THE LANCET

Supplementary appendix

This appendix formed part of the original submission and has been peer reviewed. We post it as supplied by the authors.

Supplement to: Cotton MF, Violari A, Otwombe K, et al, on behalf of the CHER Study Team. Early time-limited antiretroviral therapy versus deferred therapy in South African infants infected with HIV: results from the children with HIV early antiretroviral (CHER) randomised trial. *Lancet* 2013; published online Aug 22. http://dx.doi.org/10.1016/S0140-6736(13)61409-9.

Other exclusion criteria were:

- Parent or legal guardian unable or unwilling to attend regularly scheduled study visits
- Disallowed medications
    - All investigational drugs
    - All prohibited medications listed in protocol section 5.6

        Package inserts of antiretroviral and concomitant agents should be referred to whenever a concomitant medication is initiated or the dose is changed to avoid drug interaction adverse events. Please refer to the following website for the most current list of drug interactions: http://www.hivinsite.com/InSite.jsp?page=ar-00-02. The following medications are disallowed:

        - Disallowed with NNRTIs
            - Antihistamines (with EFV)
                - Astemizole
                - Terfenadine
            - Anti-infectives
                - Ketoconazole
            - GI Motility
                - Cisapride
            - Sedative/hypnotic
                - Midazolam (can give a single dose only if adequate monitoring)
                - Triazolam
            - Ergot alkaloids and derivatives
            - Calcium Channel Blockers
                - Bepridil
            - Neuroleptic
                - Pimozide
        - Disallowed with PIs
            - Antiarrhythmics
                - Amiodarone
                - Flecainide
                - Propafenone
                - Quinidine
            - Anticonvulsants
                - Phenytoin
            - Antihistamines
                - Astemizole
                - Terfenadine
            - Anti-infectives

        - Systemic Itraconazole
    - GI motility
        - Cisapride
    - HMG-CoA Reductase Inhibitors
        - Lovastatin
        - Simvastatin
    - Psychiatric medication
        - Pimozide
    - Sedative/hypnotic
        - Midazolam (Oral midazolam is contraindicated. If parenteral midazolam is co-administered in subjects on Lopinavir/ritonavir, close monitoring for respiratory depression and/or prolonged sedation should be exercised and dosage adjustment should be considered.)
        - Triazolam
    - Ergot alkaloids and derivatives
- Inability to tolerate oral medication

CIPRA ZA 002
DAIDS-ES ID 10404

A Phase III, Randomised, Open-label Trial to Evaluate Strategies for Providing Antiretroviral Therapy to Infants Shortly after Primary Infection in a Resource Poor Setting

A Limited Center Trial of the Comprehensive International Program of Research on AIDS - South Africa

Children with HIV Early Antiretroviral Therapy (CHER)

Pharmaceutical Support provided by:

GlaxoSmithKline
The South African Department of Health

Sponsored by:

The National Institute of Allergy and Infectious Diseases
Division of AIDS (DAIDS)

IND Number: IND 71,494

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TABLE OF CONTENTS

ABBREVIATIONS AND ACRONYMS

3TC	lamivudine or Epivir®
ABC	abacavir or Ziagen®
AE	adverse event
AIDS	Acquired Immunodeficiency Syndrome
ALT	alanine transaminase
ARV	antiretroviral
ART	antiretroviral therapy
AST	aspartate transaminase
AUC	area under the curve
BSA	body surface area
AZT	zidovudine or Retrovir®
CDC	(United States) Centers for Disease Control and Prevention
CHBH	Chris Hani Baragwanath Hospital
CHER	Children with HIV Early Antiretroviral Therapy
CIPRA	Comprehensive International Program of Research on AIDS

CRF	case report form
d4T	stavudine or Zerit®
DAIDS	(United States) Division of AIDS
ddI	didanosine or Videx®
DNA	deoxyribonucleic acid
DOB	date of birth
DSMB	Data Safety Monitoring Board
EAE	expedited adverse event
EFV	efavirenz or Stocrin®
EKG	**Electrocardiogram**
EPI	Expanded Program for Immunization
FDA	(United States) Food and Drug Administration
FTT	Failure To Thrive
GCP	Good Clinical Practices
GI	Gastrointestinal
HAART	Highly active antiretroviral therapy
HDL	High density lipoproteins
HLA	Human Leukocyte Antigen
HibCV	Haemophilus influenzae type b conjugate vaccine
HIV	Human Immunodeficiency Virus
hpf	high power field
HPPMCS	HIV Pediatric Prognostic Markers Collaborative Study Group
HSR	hypersensitivity reaction
ICH	International Conference on Harmonization
IEC	Independent Ethics Committee
IL	interleukin
INH	isoniazid
IRB	Institutional Review Board
IRIS	Immune Reconstitution Inflammatory Syndrome
KID-CRU	Children's Infectious Diseases Clinical Research Unit
LDL	Low Density Lipoproteins
LFT	liver function tests
LIP	Lymphocytic interstitial pneumonia
LPV/r	lopinavir/ritonavir or Kaletra®
LTX	Long term treatment
MCC	(South Africa) Medicines Control Council
MOP	Manual of Procedures
MTCT	mother-to-child-transmission
NFV	nelfinavir or Viracept®
NIAID	(United States) National Institute of Allergy and Infectious Diseases
NIH	(United States) National Institutes of Health
NNRTI	non-nucleoside reverse transcriptase inhibitor
NRTI	nucleoside reverse transcriptase inhibitor
NSAI	non-steroidal anti-inflammatory
NVP	nevirapine or Viramune®
OI	Opportunistic Infection
PACTG	Pediatric AIDS Clinical Trials Group
PCP	*Pneumocystis jiroveci* pneumonia

PCR	polymerase chain reaction
PENTA	Pediatric European Network for Treatment of AIDS
PETRA	Perinatal Transmission Trials
PHRU	Perinatal HIV Research Unit
PncCV	polysaccharide-protein conjugate vaccine
PI	protease inhibitor
PK	pharmacokinetics
PMTCT	prevention of mother-to-child-transmission
PTX	Primary treatment
PY	person years
PZA	pyrazinamide
QC	Quality Control
QCA	Quality Control Associate
RSC	(DAIDS) Regulatory **Support** Center
RNA	ribonucleic acid
RTV	ritonavir or Norvir®
SAE	serious adverse event
SDMC	(PHRU) Statistical and Data Management Center
SIV	Simian Immunodeficiency Virus
SJS	Stevens-Johnson Syndrome
SOP	Standard Operating Procedure
STD	sexually transmitted disease
SUSAR	**Suspected Unexpected Serious Adverse Reaction**
TCH	Tygerberg Children's Hospital
ULN	upper limit of normal
VL	viral load
VTP	vertical transmission prophylaxis
WHO	World Health Organization

TRIAL SUMMARY

"A Phase III, Randomised, Open-label Trial to Evaluate Strategies for Providing Antiretroviral Therapy to Infants Shortly after Primary Infection in a Resource Poor Setting"

A Limited Center Trial of the Comprehensive International Program of Research on AIDS-South Africa

PRIMARY OBJECTIVE:

1. To compare time to failure of first line ART (due to clinical, **virological** or immunological disease progression, or regimen-limiting ART toxicities) or death among three randomised arms (infants who receive early ART in Arms 2 and 3 and infants in whom ART is deferred until clinical or immunological disease progression in Arm 1).

SECONDARY OBJECTIVES:

1. To compare the cumulative rate of clinical disease progression or death in the 3 randomised arms at 182 weeks (3.5 years).

