

BUDGET JUSTIFICATION PAGE MODULAR RESEARCH GRANT APPLICATION				
Initial Budget Period	Second Year of Support	Third Year of Support	Fourth Year of Support	Fifth Year of Support
\$ 200,000	\$ 175,000	\$ 175,000	\$ 175,000	\$ 175,000
Total Direct Costs Requested for Entire Project Period				\$ 900,000

Personnel

John Smith, M.D., Ph.D., Principal Investigator (30% effort) will be responsible for the overall administration and direction of the project. He will analyze reaction of soluble IV9-HLA-A*0201 complex with TCR on 68A62 anti-IV9CTL.

Helen Thomas, M.D., (25% effort) will focus on investigating the ability of various SL9-specific CD8 CTL clones from various HIV infected individuals whose T-cell receptors bind with different strength to the cognate pepMHC complex (SL9-HLA-A*0201) to suppress viral replication in HIV infected cells *in vitro*. In addition, Dr. Thomas will maintain initial preparation of the CTL clones and will characterize them on a regular basis to ensure maintenance of their initial quality.

Thomas Club, Ph.D., PostDoctoral Fellow (100% effort) will be involved in all the measurement of SD50 and SD25 for RT-and *gag*-derived peptides required to induce various responses of anti-HIV CTL. Most of her effort will be direct towards measurement of the intervals of epitope densities on target cells required for various responses of anti-HIV CTL.

Jane Jones, Ph.D., PostDoctoral Fellow (100%) will be responsible for the isolation and characterization of recombinant MHC class I molecules using *Drosophila Melanogaster* and *E. coli* expression systems and measurement of kinetics and affinity of reactions between soluble complex of immunodominant peptide SL9 with HLA-A-*0201 soluble protein and TCR on various clones of live anti-SL9 CTL. Dr. Jones will also measure level of α,β -TCR and CD8 molecules on anti-SL9 CTL.

Ms. Stephanie Wilson, Technician, (50% effort) is responsible for laboratory animal preparation and some of the biochemical analyses.

To be Appointed Technician, (25% effort) is responsible for the repair and maintenance of the equipment and will run a variety of assays.

Ms. Whitney Johnson, Graduate Student, (50% effort) in Dr. Smith's laboratory will participate in all aspects of the proposed experiments.

Equipment (*This additional narrative budget justification is provided because there is a variation in the number of modules requested.*)

Purchase of a Thermocycler (\$10,000) and HPLC Fraction Collector (\$15,000) is requested during the first year. This will increase the requested budget for the first year by one module (\$25,000). The requested equipment is necessary for this project and will be used extensively to analyze IV9-HLA-A*0201 complex.

Principal Investigator/Program Director (Last, first, middle): Smith, John W.

Consortium

Approximately \$25,000 Total Costs per year

Consortium with the University of Virginia {X} Domestic { } Foreign

James Livingston, Ph.D., will devote 10%. Dr. Livingston will be responsible for establishing CTL clones from HIV-infected subjects, and propagating previously isolated CTL clones. Dr. Livingston will also be responsible for planning and overseeing all functional cell assays.

Norman Cross, 50% Research Technologist will perform all CTL assays, proliferation assays, ELISA assays and assays designed to measure inhibition of HIV-1 replication by virus-specific CTL clones.

Fee (SBIR/STTR Only)