# Effect of N-803 on B Cell Follicles in Antiretroviral Treated HIV Disease

**Sponsored by:**
NIH

**DAIDS-ES Document Number:** DAIDS-ESID 38604 (DARE IL-15)

**NIAID Funding Mechanism:** NIH 5UM1AI126611

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ImmunityBio., Inc.

**IND Sponsor:**
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**IND Number:**
125191

**STUDY PHASE:**
**1B**

**Version Number:**
Version 4.4

**Day Month Year**
21 January 2022

# PROTOCOL VERSION HISTORY

| Version Number | Date | Significant Changes |
|---|---|---|
| 1.0 | 10/17/2018 | Original document |
| 1.1 | 01/07/2019 | Changes to background and other modifications requested by DAIDS |
| 1.2 | 04/01/2019 | Changes per DAIDS CSRC comments |
| 1.3 | 07/16/2019 | Changes per DAIDS CSRC May 20, 2019 comments |
| 1.4 | 08/20/2019 | Added EKG evidence for ischemia to early stopping rules and more detail re: prolonged QTC interval per ProPEP April 1, 2019 comments |
| 1.5 | 10/01/2019 | Changes per DAIDS Full regulatory review from 9/20/2019 |
| 1.6 | 10/10/2019 | Added Nant Reporting Requirements |
| 1.7 | 01/16/2020 | Removed UCSF as study site, added Hennepin Healthcare study site, replaced NantKwest with ImmunityBio |
| 1.8 | 04/09/2020 | Per IRB stipulations, noted the study personnel performing study specific procedures. |
| 1.9 | 08/28/2020 | Per FDA, added additional EKG testing at specified visits, clarified language re: safety reporting of all enrolled participants, (7.16)<br>Personnel change for Biostatistician<br>Added COVID-19 testing protocol (7.18)<br>Indicated external monitor will conduct site initiation visit |
| 2.0 (formerly 1.10) | 10/16/2020 | Modified details about COVID-19 testings |
| 2.1 | 11/10/2020 | Added Joshua Rhein as Co-I; revised instructions about drug administration (5.2.3) |
| 2.2 | 11/19/2020 | Addressed comments from NIH: added instructions to match the ICF regarding isolating at home after PCR test and before injections and biopsies |
| 3.0 | 1/6/2021 | Added rectal swabs and stool collection at baseline, day 7, day 28, day 49, post study drug procedures, and 90 days after 3rd dose follow-up visits; added windows for the follow-up visits |
| 4.0 | 1/25/2021 | Removed community member from the Safety Monitoring Committee (SMC) in section 14.7 |
| 4.1 | 2/8/2021 | Changed the inclusion criteria to be on ART for at least 24 months rather than between 12 and 48 months; added the SARS-CoV-2 vaccine in sections 4.3 and ll.4 |
| 4.2 | 4/9/2021 | Contraindications clarified in section 6.2; added a range of colonoscopy of 14-18 tissue biopsies in section 7.13; added collection of adjacent adipose tissue during the LN biopsy in section 7.14; added storage and DNA and RNA in section 7.17; removed rectal swabs and modified the stool collection schedule; added details about the screening blood tests in section 8.1 |

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| 4.3 | 11/29/2021 | Added a large volume blood draw if the participant is not eligible for leukapheresis in sections 4.3, 7.12, 8.2, 8.9 |
| 4.4 | 1/21/2022 | Modified details about COVID-19 testing, including an exception to COVID-19 screening for 90 days following a positive diagnosis. |

# SIGNATURE PAGE

I have read the protocol specified below. In my formal capacity as Investigator, my duties include ensuring the safety of the study participants enrolled under my supervision, as outlined in the protocol. It is understood that all information pertaining to the study will be held strictly confidential and that this confidentiality requirement applies to all study staff at this site. I will conduct the study in accordance with the provisions of this protocol and all applicable protocol-related documents. I agree to conduct this study in compliance with United States (US) Health and Human Service regulations (45 CFR 46); applicable U.S. Food and Drug Administration regulations; standards of the International Conference on Harmonization Guideline for Good Clinical Practice (E6); Institutional Review Board/Ethics Committee determinations; all applicable in-country, state, and local laws and regulations; and other applicable requirements (e.g., US National Institutes of Health, Division of AIDS) and institutional policies.

Principal Investigator: Timothy W. Schacker
Print/Type

Signed:  Date: 1/21/2022
Name/Title

## Table of Contents

# LIST OF ABBREVIATIONS

| | |
|---|---|
| ACC/AHA | American College of Cardiology/American Heart Association |
| ADCC | Antibody-dependent cell-mediated cytotoxicity |
| AE | Adverse Event |
| AIDS | Acquired Immune Deficiency Syndrome |
| ALT/SGPT | Alanine Aminotransferase |
| ART | Antiretroviral Therapy |
| ASCVD | Atherosclerotic Cardiovascular Disease |
| AST/SGOT | Aspartate Aminotransferase |
| BP | Blood Pressure |
| BUN | Blood Urea Nitrogen |
| CA | Cell-associated |
| CLIA | Clinical Laboratory Improvement Amendments |
| ConA | Concanavalin A |
| CD | Cluster of Differentiation |
| cDNA | Complementary DNA |
| CFR | Code of Federal Regulations |
| CMV | Cytomegalovirus |
| CNS | Central Nervous System |
| COVID-19 | Coronavirus Disease 2019 |
| CRF | Case Report Form |
| CRP | C-reactive Protein |
| CS | Clinical Significance |
| CSR | Clinical Study Report |
| CSRC | DAIDS Clinical Science Review Committee |
| CTL | Cytotoxic T Cell |
| CXCR5 | CXC chemokine receptor type 5 |
| DAIDS | Division of Aids, NIH |
| DARE | Delaney AIDS Research Enterprise |
| DNA | Deoxyribonucleic Acid |
| E/CIA | HIV Enzyme or Chemiluminescence Immunoassay |
| EDITS | Envelope Detection by Induced Transcription-based Sequencing |
| EKG | Electrocardiogram |
| FACS | Fluorescence-activated cell sorting |
| FDA | Food and Drug Administration |
| FDC | Follicular Dendritic Cells |
| GALT | Gut Associated Lymphoid Tissue |
| GCP | Good Clinical Practice |
| HCG | Human chorionic gonadotropin |
| HIPAA | Health Insurance Portability and Accountability Act |
| HIV | Human Immunodeficiency Virus |
| HLA | Human Leukocyte Antigen |
| IB | Investigator's Brochure |
| ICF | Informed Consent Form |
| ICH | International Conference on Harmonisation |
| Id2 | DNA-binding protein inhibitor |

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| IEC | Independent Ethics Committee |
| Ig | Immunoglobulin |
| IL-2 | Interleukin-2 |
| IL-15 | Interleukin-15 |
| IND | Investigational New Drug |
| IRB | Institutional Review Board |
| ISM | Independent Safety Monitor |
| IUD | Intrauterine device |
| IV | Intravenous |
| Kg | Kilogram |
| KIRS | Killer-cell Immunoglobulin Receptors |
| LDMS | Laboratory data management system |
| LN | Lymph node |
| LT | Lymphatic Tissues |
| MHC-I | Major Histocompatibility Complex Class 1 |
| mL | Milliliter |
| µg | Microgram |
| MO | DAIDS Medical Officer |
| MOLT | Cell line |
| mQVOA | Quantitative Viral Outgrowth Assay |
| MTD | Maximum Tolerated Dose |
| N-803 | IL-15 superagonist (Nant-803, formerly ALT-803) |
| NCI | National Cancer Institute |
| Nef | Negative Regulatory Factor |
| NGS | Next Generation Sequencing |
| NHP | Non-human Primate Model |
| NIAID | National Institute of Allergy and Infectious Diseases |
| NIH | National Institutes of Health |
| NK | Natural Killer |
| PB | Peripheral Blood |
| PBMC | Peripheral Blood Mononuclear Cell |
| PCR | Polymerase Chain Reaction |
| PD-1 | Programmed Cell Death Protein 1 |
| PE | Physical Exam |
| PFT | Pulmonary Function Test |
| PI | Principal Investigator |
| PO | DAIDS Program Officer |
| PRD | Participant Reminder Diary |
| pVL | Plasma Viral Load |
| qPCR | Quantitative polymerase chain reaction |
| QT/QTc | Heart-rate Corrected QT(QTc) Interval |
| RNA | Ribonucleic Acid |
| SAE | Serious Adverse Event |
| SAP | Statistical Analysis Plan |
| SEM | Standard error of the mean |
| SIV | Simian Immunodeficiency virus |

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| SMC | Safety Monitoring Committee |
| SQ | Subcutaneous |
| TB | Tuberculosis |
| TEM | Effector T cell Memory Subset |
| TFh | T Follicular Helper cell |
| TIA | Transient Ischemic Attack |
| TID | Three times a Day |
| TIGIT | T-cell immunoreceptor with Ig and ITIM domains |
| Treg | Regulatory T cell |
| TTM | Transitional T cell Memory Subset |
| UCSF | University of California, San Francisco |
| UMN | University of Minnesota |
| vRNA | Viral RNA |

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# PROTOCOL SUMMARY

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|---|---|
| **TITLE** | Effect of N-803 on B Cell Follicles in Antiretroviral Treated HIV Disease |
| **SPONSOR** | University of Minnesota |
| **PRINCIPLE INVESTIGATOR** | Timothy Schacker, MD |
| **FUNDING ORGANIZATION** | NIAID: NIH 5UM1AI126611 |
| **NUMBER OF SITES** | University of Minnesota<br>Hennepin Healthcare |
| **RATIONALE** | HIV-specific CD8+ T cells are the primary mechanism by which the virus is controlled. CD8+ T cells and other effector cells, including NK cells, are excluded from B-cell follicles. As a consequence, HIV replicates at higher titers in the follicles. This reservoir for the virus and may prove to be difficult to target with immune-based curative interventions but therapies that allow effector cells (i.e., CD8+ T cells or NK cells) to access this reservoir will likely be needed for many of the emerging curative approaches.<br>The interleukin-15 (IL-15) superagonist N-803 (previously ALT-803) induces the expression of CXCR5 on CD8+ T cells, allowing them to access to B cell follicles. In a recent study by Webb et al, N-803 administration to SIV infected macaques was associated with increased frequency of SIV specific CD8 T cells in B cell follicles in spatial proximity to SIV infected T follicular helper (TFh) cells.<br>In an ongoing Phase 1 dose escalation study of N-803 in HIV-infected adults on effective ART, we have thus far found in peripheral blood:<br>1. There is activation and proliferation of CD4 and CD8 T cells and NK cells.<br>2. HIV transcription is induced at all doses tested.<br>3. There was a significant reduction in the frequency of cells with an inducible provirus after 3 doses of drug that persisted up to 6 months.<br>Based on these findings, we hypothesize that the mechanism of reservoir reduction is proliferation and activation of HIV specific CD8 T cells that migrate into follicles and clearance of virus producing cells. |
| **STUDY DESIGN** | A phase 1B single arm study of 10 HIV-infected adults on effective ART will be performed. This is a two-center, non-randomized, open label, and uncontrolled study. |

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| | All participants will undergo an extensive baseline evaluation 7-14 days before the first dose that will include an excisional biopsy of a lymph node, colonic biopsies, and leukapheresis. Participants will then receive three doses of N-803 administered every 21 days. A second excisional biopsy will be performed between 7-14 days after the final dose. The study drug N-803 will be administered at 6 mcg/kg, which is the maximum tolerated dose determined in a recently completed dose-escalation study. |
| **PRIMARY OBJECTIVE** | • To determine the impact of N-803 on the frequency and function of CD8+ T cells in B cell follicles.<br>• To determine the safety of N-803 HIV+ ART suppressed individuals by assessment of adverse events experienced by participants during the conduct of the trial. |
| **SECONDARY OBJECTIVES** | • To determine the impact of IL-15 on the frequency, location, and phenotype of vRNA+ and vDNA+ cells in lymphoid tissues. |
| **EXPLORATORY OBJECTIVES** | • To determine the effect of N-803 on frequency of T follicular helper cells harboring intact HIV genomes<br>• To determine the effect of N-803 on frequency and activation status of NK cells in tissue and blood<br>• Impact of N-803 on Antibody-dependent cell-mediated cytotoxicity (ADCC) in tissues and blood.<br>• Impact of N-803 on HIV specific cytotoxic T cell (CTL) in tissues and blood.<br>• Impact of N-803 on detection of HIV RNA in plasma |
| **RATIONALE FOR TARGET POPULATION** | We carefully considered the ideal target population for this trial to maximize the impact of the study.<br>The role of the B cell follicle as a potential sanctuary for HIV replication and persistence has been demonstrated in untreated SIV-infected macaques, particularly those with low-level viremia ("elite" controllers). Data in humans are more limited, but Banga and colleagues found replication-competent HIV to be highly enriched in the PD-1 expressing TFh cells that reside in the germinal centers of the B cell follicle. These inflammatory centers are more readily apparent during early ART.<br>To enhance our capacity to characterize the impact of IL-15 on follicular integrity and the size of the reservoir we will enroll individuals who started therapy during chronic infection (and hence likely have a relatively high reservoir size). |

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| **NUMBER OF PARTICIPANTS** | Ten HIV-infected adults on short-term effective ART will be studied. |
| **SUBJECT SELECTION CRITERIA** | Key Inclusion Criteria:<br>1. Male or female, age ≥ 18 and ≤ 65 years<br>2. HIV-1 infection (see section 4.2)<br>3. On continuous antiretroviral therapy for at least 24 months without any interruptions of greater than 14 consecutive days, without plans to modify ART during the study period<br>4. Plasma HIV RNA levels < 20 copies/mL at screen and on at least one determination in past 12 months (isolated single values ≥ 20 but < 200 copies/mL will be allowed if they were preceded and followed by undetectable viral load determinations)<br>5. Screening CD4+ T-cell count ≥ 350 cells/$mm^3$ and nadir CD4+ T cell count of >200 per participant report<br>Key Exclusion Criteria:<br>1. Pregnant, breastfeeding, or unwilling to practice birth control during participation in the study<br>2. Active or recent malignancy requiring systemic chemotherapy or surgery in the preceding 36 months or for whom such therapies are expected in the subsequent 12 months; minor surgical removal of localized skin cancers (squamous cell carcinoma, basal cell carcinoma) are not exclusionary<br>3. Chronic liver disease defined as Class B and C on the Child-Pugh scale<br>4. Active and poorly controlled atherosclerotic cardiovascular disease (ASCVD), as defined by 2013 ACC/AHA guidelines, including a previous diagnosis of any of the following: (a) acute myocardial infarction, (b) acute coronary syndromes, (c) stable or unstable angina, (d) coronary or other arterial revascularization, (e) stroke, (f) transient ischemic attack (TIA), or (g) peripheral arterial disease presumed to be of atherosclerotic origin.<br>5. History of potential immune-mediated medical conditions requiring concomitant treatment with immunomodulatory drugs, and/or exposure to any immunomodulatory drug in the 30 days prior to study enrollment<br>6. Exposure to any experimental therapies within 90 days of study screening. Exposure to long acting injectable ART therapies is not exclusionary.<br>7. Clinical vaccination administered within 14 days of screen |

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| **TEST PRODUCT, DOSE, AND ROUTE OF ADMINISTRATION** | • N-803, provided by ImmunityBio., Inc. administered subcutaneously at a dose 6 mcg/kg on Days 0, 21, and 42 (with a dosing window of up to 14 days post the planned dosing day) |
| ***DURATION OF SUBJECT PARTICIPATION AND DURATION OF STUDY*** | Participants will be observed on study for up to six months. |
| **CONCOMITANT MEDICATIONS** | All participants should be maintained on a stable antiretroviral drug regimen through the end of the study. |
| **PRIMARY ENDPOINTS** | • Frequency of CD8+ T cells in follicles (and their phenotypes) before and after N-803 therapy will be measured using quantitative image analysis techniques we have developed in our lab (10, 11, 15, 39-41). Briefly we will stain cells with antibodies to CD8 and determine their frequency per unit area. We have used these techniques to account for changes in CD4 T cell populations in the parafollicular T cell zone after antifibrotic therapy (42).<br>• Safety of N-803 given at this dose and frequency in this population. Any participant who receives an injection of N-803 will be followed for AEs. Clinical and laboratory adverse events will be recorded in the CRF and the study will have quarterly monitoring by the team of monitors in the University of Minnesota Clinical and Translational Science Institute Regulatory Core. This group monitors all FDA trials at the University of Minnesota and routinely monitors multi-site trials at different institutions. Every 14 days, the co-investigators will review any and all clinical and laboratory adverse events to ensure compliance with the study pause criteria listed in section 11.4. |
| **SECONDARY ENDPOINTS** | • Frequency, location, and phenotype of vRNA+ and vDNA+ cells in lymphoid tissues.<br>• Frequency of vRNA+ TFH cells in B cell follicles before and after N-803 therapy.<br>• Amount of HIV RNA/DNA detected in follicles before and after N-803 therapy. |
| **EXPLORATORY ENDPOINTS** | • Changes in frequency and activation status of NK cells in tissue and blood<br>• Impact of N-803 on ADCC<br>• Impact of N-803 on HIV specific CTL in tissues and blood<br>• Structure and integrity of the FDC network before and after N-803 therapy |

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| **SAFETY EVALUATIONS** | All participants will be followed for possible adverse events (AEs) throughout their involvement in the study. Routine blood work will be performed on a regular basis (see sections 7.5 and 8). AEs will be graded according to Corrected Version 2.1 (July 2017) of the NIH/NIAID Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events. |
| **PLANNED INTERIM ANALYSES** | A Safety Monitoring Committee (SMC) will be convened and will meet biannually to review all adverse events and the conduct of the study. Serious adverse events will be monitored by the committee on an ongoing basis throughout the study. |

# 1. INTRODUCTION / BACKGROUND AND SCIENTIFIC RATIONALE

## 1.1. Overview

N-803 (formally ALT-803) is an IL-15 receptor super-agonist (IL-15/IL-15Rα-Fc) that is in clinical trials for hematologic and other malignancies. It is a potential candidate for HIV reservoir reduction studies as it can activate CD4 T cells, reactivating virus from latency, but also expand and activate CD8 and NK cells providing an increased pool of potential effector cells. Important to this proposal it also has been shown in at least one model of a non-human primate model of SIV infection to increase the frequency of SIV specific CD8 T cells in B cell follicles of secondary lymphoid tissues (LT). N-803 was used in a Phase 1 trial for HIV infection at the University of Minnesota with Dr. Schacker as the PI. This trial in individuals who are HIV-infected was a dose-escalation trial to establish the safety and tolerability of the drug in this population. There are 3 formulations of IL-15 that are in clinical trials for treatment of various solid and hematologic malignancies (**Fig 1**). The first is a formulation from NCI that is just the IL-15 molecule, the second is the IL-15 molecule that is fused to the IL-15 receptor, which is in clinical trials at Novartis, and third is the ImmunityBio., Inc. molecule that is an IL-15/IL-15Rα-Fc superagonist complex that contains a mutation that increases avidity of the molecule for IL-2Rβγ on NK cells and also includes the IgG1Fc domain (1-3). These modifications in N-803 increase the half-life and stability of the drug and also provide better penetration of the drug into lymphatic tissues (LT) (4) where > 99% of the virus reservoir resides (5). All of the data presented here were generated using N-803.

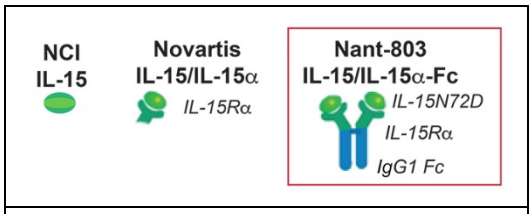


**Fig 1: IL-15 drugs in clinical trials**.

N-803 is ideally suited to be part of an HIV cure strategy for several reasons. First, we have shown in our dose escalation study, described below, that even at the lowest doses studied, N-803 can induce HIV transcription. Secondly, N-803 is associated with increased frequency of activated NK and CD8 cells (6) to clear virus infected cells. In addition, in the non-human primate model (NHP) of SIV infection, N-803 is associated with an increased frequency of SIV-specific CD8-T cells in B cell follicles where virus infected TFh reside(7). Thus, we have identified a single intervention with potential to both induce virus from latency and to clear clear virus infected cells. In this study, we focus on the effect of N-803 on B cell follicle structure and function and on the ability of the drug to reduce the reservoir of infected cells in this specific virus reservoir.