2. To compare the 3 randomised arms in terms of the development of Grade 3 or 4 (laboratory or clinical) adverse events.

3. To compare the 3 randomised arms in terms of time from randomisation to starting or needing to start continuous therapy.

4. To compare hospital admission rates, time to first hospital admission, and duration of hospitalization (for non-elective procedures) in the 3 randomised arms.

5. To compare the cumulative rate of development of viral resistance at the time of failure of first line therapy in the 3 arms. In addition, the development of new mutations to NVP during the trial will be compared to baseline in children exposed and not exposed to NVP in the perinatal period.

6. To compare the 3 randomised arms in terms of time to death alone or death plus life threatening stage C events or HIV events associated with permanent end-organ damage. Life-threatening Stage C events will be defined as a malignancy (lymphoma or Kaposi sarcoma), progressive multifocal leukoencephalopathy or intercurrent infection requiring admission to an intensive care unit. HIV events associated with permanent end-organ damage will be assessed at the end of the trial and will include HIV encephalopathy, chronic lung disease, cardiomyopathy and nephropathy where the parameters used for making the initial diagnosis have not reverted to normal.

DESIGN

The CHER trial is a two-center, Phase III, randomised, open-label trial comparing three treatment strategies in infants with perinatally acquired HIV-1 infection diagnosed between 6 and 12 weeks of age. Study participants with a baseline CD4 percentage ≥25% and HIV-1 infection diagnosed between 6 and 12 weeks will be randomised in Part A **to** early therapy for 40 weeks (from ≤12 weeks of age until approximately the first birthday (Arm 2)) or early therapy for 96 weeks (from ≤12 weeks of age until approximately the second birthday (Arm 3)). Randomisation to the deferred therapy (Arm 1) is closed. Study participants with a baseline CD4 percentage <25% and HIV-1 infection diagnosed between 6 and 12 weeks will go onto continuous ART. First line ART will be started in Arm 1 or restarted after interruption in Arms 2 and 3 **if the criteria below are met.** Second line therapy may be started for participants when failure of first line therapy occurs, as a result of either clinical, **virological** or immunological disease progression, or regimen-limiting ART toxicities.

1) The CD4% falls below 25% or the CD4 count is less than 1000cells/mm^3 regardless of CD4% for children if <12months of age;

2) The CD4% falls below 20% for children 12-**59** months of age*

3) **The CD4% falls below 20% or the CD4 count <350cells/mm^3 for children ≥ 60 months of age**

4) Occurrence of a severe CDC Stage B or Stage C disease.

* If there is discordance between CD4% and count the investigator may use an absolute CD4 count to reintroduce ART: 12-35 months CD4% <20% or CD4 count <750 cells/ mm^3; **36-59 months** CD4% <20% or CD4 count <500 cells/ mm^3

POPULATION AND SAMPLE SIZE

Approximately 4**51** HIV-1 infected infants will be enrolled at the Perinatal HIV Research Unit (PHRU), Chris Hani Baragwanath Hospital, Johannesburg, or at the Children's Infectious Diseases Clinical Research Unit (KID-CRU), Tygerberg Children's Hospital, Cape Town, South Africa. All eligible participants will be enrolled in Part A (n=4**11**) or Part B (n≈40) depending on CD4% at screening. Sample size calculations are based on the number of infants required in Part A. Part A deferred Arm 1 and Part B are closed to accrual.

- Part A (CD4 percentage ≥25%): 125 participants already enrolled in arm 1 and 14**3** in Arm 2 and 14**3** in Arm 3

- Part B (CD4 percentage <25%): 40 participants on continuous therapy

PRIMARY ENDPOINT

1. Time to death or failure of first line therapy as a result of either clinical, **virological** or immunological disease progression, or regimen-limiting ART toxicities.

SECONDARY

1. Occurrence of severe CDC Stage B or Stage C disease or death (cumulative after 3.5 years).

2. Occurrence of Grade 3 or 4 (clinical or laboratory) adverse events.

3. Time from randomisation to starting or needing to start continuous therapy.

4. Hospital admissions, time to first hospital admission, and duration of hospitalization.

5. Cumulative viral resistance mutations at the time of failure of first line therapy.

6. Time to death alone or death plus life threatening stage C events or HIV events associated with permanent end-organ damage.

TREATMENT

First line: zidovudine (AZT) + lamivudine (3TC) + lopinavir/ritonavir (LPV/r)

Second line: didanosine (ddI) + abacavir (ABC) + nevirapine (NVP) (if <3 years or <10 kg) or efavirenz (EFV) (if >3 years and >10 kg)

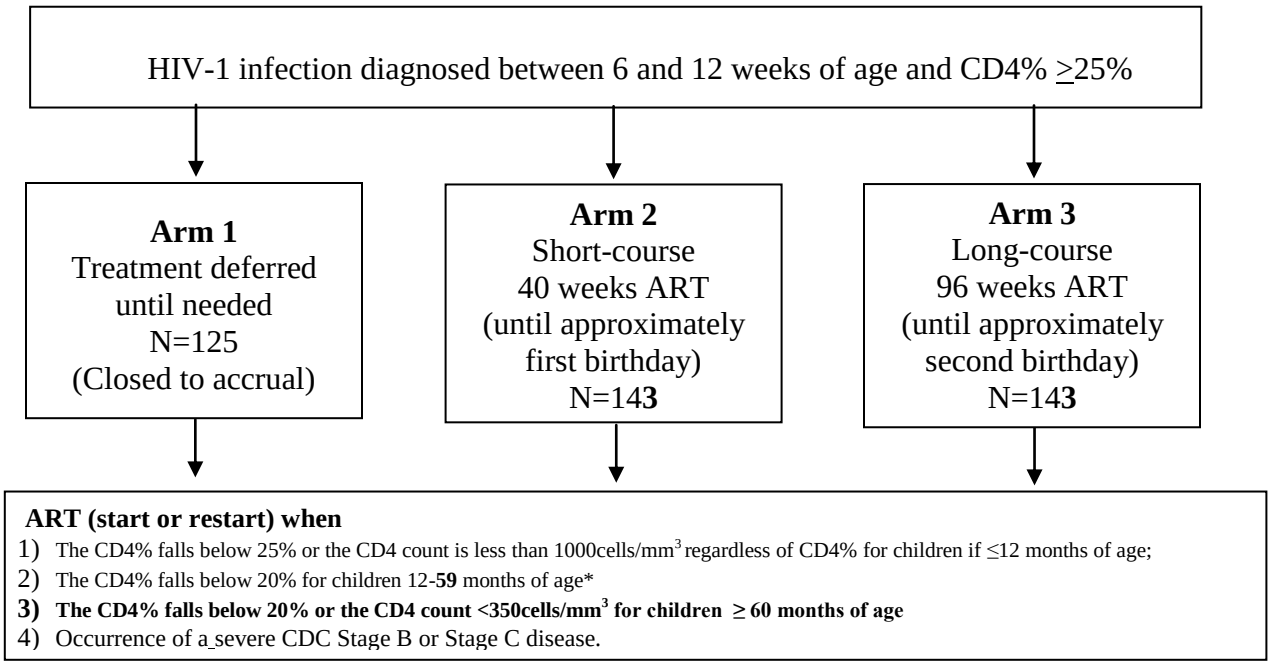
The dosage for each treatment drug is provided in Section 5.1 and Appendix VIII.

