University of California, San Francisco

Clinical Research Protocol
IMPACT OF EVEROLIMUS ON HIV PERSISTENCE POST KIDNEY (AND KIDNEY/PANCREAS) OR LIVER TRANSPLANT
VERSION 3.0

Protocol Number:	HIVTR-EVE
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Investigational Product:	Zortress (everolimus)
Development Phase:	exploratory
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Approval:

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PI or Sponsor Signature (Name and Title) *Date*

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LIST OF ABBREVIATIONS

AE	adverse event
ART	Antiretroviral therapy
BUN	blood urea nitrogen
CFR	Code of Federal Regulations
CRF	case report form
DMC	Data Monitoring Committee
DSMB	Data Safety Monitoring Board
FDA	Food and Drug Administration
GCP	Good Clinical Practice
HIPAA	Health Insurance Portability and Accountability Act of 1996
HIV	Human Immunodeficiency Virus
ICF	informed consent form
ICH	International Conference on Harmonisation
IEC	Independent Ethics Committee
IRB	Institutional Review Board
PI	Principal Investigator
SAE	serious adverse experience
SAP	Statistical Analysis Plan
TCR	T-Cell Receptor
TILDA	Tat/Rev Induced Limiting Dilution Assay

PROTOCOL SYNOPSIS

TITLE	Impact of everolimus on HIV persistence post kidney (and kidney/pancreas) or liver transplant
SPONSOR	University of California, San Francisco
FUNDING ORGANIZATION	American Foundation for AIDS Research (amfAR)
STUDY DRUG	Zortress (everolimus)
NUMBER OF SITES	1
RATIONALE	We hypothesize that the addition of an mTOR inhibitor (everolimus) to the immunosuppressive regimens of HIV infected transplant recipients will decrease HIV persistence in CD4+ lymphoctyes.
STUDY DESIGN	This is an open-label, single arm, exploratory study
PRIMARY OBJECTIVE	To determine the effect of everolimus on HIV DNA and RNA in CD4+ T cells in HIV infected patients on stable antiretroviral regimens.
SECONDARY OBJECTIVES	1. To determine the effect of everolimus on plasma HIV-1 RNA when added to a stable antiretroviral regimen. 2. To determine the safety and tolerability of everolimus at standard immunosuppressive doses in HIV-infected transplant recipients who are on stable antiretroviral drugs and immunosuppressive regimens.
NUMBER OF SUBJECTS	10
SUBJECT SELECTION CRITERIA	<u>Inclusion Criteria</u>: 1. Solid organ (kidney, kidney/pancreas, or liver) transplant recipient 2. Male or female ≥ 18 years of age. 3. Documentation of HIV-1 infection diagnosis as evidenced by any licensed ELISA and confirmation by Western Blot, or documented history of detectable HIV-1 RNA 4. HIV-1 plasma RNA <50 copies/ml for at least 2 years with at least one measurement per year and most recent viral load within 16 weeks of enrollment and study drug initiation. Episodes of a single HIV plasma RNA 50 - 500 copies/ml will not exclude participation if the subsequent HIV plasma RNA was <50 copies/ml. 5. CD4+ T cell count greater than 200 cell/µl within 16 weeks of enrollment and study drug initiation. 6. Receiving combination antiretroviral therapy (at least 3 agents) 7. Written informed consent obtained from subject or subject's

	legal representative and ability for subject to comply with the requirements of the study. Exclusion Criteria: 1. Pregnant, breastfeeding, or unwilling to practice birth control during participation in the study. 2. Presence of a condition or abnormality that in the opinion of the Investigator would compromise the safety of the patient or the quality of the data. 3. Patients who are intending to modify antiretroviral therapy in the next 6 months for any reason. 4. Serious illness requiring hospitalization or parenteral antibiotics within preceding 3 months. 5. A screening hemoglobin below 11.5 g/dL. 6. A screening TSH consistent with hypothyroidism. 7. Significant renal disease (eGFR < 60 ml/min) or acute nephritis 8. Clinically active hepatitis as evidenced by clinical jaundice or Grade 2 or higher liver function test abnormalities. 9. Hepatic cirrhosis or decompensated chronic liver disease. 10. Concurrent treatment with immunomodulatory drugs, such an interferon-alpha, or exposure to any immunomodulatory drug in past 16 weeks (outside of standard immunosuppression).
TEST PRODUCT, DOSE, AND ROUTE OF ADMINISTRATION	Everolimus will be added to subject's immunosuppressive regimen at .5 mg – 1.5 mg oral twice a day. Dose will depend on a target trough level between 3-8 ng/ml). Calcineurin inhibitors will be decreased to obtain a 50% reduction in trough levels with the addition of everolimus. Subjects will be maintained on that regimen for 6 months.
DURATION OF SUBJECT PARTICIPATION AND DURATION OF STUDY	Subjects will be on study for up to 53 weeks **Screening:** up to 7 days **Treatment:** 182 days (6 months) **Follow-up:** 182 days (6 months) The total duration of the study is expected to be 2 years. 12 months for subject recruitment and 12 for final subject follow-up.