**N-803 induces HIV transcription**

As mentioned above, we have completed an IRB and FDA approved Phase 1 clinical trial (IND 125191) to determine the safety and tolerability of N-803 in HIV infected people where Dr. Schacker was the Principal Investigator. The study was a dose-escalation trial to establish the maximum tolerated dose (MTD) of N-803 in HIV infected ART suppressed individuals. Two individuals were studied at 0.3 mcg/kg. A total of 3 individuals were studied at each subsequent dose level with the first dose level: 1, 3, and 6 mcg/kg. The protocol design allowed an increase to 10 mcg/kg as the final dose tested however the study was concluded after the 6 mcg/kg dose cohort because of the increased frequency of myalgia's and other flu-like symptoms at the higher doses tested. The protocol was initially designed for drug to be administered intravenously however while we were completing the 0.3 mcg/kg dose level it was learned that subcutaneous (SQ) administration of N-803 provided better penetration into lymph nodes and was

associated with fewer systemic symptoms of fever, myalgia. The 1, 3, and 6 mcg/kg doses of N-803 were administered SQ.

In addition to the primary safety endpoints, additional virologic and immunologic data was obtained to determine the immunologic activity of N-803 in these participants and if there was an impact on HIV reservoirs. We first noted that measures of plasma HIV-RNA became intermittently detectable after doses of N-803. This observation was not consistent nor was it dose dependent. However, Dr. Karn of Case Western Reserve University, a co-investigator on these studies, has recently developed a novel Next Generation Sequencing (NGS)-based protocol, called EDITS (Envelope Detection by Induced Transcription-based Sequencing), to measure inducible cell-associated HIV RNA (8). Sequences of envelope are detected using a nested PCR on 1.25 x$10^6$ resting memory cells from aviremic HIV-1-infected participants before and after ConA stimulation. Samples from different patients are bar-coded, pooled, and sequenced simultaneously which saves both time and sequencing costs, and allows accurate comparisons since input cDNA levels are effectively normalized. All EDITS experiments include control reactions set up without the addition of reverse transcriptase, as well as no template controls. In addition, this assay is quantitative because latently infected cells carry, on average, only one provirus (9) and therefore the frequency of inducible RNAs is proportional to the numbers of inducible cells in each of the wells. Comparing the reads obtained in the EDITS assay after maximal activation of cells by treatment with the mitogenic lectin ConA to a standard curve obtained by sorting known numbers of productively infected cells (**Fig 2**), provides an accurate measurement of the number of latently infected cells in any patient sample. Using this method, we are able to accurately detect 20% changes in the pool of cells with inducible HIV RNA.

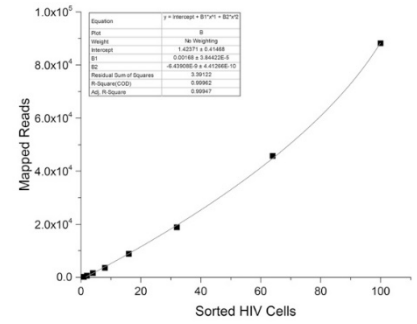


**Fig 2. Edits assay to measure inducible cell-associated HIV RNA.**

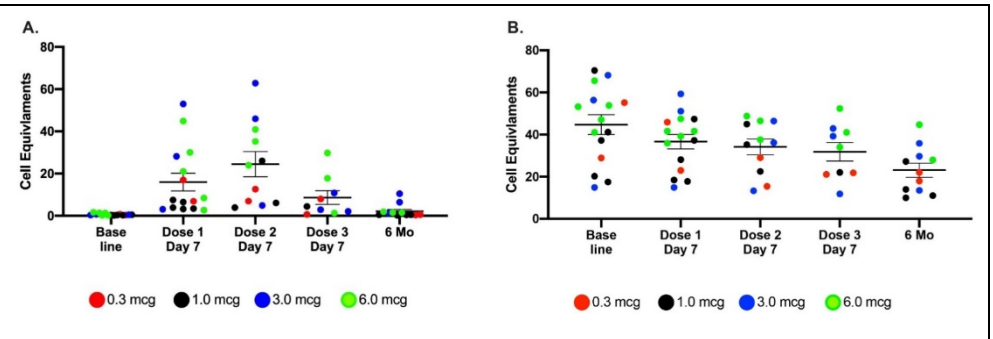


**Fig. 3: Mean frequency and SEM of transcription events in PBMC from 8 participants receiving N-803 before (A) and after stimulating PBMCs with ConA (B).**

The EDITS assay was performed on PB samples collected at baseline and again at 7 days after receiving a dose of N-803 from all 15 participants. Another sample was obtained at least 6 months after the last dose of N-803. Prior to Con A stimulation (**Fig 3A**) we measured a significant increase in HIV transcription after the first 2 doses but after dose 3 there were fewer transcription events. At the 6-month time-point the frequency of cells transcribing HIV was back to baseline. With ConA stimulation, (**Fig 3B**) the population of inducible cells was largest at baseline (before N-803) and trended lower after each subsequent dose and remained stable at the 6-month time-point. The 6 mcg/kg dose gave the most consistent increase in transcription and is the reason we are using 6 mcg/kg in this trial. Note, the interval between doses was 2 weeks for the 6 mcg/kg dose and 1 of the participants receiving 3.0 mcg/kg because

of the longer duration of the rash and adenopathy at the higher dose. In Figure 3 A and B we combine the data for simplicity in presentation of data to justify the 6 mcg/kg dose.

**N-803 induces CD4, CD8, and NK cell activation and proliferation**

We are interested in studying the impact of N-803 on B cell follicle function for several reasons. The first is that the lymphoid tissue (LT) compartment is where the primary reservoir resides (5, 10-16). While many of the vRNA+ cells are located in the parafollicular T cell zone (TZ), a significant proportion of these cells are in the B cell follicle in the T follicular helper cells (Tfh). Importantly, effector cells are excluded from the B cell follicle (7) but at least one study in NHP showed that animals treated with N-803 had an increased frequency of SIV specific CD8 T cells in the follicles. N-803 has been shown to activate CD4 and CD8 T cells. In vitro, Sekaly and colleagues showed that IL-15 increases CD8 T cell proliferation by up-regulating the expression of Id2, a transcriptional adaptor that is critical for inducing the proliferation and differentiation of CD8 memory T cells (17, 18). IL-15 induces proliferation of all CD8 T cell memory subsets and the differentiation and activation of the transitional (TTM) and effector (TEM) memory subsets (18).

N-803 has been shown to activate NK cells which is important in HIV infection. There is an association between possession of specific killer-cell immunoglobulin-like receptors (KIRs) on NK cells and control of HIV infection (19-22). HIV selectively modulates MHC-I molecules on the infected cell surface which, in turn, impairs NK cell killing. The HIV protein Nef down-regulates HLA-A and –B, but not HLA-C or –E (23, 24) which reduces the susceptibility of infected cells to recognition by most virus-specific T cells. The retention of HLA-C and –E impairs the response by NK cells that express the inhibitory KIRs KIR2DL (KIR2DL1/2/3) and CD94/NKG2A, which are the receptors for HLA-C and –E (respectively) (25, 26). However, activation of NK cells through cytokine stimulation can lower the activation threshold and overcome this inhibitory receptor dampening to induce NK mediated target cell killing and cytokine secretion (27). While there are many cytokines and combinations of cytokines that will activate NK cells (e.g., IL-2 & IL-12)(27, 28), IL-15 is particularly appealing as it is necessary for NK cell homeostasis (29-31) and will not cause expansion of regulatory T cells (Tregs) that can suppress NK cell responses like other cytokines might (e.g., IL-1) (32, 33).

N-803 may also restore CD8 T cell responses and cause activation and proliferation of both CD4 T and CD8 Tcells. In vitro, Sekaly and colleagues showed that IL-15 increases CD8 T cell proliferation by up-regulating the expression of Id2, a transcriptional adaptor that is critical for inducing the proliferation and differentiation of CD8 memory T cells (17, 18). IL-15 induces proliferation of all CD8 T cell memory subsets and the differentiation and activation of the transitional (TTM) and effector (TEM) memory subsets (18).

In SIV+ Rhesus macaques, N-803 therapy is associated with significant reductions in plasma viremia. In Rhesus macaques infected with SIVmac239 for > 1 year, who were given N-803 at 0.1 mg/kg weekly for 3 weeks, there was a rapid decline in plasma viremia to undetectable 1 week after the first dose (**Fig 4**) that correlated with rapid increases of CD8 T cell and NK cell numbers suggesting that NK cells and CD8 T cells may have been responsible for the rapid reduction in plasma viremia (34).

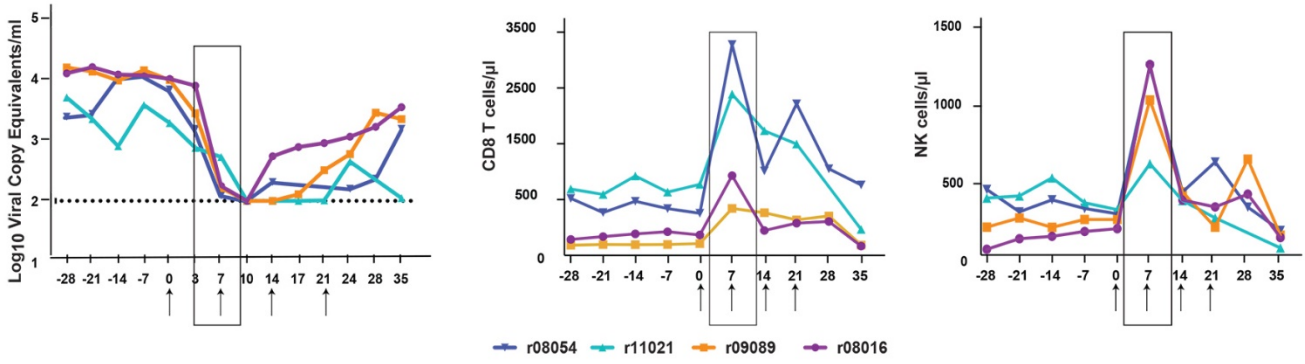


**Fig 4: N-803 reduces viral loads in the absence of ARVs**. The effect is temporally associated with expansion of the CD8 T cell and NK cell population, each arrow represents a dose of N-803. The x-axis represents days.

**Side Effect Profile and adverse events associated with N-803 in patients with cancer**

To date a total of 250 patients in 20 cancer related clinical trials have received multiple doses of N-803. The dosing range was 0.1 mcg/kg – 20 mcg/kg and included both IV and SQ routes of administration. These trials included patients with pancreatic cancer, refractory multiple myeloma, non-Hodgkin's lymphoma, bladder cancer, metastatic non-small cell lung cancer, CEA expressing cancer, Merkel Cell carcinoma, hematologic malignancies, and ovarian cancer. Of those 20 trials, 7 have completed sufficient accrual to report preliminary efficacy data. Most of these trials were N-803 in combination with other interventions (e.g., checkpoint inhibitors, rituximab, etc). All of the trials reporting efficacy data were dose-escalation trials, typically starting at very low doses and there is no data reported about dose-effect. The reported data are in **Table 1**.

**Table 1: Efficacy data for N-803 in cancer trials that has been reported to date**

| NCT# | Tumor Type | N | CR | PR | SD | DP |
|---|---|---|---|---|---|---|
| 01946789 | Solid Tumors | 26 | 0 | 0 | 0 | |
| 0255967 | Pancreatic | 7 | 0 | 1 | 1 | |
| 02138734 | Bladder* | 9 | 100 | | | |
| 02099539 | Refractory Myeloma | 19 | | | | 15 |
| 02384954 | Non-Hodgkin's Lymphoma | 21 | 7 | 3 | 8 | 3 |
| 01885897 | Relapsing hematologic malignancy | 33 | 1 | 5 | | |
| 02523469 | Non-small cell lung cancer | 23 | 6 (reported as overall response rate) | | | |

CR=complete response, PR=partial response, SD=stable disease, and DP=disease progression

* Bladder study data show % response, not number of subjects. Due to nature of the study and the types of cancer seen in subjects as part of this study, there is no such thing as a CR. 9/9 subjects had either a CR or a lack of recurrence/progression

A total of 3 of the studies reported data on the effect of N-803 on CD8 T cells and NK cells (no data reported on CD4 T cells). In each of the 3 trials CD8 T cell numbers in PBMC modestly expanded and NK cells were significantly increased.

In **Table 2** we present the clinical AEs that were observed in the trial for people with solid tumors (NCT 01946789). The dose escalation scheme was similar to our dose-escalation trial in people with HIV infection and representative of the other AEs reported in the other cancer trials. There has been a total of 37 serious AEs reported in the cancer trials. Of those, 9 were associated with IV infusion, 24 with subcutaneous injection, and 4 by intravesical instillation (for bladder cancer). In **Table 3** we present adverse events encountered in a healthy volunteer study conducted by the pharmaceutical company. Note, we do not have attribution data to know if these adverse events were felt to be associated with N-803.

**Table 2: Clinical Adverse Events Associated with N-803 in patients with cancer, note that this table shows the highest grade value AE observed among all reported incidents for a particular AE within each dose cohort.**

| **Intravenous N-803** | | | **Highest AE Grade/Dose Cohort** | | | |
|---|---|---|---|---|---|---|
| Adverse Event | **# IV Subjects Affected, Total (N=11)** | **# IV Subjects Affected, 3 & 6 μg/kg (N=6)** | **0.3-0.5 μg/kg** | **1 μg/kg** | **3 μg/kg** | **6 μg/kg** |
| Fatigue | 6 (55%) | 3 (50%) | 1 | 2 | 1 | 1 |
| Nausea | 6 (55%) | 3 (50%) | 2 | 1 | 1 | 1 |
| Vomiting | 4 (36%) | 2 (33%) | 1 | 2 | 1 | - |
| Chills | 4 (36%) | 1 (17%) | 1 | 1 | - | 2 |
| Fever | 3 (27%) | 2 (33%) | 1 | - | 1 | 2 |

**Table 2 continued**

| **Subcutaneous N-803** | | | **Highest AE Grade/Dose Cohort** | | | |
|---|---|---|---|---|---|---|
| **Adverse Event** | **# SubQ Subjects Affected, Total (N=13)** | **# SubQ Subjects Affected, 15 & 20 μg/kg (N=7)** | **6 μg/kg** | **10 μg/kg** | **15 μg/kg** | **20 μg/kg** |
| Injection site reaction | 11 (85%) | 7 (100%) | 2 | 2 | 2 | 1 |
| Fatigue | 7 (54%) | 4 (57%) | - | 2 | 2 | 2 |
| Hypoalbuminemia | 6 (46%) | 4 (57%) | 2 | 2 | 2 | 2 |
| Anemia | 5 (38%) | 4 (57%) | - | 2 | 3 | 2 |
| Fever | 5 (38%) | 2 (29%) | 1 | 2 | 2 | 2 |
| Lymphocyte count decreased | 4 (31%) | 3 (43%) | - | 3 | 4 | 2 |
| Limb Edema | 3 (23%) | 3 (43%) | - | - | 1 | 1 |
| Anorexia | 3 (23%) | 2 (29%) | - | 2 | 2 | 2 |
| Arthralgia | 3 (23%) | 2 (29%) | - | 2 | 1 | - |

| | | | | | | |
|---|---|---|---|---|---|---|
| Vomiting | 3 (23%) | 2 (29%) | - | 2 | 1 | - |

**Table 3: Incidence of Treatment-Emergent Adverse Events by Study Period, MedDRA System Organ Class, and Preferred Term (Occurring in >1 Subject) in Healthy Volunteers– note that all incidences of AEs reported in this table are graded ≤2**

| | **Study Period 1 10 µg/kg** | | **Study Period 2 20 µg/kg** | |
|---|---|---|---|---|
| **System Organ Class Preferred Term** | **N-803 1.0 mg/mL n = 10** | **N-803 2.0 mg/mL n = 10** | **N-803 1.0 mg/mL n = 7** | **N-803 2.0 mg/mL n = 7** |
| Subjects with at least 1 AE | 10 (100%) | 10 (100%) | 7 (100%) | 7 (100%) |
| Blood and lymphatic system disorders | 2 (20%) | 2 (20%) | 1 (14%) | 1 (14%) |
| Lymphadenopathy | 2 (20%) | 2 (20%) | 1 (14%) | 1 (14%) |
| Gastrointestinal disorders | 3 (30%) | 8 (80%) | 6 (86%) | 4 (57%) |
| Abdominal pain lower | 3 (30%) | 7 (70%) | 5 (71%) | 3 (43%) |
| Vomiting | 0 | 0 | 0 | 3 (43%) |
| General disorders and administration site conditions | 10 (100%) | 10 (100%) | 7 (100%) | 7 (100%) |
| Axillary pain | 2 (20%) | 1 (10%) | 0 | 1 (14%) |
| Chest pain | 0 | 0 | 2 (29%) | 1 (14%) |
| Chills | 8 (80%) | 8 (80%) | 6 (86%) | 7 (100%) |
| Fatigue | 2 (20%) | 0 | | |
| Feeling of body temperature change | 3 (30%) | 7 (70%) | 2 (29%) | 4 (57%) |
| Injection site reaction | 10 (100%) | 10 (100%) | 7 (100%) | 7 (100%) |
| Malaise | 0 | 3 (30%) | 0 | 1 (14%) |
| Pyrexia | 4 (40%) | 4 (40%) | 6 (86%) | 5 (71%) |
| Musculoskeletal and connective tissue disorders | 4 (40%) | 4 (40%) | 3 (43%) | 5 (71%) |
| Back pain | 1 (10%) | 0 | 0 | 4 (57%) |
| Myalgia | 3 (30%) | 4 (40%) | 3 (43%) | 2 (29%) |
| Nervous system disorders | 6 (60%) | 6 (60%) | 4 (57%) | 6 (86%) |
| Dizziness | 0 | 0 | 1 (14%) | 2 (29%) |
| Headache | 6 (60%) | 6 (60%) | 4 (57%) | 6 (86%) |
| Respiratory, thoracic and mediastinal disorders | 2 (20%) | 2 (20%) | 4 (57%) | 1 (14%) |
| Cough | 2 (20%) | 1 (10%) | 3 (43%) | 1 (14%) |
| Skin and subcutaneous tissue disorders | 3 (30%) | 1 (10%) | 1 (14%) | 2 (29%) |
| Night sweats | 2 (20%) | 1 (10%) | 1 (14%) | 2 (29%) |

**Pharmacokinetic Profile in Cancer Studies**

Pharmacokinetic data were obtained pre-dose and at 30 minutes, 2, 4, 8 and 24 hours after N-803 dosing in the solid tumor trial discussed above (NCT01946789) where participants received either IV or SQ dosing. PK data were obtained from 2 participants in each dose cohort according to the study protocol (**Fig 5**). The $T_{max}$ occurred consistently at 30 minutes after IV administration. By contrast, SQ N-803 exhibited a very gradual increase in serum concentration, with maximal serum levels >100-fold less than the average peak after IV N-803. In 4 of 8 SQ subjects, the serum concentration peaked at 4 to 8 hours, but in the remaining 4 subjects, the highest concentration measured occurred at 24 hours, the last time point tested. Therefore, it is possible that the $T_{max}$ occurred either at or after 24 hours in these 4 subjects.