FOLLOW-UP

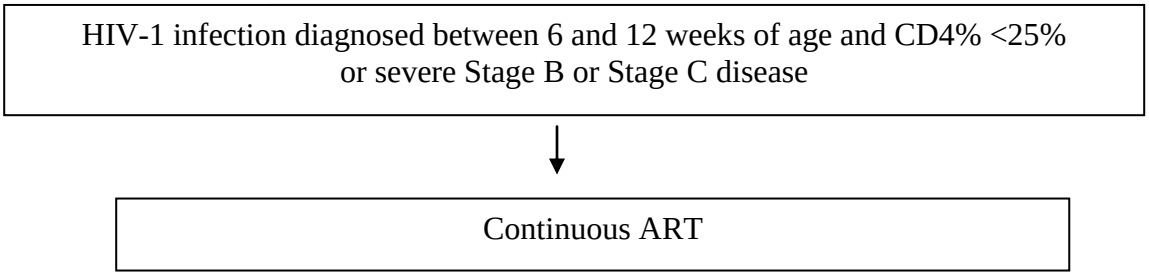
Enrolment **took** place over 78 weeks (1.5 years), **but was reopened to increase the sample size and power due to a change in the criteria for starting therapy in line with the WHO guideline at the time. The** minimal follow-up will be for at least 182 weeks (3.5 years). Therefore, the first infant enrolled will be followed for **up to 330** weeks (**6** years) and the last infant enrolled will be followed for at least 182 weeks (3.5 years). The pre-screening visit **took** place from four weeks of age with screening from five weeks and randomisation to therapy or the deferred therapy arm from six weeks of age. Randomisation and initiation of ART **was** permitted until 12 weeks of age.

Figure 1. Schematic representation of first line therapy

Part A: N=411



Part B: N≈40



* If there is discordance between CD4 % and count the investigator may use an absolute CD4 count to reintroduce ART: 12-35 months CD4% <20% or CD4 count <750 cells/ mm^3; **36-59 months** CD4% <20% or CD4 count <500 cells/ mm^3

1.0 INTRODUCTION

1.1 Background

1.1.1 Epidemiology and Impact of Vertical Transmission Prophylaxis

There are 1.1 million births in South Africa each year (Actuarial Society of South Africa, 2004). A 24.6% seroprevalence rate in pregnant women translates into approximately 250,000 infants exposed to HIV-1 each year (Department of Health, 2000). The vertical transmission rate in South Africa, in the absence of vertical transmission prophylaxis (VTP), is between 25% and 40%, and would result in a minimum of 62,500 HIV-1 infected infants born every year (Bobat, 1996). VTP using nevirapine (NVP) is associated with a 13% transmission rate in the absence of breastfeeding and would be expected to result in a minimum of 32,500 HIV-1 infected infants per year (Guay, 1999; Moodley, 2003). If a dual therapy regimen were introduced, such as combining zidovudine (AZT) and NVP, it is expected that the transmission rate would be on the order of 7%, resulting in

approximately 17,500 HIV-1 infected infants per year (Dabis, 2002; Lallemant, 2002). Finally, under optimal circumstances, a triple therapy-based vertical prophylaxis regimen for mothers with plasma HIV-1 RNA levels <1,000 copies/mL has a transmission rate of 1% and will result in 2,500 HIV-1 infected infants per year (Cooper, 2002).

Thus, even with the best scenario and uniformly good application of complex VTP regimens, relatively large numbers of HIV-infected infants will be born and will require optimal antiretroviral therapy.

1.1.2 Natural History of HIV Infection in Children

The HIV-1 epidemic has had a profound impact on child mortality. Chris Hani Baragwanath Hospital (CHBH) admits 5,500 children annually. In 1992, about 7% of pediatric deaths for children under 15 months of age were HIV-1 related, rising to over 46.1% in 1996 (Zwi, 2000). HIV-1 related pediatric admissions increased from 30% in 1998 to 38% by 2000, with 48% of children under 15 months of age (Zwi, 1999; Meyers, 2000). At Tygerberg Children's Hospital (TCH), the prevalence of HIV infection is lower than at CHBH, but is still significant. HIV-1 infected children occupied 10% of pediatric beds in a point prevalence survey conducted in 1999 (Roux, 2000) and 17.6% in March 2003 (M. Cotton, personal communication, March 28, 2003). The number of HIV-positive admissions per year increased from 52 in 1992 to 151 in 2002. The overall case fatality rate for all admissions is 5% as opposed to 14% for HIV-1 infected infants and children.

Most early natural history studies conducted prior to the introduction of highly active antiretroviral therapy (HAART) in children were conducted in Europe and the United States (US). For example, an Italian cohort study reported that 20 to 25% of HIV-1 infected children progressed rapidly to AIDS or died during infancy, with slower disease progression thereafter (Tovo, 1992). In this study, the mean age of onset of HIV-1 related signs or symptoms was 5.2 months. These signs appeared significantly earlier in the 102 children who died of HIV-1-related illness than in those who were still alive (median three versus six months); the probability of remaining asymptomatic at one year of age was 19% and at five years was 6.1%. The mean survival of those not progressing rapidly in the first year of life, in the absence of ART, is nine to 10 years. In another prospective cohort study, 23% of infants had developed AIDS-defining conditions by one year and 40% by four years (Newell, 1994).

In a Cape Town study of children attending outpatient clinics, median survival of untreated infants presenting with symptomatic disease less than one year of age was between 32 and 36 months (Hussey, 1998). By that time, pneumocystis jiroveci pneumonia (PCP) prophylaxis was firmly established and contributed to improved survival. In a retrospective analysis of HIV-1 infected children hospitalized between 1991 and 1996 at TCH, 36 of 91 (40%) children died (Cotton, 1998).

1.1.3 Immune System and HIV Replication in Primary Infection

Perinatally acquired HIV-1 infection should be regarded as primary infection, as it occurs most commonly during birth and can therefore be detected shortly after infection has occurred. Studies of perinatal transmission intervention show an *in utero* infection rate of five to seven percent (Moodley, 2003; PETRA Study Team, 2002). *In utero* infection is defined as having a positive virological test for HIV-1 at or before 48 hours of age (Bryson, 1992). Infant rhesus macaques, infected after mucosal exposure to simian immunodeficiency virus (SIV), have detectable virus in their blood after four to seven days (Staprans, 1999). For this reason, much of the *in utero* infection may have occurred within the last two to three weeks of pregnancy and should still be regarded as primary infection. It may respond to early antiretroviral therapy in the same way as with infection during the birth process. Longitudinal studies have confirmed that *in utero* and perinatal acquisition of HIV-1 have similar outcomes (Shearer, 1997).

Neonatal T-cells express a limited repertoire of lymphokines in response to activation. Interferon-gamma and interleukin-4 (IL-4) production are reduced more than tenfold compared to adults. These cytokines participate in the maturation of cytotoxic cells, activation of macrophages and the maturation of B cell function (Wilson, 1992). In children, the immune system continues to mature during the first four to six years of life (Adderson, 2001; Holt, 1995). Examples of immunological immaturity include an inability to produce T-cell dependent antibodies to polysaccharide capsular antigens from Streptococcus pneumoniae and Haemophilus influenzae type B, in the absence of protein conjugation.

Cytotoxic T-lymphocytes specific for HIV-1 infection are detected in adults within weeks of HIV infection (Cao, 2003). Although this response is not well documented in vertically infected children, the majority can demonstrate HIV specific responses by two years of age (Wilfert, 1994).

Age at time of infection appears to be an extremely important determinant of outcome of HIV-1 infection. Infection at a time of inability to generate virus-specific immune responses may contribute to diminished control of viral replication. For example, nef-deleted SIV, while not virulent in adult macaques, is highly pathogenic in neonatal macaques (Baba, 1995). Factors permitting high replication in infants include a large pool of CD4+ T-cells, a large thymus, delayed development of cytotoxic lymphocyte activity and antibody dependent cytotoxicity (Luzuriaga, 2000a).

