Identification of HIV-1 Infected But Seronegative Infants, Children, and Youth

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Abstract
The report of a perinatally infected infant who was thought to have achieved a “functional cure” of HIV infection prompted the investigation as to whether this outcome can be duplicated. Several recent reports indicate that in HIV infected adults treated early in the course of acute infection with potent antiretroviral treatment (ART) given for a substantial period of time, the size of the HIV reservoirs can be reduced. Moreover, with durable virologic control, seroreversion can occur. In HIV infected persons, it is believed that low level virus replication is a continuous source of stimulation of B cells for producing HIV Ab. However, with potent ART, the suppression of virus to undetectable levels results in lack of antigenic stimulation and seroreversion. We have a cohort of HIV infected children, adolescents and youth, and have recently noted an increase in HIV pregnant women presenting for the first time at term without having taken ART during pregnancy which reduces perinatal transmission of the virus. Based on this premise, we postulate that infants, children and youth who are infected with HIV may have seronegativity due to failure of generation of a full antibody response due to early initiation of ART, from achievement of a sustained virologic suppression. The purpose of this proposal is to lay the basis for further investigating these phenomena. It is our interest to determine if infants, children, and youth who are infected with HIV-1 may have negative serologic tests for the virus after being in virologic control and if there is a pattern of the present Western blot bands that correlate with disease stage or viral suppression.

Background
The report of a seemingly cured perinatally HIV infected infant (“the Mississippi baby”) prompted the question as to whether this outcome can be duplicated. This child was able to remain off ART for two years and maintain the virus quiescent for that length of time. We are in a unique position to investigate new approaches for early intervention to prevent establishment of HIV reservoirs because we have noticed a large number of women who fail to access care and present to the clinic at term, without having received ART during pregnancy. In such infants who are identified as being infected, and treated early, serologic testing for HIV antibody after the period of decay of maternal Ab will be a critical diagnostic test to ascertain the possibility of functional cure, like in the Mississippi baby.

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In this pilot project we propose to 1. determine HIV serostatus of ART treated patients with perinatal HIV infection, 2. determine HIV reservoirs among ART treated patients with perinatal HIV infection, and 3. collect peripheral blood mononuclear cells (PBMC)/plasma samples for a repository for future virologic and immunologic assays based on developments in the field of HIV reservoir, persistence, and cure.

Study population: Target population: Patients from pediatric ID clinic at UM. Patients with perinatally acquired HIV infection from birth to 24 years of age qualify if they have virologic suppression with a viral load <1000 copies/mL for at least one year prior to enrollment. Charts will be reviewed to obtain demographics, when were medications started, and duration of viral load suppression.

Serologic response: All patients consenting will have HIV ELISA and Western blot (WB) determination. The WB band profiles will be analyzed and correlated with ELISA.

Analysis of HIV reservoirs: PBMC were cryopreserved in the CFAR repository for future immunologic assays, including immune activation by flow cytometry.

Study ID Age Race/Ethnicity Gender ART at Enrollment (NEEDS ATRT SPECIFICATION) VL (copies/mL) Age at entry (months) Weight (kg) Last weight (kg) Height (cm) Last height (cm) Age last weight (cm) Last height (cm) Age last weight (cm) Last height (cm) Age last weight (cm) Last height (cm)

We enrolled 52 patients, all of them from minority ethnic groups, all but three of them had viral loads over 1000, but less than 3000 copies/mL, on the day of enrollment. Samples were submitted for serologic testing with HIV ELISA and Western blot determination. Results are awaited.

Evaluation for HIV reservoirs will be performed in PBMC, purifying total DNA, extrachromosomal DNA, chromosomal DNA and low molecular weight DNA from PBMC. We will be using primers and fluorogenic probes to quantitate total HIV-1 clade B genomes, integrated proviruses, 2 LTR episomal DNA, human CCR5 and mitochondrial DNA to measure target molecules by a nested PCR using a LTR primer for first round amplification. The single-copy human CCR5 gene is quantified to measure the number of cell equivalents in DNA samples for standardization.

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Results

Characteristics of the participants

We acknowledge the CFAR pilot award and support from the CFAR laboratory sciences core. The Miami Center for AIDS Research (CFAR) at the University of Miami Miller School of Medicine funded by a grant (P30AI073961) from the National Institutes of Health (NIH), which is supported by the following NIH Co-Funding Participating Institutes and Centers: NIAID, NCI, NICHHD, NINDS, NIMH, NIA, NIDDK, NIGMS, FIC AND OAR