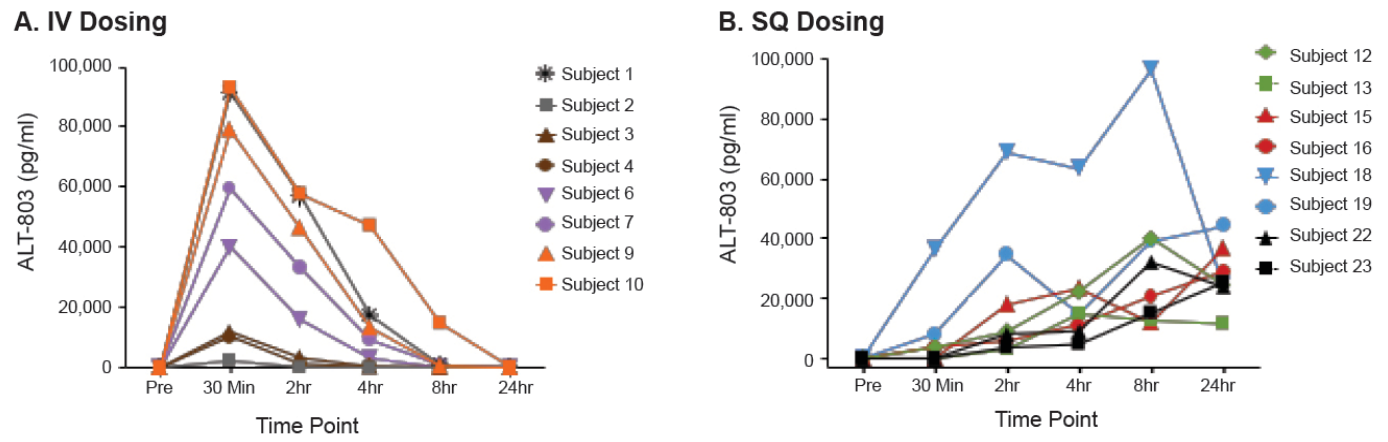


**Fig 5: Panel A ALT-803** (AKA N-803) IV dose levels for subjects 1 – 10 are as follows: 1 = 0.3 µg/kg; 2 = 0.5 µg/kg; 3 and 4 = 1 µg/kg; 6 and 7 = 3 µg/kg; 9 and 10 = 6 µg/kg. (4.2-2) ALT-803 (AKA N-803) **Panel B** SQ dose levels for subjects 12 – 23 are as follows: 12 and 13 = 6µg/kg; 15 and 16 = 10 µg/kg; 18 and 19 = 15 µg/kg; 22 and 23 = 20 µg/kg.

**Side-effect profile and adverse events associated with N-803 in HIV infected people**

Clinically the drug has been relatively well-tolerated. All participants dosed SQ experience an injection site reaction that resolves in 7-10 days. All of the HIV + patients dosed at 1 mcg/kg or higher have experienced lymphadenopathy in the draining LN and the one participant dosed at 3 mcg/kg experienced bilateral inguinal and axillary adenopathy. The adenopathy resolves in 7-10 days. There is a risk of QTc prolongation. Note: Two individuals had a grade 1 prolonged QTc interval prior to receiving study drug that persisted through follow-up. One individual experienced grade 1 prolonged QTc interval that persisted through follow-up. One individual experienced grade 1 and 2 prolonged QTc intervals that resolved. At this point in time, there is no clear connection between the administration of N-803 and these minor abnormalities.(See Table 6 below).

In **Table 4** below we present the AE data for clinical and laboratory abnormalities. The primary clinical AE was an injection site reaction that was dose-dependent and persisted for a longer period of time but which resolved in 5-7 days. These were all considered to be Grade 3 AEs by DAIDS grading criteria.

In **Table 5** we present measures of serum creatinine and the eGFR collected during the dose escalation trial in HIV infected individuals. These values are reported as many HIV infected people on ARTs, who are otherwise doing clinically well and would be qualified for studies of IL-15, often have baseline

abnormalities in renal function that would qualify as a grade 1 or 2 abnormality using the DAIDS adverse event criteria.

**Table 4: Adverse events data for clinical and laboratory abnormalities for N-803**

(Note: For each dosing arm, the instances documented for each AE are shown by grade (G1, G2, and G3). The overall prevalence of the AE among the participants for each dosing arm is represented in the column of # Affected)

**Clinical AEs**

| | **0.3 mcg/kg IV N-803** (N = 2) | | | **1 mcg/kg Subcutaneous N-803** (N = 6) | | | | **3 mcg/kg Subcutaneous N-803** (N = 3) | | | | **6 mcg/kg Subcutaneous N-803** (N = 5) | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Adverse Event | **G1** | **G2** | **#** | **G1** | **G2** | **G3** | **#Affected** | **G1** | **G2** | **G3** | **#Affected** | **G1** | **G2** | **G3** | **#Affected** |
| Injection Site Erythema | - | - | - | 11 | 11 | 9 | 6 (100%) | 7 | 6 | 5 | 3 (100%) | 9 | 7 | 9 | 5 (100%) |
| Injection Site Induration | - | - | - | 3 | - | 1 | 4 (67%) | 3 | 3 | 2 | 3 (100%) | 4 | 3 | 5 | 4 (80%) |
| Injection Site Pruritis | - | - | - | 5 | - | - | 3 (50%) | 6 | - | - | 2 (67%) | 9 | - | - | 5 (100%) |
| Injection Site Pain | - | - | - | 3 | - | - | 2 (33%) | 3 | - | - | 1 (33%) | 11 | - | - | 5 (100%) |
| Injection Site Swelling | - | - | - | 2 | 4 | 2 | 3 (50%) | 2 | 2 | - | 2 (67%) | 1 | - | 1 | 1 (20%) |
| Injection Site Tenderness | - | - | - | 3 | - | - | 2 (33%) | 2 | - | - | 1 (33%) | - | - | - | - |
| Pain[1] | - | - | - | 8 | - | - | 5 (83%) | 7 | - | - | 2 (67%) | 5 | 1 | - | 4 (80%) |
| Fatigue | - | - | - | 3 | - | - | 2 (33%) | 1 | - | - | 1 (33%) | 2 | - | - | 2 (40%) |
| Myalgia | - | - | - | 2 | 1 | - | 2 (33%) | 3 | - | - | 1 (33%) | 3 | - | - | 2 (40%) |
| Nausea | - | - | - | 3 | - | - | 3 (50%) | 1 | - | - | 1 (33%) | 3 | - | - | 1 (20%) |
| Chills | - | - | - | - | - | - | - | 2 | - | - | 2 (67%) | 1 | - | - | 1 (20%) |
| Fever | - | - | - | - | - | - | - | - | 2 | - | 1 (33%) | 3 | - | - | 3 (60%) |
| Headache | - | - | - | 1 | - | - | 1 (17%) | - | - | - | - | 2 | - | - | 2 (40%) |
| Arthralgia | - | - | - | - | - | - | - | - | - | - | - | 5 | - | - | 2 (40%) |
| Bruising | - | - | - | - | - | - | - | - | - | - | - | - | 1 | - | 1 (20%) |
| Diarrhea | - | - | - | 1 | 1 | - | 1 (17%) | - | - | - | - | - | - | - | - |

| | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Rash | - | - | - | - | - | - | - | 1 | - | - | 1 (30%) | - | - | - | - |
| Syncope | - | - | - | - | 1 | - | 1 (17%) | - | - | - | - | - | - | - | - |
| Prolonged QTc Interval | 2 | 1 | 1 (50%) | 6 | 3 | - | 3 (50%) | - | - | - | - | - | - | - | - |
| Vertigo | - | - | - | 1 | 2 | - | 1 (17%) | - | - | - | - | - | - | - | - |
| Other AE not identified[2] | - | - | - | 6 | - | - | 3 (50%) | 8 | 2 | - | 3 (100%) | 4 | 4 | - | 4 (80%) |

1. Pain includes inguinal lymph node pain, lymph node tenderness, chest pain, sore throat, shoulder pain, abdominal pain, sinus pain, midback pain, knee pain, and tenderness at biopsy site
2. "Other AE not identified" includes events not covered in the DAIDS AE grading such as nasal congestion, cough, pimple, vesicles at injection site, flaky skin at injection site, greater than 20% decrease in FEV1, mitral valve insufficiency diagnosed 5 months after the last dose of N-803, dizziness, and sneezing

**Laboratory AEs**

| | **0.3 mcg/kg IV N-803** (N = 2) | | | **1 mcg/kg Subcutaneous N-803** (N = 6) | | | | **3 mcg/kg Subcutaneous N-803** (N = 3) | | | | **6 mcg/kg Subcutaneous N-803** (N = 5) | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Adverse Event | **G1** | **G2** | **#** | **G1** | **G2** | **G3** | **#Affected** | **G1** | **G2** | **G3** | **#Affected** | **G1** | **G2** | **G3** | **#Affected** |
| Albumin, Low | - | - | - | 2 | - | - | 1 (17%) | 2 | - | - | 1 (33%) | - | - | - | - |
| AST, High | 1 | - | 1 (50% | - | - | - | - | 1 | - | - | 1 (33%) | - | - | - | - |
| Calcium, Low | 2 | - | 1 (50% | 3 | - | - | 2 (33%) | - | - | - | - | 2 | - | - | 2 (40%) |
| Creatinine, High | 2 | - | 1 (50%) | - | - | - | - | - | - | - | - | 1 | - | - | 1 (20%) |
| eGFR, Low | - | - | - | - | 6 | - | 3 (50%) | - | - | - | - | - | - | 2 | 2 (40%) |
| Glucose, High | 2 | - | 1 (50%) | 6 | - | - | 5 (83%) | - | - | - | - | 2 | 1 | - | 1 (20%) |
| Glucose, Low | - | - | - | 1 | - | - | 1 (17%) | - | - | - | - | - | 1 | - | 1 (20%) |
| Increased Bilirubin | - | - | - | 1 | - | 2 | 1 (17%) | - | - | - | - | - | - | - | - |
| Potassium, Low | - | - | - | 2 | - | - | 1 (17%) | - | - | - | - | - | - | - | - |
| Absolute Neutrophil, Low | - | - | - | 1 | - | - | 1 (17%) | - | - | - | - | - | - | - | - |

| | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Hemoglobin, Low | - | - | - | 3 | 1 | - | 2 (33%) | - | - | - | - | - | - | - | - |

Table 5: Measures of serum creatinine (SCr) and the eGFR during the dose escalation trial

| | | | Dose 1 | | | Dose 2 | | | Dose 3 | | | FU[1] |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Screen | Day 0 | Day 1 | Day 4 | Day 0 | Day 1 | Day 4 | Day 0 | Day 1 | Day 4 | |
| **0.3 mcg/kg (IV)** | | | | | | | | | | | | |
| 2349 | SCr | 1.25 | 1.19 | 1.3 | 1.38 | 1.28 | 1.3 | 1.23 | 1.28 | 1.42 | | 1.18 |
| | eGFR | 59 | 62 | 56 | 52 | 57 | 56 | 60 | 57 | 51 | | 63 |
| 2354 | SCr | 1.28 | 1.27 | 1.14 | 1.23 | 1.05 | 1.14 | 1.03 | 1.24 | 1.15 | 1.05 | 1.09 |
| | eGFR | 67 | 67 | 76 | 70 | 84 | 76 | 86 | 69 | 76 | 84 | 80 |
| **1 mcg/kg (SQ)** | | | | | | | | | | | | |
| 2356 | SCr | 1.07 | 1.03 | 1.13 | 1.17 | 1.14 | 1.11 | 1.05 | 1.14 | 1.05 | 1.07 | 1.03 |
| | eGFR | 74 | 77 | 69 | 66 | 68 | 71 | 75 | 68 | 75 | 74 | 77 |
| 2383 | SCr | 0.84 | 0.87 | 0.86 | 0.76 | | | | | | | |
| | eGFR | >90 | >90 | >90 | >90 | | | | | | | |
| 2403 | SCr | 0.85 | 0.91 | 0.82 | 0.96 | 0.9 | 0.95 | 1.02 | 0.85 | 0.89 | 0.92 | 0.82 |
| | eGFR | >90 | 88 | >90 | 83 | 89 | 84 | 77 | >90 | >90 | 88 | >90 |
| 2413 | SCr | 0.95 | 0.83 | 0.93 | 1 | | | | | | | |
| | eGFR | >90 | >90 | >90 | 89 | | | | | | | |
| 2470 | SCr | 0.67 | 0.76 | 0.86 | 0.69 | 0.86 | 0.84 | 0.72 | | | | |
| | eGFR | >90 | >90 | >90 | >90 | >90 | >90 | >90 | | | | |
| 2530 | SCr | 1.14 | 1.15 | 1.05 | 1.1 | 0.94 | 0.99 | 1.06 | 1.02 | 1.07 | 1.06 | 1.19 |
| | eGFR | 73 | 72 | 80 | 76 | >90 | 86 | 79 | 83 | 78 | 79 | 69 |
| **3 mcg/kg (SQ)** | | | | | | | | | | | | |
| 2543 | SCr | 0.96 | 0.93 | 0.91 | 1.02 | 1.01 | 0.92 | | 0.91 | 0.85 | 1.05 | 1.02 |
| | eGFR | >90 | >90 | >90 | 87 | 88 | >90 | | >90 | >90 | 84 | 87 |
| 2124 | SCr | 1.05 | 0.79 | 0.9 | 0.9 | 1.08 | 0.86 | 1.1 | 1.06 | 0.9 | 0.89 | 0.9 |
| | eGFR | 70 | >90 | 84 | 84 | 68 | 88 | 66 | 69 | 83 | 85 | 84 |
| 2045 | SCr | 0.91 | 0.96 | 0.91 | 1.05 | 0.99 | 0.87 | 1.05 | 0.97 | 0.92 | 1.03 | 0.92 |
| | eGFR | 76 | 72 | 76 | 65 | 70 | 80 | 65 | 71 | 75 | 66 | 75 |
| **6 mcg/kg (SQ)** | | | | | | | | | | | | |
| 2189 | SCr | 1.27 | 1.24 | 1.28 | 1.24 | 1.4 | 1.34 | 1.18 | | | | 1.19 |

| | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | eGFR | 60 | 62 | 60 | 62 | 54 | 57 | 65 | | | | 65 |
| 2531 | SCr | 0.97 | 0.9 | 1.02 | 1.13 | | | | | | | |
| | eGFR | >90 | >90 | >90 | >90 | | | | | | | |
| 2415 | SCr | 1.14 | 0.99 | 1.04 | 1.08 | 1.04 | 1.14 | 1.1 | 1.11 | 1.18 | 1.26 | 1.11 |
| | eGFR | 68 | 80 | 76 | 72 | 76 | 68 | 71 | 70 | 65 | 61 | 70 |
| 2484 | SCr | 0.76 | 0.82 | 0.9 | 0.81 | 0.75 | 0.84 | 0.81 | 0.71 | 0.76 | 0.71 | 0.71 |
| | eGFR | >90 | >90 | >90 | >90 | >90 | >90 | >90 | >90 | >90 | >90 | >90 |
| 2603 | SCr | 0.91 | 0.86 | 0.85 | 0.96 | 0.88 | 0.86 | 0.82 | 0.82 | 0.95 | 0.97 | 0.99 |
| | eGFR | >90 | >90 | >90 | 85 | >90 | >90 | >90 | >90 | 86 | 84 | 82 |

1. Measure made 1 week after the last dose received

In **Table 6** we present the QTc measures made during the dose-escalation study. A total of 4 individuals experienced a QTc > 450 ms; 1 at 0.3 mcg/kg IV and 3 at 1.0 mcg/kg. Among these 4 individuals there were 15 EKGs with a QTc > 450 ms. Of those, 10 were a grade 1 and 5 were a grade 2 AE.

**Table 6:** Measures of QTc (ms) from the dose escalation study.
Values that are considered adverse events are marked with the grades of G1 or G2 per DAIDS criteria.

| | **Screen** | **Dose 1** | | **Dose 2** | | **Dose 3** | |
|---|---|---|---|---|---|---|---|
| **0.3 mcg/kg IV** | | **Predose** | **+ 6 hours** | **Predose** | **+ 6 hours** | **Predose** | **+ 6 hours** |
| 2349 | 413 ms | 433 ms | 407 ms | 403 ms | 422 ms | 413 ms | 416 ms |
| 2354 | **454 ms (G1)** | 436 ms | 422 ms | 420 ms | 431 ms | **479 ms (G2)** | **468 ms (G1)** |

| | **Screen** | **Dose 1** | | | | | | **Dose 2** | | | | **Dose 3** | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **1 mcg/kg SQ** | | **Predose** | **+ 6 hours** | **+ 24 hours** | **Day 3** | **Day 4** | **Day 7** | **Predose** | **+ 6 hours** | **Day 4** | **Day 7** | **Predose** | **+ 6 hours** |
| 2383 | 423 ms | **465 ms (G1)** | **463 ms (G1)** | **475 ms (G2)** | **464 ms (G1)** | 423 ms | **455 ms (G1)** | | | | | | |
| 2356 | 410 ms | 403 ms | 428 ms | 426 ms | 428 ms | 413 ms | | 441 ms | 414 ms | | | 439 ms | 392 ms |
| 2413 | 407 ms | 415 ms | 412 ms | 420 ms | 406 ms | 417 ms | 423 ms | | | | | | |
| 2470 | 405 ms | 406 ms | 440 ms | 423 ms | 435 ms | 417 ms | | **452 ms (G1)** | **459 ms (G1)** | | | | |
| 2403 | 415 ms | 447 ms | **462 ms (G1)** | 420 ms | **450 ms (G1)** | 446 ms | **477 ms (G2)** | 428 ms | 420 ms | **455 ms (G1)** | | **482 ms (G2)** | 434 ms |
| **3 mcg/kg SQ** | | | | | | | | | | | | | |
| 2543 | 408 ms | 407 ms | 394 ms | 385 ms | 406 ms | 427 ms | 425 ms | 406 ms | 386 ms | | 420 ms | 402 ms | 399 ms |
| 2124 | 373 ms | 392 ms | 397 ms | 386 ms | 403 ms | 382 ms | | 405 ms | 403 ms | | | 384 ms | 388 ms |
| 2045 | 413 ms | 420 ms | 424 ms | 411 ms | 433 | 426 | | 425 ms | 435 ms | 426 ms | | 429 ms | 430 ms |
| **6 mcg/kg SQ** | | | | | | | | | | | | | |
| 2189 | 426 ms | 418 ms | 420 ms | 415 ms | 439 ms | 439 ms | 426 ms | 436 ms | 441 ms | | | | |

| | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 2531 | 379 ms | 388 ms | 402 ms | 381 ms | 389 ms | 392 ms | | | | | | | |
| 2415 | 403 ms | 407 ms | 417 ms | 397 ms | 413 ms | 429 ms | | 428 ms | 397 ms | | | 426 ms | 421 ms |
| 2484 | 409 ms | 421 ms | 431 ms | 430 ms | 445 ms | 434 ms | | 411 ms | 446 ms | | | 426 ms | 412 ms |
| 2603 | 414 ms | 421 ms | 414 ms | 423 ms | 419 ms | 426 ms | | 434 ms | 420 ms | | | 435 ms | 426 ms |

The current dose-escalation study has been put on pause 5 times. Two of those were for protocol changes to switch the route of administration from intravenous to subcutaneous (SQ) and to modify language in the consent for the addition of lymph node biopsies and colonoscopy procedures. Three of the pauses were for adverse events. The first was for a participant receiving 1 mcg/kg SQ who experienced an injection site reaction that was a grade 3 which resolved within 5-7 days. We modified the protocol to state" "*In the case of injection site erythema or redness or injection site induration or swelling (Grades 1-3), ALT-803 will be held until the injection site reaction resolves by at least 50%. In cases where injection site reaction occurs, photographs may be taken to document the reaction and assess resolution*". We also changed the protocol to state that a grade 3 injection site reaction does not count toward any stopping rules. The second adverse event requiring a pause was a change in the FEV1 of >20% in one participant. In the original protocol, the baseline FEV1 was collected at screening which could have been several weeks prior to the initiation of dosing. We did not have any PFT measures collected just prior to the administration of the drug to be able to determine if the change was related to the drug. We amended the protocol to add PFT screening within 4 hours before the drug is given and again 12 and 24 (+/-4) hours after each dose. The third pause was due to a change in the eGFR for a participant at the 6.0 mcg/kg dose that qualified as a grade 3 toxicity. This individual had a baseline eGFR of 64 ml/min that was a grade 2 toxicity (< 90 to 60 ml/min) which was allowed in the protocol. His eGFR decreased to 59 ml/min which was determined by the SMC to be most likely related to dehydration. It returned to baseline; per protocol, he did not receive further doses of study drug.