CONCOMMITANT MEDICATIONS	Allowed: Standard therapy for HIV-1 infection and for post renal or liver transplant management are allowed except for treatments noted in the exclusion criteria described above and as noted in the prohibited medications section below. Prohibited: The following medications are prohibited during the study and administration will be considered a protocol violation. • Immunomodulatory drugs outside of standard immunosuppression.
PRIMARY VIROLOGIC ENDPOINT	The primary virologic endpoint of the study will be change from baseline in frequency of CD4+ T cell-associated HIV-1 DNA copy number.
SECONDARY ENDPOINTS	Secondary endpoints will include cell-associated total HIV RNA, plasma RNA, and changes in inducible msRNA, (TILDA) at treatment completion.
SAFETY EVALUATIONS	BUN, Creatinine levels will be monitored weekly for one month and then monthly to detect rejection and/or nephrotoxicity. AST , ALT and ALP will also be monitored weekly in liver transplant recipients. Tacrolimus and everolimus drug levels (12 hour trough levels) will be monitored weekly for one month, then monthly
STATISTICS **Primary Analysis Plan**	The primary virologic endpoint of the study will be change from baseline in frequency of CD4+ T cell-associated HIV-1 DNA copy number . Secondary endpoints will include cell-associated total HIV RNA, plasma RNA, and changes in inducible msRNA, (TILDA) at treatment completion Based on a design with ten patients and a null hypotheses that there is no change in the HIV DNA over time, we estimate that the study will have approximately 80% power to detect a 0.5 log (3.2 fold) difference in HIV DNA levels between baseline and 6-month post-treatment time point (at the 0.05 significance level using a paired t-test). We also estimate the study has 80% power to detect 0.7 log (5.0 fold) and 0.4 log (2.5 fold) differences between pre-treatment and post-treatment in cell-associated RNA and inducible msRNA (TILDA), respectively. These estimates are based on the assumption that HIV DNA, cell-associated RNA and inducible msRNA are log normally distributed and that the standard deviation of the difference in pre- and post-dose levels is 0.5 $\log_{10}$, 0.7 $\log_{10}$, and 0.4 $\log_{10}$, respectively (derived from previous work). We expect to see trends in decreasing cell-associated DNA and RNA by week 26 of everolimus treatment based on our retrospective analysis. The durability of this potential effect will be evaluated 6 months after everolimus

	discontinuation.

1 BACKGROUND

Zortress (everolimus), the 40-O-(2-hydroxyethyl)-derivative of rapamycin, is an mTOR inhibitor approved for rejection prophylaxis in kidney transplant recipients.

1.1 Overview of Clinical Studies

There are three clinical trials in HIV negative kidney transplant recipients which study the safety and efficacy of concomitant use of everolimus and tacrolimus. These three trials include the US09 trial (n=92) [1], the ASSET study (n=224) [2], and the EVEROTAC study (n=35) [3]. These three trials suggest that the use of concentration-controlled everolimus allow substantial minimization of TAC exposure to achieve good renal function without compromising efficacy or safety in de novo renal transplant recipients.

In a separate study in HIV positive kidney recipients, the impact of immunosuppressant therapy on peripheral blood measures of HIV persistence on HIV persistence was explored. Stored plasma and peripheral blood mononubear cells prior to transplantation and at weeks 12, 26, 52, and 104 post-transplant were obtained from 91 transplant recipients. In a multivariate model, a higher baseline CD4 T cell counts (p=.001) and sirolimus use (p=.04) were associated with lower post-transplant HIV DNA levels. Kidney function was not compromised in the patients on sirolimus, and was associated with a significant increase in GFR over time. [4]

2 STUDY RATIONALE

HIV-1 is a retrovirus that integrates into the host genome, primarily in memory CD4+ T cells that can harbor latent, replication-competent HIV-1 DNA for years [5-7]. This latent HIV-1 reservoir is thought to be established during acute infection [8-10], although precisely when, where, and how the reservoir is seeded remains to be determined. The reservoir has a remarkably long half-life and is resistant to antiretroviral therapy (ART), resulting in lifelong infection and viral rebound in the vast majority of HIV-1-infected individuals when ART is discontinued [10, 11]. Major research efforts are currently underway to understand the biology of the viral reservoir, the mechanism of viral latency, and the potential of various therapeutic approaches to target the reservoir.

The HIV reservoir is maintained during ART by (1) the long half-life of infected memory T cells, (2) homeostatic proliferation of these cells[12, 13] and perhaps (3) low levels of cell-to-cell virus transfer (“cryptic replication”).[14] In the absence of therapy, the frequency of activated T cells is associated with the level of viremia. During long-term suppressive ART, a similar albeit less consistent association exists, with the frequency of activated CD4+ T cells expressing HLA-DR, CCR5, and PD-1 correlated with the frequency of cells containing HIV-1 RNA or DNA.[15, 16] The mechanism for this association is not known. Theoretically, an inflammatory immune environment may contribute to the persistence of the viral reservoir by a number of mechanisms.[17] T cell activation promotes cell-to-cell virus transfer as activated cells are both more likely to produce virus and more likely to become infected. TCR engagement by cognate antigen or cytokines (e.g., IL-7) stimulates CD4+ T cell proliferation and the expansion of the infected cell population.[13] Indeeed, HIV may commandeer the pathways by which cell proliferation is controlled, leading to preferential expansion of infected cells.[18, 19] A chronic inflammatory environment would also be expected to prevent the generation of optimal HIV-1-

specific immune responses. Chronic inflammation stimulates potent and sustained immunoregulatory responses, including expansion of T regulatory cells and the up-regulation of PD-1 and other negative regulators on effector cells.[12, 20, 21]

There is intense interest in "shock and kill" strategies in which intracellular pathways that regulate DNA transcription are manipulated (the "shock"), leading to the production of virus and hopefully the death of the infected cell (the "kill"). The most effective way to stimulate HIV production from the latent reservoir is to activate CD4+ T cells. Theoretically, any activating intervention could lead to proliferation and expansion of the infected cell population. Combination approaches toward a cure might require addition of immune-modifying agents that prevent non-specific T cell proliferation and activation.