In adults and infants, primary infection is associated with high levels of replication. The best evidence of the severe consequences of primary HIV-1 infection in neonates is that the time taken for viral replication to decrease to a set point after the initial high levels seen at primary infection is far longer than in adults.

In adults, after the initial viremia, there is a steady decline to reach an individualized set point by four months (Kaufmann, 1998; Schacker, 1998; Sterling, 2001). At the same time, CD8+ suppressive and cytotoxic lymphocyte activity first appear**s** (Borrow, 1994; Musey, 1997).

In the first longitudinal study of plasma HIV-1 RNA levels in newly infected infants, Shearer and colleagues showed that plasma HIV-1 RNA levels increase rapidly after birth, peak at one to two months of age (median values at one to two months, 318,000 and 256,000 copies/mL, respectively), and then slowly decline to a median of 34,000 copies/mL at 24 months (Shearer, 1997). Continued reduction in plasma HIV-1 RNA has been observed through five to six years of age (Mofenson, 1997). Other studies have shown that plasma HIV-1 RNA levels do not decline below 10^5 copies/mL until the third year of life (McIntosh, 1996; Mofenson, 1997). Although there is considerable overlap in the first 12 months of life, plasma HIV-1 RNA levels >299,000 copies/mL are associated with more rapid progression (Shearer, 1997). Dickover *et al.*, showed that high levels of HIV-1 RNA at birth and during primary viremia are associated with early onset of symptoms and rapid disease progression to AIDS and death (Dickover, 1998). In a prospective study comparing newly infected Kenyan adults and infants, the mean peak HIV-1 RNA was significantly higher in infants than adults (6.06 versus 5.07 $\log_{10}$), as was the mean set point level (5.84 versus 4.6 $\log_{10}$). Also, the viral set point in infants infected later in life, through breastfeeding after two months of age, was far lower than that in infants infected earlier (Richardson, 2003).

1.2 Early Antiretroviral Therapy

In acute HIV-1 infection, the primary aim of treatment is preservation and reconstitution of HIV-specific and general immune function. Starting therapy early during the course of HIV-1 infection may be beneficial, as early control of viral replication following acute infection can lower the viral set point, limit dissemination of the virus and allow normal immune system development (de Rossi, 2002). This may lead to an improved clinical outcome.

Studies in SIV-infected macaques support a role for HAART in primary infection (Altfeld, 2000; Rosenwirth, 2000). Early therapy for a limited time period is associated with diminished viral replication in some macaques. For example, Rosenwirth and colleagues showed prolonged suppression of viral replication in two of six macaques treated with a single agent for eight weeks, beginning between seven and fourteen days after infection (Rosenwirth, 2000). Hel and colleagues demonstrated only transient viremia in macaques treated for six months with a combination of antiretroviral therapy and therapeutic immunization (Hel, 2000).

1.2.1 Antiretroviral Therapy in Adults

Studies of adult treatment soon after seroconversion have shown that early treatment may be beneficial in the immune control of HIV-1 infection. In a small group of adults with primary infection, therapy prior to development of HIV-1 antibodies was associated with greater reversal of CD4+ T-cell abnormalities, including improved numbers of memory CD4+ T-cells and activated CD4+ T-cells, than beginning therapy when antibody was already detectable (Keoshkerian, 2003). Combination antiretroviral therapy during primary HIV-1 infection was shown to decrease the frequency of minor opportunistic infections,

mucocutaneous disorders, and respiratory infections, and reduce disease progression (Berrey, 2001).

Rosenberg and colleagues showed that institution of HAART during acute HIV-1 infection leads to persistent, strong HIV-1-specific T-helper cell responses, analogous to those seen in persons able to control viremia in the absence of antiviral therapy (Rosenberg, 1999). They describe eight participants treated during acute infection followed by treatment interruption. Five were able to remain off therapy for a median of six months (range 5 to 8.7 months) with HIV-1 RNA of <500 HIV-1 RNA copies/mL (Rosenberg, 2000). They hypothesize that interruption may be more successful early in the course of the disease, due to the homogeneity of the virus and the relatively intact immune system (Rosenberg, 2000).

The appropriate duration of therapy prior to the interruption remains unknown, although a minimum of a year seems to be favored (E. Konnick, personal communication, September 2002).

1.2.2 Antiretroviral Therapy in Infants

Luzuriaga *et al.* recently reported the results of Pediatric AIDS Clinical Trials Group (PACTG) study 356, where HIV-1-infected children starting highly active antiretroviral therapy at or below three months of age were compared to those starting at an older age, according to age of presentation (Luzuriaga, 2004). There were 25 in the former and 27 in the latter group. Although there were no differences in virological suppression at weeks 16 and 48, fewer infants in the delayed-therapy group than in the early-therapy group (30 percent vs. 60 percent) had plasma HIV-1 RNA levels less than 400 copies/mL at 200 weeks (P=0.03). These workers had earlier reported on 17 infants in the early treatment group where the ability to respond normally to childhood vaccination and viruses such as Epstein-Barr virus, was retained, suggestive of an intact immune system. However, seroreversion (loss of antibodies to HIV) and absence of HIV-specific cell mediated immunity were also noted, and may be of concern should ARV resistance develop, permitting rapid viral replication (Luzuriaga, 2000b).

1.3 Antiretroviral Regimens in Adults

Antiretroviral drugs will be available for adults through CIPRA-SA Project 1 (CHBH) and through the South African National Treatment Plan (CHBH and TCH). For both programs, the first line regimen will be stavudine (d4T) + lamivudine (3TC) + efavirenz (EFV) or nevirapine (NVP) or lopinavir/ritonavir (LPV/r) (Kaletra®). Women of child-bearing potential will be offered NVP because of the teratogenic potential of EFV (Mofenson, 2002).

1.4 Antiretroviral Regimens in Infants and Children

ARVs suitable for young infants should preferably be in pleasant-tasting suspensions of high density (requiring small volumes), with no food restrictions, and require administration not more than twice daily. The ideal regimen should be well tolerated,

have minimal toxicity, and result in low levels of resistance, but would have a major impact on the subsequent natural history of the disease.

Drug absorption and metabolism differ in infants when compared to adults, and very high doses (particularly of protease inhibitors) are required to achieve adequate drug levels (Faye, 2002; Litalien, 2003). Pharmacokinetic data in infants are unavailable for many antiretroviral drugs and there is the potential for long-term toxicity as survival improves.

Side effects are generally transient, mild and mainly gastrointestinal. More serious side effects, related to long-term cumulative exposure, include lipodystrophy, dyslipidemia, insulin resistance, hyperlactatemia, osteopenia and growth failure (Leonard, 2003). Asymptomatic hyperlactatemia (>2 mmol/L) may occur in up to one-third of asymptomatic children receiving a combination of NRTIs and PIs (Desai, 2003).

Suitable agents include the nucleoside reverse transcriptase inhibitors (NRTI) zidovudine (AZT), lamivudine (3TC) and abacavir (ABC). Other NRTIs have minor disadvantages.

Didanosine (ddI) must be given on an empty stomach and stavudine (d4T) suspension is at a low concentration (1 mg/mL), thus requiring large volumes. Another minor disadvantage is that ddI and d4T require refrigeration once reconstituted and are only stable for one month thereafter.