Each time the study was paused due to an AE, the SMC was consulted, and the FDA and IRB were notified. Protocol changes were made in consultation with the FDA and approved by our IRB.

## 1.2. Study Rationale and Hypothesis

As discussed above, N-803 has demonstrated ability to reactivate HIV from latency and can activate T cells and NK cells to clear those cells, thus reducing the reservoir. However, a concern is that CD8 T cells may be excluded from the B cell follicles, where a significant part of the reservoir resides. Webb, et al, has shown that in SIV infected monkeys CD8 T cells in follicles increase in frequency when N-803 is administered. We hypothesize that in HIV infected humans treated with N-803 that CD8 T cells will increase in B cell follicles and that there will be a further reduction in the frequency of cells with an inducible provirus.

## 1.3. Risk / Benefit Assessment

There is no expected direct benefit for the study participant. The primary benefit is the gain in knowledge regarding a potentially effective intervention that may in future studies allow for a durable control of HIV in absence of therapy (a remission). There are several risks involved with this study. N-803 is an unlicensed molecule, and the available safety data on this molecule are based on trials with very small numbers of participants. The adverse events, directly attributable to the study drug,

experienced by HIV infected people are an injection site reaction and lymphadenopathy that typically clears in 7-10 days. As seen in the cancer and HIV trials, other adverse events that may be related to the administration of N-803 include fatigue, myalgia, arthralgia, fever, chills, and injection site pain. Other potential risks include drug interactions which are discussed in section 6. Other clinical management issues that may pose a risk to the participant are discussed in section 10. Other risks involved with participation in this study include:

1) (Medical history and information from medical record) Discussion of HIV serostatus, collection of medical history, and physical exam can cause embarrassment and discomfort. Research coordinators are trained to be sensitive to stigma and embarrassment that participants may feel, and provide support. Anticipation of a medical procedure may cause stress or anxiety. All data collection and storage of PHI will be compliant with HIPAA and techniques for Good Clinical Practice (GCP) in research.

2) (Blood draws and leukapheresis) Risk to study participants associated with blood draw procedures will be minimal. There is potential for bruising, bleeding, infection and mild discomfort from the venipuncture. To minimize this discomfort, there is required training and certification for research technicians, and they routinely use the smallest gauge butterfly (21 gauge) needle compatible with the protocol and prevention of hemolysis. Rarely, participants experience faintness (near-syncope or syncope) at the time of phlebotomy. The risks of leukapheresis include the risks associated with blood draws listed above, as well as fatigue, headache, nausea, vomiting, and hypotension and air embolism. Other risks of leukapheresis include allergic reaction, damage to red blood cells, and loss of platelets. Very rare risks include heart attack, stroke, and death. Due to citrate anticoagulation, participants may experience temporary numbness or tingling of the fingertips or around the mouth, cramping, chills, or anxiety.

3) (Lymph node biopsy procedure) Lymph node (LN) biopsies will be performed at the University of Minnesota by surgical faculty that have collaborated with Dr. Schacker's projects for over 15 years. Risks from LN biopsy include bleeding, bruising, infection, pain, development of a seroma, and scarring. Thus far they have performed over 635 LN biopsies on HIV+ patients for related studies without any significant adverse events. If complications do develop, at least one surgeon will be available to provide immediate and appropriate follow up care. Participants from both study sites will undergo the LN biopsies at the UMN study site.

4) (Colonoscopies) Colonoscopies will be performed within clinical procedure suites at the U of Minnesota by physician faculty in Gastroenterology, Alexander Khoruts, MD. Conscious sedation will be used during colonoscopies consistent with standard of care for clinical protocols, and participants will be monitored by nursing within clinical research units following the procedure. Risks from colonoscopy include discomfort, cramping, anxiety, dizziness after sedation, and bleeding. Colonoscopy is also associated with risk of colonic perforation, estimated to occur in approximately 1/3000 procedures. They have performed over 473 colonoscopies in MN in similar protocols with no significant adverse events. Participants from both study sites will undergo colonoscopies with biopsy at the UMN study site.

# 2. STUDY OBJECTIVES

## 2.1. Primary Objectives

- To determine the impact of N-803 on the frequency and function of CD8+ T cells in B cell follicles.
- To determine the safety of N-803 in HIV+ ART suppressed individuals by assessment of adverse events experienced by participants during the conduct of the trial

## 2.2. Secondary Objectives

- To determine the impact of IL-15 on the frequency, location, and phenotype of vRNA+ and vDNA+ cells in lymphoid tissues (inguinal lymph node, ileal, and rectal tissues)

## 2.3. Exploratory Objectives

- To determine the impact of IL-15 on frequency of T follicular helper cells harboring intact HIV genomes
- To determine the effect of IL-15 on frequency and activation status of NK cells in tissue and blood
- Impact of N-803 on ADCC in tissues and blood
- Impact of N-803 on HIV specific CTL in tissues and blood
- Impact of N-803 on detection of HIV RNA in plasma

# 3. STUDY DESIGN

## 3.1. Study Overview

This is a Phase 1B, single arm, two-center, non-randomized, open label, uncontrolled study to determine how administration of N-803 affects B cell follicle structure and function and the impact on measures of virus in the follicles. The two sites will be the University of Minnesota and Hennepin Healthcare. There will be a total of 10 participants. Participants will be generally healthy HIV infected men or women suppressed on ART $\geq$24 months, who have undetectable plasma viremia, and CD4 $\geq$ 350 cells/µl. The study drug N-803 will be administered at 6 mcg/kg, which is the maximum tolerated dose determined in a recently completed dose-escalation trial. At screening participants will have an EKG, medical history obtained, and blood drawn to assess if they meet entry criteria. At least 7 days (up to 14) prior to the first dose of drug, an inguinal lymph node will be obtained by excisional biopsy, colon and ileal tissue will be obtained via colonic biopsy, and a large volume of PBMCs obtained by leukapheresis. These procedures will occur at least 7 days prior to the first dose of N-803 to ensure that N-803 induced lymphadenopathy will not impair wound healing. Each participant will receive 3 doses of N-803, each dose separated by at least 3 weeks and not more than 5 weeks (see 5.3). Blood will be obtained at regular intervals to monitor for drug toxicity and changes in plasma viral load and CD4 cell count. At least 7 days after the last dose (and not more than 14 days) a second set of tissue biopsies will be obtained. The participant will return for a healing assessment within the following 7 days. The participant will be seen at monthly intervals for an additional 3 months to obtain peripheral blood to monitor for long-term toxicity, including development of antibodies for IL-15.

### 3.1.1. Rationale for Dose and Dosing Interval

In our dose escalation study with N-803 in HIV infected persons, we started with a dose of 0.3 mcg/kg IV with plans to increase to 1, 3, 6, and 10 mcg/kg IV. As we completed the 0.3 mcg/kg cohort it was

reported that giving the drug subcutaneously (SQ) had 2 benefits. First the frequency of the cytokine related symptoms of fevers, rigor, and hypotension virtually resolved and second, the drug concentrated primarily into lymphatic tissues. This is the rationale for dosing the drug SQ. For this protocol where we study the impact that N-803 will have on effector cell function in B cell follicles, we will use 6 mcg/kg. This is the maximum tolerated dose from our recently completed dose-escalation trial.

In this protocol we will administer the drug every 21 days. We have chosen this dosing interval because in cancer trials, there has been a consistent observation that shorter dosing intervals are associated with decreased proliferation of NK cells and an exhausted phenotype (35) which likely explains the loss of antiviral effect with N-803 in a macaque trial when the dosing interval was shorter (34). Those investigators found the antiviral effect was restored when there was a longer period of time between dosing intervals. An exhausted phenotype is not a novel concept and has been described with T cells (36, 37) but also in experimental models of prolonged NK cell activation with prolonged exposure to ionomycin where reduced cytolytic function and cytokine production was observed (38).
In studies in humans with cancer using N-803 at weekly intervals there was a diminished proliferative capacity of NK and CD8 T cells after repeated weekly dosing, and we saw this in our study **(Fig 6)**

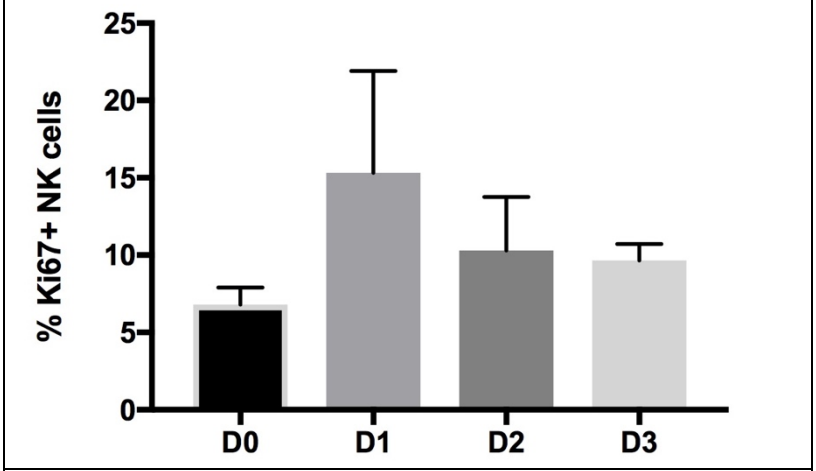


**Fig 6** The percentage of NK cells that were Ki67+ 24 hours after each dose of N-803. There was a decrease after the first dose. These data are based on the 3 individuals receiving 3 mcg/kg.

## 3.2. Criteria for Evaluation

For purposes of efficacy, only those participants who have received all 3 doses of N-803 and have tissue available from both the first and second procedure timepoints will be considered evaluable. Any participant who does not have tissue from both timepoints, and/or has not received all 3 doses of N-803 will be replaced.

### 3.2.1. Primary Endpoint

- Frequency of CD8+ T cells in follicles (and their phenotypes) before and after N-803 therapy will be measured using quantitative image analysis techniques we have developed in our lab (10, 11, 15, 39-41). Briefly we will stain cells with antibodies to CD8 and determine their frequency per unit area. We have used these techniques to account for changes in CD4 T cell populations in the parafollicular T cell zone after antifibrotic therapy (42).

- Safety of N-803 given at this dose and frequency in this population. Any participant who receives an injection of N-803 will be followed for AEs. Clinical and laboratory adverse events will be recorded in the CRF and the study will have quarterly monitoring by the team of monitors in the University of Minnesota Clinical and Translational Science Institute Regulatory Core. This group monitors all FDA trials at the University of Minnesota and routinely monitors multi-site trials at different institutions. Every 14 days, the co-investigators will review any and all clinical and laboratory adverse events to ensure compliance with the study pause criteria listed in section 11.4.

#### 3.2.2. Secondary Endpoints

- Frequency, location, and phenotype of vRNA+ and vDNA+ cells in lymphoid tissues.
- Frequency of vRNA+ TFH cells in B cell follicles before and after N-803 therapy.
- Amount of HIV RNA/DNA detected in follicles before and after N-803 therapy.

#### 3.2.3. Exploratory Endpoints

- Changes in frequency and activation status of NK cells in tissue and blood
- Impact of N-803 on ADCC in tissues and blood
- Impact of N-803 on HIV specific CTL in tissues and blood
- Structure and integrity of the FDC network before and after N-803 therapy

# 4. STUDY POPULATION

## 4.1. Study Population

Participants will be HIV infected and on ART ≥24 months, have undetectable plasma viremia, and CD4 ≥ 350 cells/µl.

## 4.2. Inclusion Criteria

1) Male or female, age ≥ 18 and ≤ 65 years
2) HIV-1 infection, documented by any licensed rapid HIV test or HIV enzyme or chemiluminescence immunoassay (E/CIA) test kit at any time prior to study entry and confirmed by a licensed Western blot or a second antibody test by a method other than the initial rapid HIV and/or E/CIA, or by HIV-1 antigen or plasma HIV-1 RNA viral load.
3) On continuous antiretroviral therapy for over 24 months without any interruptions of greater than 14 consecutive days, without plans to modify ART during the study period.
4) Screening plasma HIV RNA levels < 20 copies/mL and on at least 1 determination in past 12 months (isolated single values ≥ 20 but < 200 copies/mL will be allowed if they were preceded and followed by undetectable viral load determinations)
5) Screening CD4+ T cell count ≥ 350 cells/$mm^3$ and nadir CD4+ T cell count of >200 per participant report, if able to recall.
6) Ability to be off prednisone and other immunosuppressive drugs for at least 14 days before screen. Inhaled, nasal spray, intraarticular injections, and topical steroids are acceptable.
7) Acceptable blood pressure and heart rate parameters within normal limits (systolic = 88-140mmHg; diastolic = 50-<90mmHg; heart rate = 46-100 bpm). Treatment with antihypertensive medication is allowed. However, if someone is on a beta-blocker this must be switched to another class of medication as there is a theoretical risk for bradycardia if the participant were to experience cytokine release syndrome symptoms (which has not happened with this drug delivered SQ).
8) Sexually active females of child bearing potential and males with partners of child bearing potential

must agree to use effective contraception during study participation and for 1 month following the final study visit (4 months after final dose of study drug)

a) Acceptable birth control is defined as the following:
   i) For female participants of childbearing potential, two of the following forms of contraception are required, one of which must be a barrier method:
      (1) Condoms (male or female) with or without a spermicidal agent
      (2) Diaphragm or cervical cap with spermicide
      (3) Intrauterine device (IUD) with published data showing that expected failure rate is < 1% per year
      (4) Tubal ligation
      (5) Hormone-based contraceptive such as oral birth control pills

9) Laboratory tests performed within 14 days of study enrollment must be a grade 0 or 1 as defined by the Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events, Corrected Version 2.1, with the following exceptions:
   - Platelet counts ($\geq$ 150,000/mm$^3$)
   - Hemoglobin > 12.5 g/dL for men and > 11.5 g/dL for women. It is not acceptable for patients to be transfused to meet this requirement. The use of Epogen is permitted.
   - Estimated Cr Cl (eGFR) > 50
10) Voluntary written consent provided by the participant

## 4.3. Exclusion Criteria

1) Active or recent malignancy requiring systemic chemotherapy or surgery in the preceding 36 months or for whom such therapies are expected in the subsequent 12 months; minor surgical removal of localized skin cancers (squamous cell carcinoma, basal cell carcinoma) are not exclusionary
2) Chronic liver disease defined as Class B and C on the Child-Pugh chronic liver disease scale.
3) Active and poorly controlled atherosclerotic cardiovascular disease (ASCVD), as defined by 2013 ACC/AHA guidelines, including a previous diagnosis of any of the following: (a) acute myocardial infarction, (b) acute coronary syndromes, (c) stable or unstable angina, (d) coronary or other arterial revascularization, (e) stroke, (f) transient ischemic attack (TIA), or (g) peripheral arterial disease presumed to be of atherosclerotic origin.
4) History of potential immune-mediated medical conditions requiring concomitant treatment with immunomodulatory drugs, and/or exposure to any immunomodulatory drug in the 30 days prior to screen (e.g. corticosteroid therapy equal to or exceeding a dose of 15 mg/day of prednisone for more than 10 days, IL-2, interferon, methotrexate, cancer chemotherapy). NOTE: use of inhaled, nasal steroid or topical steroid lotions and creams is not exclusionary. Prior exposure to N-803 is not exclusionary if prior exposure occurred at least 6 months before screen.
5) Exposure to any experimental therapies within 90 days of study screen. Exposure to long acting injectable ART therapies is not exclusionary.
6) Latent TB infection or active TB disease prior to completing a standard regimen of anti-TB therapy that is defined as meeting PPD criteria for TB exposure or a positive quantiferon gold test collected at screening.
7) Active fungal infection requiring systemic antifungal therapy
8) Active herpes outbreak or varicella-zoster virus infection requiring episodic treatment
9) Chronic active hepatitis B or C. For Hepatitis B this will be defined as HBs antigen + and for Hepatitis

C this will be defined as Hepatitis C antibody positive and Hepatitis C PCR+.

10) History and/or presence of any clinically significant disease or disorder, such as cardiovascular, pulmonary, renal, hepatic, neurological, gastrointestinal and psychiatric/mental disease/disorder, which, in the opinion of the site Principle Investigator may either put the subject at risk because of participation in the study, influence the results of the study or the subject's ability to participate in the study.
11) Any degree of baseline QT/QTc interval prolongation (QTc interval > 450 msec in men and > 470 msec in women.)
12) Any ischemic changes seen in the stress treadmill test administered per the discretion of the PI in order to assess any other EKG abnormalities as discussed in section 7.17
13) History or evidence of uncontrollable CNS disease such as dementia, demyelinating disease, Parkinson's, or a CNS degenerative disease that, in the opinion of the site Principle Investigator, may either put the subject at risk because of participation in the study, influence the results of the study or the subject's ability to participate in the study.
14) Prior organ allograft or allogeneic transplantation
15) Planning or current pregnancy or breastfeeding
16) Any clinically indicated vaccination (other than influenza or SARS-CoV-2) administered within 14 days of screen

# 5. STUDY TREATMENTS / AGENTS

## 5.1. Product Characteristics, Preparation and Storage

N-803 is an investigational drug supplied to investigators by the ImmunityBio., Inc. in Culver City, CA. It will be sent to and maintained by Investigational Drug Services (IDS) using established procedures. Sufficient study drug will be available for this protocol to treat all of the projected enrolled patients. The Principal Investigator will be responsible for the maintenance of records of receipt and disposition of study drug including dates, quantities administered and availability, and patient assignment. At the end of the study, ImmunityBio's representatives will perform a full accountability of all study drugs. All unused study drug vials will be returned to ImmunityBio. All partially used or empty study drug vials will be destroyed at the study sites according to the established procedures.

Investigator agrees not to supply or administer N-803 to any person except those named as Co-Investigators on Form FDA 1572 or to individuals participating in this study, respectively.

Study drug will be stored at the trial site in a secured area at 2°C to 8°C with limited access and protection from excess light and heat.

The research pharmacy at the University of Minnesota offers in-house investigational drug services and dispensing of research drugs. The research pharmacist and credentialed pharmacy technician charged with dispensing research-related drugs receive protocol-specific training and are compliant with State Board of Pharmacy requirements, FDA Guidelines and Good Clinical Practices. Pharmacy documents, including study drug accountability logs, study drug ordering and shipping logs, and regulatory documents are maintained by the pharmacy technician.

**Assessment of adherence.** N-803 will be administered by subcutaneous injection in a research unit at the University of Minnesota for all study participants. Participants will be advised to remain adherent to their antiretroviral drugs during the conduct of this study.

## 5.2. N-803 Administration (General Guidelines)

### 5.2.1. Formulation and Composition

The biological drug product, N-803, is formulated in a phosphate buffered saline (PBS) solution. The solution appears as a clear and colorless liquid. The drug substance is produced by a recombinant mammalian cell line and is manufactured using protein-free media. Study medication is provided in a 2 mL single-dose/single-use vial containing 0.6 mL of N-803 (extractable volume is 0.5 mL) at a concentration of 2 mg/mL. The vialed quantitative composition of N-803 is listed in **Table 7**.

**Table 7: Quantitative Composition of N-803**

| *Component* | *Concentration* | *Amount / Vial* |
|---|---|---|
| N-803 | 2 mg/mL | 1.2 mg |
| Phosphate Buffered Saline (PBS) | QS | 0.6 mL |
| **PBS Formulation: Sodium Chloride (USP) 8.18 g/L; Sodium Phosphate Dibasic (USP) 1.43 g/L; Potassium Phosphate Monobasic (NF) 1.36 g/L pH 7.4.** | | |

### 5.2.2. Storage and Handling

Vials are packaged in cartons and shipped to the clinical site. Study medication must be maintained at a temperature between 2˚C and 8˚C. The drug will be maintained in the Investigational Drug Service at each institution using the manufacturers guidelines for storage. The individual dose for administration to each participant will be prepared in syringes by the investigational pharmacist using protocols provided by the manufacturer and then delivered to the Clinical Research Unit at each institution. The product expires 24 hours after it is drawn in to the syringe. The syringe is labelled with the date and time of its expiration, and is then stored in a refrigerator until its appropriate use or expiration.

