ANTIMYCOBACTERIAL IMMUNE RESPONSES IN HIV-INFECTED CHILDREN STARTING ANTIRETROVIRAL THERAPY IN LUSAKA, ZAMBIA

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LIST OF ACRONYMS

AIDS……..Acquired immunodeficiency syndrome

ART ……..Antiretroviral therapy

CIDRZ…….. Centre for Infectious Disease Research in Zambia

HIV……..Human immunodeficiency virus

IFNγ ……..Interferon gamma

PBMC s ……Peripheral blood mononuclear cells.

PPD……..Purified protein derivative

TB ……Tuberculosis

UTH …….. University Teaching Hospital

DEDICATION

I dedicate this thesis to my late father Manasseh Nkamba and my late mother Stella Muchinda Nkamba who from an early age instilled in me the importance of education. I also dedicate it to my dear wife and friend, Sithabile Lubinda Nkamba with all my love and gratitude for showing great patience, support and understanding, my children, Caleb Nkamba and Shalom Nkamba who give me great joy and motivation, to my aunt, Justina Muchinda, may Almighty God bless you richly for being our mother.

DECLARATION

I, Hope C. Nkamba, do hereby declare that the work presented in this dissertation for the Masters in Public Health has not been presented either wholly or in part for a degree or Diploma in any other University and currently not being presented for any other degree.

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APPROVAL

This dissertation of Hope C. Nkamba is approved in partial fulfillment of the requirements for the award of the degree of Master of Public Health (MPH) by the University of Zambia.

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CERTIFICATE OF COMPLETION OF DISSERTATION

I…… here by certify that this dissertation is the product of my work in submitting it for my Master of Public Health degree program, further attest that it has not been submitted in part or whole to another university.

I/We…….having supervised and read this dissertation am/are satisfied that this is the original work of the author under whose name it is being presented. I/We confirm that this work is completely satisfactory and ready for presentation to the examiners.

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DEFINITION OF TERMS AND KEY CONCEPTS

In order to shade more light on the introduction and literature review, the following terms and concepts are defined:

T Cells

T cells or T lymphocytes belong to a group of white blood cells known as lymphocytes. These cells play a central role in cell mediated immunity. There are a number of different T cell subsets, each with a distinct function. T helper cells also known as CD4 T cells assist or help other white blood cells in mounting an immune response once the body is invaded by pathogens (antigens). Among other functions, CD4 T cells are crucial in the maturation of B cells into plasma cells and activation of cytotoxic T cells and macrophages. The other type of T cells is called cytotoxic T cells or CD8 T cells, which destroy infected cells and tumor cells.

Naïve T cells

Naïve T cells are considered to be mature T cells except, unlike activated T cells or memory T cells, they have not encountered antigen. Having adequate numbers of naïve T cells is essential for the immune system to continuously respond to new pathogens.

Activated T cells

These are T cells that have encountered antigen. These are identified by surface markers CD38 and HLA-DR.

Effector T cells

These are T cells that are produced in response to a given antigen. These can be derived from activated T cells and memory T cells. Many effector T cells are short lived.

Memory T cells

These are T cells that have previously encountered and responded to antigens. These T cells can recognize foreign invaders such as viruses, bacteria and cancer cells. Memory T cells have become “experienced” by having encountered antigen during prior infection, encounter with cancer or previous vaccination. At a second encounter with the invader, memory T cells can

mount a faster and stronger immune response than the first time the immune system responded to the invader. Memory T cells can be grouped into effector memory and central memory T cells.

Effector memory T cells

These can rapidly mature into effector T cells and secrete large amounts of cytokines early after restimulation. They seem to be specialized for quickly entering inflamed tissues.

Central memory T cells

These cells take longer to differentiate into effector cells and thus do not secrete as much cytokines as effector memory T cells do early after restimulation. They however recirculate more easily to the T cell zones of peripheral lymphoid tissues.

PPD

PPD is an extract of *Mycobacterium tuberculosis*, the bacteria that causes tuberculosis in humans. This extract is similar to a protein expressed by *Mycobacterium tuberculosis* that the immune system responds to and recognizes upon a second encounter.

Interferon gamma (IFN-γ)

IFN-γ is a cytokine that is produced by T cells when antigen-specific immunity develops and is important in the immune system because it is involved in fighting viral and intracellular bacterial infections and in tumor control. It has the ability to inhibit viral replication directly and is involved in stimulating and modulating the immune system.

ABSTRACT

Background: Children infected with HIV are at risk of developing TB, this may be attributed to the fact that HIV infected children are immune compromised. ART has been linked to improved immune responses to TB. With the scaling up of ART in Zambia, more children will have access to treatment. The objectives of this study were to determine the magnitude and quality of immune reconstitution in HIV-infected children receiving antiretroviral therapy (ART) and to determine pathogen-specific immune reconstitution to *Mycobacterium tuberculosis*.

Methods: A total of 59 children of age 9 months to 5 years initiating ART with a history of BCG vaccination from Matero Reference Clinic in Lusaka were enrolled in a prospective cohort study. Demographic and clinical data were collected using questionnaires. Blood samples were drawn before starting ART, at 3 months and 6 months for measurement of T cell subsets and PPD stimulation for intracellular cytokine staining.

Results: After 6 months of ART, the median CD4 T cell percentage increased from 9.4% at baseline to 25.9% ($p< 0.001$). Total CD8 T cell percentage decreased from 42.8% pre-ART to 36.5% after 6 months of ART ($p = 0.010$). However, naïve CD8 T cells increased within the same period ($p = 0.038$). Both activated CD4 and CD8 T cells decreased after 6 months of ART ($p < 0.001$). On the other hand, both central memory CD4 and CD8 T cells increased after 6 months of ART ($p = 0.029$ and 0.021, respectively), while effector memory CD8 T cells decreased ($p = 0.006$). After 3 months of ART, CD4 T cells expressing IFN-γ decreased ($p = 0.033$) but after 6 months of ART the percentage increased to pre-ART levels.

Conclusion: ART has a positive impact on HIV-infected children, likely reducing the risk of tuberculosis as evidenced by the increases in CD4 T cells critical to an effective immune response against TB. Before starting ART, anti-mycobacterial immune responses seem to be primarily driven by effector memory T cells while after ART by central memory T cells. Therefore, central memory T cells appear to be the primary cells in restoring specific immune responses. These findings have valuable implications for TB vaccine development strategies in HIV-infected children.