Of the non-nucleoside reverse transcriptase inhibitors (NNRTIs), NVP has suitable physical characteristics, but its major disadvantage is that it is currently used as monotherapy for VTP in South Africa. In an examination of viral isolates from women and infants who received single doses of NVP in the HIVNET 012 study, resistance mutations, the most common of which was K103N, were detected in 21 of 111 (19%) women. Mutations were detected in a subset of 11 out of 24 (46%) infants infected by six to eight weeks of age, the most common of which was Y181C, which faded from detection by 12 months of age in all seven evaluable infants (Eshleman, 2001). The frequency of mutations in infected infants, when NVP is part of triple therapy in pregnant women, is not known. EFV is currently not permitted in children less than three years of age and weighing less than 10 kg. Of note, while Y181C mutation confers with a high level resistance to NVP, EFV resistance appears to be several times lower (Stanford HIV drug resistance database, 2003), suggesting a possible role for EFV in second line therapy in infants.

Of the protease inhibitors (PI), only ritonavir (RTV) and lopinavir (LPV/r) have liquid formulations. RTV is registered for use in infants from one month of age, but is often associated with intolerance due to its bad taste. Studies have shown poor virological efficacy thus far, but none have been conducted in ARV-naïve infants (Yogev, 2002; Wiznia, 2000; van Rossum, 2002). LPV/r is registered for infants above six months and once opened, the formulation should be refrigerated. Nelfinavir (NFV) dosage has not been adequately defined in young children, although it is used frequently. NFV tablets require crushing, which is sometimes difficult for caregivers.

LPV/r showed excellent durability at 48 weeks in 100 infants and children, with 79% having a HIV-1 RNA level <400 copies/mL (Saez-Llorens, 2003). PACTG 1030, a Phase I/II study of LPV/r in HIV-1 infected infants less than six months of age, established that

LPV 300 mg/m^2/RTV 75 mg/m^2 given orally twice daily, either with formula or a fatty food) gave adequate exposure. (Chadwick, 2008).

In the PENTA 7 study, 20 infants under three months of age received NFV in combination with d4T and ddI (Compagnucci, 2002). Among the first 16 infants, only 50% had a viral load (VL) <400 copies/mL by week 24 and 25% had VL<50 copies/mL (Faye, 2002). In PK evaluations of NFV in infants between two and eight months of age, the PK parameters (AUC and trough levels) were highly variable. With doses of 150 mg/kg/day, levels were inadequate in 27% of infants (Litalien, 2003). NFV powder was poorly tolerated and most were switched to crushed tablets.

1.4.1 Selection of Three Drug Regimen

The proposed first line regimen for this trial (AZT+3TC+LPV/r) is based on the availability of good pediatric formulations for infants, pharmacokinetic data in infancy and good provisional long-term experience with LPV/r. Many favor the inclusion of a PI-based first line regimen because of the very high HIV-1 RNA levels in infants and the relatively large number of mutations required to produce resistance. Triple therapy that included a PI was independently associated with reduced mortality in HIV-1 infected infants and children in the US, where the mortality declined from 5.3% in 1996 to 0.7% in 1999 (Gortmaker, 2001).

The proposed second line regimen is ddI + ABC + NVP (if <3 years of age or <10 kg in body weight) and ddI + ABC + EFV (if >3 years of age and >10 kg in body weight). Abacavir is chosen over d4T because of potential increased toxicity associated with ddI + d4T seen in adults (Blanco, 2003; Robins, 2002). According to the package inserts, ddI and d4T, in combination, are contraindicated in pregnancy because of an increased risk of lactic acidosis.

1.4.2 Antiretroviral Drug Exposure to Breastfeeding Infants

Untreated children who are breastfed will have some exposure to ARVs secreted in breast milk. This could affect the study outcome by suppression of viremia and selection of ARV resistant mutants, especially in infants on the observational arm. Even infants on therapy are at risk, as HIV-1 superinfection has been described (Altfeld, 2002) and could theoretically occur with maternal virus resistant to 3TC or d4T. Recent data shows significant absorption of lamivudine and nevirapine in breast-feeding infants whose mothers are receiving ART (Shapiro, 2005). In contrast, however, breast-feeding confers significant survival advantages to HIV-infected infants (Coovadia, 2007; Thior, 2006). Therefore, breast-fed infants whose mothers are on ART will not be excluded from the study despite the theoretical risk of ARV resistance in the infant. Antiretrovirals have been shown to suppress HIV RNA but not DNA in breast milk (Shapiro, 2005).

Some studies suggest that mortality in breastfeeding mothers may be higher (Nduati, 2001), others show no difference (Coutsoudis, 2001). The rate of breastfeeding in HIV-1 positive mothers in Soweto and the Western Cape/Tygerberg area is very low (5%) and a few of these mothers will be on ART. Mothers eligible for treatment will receive ARV therapy as part of Project

1 or through the South African National Treatment Plan, which was announced in July 2003. Previous studies indicate that only 15 to 20% of mothers have a CD4+ count <200 cells/mm. Therefore, the number of mothers who are both on ARV treatment and breastfeeding will be very small. In Soweto, mothers who choose not to breastfeed their infants are eligible to purchase infant formula at a subsidized price. In the Western Cape/Tygerberg area, the department of health provides free infant formula for the first six months.

1.5 Events After Interruption of ART

There is concern that infants interrupting HAART may develop acute HIV syndrome. This syndrome was first described in adults as a mononucleosis syndrome (Cooper, 1985). The prevalence of acute retroviral syndrome is not known. The onset of symptoms varies between one and three weeks after exposure to the virus, but peaks at three weeks. Signs and symptoms include fever, sweating, malaise, myalgia, anorexia, nausea, diarrhea and non-exudative pharyngitis. More than 60% have a maculopapular rash. More serious manifestations include encephalitis and ascending polyneuropathy (Chaisson, 2000). In one instance, multi-organ failure was noted after treatment interruption (Crespo, 2003).

Structured treatment interruption, proposed as a means for reducing subject exposure to antiretroviral drugs, is complicated by the possibility of acute HIV syndrome following interruption. The frequency of acute HIV syndrome after discontinuation of antiretroviral therapy in two recent studies was 4 to10% (Worthington, 2003).

Thus far, only one study, a matched case-control study, has documented this syndrome in infants. In a careful follow-up of ARV naïve infants HIV-1 infected through breastfeeding in Côte d' Ivoire, at least one sign or symptom suggestive of acute HIV syndrome was noted in 21 of 22 infants (Rouet, 2002). However, 61% of uninfected breastfeeding infants had similar signs and symptoms. A mononucleosis syndrome that included fever and a sore throat occurred in 16 of the 22 (72%) infected infants, but also in 26 of 44 (59%) uninfected infants. Generalized lymphadenopathy (≥1 cm in two or more extra-inguinal sites) was also noted significantly more often in infected infants. Other signs and symptoms included fever alone, diarrhea, dermatitis, ear or eye infections, and oral candidiasis.

In a report of 71 children (median age seven years) experiencing 82 unplanned interruptions, acute HIV syndrome was not noted (Gibb, 2004). Also of note, the median monthly decline in CD4% of 0.52% was independent of age, CD4% at HAART initiation, pre-interruption CD4+ or HIV-1 RNA levels or previous AIDS.

