENDIX D: MEMBERSHIP OF THE STUDY SCIENTIFIC COMMITTEE (SSC)

APPENDIX E: NOVIMMUNE SAE REPORTING FORM

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SERIOUS ADVERSE EVENT REPORT

I. ADVERSE EVENT INFORMATION

PATIENT INITIALS (first, last)	PATIENT ID	DATE OF BIRTH (dd/mm/yyyy)	SEX (M/F)	HEIGHT (cm)	WEIGHT (kg)	DATE OF ADVERSE EVENT ONSET (dd/mm/yyyy)

CLINICAL DESCRIPTION OF EVENT(S)
DIAGNOSIS:

SIGNS and SYMPTOMS:

INVESTIGATIONS PERFORMED (Imaging, Lab tests...)

THERAPEUTIC MEASURES

MEDICATIONS GIVEN FOR TREATING THE EVENT

Drug name	Indication/daily dose/route	start date	stop date
_____	_____	___/___/___	___/___/___
_____	_____	___/___/___	___/___/___
_____	_____	___/___/___	___/___/___

REASON FOR SERIOUSNESS

☐ Death

☐ Life-threatening

☐ Resulted in persistent or significant disability / incapacity

☐ Resulted in or prolonged inpatient hospitalisation

From:

To:

(dd/mm/yyyy)

☐ Is a congenital anomaly / birth defect

☐ Other medically important condition

SEVERITY

☐ Mild, WHO1

☐ Moderate, WHO2

☐ Severe, WHO3/4

DURATION

☐ If ≤ 24h:

_____(min or h)

☐ If > 24 h:

Date of end:

(dd/mm/yyyy)

☐ ongoing

RELATIONSHIP TO STUDY DRUG

☐ Reasonable possibility

☐ No reasonable possibility*

* If "No reasonable possibility" checked, other possible cause(s) of AE must be specified in section II, page 2.

OUTCOME

☐ Recovered

☐ Recovered with sequelae

☐ Recovering

☐ Not recovered

☐ Unknown

☐ Fatal Date of death:

(dd/mm/yyyy)

Autopsy performed: ☐ Yes ☐ No

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II. OTHER POSSIBLE CAUSE OF ADVERSE EVENT

Check all other factors that in your opinion may have contributed to this adverse event

- ☐ Disease under study
- ☐ Worsening of Disease under study
- ☐ Other medical condition(s), specify: ______
- ☐ Study drug withdrawal-effect
- ☐ Concomitant or previous medication, specify (and complete section IV): ______
- ☐ Erroneous administration of treatment
- ☐ Protocol-related procedure
- ☐ Other, specify: ______

III. STUDY DRUG INFORMATION

STUDY DRUG		**STUDY NUMBER**	**ACTION TAKEN WITH DRUG** ☐ Drug withdrawn ☐ Dose reduced ☐ Dose increased ☐ Dose not changed ☐ Unknown ☐ Not applicable
DOSE (unit)	**FREQUENCY**	**ROUTE OF ADMINISTRATION**	
UNBLINDED by Investigator: Yes ☐ No ☐ NA ☐ **If Yes, Date (dd/mm/yyyy):**			
TREATMENT DURATION: From (dd/mm/yyyy): To (dd/mm/yyyy): Ongoing ☐			**DID REACTION REAPPEAR AFTER REINTRODUCTION?** ☐ Yes ☐ No ☐ Unknown/ Not applicable

IV. CONCOMITANT DRUG(S) AND HISTORY

CONCOMITANT DRUG(S) (excluding those to treat the event)

Drug name	Indication/daily dose/route	start date	stop date	ongoing	Suspected to cause AE
______	______	__/__/__	__/__/__	☐	☐Yes☐No
______	______	__/__/__	__/__/__	☐	☐Yes☐No
______	______	__/__/__	__/__/__	☐	☐Yes☐No
______	______	__/__/__	__/__/__	☐	☐Yes☐No
______	______	__/__/__	__/__/__	☐	☐Yes☐No
______	______	__/__/__	__/__/__	☐	☐Yes☐No
______	______	__/__/__	__/__/__	☐	☐Yes☐No
______	______	__/__/__	__/__/__	☐	☐Yes☐No