### 5.2.3. General Guidelines

N-803 injections will occur in a research facility at the University of Minnesota. Each dose will be separated by at least 21 days but not more than 35 days (see section 5.3). Study drug will be provided free of charge to the study participants. The dosing interval of 21 days is to avoid the “cytokine exhaustion syndrome” that has been described using this compound in cancer trials. When the interval is too short (e.g., 7 days) the T cells and NK cells do not have the same magnitude of response to N-803 and there is less activation (as measured by CD69) or proliferation (as measured by Ki67). This phenomenon has been characterized in the mouse model (35) and is avoided when the interval is at least 14-21 days between dose. We have chosen 21 days as that is the current practice in most of the cancer trials going forward.

The study drug N-803 will be administered at 6 mcg/kg, which is the maximum tolerated dose determined in a recently completed dose-escalation trial.

N-803 dosing is calculated using a weight obtained on the day of each dose administration. For participants > 100 kilograms weight, the N-803 dose is calculated using a weight capped at 100 kg. The weight will be re-checked prior to each injection with the dose re-calculated if ≥ 10% change from the weight used for the baseline calculation. The calculated amount of N-803 will be drawn into a syringe for subcutaneous injection. The current IDS stock concentration is 2 mg/ml. Doses will be drawn directly into the syringe for injection.

Injections are given in the abdominal area. The injection site should be rotated per institutional guidelines and each injection site separated by at least 1 inch.

**Required post N-803 monitoring:** Participants will be observed for a minimum of 2 hours after each dose of N-803 for immediate adverse events. Vital signs (heart rate, blood pressure, respiration, temperature, and oxygen saturation) will be documented prior to the N-803 injection and then at 30, 60 and 120 minutes with a ± 20 minute window for each time point. If systolic blood pressure drops to less than 90, and or diastolic drops to less than 50, obtain vital signs every 5 minutes for 30 minutes. Then obtain vital signs every 15 minutes and proceed with frequent vital signs as directed by the PI and/or Co-I.

Administration of additional glucocorticoids is discouraged during the N-803 treatment period as the use of systemic steroid medications may result in loss of therapeutic effects of the study drug (see section 6.1).

**Pre and post-therapy intervention guidelines.** The intervention guidelines described in the section below can be modified by the individual study center as medically necessary or as appropriately without requiring a protocol amendment or being considered a protocol deviation. In general, these guidelines are intended to address AEs graded 3 or greater (see Note 1 in table).

| **Condition** | **Agents** | **Dose** | **Route** | **When** |
|---|---|---|---|---|
| **Fever/chills** | Acetaminophen | up to 650 mg<br>or<br>10 to 15 mg/kg recipient weight | Orally | Prior to each dose & repeat 4 hours after dosing.<br>Repeat every 4 hours if fever present |
| | AND | | | |
| | Indomethacin | 25 mg daily dose<br>or<br>50 mg when participant experienced a fever > 39.0°C in a previous dose | Orally | Prior to each dose & repeat it<br>4-6 hours after dosing<br>If persistent fever > 39.0°C, repeat 50 mg every 8 hours (if adequate renal function) with acetaminophen every 6 hours. |

| Condition | Agents | Dose | Route | When |
|---|---|---|---|---|
| **Allergies** | Diphenhydramine | 25 mg<br>or<br>up to 0.5 to 1 mg/kg recipient weight | Orally or IV | Prior to each dose and Repeat 4-6 hours after dosing.<br>Antihistamine, such as diphenhydramine or cetirizine, should be taken for one day before and one to two days after dosing. |
| | In combination with one of the following: | | | |
| | Ranitidine | 300 mg | Orally | |
| | Cimetidine | 300 mg | Orally | |
| | Famotidine | 20 mg | Orally | |
| **Hypotension** | Normal saline (0.9%) | 2 mL/kg/hr (up to 100 mL/hr)*** | Continuous IV | Begin 30 min prior to and for 6 hours after each dose |
| **Nausea/ Vomiting/** | Ondansetron | 0.2 mg/kg (up to 8 mg) | IV | 30 min prior to each dose |
| | OR other 5-HT3 antagonists at the discretion of the Investigator. | | | |
| **Pain** | Hydromorphone | 0.5 mg | IV | 30 min prior to each dose |
| | OR other opioid medications at the discretion of the Investigator | | | |

***In the last 2 hours, if BP does drops slightly, it is recommended to increase the normal saline rate to 250 mL/hr (given no evidence of fluid overload) and give over 2 hours until pressure is 90 mm systolic or more stable. Alternatively, if this has occurred previously, a higher rate of fluid administration from the start of the dosing protocol (i.e. 200 mL/hr over 6 hours) can be used as long as there are no signs of fluid overload (i.e. crackles in the lungs or peripheral edema or > 10% weight gain).

*Note 1:* Acetaminophen and diphenhydramine will be administered approximately 30 minutes prior to each dose to every participant on every dosing day. Participants will also be instructed to take another dose of acetaminophen and diphenhydramine 4 hours after the study drug is given. These medications will be provided by the study at no cost to the participant.

*Note 2:* Non-steroidal anti-inflammatory medication including acetaminophen, ibuprofen, or naproxen may be given per physician discretion following the recommended dosing thresholds:
Acetaminophen: not to exceed 3000 mg (3 grams) in 24 hours
Ibuprofen: not to exceed 2400 mg in 24 hours
Naproxen: not to exceed 1100 mg in 24 hours

*Note 3:* The use of systemic steroid medications may result in loss of therapeutic effects of the study drug and should be avoided; however, in the event of a life-threatening inflammatory reaction to N-803, the IV administration of dexamethasone or other steroid-based medication is warranted.

Based on current experience, localized skin reactions are common with subcutaneous administration. If a reaction occurs and the reaction area surrounding the N-803 injection site is > 6 cm or symptomatic (painful and/or itchy), it may be treated with topical 0.05% clobetasol propionate, 0.1% triamcinolone, or 1% hydrocortisone cream. Diphenhydramine may be administered pre- (25-50 mg TID orally) and

post-dosing (25-50 mg TID orally x 2 days) of N-803 at the discretion of the treating physician. Diphenhydramine should be eliminated if not tolerated. Generally, the reaction associated with an N-803 starts small at the site of the injection and spreads outwardly sometimes covering a large area of the abdomen. It generally resolves within 7-9 days of treatment.

### 5.3. Individual Dose Delays

N-803 administration may be delayed by up to 14 days (resulting in a maximum 35 day period between doses) if on the day of the planned dosing for any of the following situations:

- the participant has a fever (defined as a temperature of > 100.4°F (38°C))
- treatment related side effects have not resolved to grade 1 or better
- if in the opinion of the treating physician, a delay would be of benefit

However, if dosing is delayed for one of these reasons the participant will be evaluated at the end of 7 days and if the issue has resolved the subject will receive the dose as soon as possible. If the participant does not receive the dose within the drug administration window (up to 14 days post the planned dosing day), they will be discontinued and replaced.

# 6. CONCOMITANT MEDICATIONS

All participants should be maintained on the same medications throughout the entire study period, as medically feasible.

Participants must provide their own antiretroviral drugs. No restrictions will be placed on the antiretroviral drug combinations. If medically feasible, study participants will be encouraged to remain on their entry antiretroviral regimen through the entire study period. If a participant's ART regimen does change, refer to section 11.3 for the guideline concerning discontinuation.

Participation in the study will not interfere in any manner with the subject's standard of care.

Prohibited and contraindicated medications are defined below. There are no precautionary medications.

## 6.1. Prohibited Medications

1. **Glucocorticoids**:
    - These have been shown to reduce cytokine-induced side effects including fever, renal insufficiency, hyperbilirubinemia, confusion, and dyspnea, concomitant administration of these agents with N-803 may reduce the biologic effectiveness of N-803. Therefore, administration of glucocorticoids is prohibited during N-803 treatment period.
    - They should be avoided except in the event of a severe toxicity or unrelated condition requiring steroid and their use will be an indication to stop N-803.
    - Inhaled, nasal spray, and topical steroids are acceptable.
2. **Chemotherapies and interferon**:
    - Hypersensitivity reactions have been reported in patients receiving combination regimens containing sequential high dose IL-2 and antineoplastic agents, specifically, dacarbazine, cisplatin, tamoxifen and interferon-α. These reactions consisted of erythema, pruritus,

and hypotension and occurred within hours of administration of chemotherapy. Concomitant chemotherapies and interferon-α with N-803 are prohibited.

3. **IL-2 and interferon**:
    - Myocardial injury, including myocardial infarction, myocarditis, ventricular hypokinesia, and severe rhabdomyolysis appear to be increased in patients receiving IL-2 and interferon-α concomitantly. Exacerbation or the initial presentation of a number of autoimmune and inflammatory disorders has been observed following concomitantly use of interferon-α and IL-2, including crescentic IgA glomerulonephritis, oculo-bulbar myasthenia gravis, inflammatory arthritis, thyroiditis, bullous pemphigoid, and Stevens-Johnson syndrome. Therefore, concomitant interferon-α treatment with N-803 is prohibited.

## 6.2. Contraindicated Medications

All of the potential drug interactions with cytokine therapy listed below might occur with N-803 treatment. Contraindicated medications will be generally avoided when possible, but will be allowed in situations deemed low clinical risk per the discretion of study investigators. Medication dosing, frequency (intermittent for symptom alleviation vs continuous), and indication will be considered in these decisions

1. **Psychotropic drugs:**
    - Central nervous function may be affected. Therefore, interactions could occur following concomitant administration of psychotropic drugs (e.g., narcotics, analgesics, antiemetics, sedatives). Intermittent use of oxycontin (5 mg tablets) is permitted for procedure related pain or if required for trauma. Analgesics purchased over the counter are permitted. The sleep aides Ambien and Lunesta are permitted. SSRI/SNRIs are considered low risk, and if used in standard doses are allowed. Other psychotropic medications that are not currently associated with sedating or cognitive side effects and are not being used to treat symptoms related to psychosis or alterations in thought processing are also allowed.
2. **Kidney and liver**
    - Kidney and liver function may be impaired during the treatment. Use of concomitant nephrotoxic (e.g., aminoglycosides), myelotoxic (e.g., cytotoxic chemotherapy, which are excluded anyhow), cardiotoxic (e.g., doxorubicin, which is excluded anyhow) or hepatotoxic (e.g., methotrexate, asparaginase, which are excluded anyhow) medications may further increase toxicity to the kidney or liver. These medications should be generally minimized during N-803 regimen treatment period.
3. **Beta-blockers**
    - may potentiate the hypotension seen with cytokine therapy. Therefore, administration of these medications should be avoided during the N-803 portion of the treatment period. Use during a time that there is a least 72 hours gap away from hypotension and from N-803, then these may be used on the judgment of the treating physician.

# 7. STUDY PROCEDURES

Prior to conducting any study-related activities, written informed consent and the Health Insurance Portability and Accountability Act (HIPAA) authorization must be signed and dated by the subject.

Blood collection will occur at indicated visits and will be timed to stay within Red Cross Guidelines (less than 500 mL every 8 weeks).

## 7.1. Medication Review

All concomitant medications taken within 30 days prior to screening and entry and a complete history of ART, HIV-1 related vaccines, and immune-based therapies will be documented at the Screening visit.

At each subsequent visit, all additions or discontinuations of prescription medications should be recorded. Actual or estimated start and stop dates should be recorded.

## 7.2. Demographics and Medical History

Demographic information (date of birth, sex, race and ethnicity) will be recorded at Screening.

At Screening, the medical history must include all diagnoses within the past 30 days and, regardless of when the diagnosis was made, a complete history of chronic conditions, malignancies, and AIDS-defining conditions. Self-reported or documented nadir CD4+ T cell count should be recorded.

Any allergies to any medications or their formulations should also be recorded.

## 7.3. Clinical Assessment

Signs and Symptoms

At screen and baseline visits, all grades of signs and symptoms that occurred 30 days prior to the visit must be recorded.

At all subsequent visits, all grades of signs and symptoms that occurred since the previous visit must be recorded as part of an Adverse Event (AE) assessment. Duration (start and stop dates and times), severity/grade, outcome, treatment and relation to study drug will be recorded on the case report form (CRF). Criteria for participant management, dose interruptions, modifications, and discontinuation of treatment will be mandated only for toxicities attributable to the study medication(s).

Diagnoses

All clinical events and new diagnoses or changes in diagnoses should be recorded.

## 7.4. Physical Examination

Complete Physical Exam

A complete physical examination should be conducted at screening. It is to include at minimum an examination of the skin, head, mouth, and neck, auscultation of the chest, cardiac and abdominal exam, and examination of the lower extremities for edema. The complete physical exam will also include resting vital signs (temperature, pulse, respiratory rate, and blood pressure), height and weight.

Targeted Physical Exam or Assessment

A targeted physical examination should be conducted by and MD or advanced practice provider at each dosing visit. Targeted physical assessments can be conducted by an RN at all other visits except screen and dosing days. It is to include resting vital signs (temperature, pulse, respiratory rate, blood pressure) and weight, and is to be driven by any new signs or symptoms that the participant has experienced since the last visit.

## 7.5. Clinical Laboratory Measurements

All clinical laboratory evaluations will be conducted in Clinical Laboratory Improvement Amendments (CLIA) certified laboratories.

Hematology

Complete blood count with differential (CBCD) will be obtained at the indicated visits.

Chemistry

Basic metabolic chemistry panel (CBASIC) and hepatic panel (BHEPAT) will be obtained at the indicated visits. This will include serum sodium, potassium, chloride, CO2, anion gap, random glucose, blood urea nitrogen (BUN), creatinine, eGFR, aspartate aminotransferase (AST/SGOT), alanine aminotransferase (ALT/SGPT), alkaline phosphatase, total bilirubin, direct bilirubin, albumin, and total protein.

Pregnancy Testing

A serum or urine β-human chorionic gonadotropin HCG test (urine test must have a sensitivity of 25 mIU/mL) will be obtained from female participants with reproductive potential at screening, prior to each dosing visit, and anytime during the study when pregnancy is suspected.

Hepatitis B Testing

Hepatitis B antibodies will be measured, and, if positive, a Hepatitis B antigen test will be obtained at Screen.

Hepatitis C Testing

Hepatitis C antibodies will be measured and, if positive, a Hepatitis C PCR will be obtained at Screen.

Tuberculosis Testing

A quantiferon gold test will be ordered at screening. If the test is positive and the potential participant has no prior history of TB treatment they will be referred to their primary provider for further testing. If they do have a history of prior treatment they will be considered eligible for the study.

## 7.6. HIV Clinical Laboratory Measures

Blood will be obtained and sent to a CLIA-certified laboratory for real-time determination of absolute CD4+ and CD8+ T cell counts, percentages, and CD4/CD8 ratio and plasma HIV-1 RNA quantification (viral load) at the indicated visits. All efforts should be made to have collection of blood for T cell counts occur between 8am and 11am, in order to account for diurnal variation in CD4+ T cell count.

## 7.7. Cryopreservation of Plasma

Plasma will be collected and stored at the indicated visits per the appropriate laboratory standards of procedure.

## 7.8. Cryopreservation of PBMCs

PBMCs will be collected and stored at the indicated visits per the appropriate laboratory standards of procedure.

## 7.9. Collection of PAXgene Specimen

Whole blood will be collected in PAXgene tubes at the indicated visits. Blood will be collected in the following upright tubes - PAXgene Blood RNA Tube (16x200 mm / 2.5 mL). The tubes must be filled completely. Specimens will be shipped overnight on wet ice or stored at -80 freezer until ready to ship.

## 7.10. Immunologic Measures

*T cell and NK immunogenicity.* T cell immunogenicity will be measured via several different assays. *T cell activation, proliferation, and function:* Routine flow cytometric analysis of sorted T cells (CD4 and CD8) will be completed to assess markers of proliferation and activation. Briefly, purified PBMCs and LN cell suspensions from baseline and after the last dose of N-803 will be thawed, and stimulated with various HIV peptides. Cells will be incubated for 6 hours in the presence of Brefeldin A at 37°C. Antibody panels using some or all of the following surface molecules will be used to assess the quantity, phenotype, and function of responding CD4 and CD8 T cells: Aqua Blue, CD3, CD4, CD8, CD27, CD45RO, CD57, CCR7, PD-1, ICOS, CD150, CXCR5, and CCR6. These panels will allow discrimination of differentiated CD8 and CD4 memory and TFH phenotypes. In panels using fewer surface markers we will also assess function by fixing and staining for the following intracellular functional markers: IFNgamma, IL-2, TNF, MIP-1alpha, and IL-21. Following identification of small live lymphocytes and T cells, a gate will be set for each respective phenotype using combinations that provided optimal separation (central memory, effector memory, peripheral TFH, germinal center TFH). Boolean gate platform will be used to create the full array of possible functional combinations for each phenotype.

*NK activation, proliferation, function and exhaustion (Miller Laboratory, UMN):* To assess blood and LT NK cell functional activity, flow cytometric analysis for NK cell degranulation (CD107a) and type-II interferon production will be performed on NK cells isolated from all tissue compartments using K562 cells (a human leukemia cell line) as targets. In addition, we will evaluate the killing of HIV-infected targets by staining for activated caspase-3 and -7 (CellEvent Caspase-3/7 Green Flow Cytometry Assay Kit, ThermoFisher). Primary CD4s will be isolated and *in vitro* infected from patient samples with a variety of primary HIV strains obtained from the NIH AIDS Reagent Program as previously described (43). HIV-infected $CD4^+$ T cells are then enriched by taking the negative fraction from a CD4 positive selection to remove all uninfected cells. NK and HIV-infected cells are then cultured at an effector to target ratio of 5:1. As shown in (**Fig 7**) resting autologous NK cells degranulate 3-fold greater against HIV-infected T cells than uninfected CD4's. This suggests that NK cells can recognize HIV-infected $CD4^+$ T cells presumably through upregulation of NKG2D ligands ULBP1/2, as shown previously (44). The ability of NK cells from N-803 treated participants to mediate ADCC will also be evaluated. NK cells isolated from participant PBMC will be exposed to the CD20 expressing B-cell line, Raji, in the presence and absence of the anti-CD20 monoclonal antibody, Rituxin. Rituxin bound Raji cells are a potent inducer of ADCC through Rituxin's Fc portion bound to CD16. ADCC will be evaluated by NK cell expression of CD107a after exposure to targets.

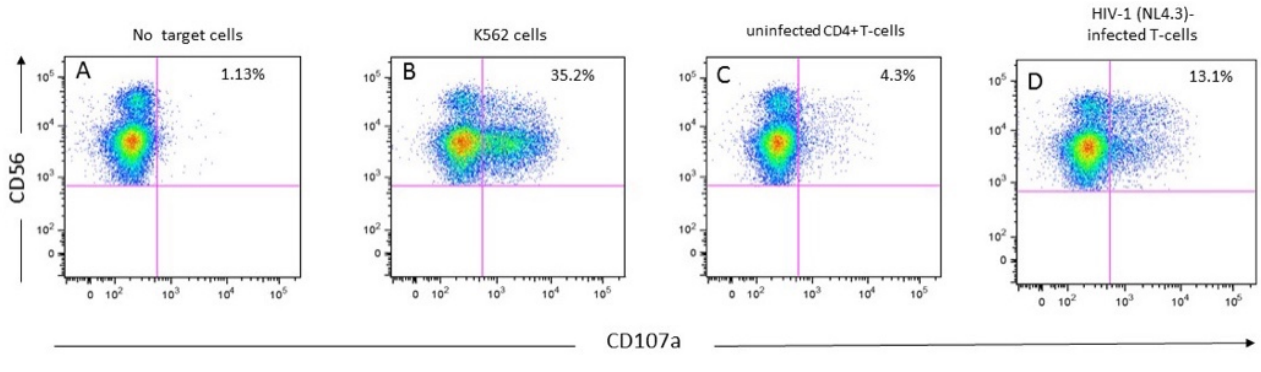


**Fig 7: Functional response of primary NK cells to autologous HIV-infected T cells.** Purified primary NK cells and CD4$^+$ T cells were isolated from the same healthy donor using magnetic columns. NK cells were co-cultured (E:T ratio of 5:1 for 4 hours) with autologous CD4+ T cells or CD4+ T cells in vitro infected with HIV. Cells were then stained for CD56 and CD107a and analyzed by flow cytometry to determine the NK cell response to HIV-infected CD4+ T cells or uninfected controls.