1.6 Plasma HIV-1 RNA and CD4 Percentage as Surrogate Markers for Disease Progression and Antiretroviral Efficacy

Numerous studies have documented the importance of HIV-1 RNA and CD4+ T-cell percentage as surrogate markers for disease progression in children (McIntosh, 1996; Mofenson, 1997; Palumbo, 1998). These findings have been confirmed in an African setting (Taha, 2000). Although CD4+ T-cell percentages and HIV-1 RNA levels are independent markers of disease progression, the combination may be more accurate

(Mofenson, 1997; Palumbo, 1998). HIV-1 RNA and CD4+ T-cell counts have also become the standard for evaluating the efficacy of ART in clinical trials (Palumbo, 1998; van Rossum, 2002) and in clinical practice (Guidelines for the use of antiretroviral agents, 2003). In children, as in adults, HIV-1 RNA levels have been reported to be a marker of disease progression, independent of CD4+ count (Mofenson, 1997; Palumbo, 1998). During the first year of life, however, the positive predictive value of viral load alone may be low (Mofenson, 1997; Shearer, 1997).

CD4+ counts and percentages predict risk of disease progression, response to therapy and the risk for opportunistic infections (OIs). The CD4+ count is highly variable in young children and decreases with age in HIV-uninfected children until five to six years of age. CD4% is less variable and decreases less with age. Significant decreases in CD4% in infected children may occur as early as two months among symptomatic, rapidly progressing children (de Martino, 1991).

The HIV Pediatric Prognostic Markers Collaborative Study (HPPMCS), a report on European and US cohort studies and randomised trials of ART or immune therapies in HIV-1 infected children, recently evaluated the risk of death or AIDS in 3,371 children, representing 7,297 years of follow-up. All children were either antiretroviral naïve or on AZT monotherapy (HPPMCS Group, 2003). The study confirmed a higher risk of progression for an equivalent CD4% in infants less than 2 years of age than at over two years of age. In older children, the risk of death increases sharply when CD4% is below 10% and the risk of AIDS when CD4% is below 15%. At the same CD4%, children less than 2 years of age have a worse prognosis than older children. To illustrate this concept, for a CD4% of 20%, the risk of AIDS within the next 12 months is 31% for an infant aged six months, 21% for a one-year old and 12% for a two-year old. For HIV-1 RNA levels, a similar but less pronounced effect of age is seen. For HIV-1 RNA of 10^6 copies/mL, the risk for AIDS at six months is 24%, at one year is 21% and at two years it is 19%. (See Table 1 below).

Table 1. Estimated risk of AIDS and death within 12 months at selected values of age and CD4% or viral load. 95% confidence interval shown in parenthesis.

(A) CD4%

Age (years)	Endpoint	CD4 percentage			
		10%	20%	30%	40%
0.5	AIDS	51 (45-57)	31 (27-35)	20 (18-23)	16 (14-17)
	Death	30 (26-35)	12 (10-15)	6.4 (5.3-7.8)	4.6 (3.8-5.5)
1	AIDS	40 (45-57)	21 (18-23)	13 (12-14)	9.9 (8.5-11.4)
	Death	20 (18-23)	6.8 (5.6-8.4)	3.3 (2.8-3.9)	2.5 (2.0-3.1)
2	AIDS	29 (26-31)	12 (11-14)	7.2 (6.4-8.2)	5.9 (4.9-7.1)
	Death	12 (11-14)	3.1 (2.6-3.7)	1.5 (1.2-1.9)	1.2 (0.9-1.6)
5	AIDS	15 (12-18)	4.7 (3.9-5.7)	3.1 (2.5-4.0)	2.9 (2.1-3.8)
	Death	4.9 (3.8-5.9)	0.9 (0.7-1.2)	0.5 (0.3-0.7)	0.5 (0.3-0.7)
10	AIDS	7.4 (5.0-10.8)	2.2 (1.6-2.8)	1.8 (1.2-3.0)	1.7 (1.1-3.1)
	Death	2.1 (1.3-3.0)	0.3 (0.2-0.5)	0.2 (0.1-0.4)	0.2 (0.1-0.4)

(B) Viral load

Age (years)	Endpoint	Viral load (copies/ml)		
		10^4	10^5	10^6
0.5	AIDS	11 (7-16)	14 (10-16)	24 (16-27)
	Death	2.7 (0.9-4.1)	4.1 (2.9-5.4)	9.7 (8.1-12.0)
1	AIDS	7.8 (4.4-12.1)	11 (8-12)	21 (12-24)
	Death	1.7 (0.8-2.8)	3.1 (2.4-4.0)	8.8 (7.2-11.0)
2	AIDS	5.3 (3.2-8.5)	8.1 (6.5-9.3)	19 (8-22)
	Death	1.1 (0.6-1.8)	2.5 (1.8-3.1)	8.2 (6.4-10.4)
5	AIDS	3.2 (2.1-4.9)	6.0 (4.5-8.0)	17 (5-21)
	Death	0.7 (0.4-1.0)	2.1 (1.4-2.9)	7.8 (5.9-10.2)
10	AIDS	2.2 (1.4-3.2)	5.1 (3.0-7.7)	16 (3-20)
	Death	0.6 (0.3-0.9)	2.0 (1.2-2.9)	7.7 (5.7-10.0)

(Source: *HPPMCS Group, 2003*)

1.7 Description of the Population

The children for this protocol will be recruited from the Chris Hani Baragwanath Hospital and surrounding clinics in Soweto and Tygerberg Children's Hospital, as well as the Western Cape region.

Active, well established MTCT prevention programs at each of the study sites identify HIV-1 positive pregnant women. The majority of participants will be recruited from these MTCT prevention programs. HIV-1 infected mothers will be informed of the study during pregnancy. At the time of delivery, they will be reminded of the study and will be given an appointment for an HIV-1 DNA PCR or RNA PCR test for their child when the child is 4 to 12 weeks of age. To avoid inequality, participants who are referred from other sources, such as private practitioners or hospital outpatient departments and wards, will also be eligible for screening.

It is expected that two-thirds of the participants will be recruited from the Johannesburg site and one-third from the Cape Town site. At the Soweto site, there are 30,000 deliveries each year, of which 30% of the mothers are HIV-infected. Administration of nevirapine to the women is expected to result in a 13% transmission rate, yielding a minimum of 1,170 HIV-1 infected infants per year in Soweto, Johannesburg.

In January 2005, the PMTCT regimen in the Western Cape was changed to include zidovudine from 34 weeks gestation to the mother and for a week to the neonate, together with a single dose of nevirapine to each (Lallemant, 2004). This regimen has been associated with a 5% transmission rate in selected sites (personal communication – Dr Fareed Abdullah, Western Cape Department of Health – August 22nd, 2005). In Cape Town, there are 25,000 deliveries each year, of which 15.4% of the mothers are HIV-1 infected (National HIV seroprevalence survey, 2004). Using nevirapine and zidovudine, as described above, will result in 193 HIV-1 infected infants at that site. The number of HIV-1 infected infants at both sites will be approximately 1,560 per year.

Assuming: 1) that 50% of these infants will not be eligible or their parents may not be willing to allow them to participate; and 2) that the transmission rate may change due to improved prevention measures, we anticipate that the recruitment period required to enroll approximately 436 infants out of a pool of at least 1,560 HIV-infected infants per year will not exceed 18 months. The medical care team for these infants is the same as the medical care team involved in the study. Thus, access to these participants is not a barrier to accrual into this study.

1.8 Rationale

The current trial is designed to evaluate short course ART strategies in young HIV-1 infected infants. If successful, although initially labor-intensive, the strategy may avoid the long-term complications of continuous therapy at an early age. However, eventually there would be reactivation of the disease and the need to reintroduce ART.

We hypothesize that starting ART from ≥6 to ≤12 weeks of age, followed by interruption of therapy, will be feasible and safe and will show a long-term benefit by delaying immunological and clinical progression of HIV-1 when compared with deferred ART. We have therefore designed a three-arm study to evaluate the risks and benefits of early ART for infants, followed by interruption at one or two years of age. Confining therapy to the period when the immune system is still immature may then permit growth and development without the need for ART for many years.