OTHER RELEVANT HISTORY & CONCURRENT CONDITIONS

(e.g., Disease/Surgery/Drug allergy)	start date	end date	ongoing
______	__/__/__	__/__/__	☐
______	__/__/__	__/__/__	☐
______	__/__/__	__/__/__	☐
______	__/__/__	__/__/__	☐
______	__/__/__	__/__/__	☐

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V. REPORTER IDENTIFICATION

Name
Address: Tel: Fax: E-mail: Date (dd/mm/yyyy): _____/_____/_____ Signature: _____

VI. STUDY INFORMATION

Protocol Number: Country: Initial ☐ Follow-up ☐ Final ☐ Other comments:

Please email this signed form to: drugsafety@novimmune.com

or fax to: 00 41 22 839 71 51

SPONSOR Use Only: **Date received** ____/____/____ **Causality assessment:** **Comments:**

APPENDIX F: NOVIMMUNE PREGNANCY FORM

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PREGNANCY FORM
HISTORY AND START OF PREGNANCY

Page 1/2

Study Number:

I. Patient and medication(s) details

Initials (first name, family name) _____ Date of birth dd/mm/yyyy __/__/____ Age _____ Bodyweight before pregnancy _____ Kg Height _____ cm

NovImmune drug taken during the course of the pregnancy: _____

Indication: _____ Dose (please specify frequency): _____

Dates of administration (dd/mm/yyyy): Start date __/__/____ Stop date __/__/____

Duration of therapy (please specify the time unit): _____

Other medications taken during the course of the pregnancy

Medication	Dates of therapy	Dose/day	Route	Indication

II. Patient medical history

Details of ongoing medical conditions:

_____

_____

_____

Obstetric History:

Gravida _____ Para _____ Abortus _____

If any, please give date (year) below:

Full term births _____ Premature births _____

Voluntary abortion _____ Stillbirth _____ Spontaneous abortions _____

Congenital anomalies _____

Points of note in previous pregnancies

Yes ☐ No ☐

If yes, please specified for which pregnancy by indicating the date (year):

Chronic alcohol consumption (___)	Nicotin (___)	Eclamptic toxemia (___)
Failure/insufficiency of the cervix (___)	Diabetes mellitus (___)	Organic diseases (___)
Severe anemia caused by pregnancy (___)	Malposition (___)	Obesity (___)
Infectious diseases (___)	Placenta praevia (___)	Other medication (___)
Arterial hypertension (___)	Vascular disease (___)	Metabolic disorder (___)

Serological test results:

Rubella _____ Syphilis _____ Varicella _____

Toxoplasmosis _____

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III. Pregnancy details

Last menstrual period ___/___/_____ dd/mm/yyyy

Irregular bleeding during conception cycle: Yes ☐ No ☐ Unknown ☐

Probable date of conception ___/___/_____ dd/mm/yyyy

Estimated date of delivery ___/___/_____ dd/mm/yyyy

Diagnosis of pregnancy ___/___/_____ dd/mm/yyyy _____ confirmed via (e.g. ultrasound)

Points of note in current pregnancy

Yes ☐ No ☐

If yes, please provide more details:

☐ Chronic alcohol consumption	☐ Nicotine	☐ Eclamptic toxemia
☐ Diabetes mellitus	☐ Failure/insufficiency of the cervix	☐ Organic diseases
☐ Severe anemia caused by pregnancy	☐ Malposition	☐ Obesity
☐ Infectious diseases *	☐ Placenta praevia	☐ Other medication
☐ Arterial hypertension	☐ Vascular disease	☐ Metabolic disorder

* Please specify: _____

IV. Reporter identification

Please fill out name, specialty, address and contact (phone, e-mail, etc.)