mTOR inhibition and the reservoir during ART: T cell activation and proliferation are controlled by a number of signalling pathways, including those involving the mammalian target of rapamycin (mTOR), signal transducer and activator of transcription 5a (STAT5a) and forkhead box O3a (FOXO3a).[22] Specific inhibitors of these pathways might therefore reduce the size of the latent viral reservoir. For example, sirolimus (rapamycin) is a naturally occurring macrolide that inhibits mTOR, and as a consequence it blocks cell cycle progression from G1 to S phase in activated T cells and reduces CCR5 expression.[23] Dendritic cells exposed to mTOR inhibitors *in vitro* failed to make interferon-alpha when exposed to deactivated HIV.[24] mTOR inhibitors also block the negative effect HIV has on autophagy, which might result in a less favorable environment for HIV replication.[25] Finally, mTOR inhibitors enhance CD8+ T cell memory formation and improve vaccine responsiveness in murine and non-human primate models, suggesting it might in humans enhance effector response to HIV and other pathogens.[26]

In order to define the potential role of immunosuppressive drugs on reservoir during effective antiretroviral therapy, we explored the impact of immunosuppressant therapy on peripheral blood measures of HIV persistence following kidney transplantation. A higher baseline HIV DNA ($p<.0001$) was significantly associated with higher HIV DNA levels post-transplant, while higher CD4+ T cell count ($p=0.001$), sirolimus use ($p=0.04$) and a longer duration of follow-up ($p=0.06$) were associated with lower post-transplant HIV DNA levels.[4] Although lacking a control group in this retrospective analysis, the apparent decrease in HIV DNA levels over time post-transplant suggest that mTOR inhibitors may impact HIV persistence during antiretroviral therapy.

Like sirolimus, everolimus inhibits mTOR, although it inhibits both mTORC1 and mTORC2 (sirolimus only inhibits mTORC1). Everolimus is being using with increasing frequency in immunosuppressive regimens in transplant recipients as obtaining therapeutic drug levels is facilitated by more favorable pharmacokinetics as compared to sirolimus. In this prospective study, we will initiate everolimus in a transplant population and perform a comprehensive assessment of the CD4 T-cell reservoir. Multiple measurements of the reservoir will be measured, including: 1) quantitative measurements of CD4+ T cell-associated HIV-1 DNA (total, integrated and 2-LTR circle DNA); 2) HIV-1 total cellular RNA quantification providing a snapshot of HIV-1 "transcriptional activity" of the target infected cells; and 3) A cellular HIV RNA induction assay, the Tat/Rev Induced Limiting Dilution Assay (TILDA) to measure

changes in transcriptionally latent, but inducible HIV. Blood plasma RNA will be monitored using a commercial real-time PCR assay modified for single copy detection.[27]

We hypothesize that the addition of an mTOR inhibitor (everolimus) to the immunosuppressive regimens of HIV infected transplant recipients will decrease HIV persistence in CD4+ lymphoctyes.

2.1 Risk / Benefit Assessment

The risk of minimizing tacrolimus (dose reduction 50%) and adding everolimus is principally related to the risk of rejection and potential nephrotoxicity related to drug-drug interactions. In three large trials [1-3] in HIV negative patients, the use of concentration-controlled everolimus allowed substantial minimization of TAC exposure to achieve better renal function without compromising efficacy or safety in de novo renal transplants. Similarly, the use of another TOR inhibitor (sirolimus) in 91 HIV positive kidney transplant recipients was actually associated with an increase in eGFR over time and a reduction in HIV DNA levels. [4]

To mitigate against the potential adverse impact of drug-drug interactions between the anti-retroviral agents (most notably the protease inhibitors) and immunosuppressive agents (most notably the calcineurin inhibitors and TOR inhibitors), trough levels will be monitored weekly during the first month of the study. Dose adjustments will be made based on previous pharmacokinetic studies in the HIV infected kidney transplant recipients. [28-30]

3 STUDY OBJECTIVES

3.1 Primary Objective

To determine the effect of everolimus on HIV DNA and RNA in CD4+ T cells in HIV infected patients on stable antiretroviral regimens.

3.2 Secondary Objectives

1. To determine the effect of everolimus on plasma HIV-1 RNA when added to a stable antiretroviral regimen.
2. To determine the safety and tolerability of everolimus at standard immunosuppressive doses in HIV-infected transplant recipients who are on stable antiretroviral drugs and immunosuppressive regimens.

4 STUDY DESIGN

4.1 Study Overview

We will perform an open-label, single arm, exploratory study in which antiretroviral-treated post-solid organ transplant adults will add everolimus to their immunosuppressive regimen. We will enroll antiretroviral treated HIV-infected adults who are doing well post-liver, post-kidney or post kidney/pancreas transplant at UCSF who are eligible and willing to add everolimus to their immunosuppressive regimen (with a target trough level between 3-8 ng/ml). Calcineurin inhibitors will be decreased to obtain a 50% reduction in trough levels with the addition of everolimus. Subjects will be maintained on that regimen for 6 months. Novartis will provide study drug.

Biologic specimens for intensive immunology and virology studies will be obtained before, during and after exposure to everolimus. Samples will be analyzed at screening, baseline (prior to addition of everolimus), and at weeks 8 and 26 (while on everolimus), and week 52 (6 months post everolimus discontinuation).

5 CRITERIA FOR EVALUATION

5.1 Primary Efficacy Endpoint

The primary virologic endpoint of the study will be change from baseline in frequency of CD4+ T cell-associated HIV-1 DNA copy number.