Recognizing the resource constraints in the African setting, we have chosen to use CD4 percentage throughout the study as the marker of disease progression rather than HIV-1 RNA, because T-cell subset measurement is affordable and CD4 percentage can identify the risk for developing specific OIs, particularly after the first year of life.

Receiving antiretroviral therapy until two rather than one year of age may allow better development of the immune system. However, this strategy increases the chances of resistant virus developing because of longer duration of therapy and possible difficulties in sustaining adequate adherence.

Thus far, no trials have been designed to determine when to initiate therapy in infants. Because of the rapid progression of disease in 20 to 30% of infants in the first year of life, coupled with high levels of viral replication, the current approach in the US is to treat all symptomatic infants with CD4% <25% and to consider treatment for asymptomatic infants with CD4% ≥25% *(*Guidelines for the use of antiretroviral agents, October 2006). By treating aggressively, enormous improvements in survival and reduction in HIV-related morbidity have been seen (de Martino, 2000; Gortmaker, 2001; Abrams, 2003).

Nevertheless, HAART in infants is not easy to sustain and requires continued motivation of the caregiver to maintain adherence. Failure to give the medications correctly results in virological failure and resistance. As children survive for longer periods, resistance will reduce the therapeutic options available. All ARVs are associated with toxicity, which increases with prolonged exposure to the drugs. Similar to adults, up to 10% of children will be long-term non-progressors and may not require therapy for a prolonged period.

Annual progression to AIDS or death in untreated children over 12 months of age with CD4% >20% is low (HPPMCS Group, 2003). Because of the above considerations, the US guidelines are currently being revised.

Other approaches have been developed. For example, the PENTA guidelines, followed in Europe and the UK, advocate a less aggressive approach, especially in the first year of life. Although ART is recommended for all infants with CDC Stage C and most with Stage B disease, for those who are relatively asymptomatic, therapy is recommended only if the viral load exceeds 10^6 copies/mL or the CD4% falls below 25 to 35% (PENTA, 2004).

The World Health Organization has released guidelines for ART use in resource-limited settings that advocates a public health approach (WHO, 2002). The approach is conservative, restricting ART for those most symptomatic or with severe CD4+ T-cell depletion. They also advocate using CD4+ assays but not HIV-1 RNA levels for both initiating and monitoring therapy because of cost and infrastructure constraints.

There is a need to develop strategies that, in the long term, will save resources. With the scale-up in vertical transmission prophylaxis programs worldwide, infants who are infected despite interventions are increasingly being identified early in life. This contrasts with adults where primary infection is rarely identified.

Because large numbers of HIV-infected infants are likely to be born in less developed countries, despite improving vertical transmission prophylaxis programs, initiating therapy early and continuing for life will result in an enlarging pool of children on therapy, placing the public health infrastructure under increasing strain. Conversely, adopting the WHO approach will result in fewer infants starting on therapy early, but once it has begun, the health care services will again have to cope with expanding numbers of children on therapy.

On June 20th 2007, the DSMB for the CHER trial noted that ART commenced before 12 weeks of age (Arms 2 and 3) reduced mortality by 75% compared to deferred ART (Arm 1). Therefore, the DSMB recommended that no additional subjects be randomized to deferred ART and that all subjects in this Arm be urgently reviewed for ART. In accordance with the DSMB recommendation, the results of Early versus Deferred ART were released (Violari A, Cotton MF, Gibb D, *et al.* Antiretroviral therapy initiated before 12 weeks of age reduces early mortality in young HIV-infected infants: evidence from the Children with HIV Early Antiretroviral Therapy (CHER) Study. In: 4th IAS conference on pathogenesis, treatment and prevention Sydney, Australia: International AIDS Society; 2007).

2.0 STUDY OBJECTIVES

2.1 Primary Objective

1. To compare time to failure of first line ART (due to clinical, **virological** or immunological disease progression, or regimen-limiting ART toxicities) or death among three randomised arms (infants who receive early ART in Arms 2 and 3 and

infants in whom ART is deferred until clinical or immunological disease progression in Arm 1).

2.2 Secondary Objectives

1. To compare the cumulative rate of clinical disease progression or death in the 3 randomised arms at 182 weeks (3.5 years).

2. To compare the 3 randomised arms in terms of the development of Grade 3 or 4 (laboratory or clinical) adverse events.

3. To compare the 3 randomised arms in terms of time from randomisation to starting or needing to start continuous therapy.

4. To compare hospital admission rates, time to first hospitalization, and duration of hospitalization (for non-elective procedures) in the 3 randomised arms.

5. To compare the cumulative rate of development of viral resistance at the time of failure of first line therapy in the 3 arms. In addition, the development of new mutations to NVP during the trial will be compared to baseline in children exposed and not exposed to NVP in the perinatal period.

6. To compare the 3 randomised arms in terms of time to death alone or death plus life threatening stage C events or HIV events associated with permanent end-organ damage. Life-threatening Stage C events will be defined as a malignancy (lymphoma or Kaposi sarcoma), progressive multifocal leukoencephalopathy or intercurrent infection requiring admission to an intensive care unit. HIV events associated with permanent end-organ damage will be assessed at the end of the trial and will include HIV encephalopathy, chronic lung disease, cardiomyopathy and nephropathy where the parameters used for making the initial diagnosis have not reverted to normal.

3.0 STUDY DESIGN

The CHER trial is a two-center, Phase III, randomised, open-label trial comparing three treatment strategies (described below and illustrated in Figure 2) in infants with perinatally acquired HIV-1 infection diagnosed between 6 and 12 weeks of age. The study is being conducted at the Perinatal HIV Research Unit, Chris Hani Baragwanath Hospital, Soweto, Johannesburg, and the KID-CRU Clinic, Tygerberg Children's Hospital, Cape Town, South Africa.

The trial will have two parts: Part A and Part B. **Four hundred and eleven** HIV-1 infected infants, with a CD4 percentage ≥25% at screening, will be enrolled in Part A. Approximately forty infants with CD4 percentage <25% at screening will be enrolled in Part B. Infants in Part A initially randomised in a 1:1:1 ratio to deferred ART (Arm 1), ART for 40 weeks (Arm 2) or ART for 96 weeks (Arm 3) will now only be randomized in a 1:1 ratio to ART for 40 weeks (Arm 2) or ART for 96 weeks (Arm 3). See below for full description of Arms. All infants in Part B will receive continuous therapy. The sample size calculations are based on Part A only. Part A deferred Arm 1 and Part B are closed to accrual.

Randomisation will be stratified by site and by entry into Part A or B of the trial. Enrolment **took** place over 78 weeks (1.5 years), **but was reopened to increase the sample size and power due to a change in the criteria for starting therapy in line with the WHO guideline at the time. The** minimal follow-up will be for at least 182 weeks (3.5 years). Therefore, the first infant enrolled will be followed for **up to 330** weeks (**6** years) and the last infant enrolled will be followed for at least 182 weeks (3.5 years).

PART A: 4**11** infants with definitive HIV-1 infection and minimal immune suppression (CD4 percentage ≥25%) ≤12 weeks of age will be randomised in a 1:1:1 ratio to one of 3 arms. The Steps referred to below correspond to the different phases of the treatment strategies, as illustrated in Figure 2.

Arm 1: Deferred ART

Arm 2: 40 weeks of ART (from between 6 and 12 weeks of age until approximately first birthday) and interrupt

Arm 3: 96 weeks of ART (from between 6 and 12 weeks of age until approximately second birthday) and interrupt

Arm 1: Infants randomised to Arm 1 will not start ART at randomisation (Step 1a). They will start first line therapy (AZT+3TC+LPV/r) **according to the criteria below.** First line ART will then be continuous until the participant reaches a study endpoint. They may then change to second line therapy (Step 3). (See Section 7.3.3).

