

1.0 SCIENTIFIC ABSTRACT

The primary target of HIV infection is the CD4⁺ T helper lymphocyte (T_h). Infection with HIV leads to cytopathic destruction of cells and the spread of virus to other CD4⁺ cells, thereby progressively depleting the host of CD4⁺ T cells. The extent of CD4⁺ T cell loss correlates with the impairment in host immunity, development of opportunistic infections, and survival. Improvements in CD4⁺ T cell counts are often observed after the initiation of anti-retroviral drug therapy; however, the anti-viral effects of these agents are limited both by the evolution of drug resistant strains of HIV and adverse reactions in treated individuals. Advances in our understanding of molecular events in the HIV life cycle have suggested an alternative approach to pharmacological agents for improving cellular resistance to viral infection. This approach has been termed intracellular immunization (IC-imm). Several IC-imm strategies have been developed that target obligate steps in the HIV replication cycle and most have demonstrated anti-viral activity *in vitro*. These genes can be inserted into antigen-specific CD4⁺ T cells to protect these cells against loss and enhance immunity in HIV-infected individuals.

The study proposed in this application will evaluate the safety of transferring CD4⁺ T cell clones to individuals infected with HIV and compare the *in vivo* survival and function of genetically modified CD4⁺ T cells. This protocol will be a single center, phase 1, open-label, non-randomized study of 10 HIV-seropositive individuals to determine the ability of IC-imm genes to protect CD4⁺ T cell clones. Two IC-imm genes, (1) a poly-TAR decoy, and (2) an RRE-polyTAR decoy, will be evaluated and compared to a control vector. All 10 individuals will receive a minimum of two infusions of autologous CMV-specific T_h clones, each comprising an equal number of T_h subclones transduced with each of the two IC-imm genes and control vector. The initial infusion, designated as the "test dose," will deliver 1 x 10⁸ T cells/m² of each subclone (total dose of 3x10⁹ T cells/m² to permit an assessment of toxicity at a low cell dose. Two weeks later, a second infusion will deliver 1 x 10⁹ T cells/m² of each subclone (total dose of 3x10⁹ T cells/m²). Recipients will be evaluated for acute toxicity associated with the infusion and significant progression of HIV disease. If toxicity develops, no further doses will be given. Recipients will be monitored for 30 weeks after infusion for treatment related toxicity and persistence of transferred CD4⁺ T cell clones. If, after the second infusion, the numbers of recoverable transferred T cells are too low to statistically compare between cells containing each of the IC-imm genes and control vector, a third infusion will be administered (four weeks after the second dose) with a cell dose of 1 x 10¹⁰ T cells/m² of each subclone (total dose of 3x 10¹⁰ T cells/m²). Peripheral blood will be obtained at specific times during the study to assay both for the presence and proportion of T_h cells containing IC-imm genes or a control vector, and for functional T_h cells based on proliferative responses and cytokine production to antigen stimulation. The results of this study will provide insight into the feasibility of protecting CD4⁺ T cells from HIV infection *in vivo* with IC-imm genes, and thereby permit the reconstitution of host cellular immunity. This study will serve to provide a foundation for future studies that will focus on augmenting HIV specific CD4⁺ T cell responses, and reconstituting hematopoietic cells by the transfer of stem cells containing IC-imm genes.