5.2 Secondary Efficacy Endpoints

Secondary endpoints will include cell-associated total HIV RNA, plasma RNA, and changes in inducible msRNA, (TILDA) at treatment completion

5.3 Safety Evaluations

- BUN, Creatinine levels will be monitored weekly for one month and then monthly to detect rejection and/or nephrotoxicity. AST , ALT and ALP will also be monitored weekly in liver transplant recipients.
- Tacrolimus and everolimus drug levels (12 hour trough levels) will be monitored weekly for one month, then monthly

6 SUBJECT SELECTION

6.1 Study Population

We will study solid organ (kidney, kidney/pancreas, or liver) transplant recipient who are doing well on ART and who meet the criteria for everolimus therapy. We will exclude individuals on mTOR inhibitors or related drugs. We will exclude individuals who intend to modify ART or who have significant kidney, liver and/or thyroid dysfunction.

6.2 Inclusion Criteria

1. Solid organ (kidney, kidney/pancreas, or liver) transplant recipient who is at least 6 months post transplant.
2. Male or female ≥ 18 years of age.
3. Documentation of HIV-1 infection diagnosis as evidenced by any licensed ELISA and confirmation by Western Blot, or documented history of detectable HIV-1 RNA)
4. HIV-1 plasma RNA <50 copies/ml for at least 2 years with at least one measurement per year and most recent viral load within 16 weeks of enrollment and study drug initiation.

Episodes of a single HIV plasma RNA 50 - 500 copies/ml will not exclude participation if the subsequent HIV plasma RNA was <50 copies/ml.

5. CD4+ T cell count greater than 200 cell/µl within 16 weeks of enrollment and study drug initiation.
6. Receiving combination antiretroviral therapy (at least 3 agents)
7. Written informed consent obtained from subject or subject's legal representative and ability for subject to comply with the requirements of the study.

6.3 Exclusion Criteria

1. Pregnant, breastfeeding, or unwilling to practice birth control during participation in the study.
2. Presence of a condition or abnormality that in the opinion of the Investigator would compromise the safety of the patient or the quality of the data.
3. Patients who are intending to modify antiretroviral therapy in the next 6 months for any reason.
4. Serious illness requiring hospitalization or parenteral antibiotics within preceding 3 months.
5. A screening hemoglobin below 11.5 g/dL.
6. A screening TSH consistent with hypothyroidism.
7. Significant renal disease (eGFR < 45 ml/min for kidney or kidney/pancreas transplant recipients or eGFR < 60 ml/min for liver transplant recipients) or acute nephritis
8. Clinically active hepatitis as evidenced by clinical jaundice or Grade 2 or higher liver function test abnormalities.
9. Hepatic cirrhosis or decompensated chronic liver disease.
10. Concurrent treatment with immunomodulatory drugs, such an interferon-alpha, or exposure to any immunomodulatory drug in past 16 weeks (outside of standard immunosuppression).

7 CONCURRENT MEDICATIONS

All subjects should be maintained on the same medications throughout the entire study period, as medically feasible, with no introduction of new chronic therapies.

7.1 Allowed Medications and Treatments

Standard therapy for HIV-1 infection and for post renal or liver transplant management are allowed except for treatments noted in the exclusion criteria described above and as noted in the prohibited medications section below.

Prohibited Medications and Treatments

The following medications are prohibited during the study and administration will be considered a protocol violation.

- Immunomodulatory drugs such as interferon-alpha (outside of standard immunosuppression)

8 STUDY TREATMENTS

8.1 Formulation of Test and Control Products

Study drug: Zortress (everolimus) manufactured by Novartis.

8.1.1 Formulation of Test Product

Formulation of Zortress (everolimus)

	Zortress (everolimus)
Active Ingredient	everolimus
Other ingredient	butylated hydroxytoluene, magnesium stearate, lactose monohydrate, hypromellose, crospovidone and lactose anhydrous

8.1.2 Formulation of Control Product

No control product.

8.1.3 Packaging and Labeling

Zortress (everolimus) will be packaged and labeled by Novartis. As mentioned previously the study will not be blinded and no placebo is being used.

Study drug is supplied in cartons containing 32 single use tablets (0.25 mg per tablet). The tablets will be packaged in sets of 4 enclosed within a laminated foil pouch. Eight pouches will be contained in each carton (1 extra pouch containing 4 tablets in the event of breakage).

Each carton (kit) of study drug will be labeled with the required FDA warning statement, the protocol number, a treatment number, the name of the sponsors, and directions for patient use and storage.

8.2 Supply of Study Drug at the Site

Novartis will ship Study Drug to the UCSF investigational pharmacy. The initial study drug shipment will be shipped after study activation (i.e., all required regulatory documentation has been completed and the contract is signed). Subsequent study drug shipments will be made after site request for resupply.

8.2.1 Dosage/Dosage Regimen

Everolimus will be added to subjects immunosuppressive regimen at .5 mg – 1.5 mg oral twice a day. Dose will depend on a target trough level between 3-8 ng/ml). Calcineurin inhibitors will be decreased to obtain a 50% reduction in trough levels with the addition of everolimus. Subjects will be maintained on that regimen for 6 months.

The study drug will be dispensed to the subject by the investigator, and will be taken at home twice daily.

8.3 Supply of Study Drug at the Site

Study drug will be supplied to the UCSF investigational pharmacy by Novartis at study initiation. Study drugs must be received by a designated person at UCSF, handled and stored safely and properly, and kept in a secured location to which only the investigator and designated assistants have access. Upon receipt, all study drugs should be stored according to the instructions specified on the drug labels. Clinical supplies are to be dispensed only in accordance with the protocol.

8.4 Study Drug Accountability

An accurate and current accounting of the dispensing and return of study drug for each subject will be maintained on an ongoing basis by a member of the study site staff. The number of study drug dispensed and returned by the subject will be recorded on the Investigational Drug Accountability Record. At the conclusion of the study, and as appropriated during the course of the study, the investigator will return all unused study drug, packaging, drug labels, and a copy of the completed Drug Accountability Record to the Novartis address provided in the investigator folder.