To measure the possibility of NK cell exhaustion after N-803 treatment, we will compare NK cells isolated from PBMC prior to and 7 days after each N-803 dose for the following analyses validated in our lab: 1) NK cell degranulation measured after 4 hour exposure to K562 cells; 2) NK cell proliferation by measure of CellTrace dye dilution after cytokine activation; 3) NK cell expression of transcription factors T-bet and Eomesodermin by intracellular flow; 4) NK cell expression of checkpoints PD-1 and TIGIT expression; 5) NK cell metabolic exhaustion by measure of intracellular CPT1a expression. CPT1a is a key component of FAO machinery, shown by us to be downregulated in functionally exhausted NK cells induced by repeated exposure of IL-15. We will also look for IL-15 receptor components (IL-15Rα, β, and γ) to look for modulation after N-803 therapy as well as indirect activation of monocytes and macrophages.

We will also explore if the newly described "adaptive NK cells" are stimulated by N-803 and if they play a role in reservoir reduction. Adpative NK cells lack the HLA-E specific inhibitory receptor, NKG2A, but posess the activating receptor NKG2C (45). These cells also lack the CD16 adaptor molecule, FcεR1γ, but retain CD3ζ for potent signaling. While the mechanism is still not well understood, this population appears to be induced or expanded in the context of past CMV infection and is enhanced with CMV reactivation as often happens in immunosuppressed people. We have shown that "adaptive" NK cells have increased function over conventional NK cells (46) and that these cells have a hypomethymation signature similar to CD8 T cells, are distinct from conventional NK cells and exhibit properties of immune memory. One striking difference between adaptive NK cell and T cell responses are that adaptive NK cells have enhanced responses to targets different from the antigens that induced their initial development (47). While the proportion of adaptive NK cells needed to erradicate disease is not known, we hypothesize that N-803 will enhance the function of this unique subset and we will explore if this is important for reservoir reduction. NK cell phenotype will be assessed for CD56, NKG2C, NKG2A, CD16, FcεR1γ and CD3ζ by flow cytometry in participant samples before and at various timepoints during the course of N-803 therapy to determine if N-803 induces expansion of the "adaptive" NK cell subset. Considering the enhanced function of "adaptive" NK cells and the importance of CD16 expressing NK cells in mediating ADCC, it is important to understand how these subsets respond to N-803 stimulation as significant loss of these populations may negatively impact responses to HIV infected cells. Conversely, expansion or functional enhancement of these NK cell subsets would positively affect surveillance of infected cells. Polyclonal

NK cells (mixed conventional and adaptive from $CMV^+$ donors) degranulate and produce type-II interferon in response to the HIV-infected T cell lines (HIVIIIB and ACH-2) compared to their uninfected counterparts (H9 and CEM CD4). Thus, endogenous NK cells or healthy donor NK cells alone may be good enough to eradicate HIV reservoir infected cells. However, it is possible that other signals will be needed.

## 7.11. Virologic Measures

We will apply a broad range of virologic assays to the samples collected to measure changes to the virus reservoir, by compartment. Our focus is on the B cell follicle, especially infected T follicular helper cells (TFh).

*EDITS assay (Karn Laboratory, Case Western Reserve University):* Dr. Karn has recently developed a novel Next Generation Sequencing (NGS)-based protocol, called EDITS (Envelope Detection by Induced Transcription-based Sequencing), to measure inducible cell-associated HIV RNA (8). This assay is extremely sensitive and will allow us to both determine the extent of proviral reactivation in response to N-803 *in vivo* and its impact on the overall latent reservoir in the peripheral circulation and lymphatic tissues. Samples from PB, LN, and GALT will be analyzed with and without ConA stimulation and the number of cell equivalents that are transcribing HIV will be determined. Sequences of envelope are detected using a nested PCR on 1.25 $x10^6$ resting memory cells from participants before and after ConA stimulation. Samples from different participants are bar-coded, pooled, and sequenced simultaneously which saves both time and sequencing costs, and allows accurate comparisons since input cDNA levels are effectively normalized. All EDITS experiments include control reactions set up without the addition of reverse transcriptase, as well as no template controls. In addition, this assay is quantitative because latently infected cells carry, on average, only one provirus (9) and therefore the frequency of inducible RNAs is proportional to the numbers of inducible cells in each of the wells. Comparing the reads obtained in the EDITS assay after maximal activation of cells by treatment with the mitogenic lectin ConA to a standard curve obtained by sorting known numbers of productively infected cells provides an accurate measurement of the number of latently infected cells in any participant sample. Using this method, we are able to accurately detect 20% changes in the pool of cells with inducible HIV RNA.

Ultrasensitive qPCR diagnostics will be used to quantify HIV RNA to single-copy levels. Cell-associated (CA) total RNA (48) and integrated DNA (49) will be quantified using qPCR assays where nucleic acid input is normalized to cell number. These or similar techniques will be used to identify HIV viral reservoir in adipose tissue.

## 7.12. Leukapheresis

Leukapheresis will be performed at Day -14 to -7 before first dose and between 7-14 days after the last dose. The UMN group has several IRB approved protocols that include leukapheresis. The collection will be performed by the University of Minnesota Medical Center - Fairview Transfusion Service staff following standard clinic policies and procedures. Leukapheresis will occur in the apheresis center and will require an apheresis consult visit. Leukapheresis is not a study-specific procedure. If the participant is not eligible for leukapheresis based on the apheresis consultation appointment, an ultrasound-guided large volume (60 mL) blood draw be scheduled at the CRU instead.

## 7.13. Colonoscopy with Biopsy

Colonoscopy with ileal and rectal biopsy will be performed at Day -14 to -7 before the first dose and between 7-14 days after the last dose for all participants. The procedure will be done in the Endoscopy Unit of the University of Minnesota Medical Center by a senior faculty Gastroenterologist, Alexander Khoruts, MD, under conscious sedation. Subjects will complete the required bowel regimen the night prior to their scheduled procedure. For the procedure, the participant will lie on their left side and the colonoscope will be inserted and guided through the colon. After 14-18 small tissue biopsies of the ileum and rectum are obtained, the colonoscope will be removed. To date we have safely performed this procedure on > 100 HIV infected participants in clinical trials at UMN.

## 7.14. Inguinal Lymph Node Biopsy

Inguinal LN biopsy will be performed at Day -14 to -7 before the first dose and between 7-14 days after the last dose on all participants from both study sites. The procedure will be performed by a general surgeon in an outpatient setting using standard surgical procedures under local anesthesia at the University of Minnesota study site. Ultrasound will be used to identify the appropriate lymph nodes. The surgeon will clean and sterilize the inguinal area, inject a local anesthetic and make a small (5 cm) incision over the lymph node. The lymph node and adjacent adipose tissue will be removed and the incision closed. Participants will be observed for 4 hours post-surgery for any side effects. If none are observed, participants will be allowed to leave. We have safely collected > 300 lymph nodes using this procedure with only minor adverse events (incisional pain and 2 minor infections requiring oral antibiotics).

## 7.15. Pulmonary Function Test (PFT)

A pulmonary function test will be done at Day -14 to -7 before the first dose and between 7-14 days after the last dose. A PFT is a non-invasive breathing test that shows how well the lungs are working. The test measures lung volume, capacity, rate of flow, and gas exchange. They will be performed in the research unit by trained staff. If there is a significant abnormality or concern, the subject will be referred back to their primary care physician.

## 7.16. EKG

A standard 12 lead EKG will be obtained at screening, before study drug is administered on Days 0, 21, and 42, and one week post each drug administration on Days 7, 28, and 49. Minor abnormalities or inconclusive results can be evaluated with a standard treadmill stress test or a stress echocardiogram evaluation at the discretion of the PI. Ischemic changes identified by stress treadmill test will be exclusionary. If no significant abnormalities are detected the individual can participate. If there is a significant abnormality or concern, the subject will be referred back to their primary care physician.

## 7.17. Sample Processing and Specimen Management

Samples obtained from participants at UMN will be managed using established protocols in the Schacker Laboratory. Blood, LN, and gut samples are transported on ice to the Schacker Lab shortly after the blood draw or tissue harvest. Processing and storage for preservation of live cells, DNA and RNA or FFPE tissues occurs as soon as the samples arrive in the lab. Samples preserved for preservation of live cells are maintained in liquid nitrogen in a freezer facility at the Microbiology Research Facility that is monitored by a programmable scanning alarm system wired into the university's telephone system.

### 7.18. COVID-19 Screening and Testing

During the COVID-19 pandemic, we will screen for COVID-19 symptoms before the patients arrive for visits and get a Polymerase chain reaction (PCR) 2-4 -days prior to any injections or procedures (with noted exceptions below). Participants will be asked to isolate at home after their PCR test and prior to injections and biopsy visits.

If COVID-19 is diagnosed at any time during the study, the patient will be instructed to isolate according to current recommendations provided by the Centers Disease Control and Prevention (CDC). Any missed study visits resulting from COVID-19 isolation will be rescheduled, and complete recovery from COVID-19 will be documented before proceeding with study procedures. We will continue to follow guidelines established by M Health Fairview around clinic visits and endoscopy regarding testing after COVID-19. It has been observed throughout the pandemic that persons with prior COVID-19 are protected from reinfection for at least 90 days, but may have persistent/recurrent positive PCR tests during that period despite resolution of infection. Based on these observations, **participants with a prior documented history of COVID-19 will be excluded from PCR screening for COVID-19 for a period of 90 days from time of diagnosis.** This is consistent with established local and national guidelines. We will continue screening for COVID-19 symptoms at all study visits.

### 7.19. Stool Collection

Stool will be collected at baseline, day 28, post study drug procedures, and 90 days after 3$^{rd}$ dose follow-up visits. Participants will be sent home with a collection kit at screening, day 21, day 49 and 60 days after 3$^{rd}$ dose follow-up visits; and asked to collect sample within 3 days of the next visit, and bring back on their next visit with the container provided.

# 8. STUDY SCHEDULE BY VISIT

Participants will be consented by the Principal Investigator or the research team before any procedures take place. All efforts will be made to have all visits occur between 8am and 11am, in order to account for diurnal variation in CD4+ T cell count and other assays and to allow for prompt specimen processing.

Once a participant is identified as potentially eligible by phone screening and is to be scheduled for a screening visit, a unique identifier will be assigned. New participants will be assigned a unique number Once a unique ID is utilized, it cannot be reassigned to another subject.

Participants who satisfy all inclusion/exclusion criteria will be enrolled into the study, which is summarized in the Schedule of Events below.
As stated in section 5.3, N-803 administration may be delayed by up to 14 days (resulting in a maximum 35 day period between doses).
Scheduled evaluations, other than N-803 dose administration days or procedure days, during the course of the study may be performed +/-3 days from the targeted date.
Baseline procedures are planned for Day -7, but can occur as earlier as Day -14. Final procedures are planned for Day 49, but can occur as late as Day 56.

| | Screening Period | Baseline Procedures | Dose 1 | | Dose 2 | | Dose 3 | | Post Study Drug Procedures | Post-biopsy F/U | 1 Month F/U | 2 Month F/U | 3 Month F/U (final visit) | Early Discontinuation |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Day -60 to -14 | Day -14 to -7 | Day 0 | Day 7 | Day 21 | Day 28 | Day 42 | Day 49 | Day 49-56 | 7 days after LN biopsy | Day 77-81 | Day 107-111 | Day 137-141 | |
| Informed Consent | X | | | | | | | | | | | | | |
| Medical History | X | | | | | | | | | | | | | |
| Complete Physical Exam[1] | X | | | | | | | | | | | | | |
| Targeted Physical Exam or Assessment[1] | | X | X | X | X | X | X | X | X | X | | | | X |
| Medication Review[2] | X | X | X | X | X | X | X | X | X | X | X | X | X | X |
| Vitals[3] | X | X | X | X | X | X | X | X | X | X | X | X | X | X |
| AE Assessment | | X | X | X | X | X | X | X | X | X | X | X | X | X |
| CBCD[4] | X | | X | X | X | X | X | X | X | | X | X | X | X |
| Serum chem and hepatic panel[4] | X | | X | X | X | X | X | X | X | | X | X | X | X |
| T cell subset profile[5] | X | X | X | X | X | X | X | X | X | | X | X | X | X |
| Plasma viral load[5] | X | | X | X | X | X | X | X | X | | X | X | X | X |
| Quantiferon gold[4] | X | | | | | | | | | | | | | |
| Hepatitis B ab/ag[4] | X | | | | | | | | | | | | | |
| Hepatitis C ab/PCR[4] | X | | | | | | | | | | | | | |
| PBMC/plasma for storage | | X | X | X | X | X | X | X | X | | X | X | X | X |

| | Screening Period | Baseline Procedures | Dose 1 | | Dose 2 | | Dose 3 | | Post Study Drug Procedures | Post-biopsy F/U | 1 Month F/U | 2 Month F/U | 3 Month F/U (final visit) | Early Discontinuation |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Day -60 to -14 | Day -14 to -7 | Day 0 | Day 7 | Day 21 | Day 28 | Day 42 | Day 49 | Day 49-56 | 7 days after LN biopsy | Day 77-81 | Day 107-111 | Day 137-141 | |
| Serum or urine B-HCG[4] | X | | X | | X | | X | | | | | | | X |
| PAXgene tube | | X | X | X | X | X | X | X | X | | X | X | X | |
| EKG[6] | X | | X | X | X | X | X | X | | | | | | |
| LN biopsy[7] | | X | | | | | | | X | | | | | |
| Colon biopsy[8] | | X | | | | | | | X | | | | | |
| Leukapheresis[9] | | X | | | | | | | X | | | | | |
| PFTs[10] | | X | | | | | | | X | | | | | |
| N-803 dose[11] | | | X | | X | | X | | | | | | | |
| COVID-19 Screen[12] | X | X | X | X | X | X | X | X | X | X | X | X | X | X |
| COVID-19 PCR test[12] | | X | X | | X | | X | | X | | | | | |
| Stool Collection | | X | | | | X | | | X | | | | X | |

1. See section 7.4
2. See section 7.1
3. Vitals to include temperature, blood pressure, pulse, height, weight, and respiratory rate
4. See section 7.5
5. See section 7.6
6. See section 7.16
7. See section 7.14
8. See section 7.13
9. See section 7.12
10. See section 7.15
11. See section 5.2

| | Screening Period | Baseline Procedures | Dose 1 | | Dose 2 | | Dose 3 | | Post Study Drug Procedures | Post-biopsy F/U | 1 Month F/U | 2 Month F/U | 3 Month F/U (final visit) | Early Discontinuation |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Day -60 to -14 | Day -14 to -7 | Day 0 | Day 7 | Day 21 | Day 28 | Day 42 | Day 49 | Day 49-56 | 7 days after LN biopsy | Day 77-81 | Day 107-111 | Day 137-141 | |
| 12. COVID-19 PCR will not be performed for 90 days following COVID-19 diagnosis. See section 7.18 | | | | | | | | | | | | | | |

## 8.1. Screening

Participants will be consented by the investigator or qualified research study staff before any procedures take place. Participants will be screened for COVID-19 symptoms before coming in for their visit.

The following procedures will be completed to confirm eligibility:

- Medical history
- Medication review
- Complete physical exam
- Vitals
- EKG to measure QT interval (see section 7.17)
- Screening blood tests (parameters considered in evaluating eligibility criteria in parentheses):
    - CBCD (WBC, hemoglobin, platelet count, ANC)
    - Hepatitis B surface antigen (if hepatitis B surface antibody positive) and Hepatitis C antibody
    - Quantiferon gold for TB
    - Serum or urine β-HCG (females of childbearing potential only)
    - Plasma HIV RNA
    - CD4+/CD8+ T cell counts
    - CBASIC, BHEPAT (sodium, potassium, bicarbonate, creatinine, eGFR, glucose, calcium, albumin, total bilirubin, alkaline phosphatase, ALT, AST)

Screening evaluations should be completed within 60 days prior to administration of first N-803 dose.

## 8.2. Baseline Procedures: Days -14 to -7

The following procedures will be performed between days -14 and -7 prior to administration of first N-803 dose:

- Targeted physical exam/assessment
- Vitals
- Medication review and AE assessment
- PFTs
- Leukapheresis or large-volume blood draw if the participant is ineligible for leukapheresis
- Colonoscopy with biopsy
- Inguinal LN biopsy
- CD4+/CD8+ T cell counts
- Plasma and PBMCs collected for research storage
- PAXgene tube
- Stool collection

All procedures must be performed within the window, but not necessarily on the same visit day. During the COVID-19 pandemic, participants will be screened for COVID-19 symptoms before coming in for their visit. In addition, a PCR test will be collected 2-4 days before any procedure takes place, except in cases when COVID-19 has been diagnosed in the last 90 days. Participants will be asked to isolate at home after their PCR test and prior to biopsy visits.

In order for a participant to proceed with the administration of study drug, there must be tissue available from the baseline procedures. If there is not tissue available, the participant will be replaced for purposes of efficacy (see section 3.2)

## 8.3. Dose 1 (Day 0)

The following procedures will be performed:

- Targeted physical exam
- Vitals
- Medication review and AE assessment
- EKG
- Blood collection:
    - CBCD
    - CBASIC, BHEPAT
    - CD4+/CD8+ T cell counts
    - Plasma HIV RNA
    - Serum or urine β-HCG (females of childbearing potential only)
    - Plasma and PBMCs for research storage
    - PAXgene tube
- N-803 injection: 6 mcg/kg SQ
    - Prior to receiving N-803 injection, participants will have the following pre-medication administered:
        - Acetaminophen 650 mg PO and diphenhydramine 25 mg PO/IV approximately thirty minutes before.
    - Participants will have their N-803 injection and be required to stay for 2 hours for observation for toxicity. Vital signs (heart rate, blood pressure, respiration, temperature, and oxygen saturation) will be documented prior to each N-803 injection and then at 30, 60, and 120 minutes with a ± 20 minute window for each time point. If they do not have signs of toxicity they will be sent home and instructed to call the on-call physician if symptoms occur while at home.
    - After they are discharged from observation, participants will be instructed to take acetaminophen 650 mg PO and diphenhydramine 25 mg PO 4 hours after receiving the N-803 injection. These medications will be provided by the study at no cost to the participant.

During the COVID-19 pandemic, participants will be screened for COVID-19 symptoms before coming in for their visit. In addition, a PCR test will be collected 2-4 days before any study drug is given, except in cases when COVID-19 has been diagnosed in the last 90 days. Participants will be asked to isolate at home after their PCR test and prior to injections.

## 8.4. Dose 1 (Day 7)

The following procedures will be performed:

- Targeted physical assessment
- Vitals
- Medication review and AE assessment
- EKG
- Blood collection:

- CBCD
- CBASIC, BHEPAT
- CD4+/CD8+ T cell counts
- Plasma HIV RNA
- Plasma and PBMCs for research storage
- PAXgene tube

During the COVID-19 pandemic, participants will be screened for COVID-19 symptoms before coming in for their visit.