_____

_____

_____

Date : Signature :

PLEASE FAX THIS FORM TO NOVIMMUNE 00 41 22 839 71 51

Report details (for internal use only)

Receipt Date: ___/___/___

Dates of subsequent follow-up information ___/___/___ ___/___/___ ___/___/___

Case Number: _____

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PREGNANCY FORM
COURSE AND OUTCOME OF PREGNANCY

Page 1/2

Study Number:
Patient Number: _____ Initials: _____ Date of birth: ___/___/____

I. Course of pregnancy

Exposure:
Tobacco ☐ _____cig/day Alcohol ☐ ____units/day Substance abuse ☐ Details:

Illness during pregnancy:

PET* ☐ Diabetes ☐ Infection ☐ Other ☐

* PET: Pre-eclamptic toxemia
Other medication taken during pregnancy:

Were treatments at the start of pregnancy continued? No ☐ Yes ☐

Details:

Medication	Dates of therapy (dd/mm/yyyy)	Dose/day	Route	Indication

Hospitalization during pregnancy: Reason(s):

Ultrasound:

Dates and results:

Specific tests — Results:

Retarded growth *in utero*: No ☐ Yes ☐

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II. Delivery

Live newborn: No ☐ Yes ☐ (see below)

	Date (dd/mm/yyyy)		Date (dd/mm/yyyy)
Voluntary abortion	/ /	Spontaneous abortion	/ /
Death *in utero*	/ /	Miscarriage	/ /

Other: _____

Full term birth	/ /	Premature birth	/ /

Specify the type of delivery and treatment(s) received during delivery:

Spontaneous delivery ☐ Caesarean ☐ Treatment(s) received: _____

_____

III. Condition of newborn*

Date of birth (dd/mm/yyyy) / / Gestational age _____ weeks

Sex Male ☐ Female ☐

Initials _____ Weight at birth ____|____ kg

Length _____ cm Head circumference _____cm

APGAR – Index _____ _____

at 1 min at 5 mins

*** In case of multiple births, please provide the information for each baby by using an additional page as required**

IV. Findings at birth

State of health Normal ☐ Abnormal ☐

If abnormal, more details (diagnostic findings to be attached) _____

Details of any special treatment required _____

Newborn followed-up by Dr: _____

Breast-feeding No ☐ Yes ☐

V. Reporter identification

Please fill out name, specialty, address and contact (phone, e-mail, etc.)

_____

_____

_____

Date : Signature :

PLEASE FAX THIS FORM TO NOVIMMUNE 00 41 22 839 71 51

Report details (for internal use only)

Receipt Date: / /

Case Number: _____

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NEWBORN INFANT FORM
COURSE OF FIRST YEAR OF LIFE

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Study Number:
Patient Number: _____ Initials: _____ Date of birth: ___/___/____

I. Course of First year of life

Breastfeeding: No ☐ Yes ☐ Duration: _____

Developmental stages:

Growth: Normal ☐ Abnormal ☐ **Weight gain**: Normal ☐ Abnormal ☐

Physical examination: Normal ☐ Abnormal ☐ _____

Sensory screening: Vision: Normal ☐ Abnormal ☐ Hearing: Normal ☐ Abnormal ☐

Developmental/neurological assessment: Normal ☐ Abnormal ☐

Any Lab results (attach) : At week 6: _____

At week 24: _____

Immunization: _____

Hospitalizations : Yes ☐ No ☐

If yes, reason for each: _____

_____

_____

Medication taken by the infant:

Medication	Dates of therapy	Dose/day	Route	Indication
_____	_____	_____	____	_____
_____	_____	_____	____	_____
_____	_____	_____	____	_____

II. Reporter identification

Please fill out name, specialty, address and contact (phone, e-mail, etc.)

_____

_____

_____

Date: Signature :

PLEASE FAX THIS FORM TO NOVIMMUNE 00 41 22 839 71 51

Report details (for internal use only)

Receipt Date: ___/___/____

Case Number: _____

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