8.5 Measures of Treatment Compliance

Subjects will be asked to keep a patient diary noting the day and date they take their study drug (or noting variances to the required daily dose) and any adverse events. They will be asked to bring their patient diary to each study visit along with all used and unused study drug containers.

9 STUDY PROCEDURES AND GUIDELINES

A Schedule of Events representing the required testing procedures to be performed for the duration of the study is diagrammed in Appendix 1.

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.

9.1 Clinical Assessments

9.1.1 Immunosuppression and ARV Medication Logs

All ARV and Immunosuppression medications from Visit 1 – 5 will be recorded in the source documentation for each study subject. Any changes in drug, dose or frequency through Visit 5will be documented.

9.1.2 Concomitant Medications

All concomitant medication and concurrent therapies will be documented at Visits 1 – 5. However, only document concomitant medications that subject is on at the study visit. Dose, route, and unit frequency will be captured.

9.1.3 Subject Diary

All subjects will be asked to keep a written Subject Diary to record any deviations in prescribed medications (i.e. missed doses, etc). The subject will also record any adverse events. The Subject

Diary will be initiated at Visit 2, and will be reviewed by someone on the study team at Visits 2 – 5.

9.1.4 Medical History and Demographics

Demographic information (date of birth, gender, race) as well as relevant medical history, including history of current disease, other pertinent respiratory history, and information regarding underlying diseases will be recorded at Visit 1.

9.1.5 Physical Examination

A complete physical examination will be performed by either the investigator or a sub-investigator who is a physician at Visit 1. Qualified staff (MD, NP, RN, and PA) may complete an abbreviated physical exam at visits 11.

9.1.6 Adverse Events

Information regarding occurrence of adverse events will be documented throughout the study. Duration (start and stop dates), severity/grade, outcome, treatment and relation to study drug will be recorded on the case report form (CRF).

9.2 Clinical Laboratory Measurements

9.2.1 Pregnancy Test

A urine pregnancy test will be obtained from female subjects who are of childbearing age prior to their participation in the study, prior to initiation of study drug on Visit 2.

9.2.2 Safety Labs

Blood will be obtained for the following safety labs at the specified Visits 1 – 12 as outlined in the schedule of events. Kidney/pancreas transplant recipients are followed the same as kidney only transplant recipients:

- Complete blood count (hemoglobin, hematocrit, red blood cell count, white blood cell count, white blood cell differential, and platelet count)
- Creatinine
- BUN
- Sodium
- Potassium
- Glucose
- Calcium (kidney subjects)
- CO2 (kidney subjects)
- Magnesium (kidney subjects)
- Phosphorous (kidney subjects)
- T Bili (liver subjects)
- SGOT/AST (liver subjects)
- SGOT/ALT (liver subjects)
- Alkaline phosphatase (liver subjects)
- Tacrolimus and everolimus trough levels

9.2.3 HIV RNA (standard) and CD4+/CD8+ T cell counts

HIV RNA (standard) and CD4+/CD8+ T cell counts will be recorded at Visits 1 – 5. However, since insurance only covers these 4 times a year, the research team will contact the subject's primary provider to obtain standard of care labs, which can be used for the closest target visit date.

9.3 Research Laboratory Measurements for Mechanistic Studies

All Research Laboratory Measurements outlined in this section will require 10 x 10cc EDTA tubes of blood from each designated study visit.

9.3.1 HIV-1 RNA: Single copy assay

Measured at Baseline, Screening and at weeks 8, 26, and 52

9.3.2 Cell associated HIV RNA and DNA in CD4+ T-cells (blood)

Measured at Baseline, Screening and at weeks 8, 26, and 52. There is no well-accepted gold-standard for measuring the size of the replication-competent reservoir. Given the proposed mechanism of action for everolimus and our preliminary data with sirolimus we will use a quantitative measurement of HIV DNA in isolated CD4+ T cells as our primary outcome measure. We will also quantify HIV RNA within these cells. Peripheral blood CD4 T cells will be enriched by negative selection using antibody-coupled magnetic beads (Stem Cell Technologies) prior to simultaneous RNA and DNA isolation using cell-sparing protocols (AllPrep, Qiagen). Total DNA, integrated proviral and unintegrated 2-LTR circle species will be measured using calibrated quantitation standards following published qPCR procedures that have been validated for single copy detection.[31, 32] While 2-LTR circles are often undetectable in long-term ART-suppressed subjects, a transient increase has been seen with drug intensification studies[3] and warrants further investigation with immunosuppressive agents. Also, the levels of total (integrated plus unintegrated) and integrated DNA are typically equivalent in suppressed patients. However independent measurements of each species are expected to fully capture potential dynamics in each component of the DNA reservoir. Finally, cell-associated RNA measurements, coupled with DNA measurements (RNA:DNA ratio) will be used to assess the transcriptional activity of the HIV reservoir.[33]

9.3.3 TILDA

Measured at Screening and week 26. Dr. Nicholas Chomont and his group at the Vaccine & Gene Therapy Institute of Florida have developed a novel assay that measures the frequency of cells harboring inducible HIV, serving as a cost effective proxy for expensive and time consuming *in vitro* viral outgrowth assays requiring large amounts of patient blood. By combining ultrasensitive detection of multiply-spliced (ms) nascent tat/rev RNAs after CD4 T cell stimulation and limiting dilution, this method measures the frequency of cells harboring transcriptionally silent – but nonetheless inducible - viruses. While unspliced HIV RNAs are frequently detected in latently infected cells in the absence of viral production, HIV msRNAs reflect *de novo* viral production.[32, 34] To our knowledge, TILDA is the only method that measures the frequency of latently infected cells harboring inducible virus that can be performed with less than a million $CD4^+$ T cells. Dr. Chomont has kindly shared the procedures pre-publication and will advise on the technical and analytical aspects of the assay.