## 8.5. Dose 2 (Day 21)

The following procedures will be performed:

- Targeted physical exam
- Vitals
- Medication review and AE assessment
- EKG
- Blood collection:
  - CBCD
  - CBASIC, BHEPAT
  - CD4+/CD8+ T cell counts
  - Plasma HIV RNA
  - Serum or urine β-HCG (females of childbearing potential only)
  - Plasma and PBMCs for research storage
  - PAXgene tube
- N-803 injection: 6 mcg/kg SQ
  - Prior to receiving N-803 injection, participants will have the following pre-medication administered:
    - Acetaminophen 650 mg PO and diphenhydramine 25 mg PO/IV approximately thirty minutes before.
  - Participants will have their N-803 injection and be required to stay for 2 hours for observation for toxicity. Vital signs (heart rate, blood pressure, respiration, temperature, and oxygen saturation) will be documented prior to each N-803 injection and then at 30, 60, and 120 minutes with a ± 20 minute window for each time point. If they do not have signs of toxicity they will be sent home and instructed to call the on-call physician if symptoms occur while at home.
  - After they are discharged from observation, participants will be instructed to take acetaminophen 650 mg PO and diphenhydramine 25 mg PO 4 hours after receiving the N-803 injection. These medications will be provided by the study at no cost to the participant.

During the COVID-19 pandemic, participants will be screened for COVID-19 symptoms before coming in for their visit. In addition, a PCR test will be collected 2-4 days before any study drug is given, except in cases when COVID-19 has been diagnosed in the last 90 days. Participants will be asked to isolate at home after their PCR test and prior to injections.

## 8.6. Dose 2 (Day 28)

The following procedures will be performed:

- Targeted physical assessment
- Vitals
- Medication review and AE assessment
- EKG
- Blood collection:
    - CBCD
    - CBASIC, BHEPAT
    - CD4+/CD8+ T cell counts
    - Plasma HIV RNA
    - Plasma and PBMCs for research storage
    - PAXgene tube
- Stool collection

During the COVID-19 pandemic, participants will be screened for COVID-19 symptoms before coming in for their visit.

## 8.7. Dose 3 (Day 42)

The following procedures will be performed:

- Targeted physical exam
- Vitals
- Medication review and AE assessment
- EKG
- Blood collection:
    - CBCD
    - CBASIC, BHEPAT
    - CD4+/CD8+ T cell counts
    - Plasma HIV RNA
    - Serum or urine β-HCG (females of childbearing potential only)
    - Plasma and PBMCs for research storage
    - PAXgene tube
- N-803 injection: 6 mcg/kg SQ
    - Prior to receiving N-803 injection, participants will have the following pre-medication administered:
        - Acetaminophen 650 mg PO and diphenhydramine 25 mg PO/IV approximately thirty minutes before.
    - Participants will have their N-803 injection and be required to stay for 2 hours for observation for toxicity. Vital signs (heart rate, blood pressure, respiration, temperature, and oxygen saturation) will be documented prior to each N-803 injection and then at 30, 60, and 120 minutes with a ± 20 minute window for each time point. If they do not have signs of toxicity they will be sent home and instructed to call the on-call physician if symptoms occur while at home.
    - After they are discharged from observation, participants will be instructed to take acetaminophen 650 mg PO and diphenhydramine 25 mg PO 4 hours after receiving the N-803 injection. These medications will be provided by the study at no cost to the participant.

During the COVID-19 pandemic, participants will be screened for COVID-19 symptoms before coming in for their visit. In addition, a PCR test will be collected 2-4 days before any study drug is given, except in cases when COVID-19 has been diagnosed in the last 90 days. Participants will be asked to isolate at home after their PCR test and prior to injections.

## 8.8. Dose 3 (Day 49)

The following procedures will be performed:

- Targeted physical assessment
- Vitals
- Medication review and AE assessment
- EKG
- Blood collection:
    - CBCD
    - CBASIC, BHEPAT
    - CD4+/CD8+ T cell counts
    - Plasma HIV RNA
    - Plasma and PBMCs for research storage
    - PAXgene tube

During the COVID-19 pandemic, participants will be screened for COVID-19 symptoms before coming in for their visit.

## 8.9. Post Study Drug Procedures (Day 49 - 56)

The following procedures will be performed as early as day 49 and up to day 56 (7-14 days post administration of final N-803 dose):

- Targeted physical exam
- Vitals
- Medication review and AE assessment
- PFTs
- Leukapheresis or large-volume blood draw if the participant is ineligible for leukapheresis
- Colonoscopy with biopsy
- Inguinal LN biopsy
- CD4+/CD8+ T cell counts
- CBCD
- CBASIC, BHEPAT
- Plasma HIV RNA
- Plasma and PBMCs collected for research storage
- PAXgene tube\
- Stool collection

All procedures must be performed within the window, but not necessarily on the same visit day. During the COVID-19 pandemic, participants will be screened for COVID-19 symptoms before coming in for their visit. In addition, a PCR test will be collected 2-4 days before any procedure is performed, except in cases when COVID-19 has been diagnosed in the last 90 days. Participants will be asked to isolate at home after their PCR test and prior biopsy visits.

## 8.10. Post Biopsy Follow Up (within 7 days of final LN biopsy)

The following procedures will be performed after the final LN biopsy:

- Targeted physical exam/assessment
- Vitals
- Medication review and AE assessment

During the COVID-19 pandemic, participants will be screened for COVID-19 symptoms before coming in for their visit.

## 8.11. Monthly for 3 Months (Day 77-81; Day 107-111; Day 137-141)

The following procedures will be performed at all 3 visits:

- Vitals
- Medication review and AE assessment
- CBCD
- CBASIC, BHEPAT
- Plasma HIV RNA
- CD4+/CD8+ T cell counts
- Plasma and PBMCs for research storage
- PAXgene tube

In addition, the following procedures will be performed at the final/3 month (day 137-141) visit:

- Stool collection

The 90 days after 3rd dose F/U visit will serve as the final study visit. During the COVID-19 pandemic, participants will be screened for COVID-19 symptoms before coming in for their visit.

## 8.12. Early Discontinuation Visit

If a participant is discontinued per the list outlined in section 11.1, an early discontinuation visit should occur as soon as possible.

The following procedures will be performed:

- Targeted physical exam/assessment
- Vitals
- Medication review and AE assessment
- CBCD
- CBASIC, BHEPAT
- CD4+/CD8+ T Cell counts
- Plasma HIV RNA
- Plasma and PBMCs for research storage
- Serum or urine β-HCG (if indicated for females of childbearing potential only

During the COVID-19 pandemic, participants will be screened for COVID-19 symptoms before coming in for their visit.

### 8.13. Unscheduled Visit(s)

At the discretion of the PI, participants may be asked to return for a visit not directed by the protocol if they believe such a visit is needed for additional testing to address abnormal lab values or to assess a specific side effect or symptom during the course of the study.

# 9. ADVERSE EVENT DOCUMENTATION and REPORTING

## 9.1. Definitions

The following definitions are based on the Code of Federal Regulations Title 21 Part 312.32 (21CFR312.32 (a)).

- **Adverse Event:** Any untoward medical occurrence associated with the use of a drug in humans, whether or not considered related to study agents (N-803).

- **Suspected Adverse Reaction**:
  Any adverse event for which there is a reasonable possibility that N-803 caused the adverse event.

- **Life-Threatening Adverse Event or Life-Threatening Suspected Adverse Reaction**: An adverse event or suspected adverse reaction is considered "life-threatening" if, in the view of either the investigator or sponsor, its occurrence places the patient or subject at immediate risk of death.

- **Serious Adverse Event or Serious Suspected Adverse Reaction**: An adverse event or suspected adverse reaction is considered "serious" if, in the view of either the investigator or sponsor, it results in any of the following outcomes:
  - Death
  - A life-threatening adverse event
  - Inpatient hospitalization or prolongation of existing hospitalization
  - A persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
  - A congenital anomaly/birth defect.

- Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered serious when, based upon appropriate medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.

- If either the IND sponsor or the investigator believes the event is life-threatening or serious, the event must be evaluated by the sponsor for expedited reporting (21CRF 312.32(a))

- **Unexpected adverse event or unexpected suspected adverse reaction:** An adverse event or suspected adverse reaction is considered "unexpected" if it is not listed in the investigator brochure or is not listed at the specificity or severity that has been observed, or, if an investigator brochure is not required or available, is not consistent with the risk information described in the general investigational plan or elsewhere in the current application, as amended. Thus, adverse events that

occur as part of the disease process or underlying medical conditions are considered unexpected.

- **Unanticipated (unexpected) problems/events** as defined by the University of Minnesota IRB are those that are not already described as potential risks in the consent form, not listed in the Investigator's Brochure or not part of an underlying disease.

## 9.2. AE Severity

The Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events Corrected Version 2.1 (July 2017) (http://rsc.niaid.nih.gov/clinical-research-sites/daids-adverse-event-grading-tables) should be used to assess and grade AE severity, including laboratory abnormalities judged to be clinically significant.

## 9.3. AE Relationship to Study Drug

The relationship of an AE to the study drug should be assessed using the following the guidelines in **Table 8**.

**Table 8: AE Relationship to Study Drug**

| Relationship to Drug | Comment |
|---|---|
| Related | There is a reasonable possibility that the AE may be related to the study agent(s). |
| Not related | There is not a reasonable possibility that the AE is related to the study agent(s). |

## 9.4. Reporting Procedures

The Investigator, or study staff, will probe, via discussion with the subject, for the occurrence of AEs during each subject visit and record the information in the site's source documents. Adverse events will be recorded in the participant CRF. Adverse events will be described by duration (start and stop dates and times), severity, outcome, treatment and relation to study drug, or if unrelated, the cause. All lab values will be frequently monitored by the Principal Investigator or qualified Co-Investigator in a timely manner. They will review these data, assess degree of severity, and make a relationship assessment to study drug. Study sites will document all AEs that occur (whether or not related to study drug). The collection period for all AEs will begin after informed consent is obtained and end after procedures for the final study visit have been completed.

All SAEs will be evaluated to assess if the event meets reporting criteria in accordance with the standard operating procedures and policies of the local Institutional Review Board (IRB)/Independent Ethics Committee (IEC). SAEs will also be reported to the U.S. Food and Drug Administration (FDA) per the reporting requirements specified by 21 CFR 312.32, with copy to the DAIDS Medical Officer (MO). Details of all related SAEs will be sent to the SMC and DAIDS MO no later than 3 reporting days after the investigators become aware of the event. For the purposes of expedited reporting to the SMC and the DAIDS Medical Officer, the definition of a "reporting day" in Version 2.0 (January 2010) of the DAIDS EAE Manual https://rsc.niaid.nih.gov/clinical-research-sites/manual-expedited-reporting-adverse-events-daids will be used.

**Safety Monitoring Committee Reporting**

The following adverse events will be reported to the SMC:

- Adverse events graded 3 and above;
- Any related, unexpected, and serious adverse events.

Details of all severe, life-threatening, or fatal adverse events will be sent to the SMC within 24 hours for review.

If a death occurs during the study and it is determined to be related either to the study drug(s) or to a study procedure, the Investigator or his/her designee must notify the SMC Chair (or qualified alternate) within 1 business day of knowledge of the event. The contact may be by phone or e-mail

**IRB Reporting Requirements**

Investigators must assess problems and events that occur on a study and comply with the appropriate reporting requirement.

**Events that require prompt reporting to the University of Minnesota IRB:**

Refer to the University of Minnesota's toolkit for what events are considered promptly reportable and should be submitted to the IRB via the appropriate forms within **5 working days** of when the researcher receives knowledge of the event.

## 9.5. Summary of Reporting Guidelines and Timeframes

| Agency | Criteria for Reporting | Timeframe | Form to Use | Submission address/fax numbers |
|---|---|---|---|---|
| IRB | **UPIRTSO**: any adverse event which in the opinion of the local investigator was unanticipated, reflects new or increased risk to the subjects and was possibly related to the research procedures | 5 Working Days | IRB Reporting Form | ETHOS Submission |
| IRB | **Other Problems or Events** meeting the definition of Prompt Reporting in HRP103 | 5 Working Days | IRB Reporting Form | ETHOS Submission |

| | | | | |
|---|---|---|---|---|
| FDA | Unexpected and fatal or life threatening suspected adverse | As soon as possible but no later than 7 Calendar-Days | MedWatch 3500A | U.S. Food and Drug Administration<br>Center for Biologics Evaluation and Research<br>Document Control Center<br>10903 New Hampshire Avenue<br>WO71, G112<br>Silver Spring, MD 20993-0002 |
| FDA | 1) Serious and unexpected suspected adverse reaction or<br>2) increased occurrence of serious suspected adverse reactions over that listed in the protocol or investigator<br>brochure or<br>3) findings from other sources (other studies, animal or in vitro testing) | As soon as possible but no later than 15 Calendar-Days | MedWatch 3500A | U.S. Food and Drug Administration<br>Center for Biologics Evaluation and Research<br>Document Control Center<br>10903 New Hampshire Avenue<br>WO71, G112<br>Silver Spring, MD 20993-0002 Form |
| FDA | All other events per CFR 312.33 | At time of IND annual report | Summary report | Submit as part of the IND annual report |
| SMC | All severe, life-threatening, or fatal adverse events | 24 hours | IRB report form | SMC Chair |
| SMC | All other Adverse events meeting reporting criteria per section 9.4 | 24 hours | IRB report form | SMC members |

The DAIDS MO and Program Officer (PO) will also receive all documents provided to the SMC and all reports form the SMC.

## 9.6. Procedures for Documenting Pregnancy During the Trial

Potential participants who are pregnant or expect to become pregnant during the course of the trial will be excluded from participation in the trial. Participants should not become pregnant, breastfeed, father a baby, and/or donate eggs/sperm on this research study and for four months after receiving the final dose of study drug. Should a subject become pregnant after enrolling in the trial, she will not be given any further treatments with study drug and follow-up invasive procedures (LN biopsy, leukapheresis, and colon biopsy) will not be performed. The pregnancy will be reported to the antiretroviral pregnancy registry. A Pregnancy Form will be completed by the Investigator and submitted to the study team and DAIDS MO within 5 working days after learning of the pregnancy. The Investigator will also report this event to the IRB within 5 working days of becoming aware of the pregnancy. Sites must request the subject's permission to query pregnancy outcome and follow each subject to determine the outcome of the pregnancy. When permission is received, participants will continue to be followed for safety assessments to trial discharge per protocol. Results will be summarized in the clinical study report (CSR).

Participants who become pregnant at any point during the trial will continue to be followed for safety assessments without receiving further study drug. Procedures that are contraindicated during pregnancy, including additional treatments, must not be performed. Investigators should use clinical judgment regarding subsequent trial-related blood collection based on the presence or absence of anemia in each subject.

All pregnancies that occur from the time of first screening procedure through four months after completing the study must be reported. The Investigator will monitor the participant and follow the outcome of the pregnancy. If the end of the pregnancy occurs after the trial has been completed, the outcome will be reported directly to the trial team and the DAIDS MO.

Male participants will be instructed through the Informed Consent Form to immediately inform the Investigator if their partner becomes pregnant until the end of follow-up period. A Pregnancy Form will be completed by the Investigator and submitted to the study team within 5 working days after learning of the pregnancy. Attempts will be made to collect and report details of the course and outcome of any pregnancy in the partner of a male participant exposed to study drug or device. The pregnant partner will need to sign an Authorization for Use and Disclosure of Pregnancy Health Information to allow for follow up on her pregnancy. Once the authorization has been signed, the Investigator will update the Pregnancy Form with additional information on the course and outcome of the pregnancy. An Investigator who is contacted by the male participant or his pregnant partner may provide information on the risks of the pregnancy and the possible effects on the fetus, to support an informed decision in cooperation with the treating physician and/or obstetrician.

## 9.7. Reporting Requirements to Nant Drug Safety

Serious AEs (SAEs), whether related or not related to study drug, and pregnancies must be reported to Nant Drug Safety (Nant) within 24 hours with regards to Nant drug use. SAEs must be recorded on a SAE report form or MedWatch form; pregnancies on a Pregnancy Surveillance Form (Nant to provide).

SAE Email Address: SAE.Reporting@NantBio.com
SAE Facsimile Number: 1-800-853-3497

If only limited information is initially available, follow-up reports are required. (Note: Follow-up SAE reports should include the same investigator term(s) initially reported.) If an ongoing SAE changes in its intensity or relationship to study drug or if new information becomes available, a follow-up SAE report should be sent within 24 hours to Nant using the same procedure used for transmitting the initial SAE report. All SAEs should be followed to resolution or stabilization. The Sponsor/Investigator will ensure that all SAEs in the clinical database are reported to Nant and any applicable health authority during the conduct of the study. This reconciliation will occur at least quarterly and be initiated by the sponsor/investigator.

Sponsor/investigator will request a reconciliation report from: SAE.Reporting@NantBio.com
During reconciliation, any events found to not be reported previously to Nant must be sent to: SAE.Reporting@NantBio.com.

The Sponsor/Investigator will be responsible for submitting all safety correspondence to the FDA. Nant will be provided with a simultaneous copy of all Serious adverse event submissions filed with the FDA.

All SAEs submitted to the FDA should simultaneously be faxed or e-mailed to NANT at:
Fax Number: 1-800-853-3497
Email: SAE.Reporting@NantBio.com

# 10. CLINICAL MANAGEMENT ISSUES

**Injection Site Reaction**
Reaction at the injection site, characterized as "Injection Site Erythema or Redness" and/or" Injection Site Induration or Swelling", is an expected and clinically insignificant toxicity. Per the arrangement made with the FDA during the course of the previous dose escalation study, all Grade 3 injection site reactions will be reported in the annual report. A Grade 4 injection site reaction will be reported immediately in accordance with the standard reporting procedures.

**Lymph Node Adenopathy**
Inguinal lymph node adenopathy in the draining, inguinal lymph node has been observed in HIV participants that have received N-803 via subcutaneous injection. All events have been reversible and have returned to baseline within 5-10 days of dosing. Grading for lymph node adenopathy is not contained in the DAIDS toxicity grading scale, therefore, we will grade all lymph node adenopathy with the following grading scheme:

| Parameter | Grade 0 | Grade 1 | Grade 2 | Grade 3 |
|---|---|---|---|---|
| Lymph Node Adenopathy | No increase in Adenopathy | Increase in size of nodes to >5 cm by palpation | Increase in size of adenopathy of > 5 cm that lasts > 7 days but < 14 days or; | Increase in size of nodes to >5 cm for > 14 days or; Adenitis diagnosed by |

| | | | | |
|---|---|---|---|---|
| | | | Ultrasound evidence of necrosis | CT with fever >101.5° F or;<br>Any other evidence of infection |

Administration of study drug should be put on hold until resolved if the lymphadenopathy is a Grade 3 or greater.

**Impaired kidney and liver functions**
Kidney and liver functions may be impaired during N-803 treatment. Use of concomitant nephrotoxic or hepatotoxic medications may further increase toxicity to the kidney and liver. Administration of study drug should be put on hold if Grade 4 or greater. Subject will be followed to resolution, and a consultation will be obtained if further assessment is required.

**Infection**
Antibiotics can be administered to treat infections.

**Fever and chills**
Acetaminophen, indomethacin and hydromorphone can be administered to reduce fever and chills. Administration of study drug should be put on hold if Grade 3 or greater. A clinical assessment will be done.

**Diarrhea, nausea and vomiting**
Antidiarrheals can be used to combat diarrhea; antiemetics can be used to attenuate nausea and vomiting. Steroid-based anti-emetics are not allowed.

**Pruritus and dermatitis**
Hydroxyzine or diphenhydramine can be used to control the symptoms from pruritus. Lotions, topical creams, and ointments should be applied as needed for skin manifestations (see section 5.2.3).

**Life-threatening toxicities**
Life-threatening toxicities, if they are attributed to inflammatory reactions of N-803 in the judgment of the investigators, may be ameliorated by the intravenous administration of dexamethasone or other steroid-based medications. No further injections of study drug will be administered to that participant. This would trigger a study pause.

**Other supportive care**
Other supportive care deemed necessary by the Principal Investigator will be used to ensure participant's' welfare. This will include any isolated deviations or modifications of the care plans outlined above. If it is the case that the same modifications are apparently needed more systematically, then a protocol modification will be considered in consultation with the Sponsor and co-investigators.