10 EVALUATIONS BY VISIT

10.1 Visit 1 (Screening)

1. Review the study with the subject (subject's legal representative) and obtain written informed consent and HIPAA authorization.
2. Assign the subject a unique screening number.
3. Record demographic data, medical history, including a history of HIV and kidney or liver transplant.
4. Perform a complete physical examination.
5. Perform and record vital signs.
6. Record ARV, Immunosuppression, and concomitant medication regimens.
7. Collect blood for safety labs.
8. Collect historical HIV RNA and CD4+/CD8+ T cell counts from subject or subject's primary care provider
9. Collect blood for Mechanistic Studies
10. Schedule subject for Visit 2 if eligible.

10.2 Visit 2 (Baseline/Day 0)

1. Record ARV, Immunosuppression, and concomitant medication regimens.
2. Perform Pregnancy Test (urine)
3. Initiated Subject Diary
4. Collect blood for safety labs.
5. Collect historical HIV RNA and CD4+/CD8+ T cell counts from subject or subject's primary care provider
6. Collect blood for Mechanistic Studies
7. Initiate study drug
8. Schedule subject for Visit 3

NOTE: Visit 1 and 2 can occur on the same day if subject has been pre-screened.

10.3 Visit 3 (Week 1)

1. Record ARV, Immunosuppression, and concomitant medication regimens.
2. Record any Adverse Experiences and/or Review subject diary for adverse experiences and dosing compliance.
3. Collect blood for safety labs.
4. Schedule subject for Visit 4

10.4 Visit 4 (Week 2)

1. Record ARV, Immunosuppression, and concomitant medication regimens.
2. Record any Adverse Experiences and/or Review subject diary for adverse experiences and dosing compliance.
3. Collect blood for safety labs.
4. Schedule subject for Visit 5

10.5 Visit 5 (Week 3)

1. Record ARV, Immunosuppression, and concomitant medication regimens.
2. Record any Adverse Experiences and/or Review subject diary for adverse experiences and dosing compliance.
3. Collect blood for safety labs.
4. Schedule subject for Visit 6

10.6 Visit 6 (Week 4)

1. Record ARV, Immunosuppression, and concomitant medication regimens
2. Record any Adverse Experiences and/or Review subject diary for adverse experiences and dosing compliance.
3. Collect blood for safety labs.
4. Schedule subject for Visit 7

10.7 Visit 7 (Week 8 ±7 days)

5. Record ARV, Immunosuppression, and concomitant medication regimens
6. Record any Adverse Experiences and/or Review subject diary for adverse experiences and dosing compliance.
7. Collect blood for safety labs.
8. Collect historical HIV RNA and CD4+/CD8+ T cell counts from subject or subject's primary care provider
9. Collect blood for Mechanistic Studies
10. Schedule subject for Visit 8

10.8 Visit 8 (Week 12)

1. Record ARV, Immunosuppression, and concomitant medication regimens
2. Record any Adverse Experiences and/or Review subject diary for adverse experiences and dosing compliance.
3. Collect blood for safety labs.
4. Schedule subject for Visit 9

10.9 Visit 9 (Week 16)

1. Record ARV, Immunosuppression, and concomitant medication regimens
2. Record any Adverse Experiences and/or Review subject diary for adverse experiences and dosing compliance.
3. Collect blood for safety labs.
4. Schedule subject for Visit 10

10.10 Visit 10 (Week 20)

1. Record ARV, Immunosuppression, and concomitant medication regimens
2. Record any Adverse Experiences and/or Review subject diary for adverse experiences and dosing compliance.
3. Collect blood for safety labs.
4. Schedule subject for Visit 11

10.11 Visit 11 (Week 26 ±7 days)

1. Stop study drug
2. Record ARV, Immunosuppression, and concomitant medication regimens
3. Record any Adverse Experiences and/or Review subject diary for adverse experiences and dosing compliance.
4. Perform abbreviated physical examination.
5. Collect blood for safety labs.
6. Collect historical HIV RNA and CD4+/CD8+ T cell counts from subject or subject's primary care provider
7. Collect blood for Mechanistic Studies
8. Schedule subject for Visit 12

10.12 Visit 12 (Week 52 ±7 days)

1. Record ARV, Immunosuppression, and concomitant medication regimens
2. Record any Adverse Experiences and/or Review subject diary for adverse experiences and dosing compliance.
3. Collect blood for safety labs.
4. Collect historical HIV RNA and CD4+/CD8+ T cell counts from subject or subject's primary care provider
5. Collect blood for Mechanistic Studies

10.13 Early Withdrawal Visit

1. Record ARV, Immunosuppression, and concomitant medication regimens

2. Record any Adverse Experiences and/or Review subject diary for adverse experiences and dosing compliance.
3. Collect blood for safety labs.
4. Collect blood for Mechanistic Studies

11 ADVERSE EXPERIENCE REPORTING AND DOCUMENTATION

11.1 Adverse Events

An adverse event (AE) is any untoward medical occurrence in a clinical investigation of a patient administered a pharmaceutical product and that does not necessarily have a causal relationship with the treatment. An AE is therefore any unfavorable and unintended sign (including an abnormal laboratory finding), symptom or disease temporally associated with the administration of an investigational product, whether or not related to that investigational product. An unexpected AE is one of a type not identified in nature, severity, or frequency in the current Investigator's Brochure or of greater severity or frequency than expected based on the information in the Investigator's Brochure.

The Investigator 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 patient 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.

AE Severity

The National Cancer Institute's Common Terminology Criteria for Adverse Events (CTCAE) Version 3.0 should be used to assess and grade AE severity, including laboratory abnormalities judged to be clinically significant. The modified criteria can be found in the study manual. If the experience is not covered in the modified criteria, the guidelines shown in Table 1 below should be used to grade severity. It should be pointed out that the term "severe" is a measure of intensity and that a severe AE is not necessarily serious.

Table 1. AE Severity Grading

Severity (Toxicity Grade)	Description
Mild (1)	Transient or mild discomfort; no limitation in activity; no medical intervention or therapy required. The subject may be aware of the sign or symptom but tolerates it reasonably well.
Moderate (2)	Mild to moderate limitation in activity, no or minimal medical intervention/therapy required.
Severe (3)	Marked limitation in activity, medical intervention/therapy required, hospitalizations possible.
Life-threatening (4)	The subject is at risk of death due to the adverse experience as it occurred. This does not refer to an experience that hypothetically might have caused death if it were more severe.

AE Relationship to Study Drug

The relationship of an AE to the study drug should be assessed using the following the guidelines in Table 2.

Table 2. AE Relationship to Study Drug

Relationship to Drug	Comment
Definitely	Previously known toxicity of agent; or an event that follows a reasonable temporal sequence from administration of the drug; that follows a known or expected response pattern to the suspected drug; that is confirmed by stopping or reducing the dosage of the drug; and that is not explained by any other reasonable hypothesis.
Probably	An event that follows a reasonable temporal sequence from administration of the drug; that follows a known or expected response pattern to the suspected drug; that is confirmed by stopping or reducing the dosage of the drug; and that is unlikely to be explained by the known characteristics of the subject's clinical state or by other interventions.
Possibly	An event that follows a reasonable temporal sequence from administration of the drug; that follows a known or expected response pattern to that suspected drug; but that could readily have been produced by a number of other factors.
Unrelated	An event that can be determined with certainty to have no relationship to the study drug.

11.2 Serious Adverse Experiences (SAE)

An SAE is defined as any AE occurring at any dose that results in any of the following outcomes:

- death
- a life-threatening adverse experience
- inpatient hospitalization or prolongation of existing hospitalization
- a persistent or significant disability/incapacity
- a congenital anomaly/birth defect

Other important medical events may also be considered an SAE when, based on appropriate medical judgment, they jeopardize the subject or require intervention to prevent one of the outcomes listed.

11.2.1 Serious Adverse Experience Reporting

Investigator or Study Coordinadtor will document all SAEs that occur (whether or not related to study drug) per UCSF CHR Guidelines. The collection period for all SAEs will begin after informed consent is obtained and end after procedures for the final study visit have been completed.

In accordance with the standard operating procedures and policies of the local Institutional Review Board (IRB)/Independent Ethics Committee (IEC), the site investigator will report SAEs to the IRB/IEC.

12 DISCONTINUATION AND REPLACEMENT OF SUBJECTS

12.1 Early Discontinuation of Study Drug

A subject may be discontinued from study treatment at any time if the subject, the investigator, or the Sponsor feels that it is not in the subject's best interest to continue. The following is a list of possible reasons for study treatment discontinuation:

- Subject withdrawal of consent (or assent)
- Subject 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
- Protocol violation requiring discontinuation of study treatment
- Lost to follow-up
- Sponsor request for early termination of study
- Positive pregnancy test (females)

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 subjects 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.

All subjects 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 Refer to section 10.13 for early termination procedures.

12.3 Withdrawal of Subjects 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 subjects 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, subjects who discontinue study treatment early (i.e., they withdraw prior to Visit 4) should have an early discontinuation visit. Refer to Section

10.13 for early termination procedures. Subjects who withdraw after Visit 4 but prior to Visit 5 should be encouraged to come in for a final visit (and the procedures to be followed would include those for their next scheduled visit).

12.4 Replacement of Subjects

Subjects who withdraw from the study treatment will be replaced.

Subjects who withdraw from the study will be replaced.

13 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 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. The Principal Investigator will determine if a protocol violation will result in withdrawal of a subject.

When a protocol violation occurs, it will be discussed with the investigator and a Protocol Violation Form detailing the violation will be generated. This form will be signed by a Sponsor representative and the Investigator. A copy of the form will be filed in the site's regulatory binder and in the Sponsor's files.

14 STATISTICAL METHODS AND CONSIDERATIONS

Prior to the analysis of the final study data, 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.

14.1 Data Sets Analyzed

All eligible patients who receive at least one dose of the study drug (the Safety Population) will be included in the safety and efficacy analysis.

14.2 Analysis of Primary Endpoint

The primary virologic endpoint of the study will be change from baseline in frequency of CD4+ T cell-associated HIV-1 DNA copy number. Based on a design with ten patients and a null hypotheses that there is no change in the HIV DNA over time, we estimate that the study will have approximately 80% power to detect a 0.5 log (3.2 fold) difference in HIV DNA levels between baseline and 6-month post-treatment time point (at the 0.05 significance level using a paired t-test).

14.3 Analysis of Secondary Endpoints

Secondary virologic endpoints will include cell-associated total HIV RNA, plasma RNA, and changes in inducible msRNA, (TILDA) at treatment completion We estimate the study has 80% power to detect 0.7 log (5.0 fold) and 0.4 log (2.5 fold) differences between pre-treatment and post-treatment in cell-associated RNA and inducible msRNA (TILDA), respectively. These estimates are based on the assumption that HIV DNA, cell-associated RNA and inducible msRNA are log normally distributed and that the standard deviation of the difference in pre- and post-dose levels is 0.5 log10, 0.7 log10, and 0.4 log10, respectively (derived from previous work). We expect to see trends in decreasing cell-associated DNA and RNA by week 26 of everolimus treatment based on our retrospective analysis. The durability of this potential effect will be evaluated 6 months after everolimus discontinuation.