Previous editions of the ALT-803 Investigator's Brochure relied heavily on clinical experience with the related cytokine therapeutic Proleukin® Interleukin2 to anticipate potential risks associated with ALT-803 (N-803) administration. This approach was based on the fact that N-803 and IL-2 are both γ chain cytokines and thus could reasonably be predicted to have similar immunostimulatory properties. However, the substantial accumulated information on N-803 effects in humans presented the most recent IB (version 6 dated April 2019) indicates that many side effects of IL-2 are not observed in patients treated with N-803 at the doses being used clinically. For this reason, side effects observed in subjects treated with IL-2 but not N-803, such as capillary leak syndrome, pulmonary dysfunction, acidosis, and gastritis, have been removed from the IB and are reflected in this protocol and its consent form. Refer to the current Investigator Brochure (April 2019 version 6) for additional information

# 11. DISCONTINUATION AND REPLACEMENT OF PARTICIPANTS

## 11.1. Early Removal

A participant may be removed from study treatment at any time if the participant or the study team feels that it is not in the participant's best interest to continue. The following is a list of possible reasons for study removal:

- Participant withdrawal of consent
- Participant is not compliant with study procedures
- Adverse event that in the opinion of the investigator would be in the best interest of the subject to discontinue study treatment
    - Including but not limited to a Grade 4 injection site reaction, sustained use of steroids, any life-threatening AE, a Grade 3 QTc prolongation, and/or EKG evidence for ischemia at Days 21 & 42.
- Protocol violation requiring discontinuation of study treatment
- Lost to follow-up
- Sponsor request for early termination of study
- Positive pregnancy test (females)
- A protocol violation

If a subject is withdrawn from treatment due to an adverse event, the subject will be followed and treated by the Investigator until the abnormal parameter or symptom has resolved or stabilized.
All participants who discontinue study treatment should come in for an early discontinuation visit as soon as possible and then should be encouraged to complete all remaining scheduled visits and procedures.

## 11.2. Withdrawal of Participants from the Study

A subject may be withdrawn from the study at any time if the subject, the investigator, or the Sponsor feels that it is not in the subject's best interest to continue.

All participants are free to withdraw from participation at any time, for any reason, specified or unspecified, and without prejudice.

Reasonable attempts will be made by the investigator to provide a reason for subject withdrawals. The reason for the subject's withdrawal from the study will be specified in the subject's source documents. As noted above, participants who discontinue study treatment early should have an early discontinuation visit.

### 11.3. Replacement of Participants

If a participant discontinues participation for any reason they will be replaced so that there are 10 evaluable participants. Participants will be discontinued and replaced if they:

- fail to receive all 3 injections of N-803
- fail to receive an injection of N-803 within the dose administration window as defined in section 5.3
- fail to have one or all of the procedures (i.e. colon biopsy, lymph node biopsy, and leukapheresis) at both indicated timepoints
- initiate systemic steroids or other prohibited medications per section 6.1
- have any change in ART regimen that demonstrates a loss of viral control by measurements of plasma viral load greater than 200 copies/mL on two consecutive measures separated by at least two weeks
- receive a clinical vaccination during the study period (other than influenza or SARS-CoV-2)

The safety data from all of the participants will be reported regardless of whether or not they completed the study or if they were withdrawn and/or replaced. Safety results will be reported for all enrolled and treated participants.

### 11.4. Study Pause Criteria

Enrollment of new participants and the administration of study drug for enrolled participants will be paused and the study plan re-evaluated after consultation with the SMC if:

- any single grade $\geq$4 adverse event, laboratory abnormality, and/or life-threatening toxicity.
- if > 2 participants experience a study drug-related Grade 3 AE. (Grade 3 injection site reactions are excluded from these criteria) or
- a pattern of significant symptoms, physical findings, or laboratory abnormalities (adverse events) that, although individually minor, collectively represent a safety concern in the opinion of the investigator. The team of investigators will review all AEs every 14 days to monitor for any safety concerns. Any safety concern will be reported by the PI to the SMC.

**If the study is paused for safety reasons, notify FDA immediately. Obtain written consent from the FDA before resuming the study.**

# 12. PROTOCOL VIOLATIONS

A protocol violation occurs when the subject or investigator fails to adhere to significant protocol requirements affecting the inclusion, exclusion, subject safety, and or primary endpoint criteria. Protocol violations for this study include, but are not limited to, the following:

- Failure to meet inclusion/exclusion criteria
- Use of a prohibited concomitant medication
- Failure to comply with Good Clinical Practice (GCP) guidelines will also result in a protocol

violation.

When a protocol violation occurs, it will be discussed with the study team and a Note to File detailing the violation will be generated. A copy of the note to file will be filed in the site's regulatory binder. All critical events must be written up by the study team with corrective actions. This will be filed with the DAIDS Medical Officer (MO), the institution's IRB, and in the site's regulatory binder.

# 13. STATISTICAL METHODS AND CONSIDERATIONS

Prior to database lock, a detailed Statistical Analysis Plan (SAP) will be written describing all analyses that will be performed. The SAP will contain any modifications to the analysis plan described below.

## 13.1. Data Sets Analyzed

The data set used for statistical analyses will contain hematology measures from all indicated visits; height and weight from the physical exam (for calculation of BMI); HIV clinical laboratory measures collected at indicated study visits; markers of T cell proliferation and activation from flow cytometry analysis; measures of NK activation and function from LT and blood samples; number of HIV transcribing cell equivalents from the EDITS assay measured in blood and LT; demographic information; and medical history for all participants who satisfy the inclusion criteria.

## 13.2. Safety Analysis

Any AEs will be reported in a study report that will document AEs and completeness of data. This report will include the total number of AEs and total number of participants experiencing an AE. AEs will be summarized in terms of severity and relatedness. Safety results will be reported for all enrolled and treated participants.

## 13.3. Data Analysis Plan

The primary endpoint for this study is change in frequency of CD8+ T cells in follicles as measured in the LN. Secondary endpoints include change in frequency of CD8+ T cells in GALT, change in frequency of vRNA+ TFH cells, frequency of viral RNA + cells as measured by the EDITS assay, and percent of NK cells expressing activation and ADCC function markers. Prior to analysis, measures reflecting cell count will be log-transformed and percentages will be arcsine transformed. Due to the exploratory nature of the proposed study, all analyses involving the secondary endpoints will be presented in entirety without adjustments for multiple comparisons using a significance level of 0.05.

Linear regression models will be used to assess change in measurements collected only at two timepoints, before and after administration of N-803, including: LT measures of frequency of CD8 T+ cells in the follicles, frequency of vRNA+ TFH cells, amount of HIV RNA/DNA, and percent of NK cells expressing activation and ADCC function markers. The outcome for the models will be change in measurement and baseline level of the measure will be included as a covariate. We will use two-sided tests for assessing if the model intercepts deviate from zero.

Mixed effects models with subject specific random intercepts will be used to evaluate measures collected throughout the study including: PB measures of plasma viral load, CD4+ and CD8+ T cell counts and percentages, CD4/CD8 ratios, and percent of NK cells expressing activation and ADCC function markers. These models will initially test for a linear change over time, but nonlinear trajectories using natural splines

will also be investigated. Permutation p-values will be presented for all models.

We will investigate how sex and other clinical factors, such as age, influence changes in B cell follicle structure, function, and HIV viral measures after N-803 therapy by including relevant clinical factors into our models.

### 13.4. Sample Size and Randomization

Because our study is the first of its kind to examine the impact of N-803 on CD8+ T cells in B cell follicles in humans, preliminary data is limited. In a recent study investigating changes in SIV-specific CD8+ T cells in response to N-803 (Webb et al., 2018), the investigators found that in 9 tissue samples from 6 SIV-infected macaques the frequency of SIV-specific CD8+ T cells per $mm^2$ in the B cell follicle drastically increased after N-803 administration. The average change divided by the standard deviation was approximately 1.11, resulting in a paired, 2-sided t-test p-value of 0.0102. Assuming N-803 has a similar response in humans (Cohen's d = 1.11), and using a type-I error rate of 0.05, our study with 10 participants would have approximately 87% power to detect an effect of this size or greater.

As this is an uncontrolled study, no randomization will be conducted.

## 14. DATA COLLECTION, RETENTION AND MONITORING

### 14.1. Data Collection Instruments

The Investigator will prepare and maintain adequate and accurate source documents designed to record all observations and other pertinent data for each subject treated with the study drug.
Detailed interviews will be conducted at each visit, including questions regarding current medications, medication adherence, intercurrent illnesses, and hospitalizations.

The Investigator is responsible for all information collected on participants enrolled in this study. All data collected during the course of this study must be reviewed and verified for completeness and accuracy by the Investigator.

### 14.2. Data Management Procedures

The data will be entered into a validated database. The Data Management group will be responsible for data processing, in accordance with procedural documentation. Database lock will occur once quality assurance procedures have been completed.

All procedures for the handling and analysis of data will be conducted using good computing practices meeting FDA guidelines for the handling and analysis of data for clinical trials.

### 14.3. Data Quality Control and Reporting

After data have been entered into the study database, a system of computerized data validation checks will be implemented and applied to the database on a regular basis.

All data will be managed and analyzed under the direction of Dr. Anne Eaton in the University of Minnesota Department of Biostatistics.

## 14.4. Archival of Data

The database is safeguarded against unauthorized access by established security procedures; appropriate backup copies of the database and related software files will be maintained. Databases are backed up by the database administrator in conjunction with any updates or changes to the database.

At critical junctures of the protocol (e.g., production of interim reports and final reports), data for analysis is locked and cleaned per established procedures.

## 14.5. Availability and Retention of Investigational Records

The Investigator must make study data accessible to the monitor, other authorized representatives of the Sponsor (or designee), IRB/IEC, and Regulatory Agency (e.g., FDA) inspectors upon request. A file for each subject must be maintained that includes the signed Informed Consent, HIPAA Authorization Form and copies of all source documentation related to that subject. The Investigator must ensure the reliability and availability of source documents from which the information on the CRF was derived.

All study documents (subject files, signed informed consent forms, copies of CRFs, Study File, etc.) must be kept secured for a period of two years following marketing of the investigational product or for two years after centers have been notified that the IND has been discontinued. There may be other circumstances for which the Sponsor is required to maintain study records and, therefore, the Sponsor should be contacted prior to removing study records for any reason.

## 14.6. Monitoring

The site initiation visit will be conducted by a monitor from DAIDS, not the University of Minnesota. This study will be monitored at a minimum of every 6 months by the University of Minnesota Clinical and Translational Science Institute's Clinical Trial Monitoring Program. This program will monitor both the University of Minnesota and Hennepin Healthcare study sites. The study monitor will review subject records to ensure protection of study subjects, compliance with the protocol and accuracy and completeness of the protocol. All subject records will be monitored for appropriate consent and eligibility. A minimum 10% of the case report form data for will be monitored against the source documents for data verification. The monitors also will inspect regulatory files to ensure that regulatory requirements are being followed and pharmacy to review product storage and management.

Clinical site monitoring visits will be conducted according to the U.S. CFR Title 21 Parts 50, 56, and 312 and ICH Guidelines for GCP (E6). By signing this protocol, the Investigator grants permission to the Sponsor (UMN), DAIDS, and appropriate regulatory authorities to conduct on-site monitoring and/or auditing of all appropriate study documentation.

The study data manager and statistician will prepare a quarterly report of accrual, and study discontinuations (and related reasons), baseline characteristics, Grade $\geq 2$ signs and symptoms, Grade $\geq 2$ laboratory abnormalities and reported SAEs. This report will be reviewed by the study team, including the DAIDS MO or designee, on a quarterly basis

### 14.7. Safety Monitoring

The Principal Investigator – Dr. Timothy Schacker, will be the primary person responsible for the monitoring of safety on this trial, which includes reporting of adverse events and unanticipated problems to the study Safety Monitoring Committee (SMC), University of Minnesota Institutional Review Board, and to the FDA. The committee will be composed of at least 3 independent scientists. These individuals were selected based on their expertise in either the clinical management of HIV infection and/or their expertise with clinical trials.

Approximately 1 month after enrollment of the first participant, and then biannually, the SMC will meet and review accrual (including screening and enrollment), AE summaries, including all reported Grade ≥3 AEs, retention of participants including off-study rates, and clinical laboratory data.

In addition to the regularly scheduled reviews, the SMC will perform expedited reviews of the safety data whenever one of the study pause criteria (section 11.4) occurs.

Whenever such an event as specified in 11.4 occurs, enrollment into the study and administration of study drug to enrolled participants will be paused until the SMC review has taken place and a determination has been made that enrollment can resume. The SMC will recommend, based on the results of the review, whether the study can proceed as planned, proceed with modifications, or should be discontinued. Any changes to the study design will have to be approved by the DAIDS Clinical Science Review Committee (CRSC).

The SMC may also be convened if a reason is identified by the study team, DAIDS MO, or study statistician.

The SMC will review progress towards pre-specified benchmarks of enrollment and retention of participants, completion of study procedures, and collection of viable samples. If progress towards any benchmark is not adequate, as determined by the SMC, the SMC will recommend protocol modification or stopping the study if necessary.

Any recommendation for modification of the protocol will be made to DAIDS as well as the protocol team, and any amendment to the protocol will require DAIDS Clinical Science Review Committee (CSRC) approval. All updated versions of the protocol, IB, and related documents will be provided to the DAIDS MO and DAIDS Program Officer (PO).

The DAIDS MO and DAIDS PO will receive copies of all correspondence with the FDA about this study at the time they are reported to the FDA or earlier. The DAIDS MO and DAIDS PO will also receive copies of any correspondence received from the FDA.

# 15. ADMINISTRATIVE, ETHICAL, REGULATORY CONSIDERATIONS

The study will be conducted according to the Declaration of Helsinki, Protection of Human Volunteers (21 CFR 50), Institutional Review Boards (21 CFR 56), and Obligations of Clinical Investigators (21 CFR 312).

To maintain confidentiality, all information that identifies study subjects will be handled in accordance with HIPAA regulations, US State and Federal laws, and University of Minnesota policy. Our sites are committed to ensuring that appropriate measures are taken to protect the privacy and confidentiality of all Personally-Identifiable Health Information (PHI) for which it is responsible. Subjects will sign an authorization to use or disclose protected health info for research purposes. All staff will have been trained in the use of protected health information. All study records will be kept in a locked file cabinet and code sheets linking a subject's name to a subject identification number will be stored separately in another locked file cabinet. Clinical information will not be released without written permission of the subject, except as necessary for monitoring by the FDA and other local, US or international regulatory entities. The Investigator must also comply with all applicable privacy regulations (e.g., Health Insurance Portability and Accountability Act of 1996, EU Data Protection Directive 95/46/EC).

## 15.1. Protocol Amendments

Any amendment to the protocol will be written by PI. Protocol amendments cannot be implemented without prior written IRB/IEC approval except as necessary to eliminate immediate safety hazards to subjects. Any amendment to the protocol requires CSRC approval. A protocol amendment intended to eliminate an apparent immediate hazard to subjects may be implemented immediately, provided the IRBs are notified within five working days.

## 15.2. Institutional Review Boards and Independent Ethics Committees

The protocol and consent form will be reviewed and approved by the IRB/IEC of each participating center prior to study initiation. Serious adverse experiences regardless of causality will be reported to the IRB/IEC in accordance with the standard operating procedures and policies of the IRB/IEC, and the Investigator will keep the IRB/IEC informed as to the progress of the study. The Investigator will obtain assurance of IRB/IEC compliance with regulations.

Any documents that the IRB/IEC may need to fulfill its responsibilities (such as protocol, protocol amendments, Investigator's Brochure, consent forms, information concerning participant recruitment, payment or compensation procedures, or other pertinent information) will be submitted to the IRB/IEC. The IRB/IECs written unconditional approval of the study protocol and the informed consent form will be in the possession of the Investigator before the study is initiated. The IRB/IECs unconditional approval statement will be transmitted by the Investigator to any sponsor prior to the shipment of study supplies to the site. This approval must refer to the study by exact protocol title and number and should identify the documents reviewed and the date of review.

Protocol and/or informed consent modifications or changes may not be initiated without prior written IRB/IEC approval except when necessary to eliminate immediate hazards to the subjects or when the change(s) involves only logistical or administrative aspects of the study. Such modifications will be submitted to the IRB/IEC and written verification that the modification was submitted and subsequently approved should be obtained.

The IRB/IEC must be informed of revisions to other documents originally submitted for review; serious and/or unexpected adverse experiences occurring during the study in accordance with the standard operating procedures and policies of the IRB; new information that may affect adversely the safety of the

subjects of the conduct of the study; an annual update and/or request for re-approval; and when the study has been completed.

## 15.3. Informed Consent Form

Informed consent will be obtained in accordance with the Declaration of Helsinki, ICH GCP, US Code of Federal Regulations for Protection of Human Participants (21 CFR 50.25, CFR 50.27, and CFR Part 56, Subpart A), the Health Insurance Portability and Accountability Act (HIPAA, if applicable), and local regulations.

The Investigator at each study site will prepare an informed consent form and HIPAA authorization and provide the documents to the Sponsor or designee for approval prior to submission to the IRB/IEC. The consent form generated by the Investigator must be acceptable to the Sponsor and be approved by the IRB/IEC. The written consent document will embody the elements of informed consent as described in the International Conference on Harmonization and will also comply with local regulations. The Investigator will send an IRB/IEC-approved copy of the Informed Consent Form to the Sponsor (or designee) for the study file.

A properly executed, written, informed consent will be obtained from each subject prior to entering the subject into the trial. Information should be given in both oral and written form and participants must be given ample opportunity to inquire about details of the study. If a subject is unable to sign the informed consent form (ICF) and the HIPAA authorization, a legal representative may sign for the subject. A copy of the signed consent form will be given to the subject and the original will be maintained with the subject's records.

Per the ICF, any incidental findings that, in opinion of the principal investigator (or co-investigator who is a physician), will be disclosed to the participant in a timely manner. The participant will be given the option to opt out of such information sharing within the ICF.

## 15.4. Subject Compensation

Subjects will be compensated for their time and travel costs for each study visit. Compensation amounts are specified in the study's informed consent form.

## 15.5. Publications

The preparation and submittal for publication of manuscripts containing the study results shall be in accordance with a process determined by mutual written agreement among the study Sponsor and participating institutions. The publication or presentation of any study results shall comply with all applicable privacy laws, including, but not limited to, the Health Insurance Portability and Accountability Act of 1996.

## 15.6. Biohazard Containment

As the transmission of HIV and other blood-borne pathogens can occur through contact with contaminated needles, blood, and blood products, appropriate blood and secretion precautions will be employed by all personnel in the drawing of blood and shipping and handling of all specimens for this study, as currently recommended by the Centers for Disease Control and Prevention and the National Institutes of Health.

All dangerous goods materials, including diagnostic specimens and infectious substances, must be transported according to instructions detailed in the International Air Transport Association (IATA) Dangerous Goods Regulations.

Biohazard waste that is produced in the context of this study will be disposed of in compliance with biohazard waste disposal guidelines at each study site.

### 15.7. Investigator Responsibilities

By signing the Agreement of Investigator form, the Investigator agrees to:

1. Conduct the study in accordance with the protocol and only make changes after notifying the Sponsor (or designee), except when to protect the safety, rights or welfare of participants.

2. Personally conduct or supervise the study (or investigation).

3. Ensure that the requirements relating to obtaining informed consent and IRB review and approval meet federal guidelines, as stated in § 21 CFR, parts 50 and 56.

4. Report to the Sponsor or designee any AEs that occur in the course of the study, in accordance with §21 CFR 312.64.

5. Ensure that all associates, colleagues and employees assisting in the conduct of the study are informed about their obligations in meeting the above commitments.

6. Maintain adequate and accurate records in accordance with §21 CFR 312.62 and to make those records available for inspection with the Sponsor (or designee).

7. Ensure that an IRB that complies with the requirements of §21 CFR part 56 will be responsible for initial and continuing review and approval of the clinical study.

8. Promptly report to the IRB and the Sponsor (or designee) all changes in the research activity and all unanticipated problems involving risks to participants or others (to include amendments and IND safety reports).

9. Seek IRB approval before any changes are made in the research study, except when necessary to eliminate hazards to the patients/participants.

10. Comply with all other requirements regarding the obligations of clinical investigators and all other pertinent requirements listed in § 21 CFR part 312.

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