15 DATA COLLECTION, RETENTION AND MONITORING

15.1 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.

Study personnel will enter data from source documents corresponding to a subject's visit into the protocol-specific paper CRF when the information corresponding to that visit is available. Subjects will not be identified by name in the study database or on any study documents to be collected by the Sponsor (or designee), but will be identified by a subject number.

For paper CRFs: If a correction is made on a CRF, the study staff member will line through the incorrect data, write in the correct data and initial and date the change.

The Investigator is responsible for all information collected on subjects 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.

15.2 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.

15.3 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. The study database will be updated in accordance with the resolved queries. All changes to the study database will be documented.

15.4 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.

15.5 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 and Assent 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 (patient files, signed informed consent forms, copies of CRFs, Study File Notebook, 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.

15.6 Monitoring

Monitoring visits will be conducted by representatives of the Sponsor 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 (or designee), and appropriate regulatory authorities to conduct on-site monitoring and/or auditing of all appropriate study documentation.

15.7 Subject Confidentiality

In order to maintain subject confidentiality, only a subject number will identify all study subjects on CRFs and other documentation submitted to the Sponsor. Additional subject confidentiality issues (if applicable) are covered in the Clinical Study Agreement.

16 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 laboratory specimens, evaluation forms, reports and other records will be identified by a coded number and initials only. All study records will be kept in a locked file cabinet and code sheets linking a patient's name to a patient 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. 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).

16.1 Protocol Amendments

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

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

16.3 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 Subjects (21 CFR 50.25[a,b], CFR 50.27, and CFR Part 56, Subpart A), the Health Insurance Portability and Accountability Act (HIPAA, if applicable), and local regulations.

The Investigator will prepare the informed consent form, assent 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 Harmonisation 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 subjects must be given ample opportunity to inquire about details of the study. If appropriate and required by the local IRB/IEC, assent from the subject will also be obtained. 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.

16.4 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.

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

APPENDIX 1. SCHEDULE OF STUDY VISITS

		ON STUDY DRUG										
	VISIT 1 Screening[1]	VISIT 2 BASELINE[1]	VISIT 3 WEEK 1	VISIT 4 WEEK 2	VISIT 5 WEEK 3	VISIT 6 WEEK 4	VISIT 7 WEEK 8	VISIT 8 WEEK 12	VISIT 9 WEEK 16	VISIT 10 WEEK 20	VISIT 11 WEEK 26	VISIT 12 WEEK 52
Informed Consent	X											
Record Medical History/Demographics	X											
Complete Physical Exam	X											
Abbreviated Physical Exam											X	
Pregnancy Test (Urine)		X										
Record CD4+/CD8+ T cell counts (as standard of care from primary provider)	X	X					X				X	X
Record HIV RNA: Standard assay (as standard of care from primary provider)	X	X					X				X	X
Start Study Drug		X										
Initiate Subject Diary and Review (medication deviations and adverse events)		X	X	X	X	X	X	X	X	X	X	X
Medication Review/Recording	X	X	X	X	X	X	X	X	X	X	X	X
Evaluate for Adverse Events			X	X	X	X	X	X	X	X	X	X
Stop Study Drug											X	
Safety Labs[2]												
Record Complete Blood Count (Kidney, Liver)	X	X				X	X	X	X	X	X	X
Record Creatinine (Kidney, Liver)	X	X	X	X	X	X	X	X	X	X	X	X
Record BUN (Kidney, Liver)	X	X	X	X	X	X	X	X	X	X	X	X
Record Sodium (Kidney, Liver)	X	X				X	X	X	X	X	X	X
Record Potassium (Kidney, Liver)	X	X				X	X	X	X	X	X	X
Record Glucose (Kidney, Liver)	X	X				X	X	X	X	X	X	X
Record Calcium (Kidney)	X	X				X	X	X	X	X	X	X

		On Study Drug										
	Visit 1 Screening[1]	Visit 2 Baseline[1]	Visit 3 Week 1	Visit 4 Week 2	Visit 5 Week 3	Visit 6 Week 4	Visit 7 Week 8	Visit 8 Week 12	Visit 9 Week 16	Visit 10 Week 20	Visit 11 Week 26	Visit 12 Week 52
Record CO2 (Kidney)	**X**	**X**				**X**	**X**	**X**	**X**	**X**	**X**	**X**
Record Magnesium (Kidney)	**X**	**X**				**X**	**X**	**X**	**X**	**X**	**X**	**X**
Record Phosphorous (Kidney)	**X**	**X**				**X**	**X**	**X**	**X**	**X**	**X**	**X**
Record Total Bilirubin (Liver)	**X**	**X**				**X**	**X**	**X**	**X**	**X**	**X**	**X**
Record SGOT/AST (Liver)	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**
Record SGOT/ALT (Liver)	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**
Record Alkaline Phosphatase (ALP) (Liver)	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**
Tacrolimus and Everolimus 12 hour trough levels (Kidney, Liver)	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**
MECHANISTIC STUDIES												
HIV RNA: Single copy assay	**X**	**X**					**X**				**X**	**X**
Cell associated HIV RNA and DNA in CD4+ T-cells (blood)	**X**	**X**					**X**				**X**	**X**
TILDA		**X**									**X**	

[1] Visit 1 and Visit 2 can occur on the same day if the subject has been prescreened. In these cases, all events indicated for visit 1 and for visit 2 should be performed, but only once.

[2]All safety labs are recorded from standard of care results for post kidney or liver transplant recipients except for the week 1, 2 and 3 safety labs post study drug initiation which will be performed for this protocol

[3]Everolimus levels will be performed for this study at all indicated study visits. Tacrolimus levels will be recorded from standard of care results, except for the weeks 1, 2 and 3 levels which will be performed for this protocol.

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