Arms 2 and 3: Infants randomised to Arms 2 and 3 will receive AZT+3TC+ LPV/r as first line therapy. This regimen will be initiated at randomisation (between 6 and 12 weeks of age) (Step 1b) and then interrupted at 40 and 96 weeks in Arms 2 and 3, respectively (Step 1c). Therapy will restart **according to the criteria below.** The same first line ART will be restarted (Step 2). First line ART will then be continuous until the participant reaches a study endpoint. They may then change to second line therapy (Step 3). (See Section 7.3.3).

ART (start or restart) when

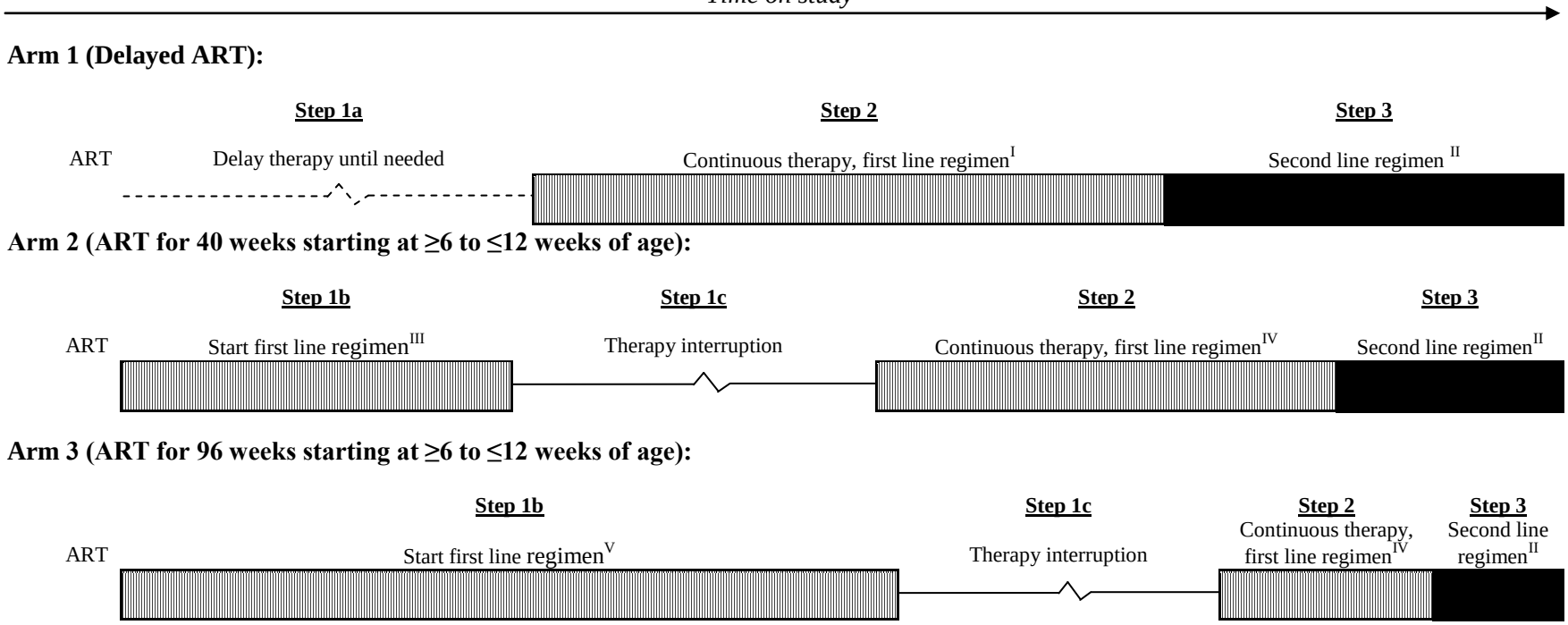
1. The CD4% falls below 25% or the CD4 count is less than 1000 cells/mm^3 regardless of CD4% for children if <12 months of age;
2. The CD4% falls below 20% for children 12-**59** months of age*
3. **The CD4% falls below 20% or the CD4 count <350 cells/mm^3 for children ≥ 60 months of age**
4. Occurrence of a severe CDC Stage B or Stage C disease.

* If there is discordance between CD4% and count the investigator may use an absolute CD4 count to reintroduce ART: 12-35 months CD4% <20% or CD4 count <750 cells/mm^3; **36-59 months CD4%** <20% or CD4 count <500 cells/mm^3

PART B: Infants with evidence of moderate to severe immune suppression (CD4 percentage <25%) at screening will all receive AZT+3TC+LPV/r as first line therapy. All Part B participants will receive continuous ongoing therapy. Participants randomised to Arm 2 or 3 and who have undergone therapy interruptions will have therapy restarted

(Step 2). Part B participants who have not been interrupted will also be assigned to Step 2, continuous ongoing therapy. Once participants reach a study endpoint, they may then change to second line therapy (Step 3).

Figure 2. Schematic representation of study treatment strategies for Part A



I Start first line regimen if CD4% falls below 25% or CD4 count <1000cells/mm^3 regardless of CD4% if <12months of age; below 20% if 12-**59** months of age*; **below 20% or CD4 count <350 cells/mm^3 if ≥ 60 months,** or participant develops a severe CDC Stage B or Stage C disease.

II Any child who has failed the first line regimen will be considered for switching to a second line therapy; these children will continue to be followed until completion of the study visits for secondary objectives.

III Start first line regimen in Arm 2 between 6 and 12 weeks of age, continuing until approximately the first birthday (40 weeks).

IV Start continuous therapy, first line regimen if CD4% falls below 25% (or CD4 count <1000cells/mm^3 regardless of CD4%) if <12months of age; below 20% if 12-**59** months of age*; **below 20% or CD4 count <350 cells/mm^3 if ≥ 60 months,** or participant develops a severe CDC Stage B or Stage C disease.

V Start first line regimen in Arm 3 at ≥6 weeks continuing until approximately second birthday (96 weeks).

* If there is discordance between CD4 % and count the investigator may use an absolute CD4 count to reintroduce ART: 12-35 months CD4% <20% or CD4 count <750 cells/ mm^3; **36-59 months** CD4% <20% or CD4 count <500 cells/ mm^3

3.1 Endpoints

3.1.1 Primary Endpoint

Time to failure of first line antiretroviral therapy or death. Failure of first line therapy includes any one of the following occurring while on first line treatment.

3.1.1.1 Failure of CD4% to reach a level ≥20% by Week 24 of therapy (initial therapy or restart), or CD4% falls below 20% on 2 occasions, within 4 weeks, at any time after the first 24 weeks of therapy (initial therapy or restart) (immunological failure).

3.1.1.2 Development of severe CDC Stage B or Stage C disease, as defined in Appendices V and VI (clinical failure).

3.1.1.3 Development of toxicity requiring more than one drug substitution within the same class or a switch to a new class of drugs (regimen-limiting toxicity failure) or requiring a permanent treatment discontinuation.

3.1.1.4 Confirmed HIV-1 RNA value ≥10,000 copies per/ml recorded on two consecutive occasions after 24 weeks of treatment (initial therapy or restart). Confirmatory HIV-RNA value should be obtained at least 1 week and up to 4 weeks from the initial value unless otherwise approved by the protocol team.

3.1.2 Secondary Endpoints

3.1.2.1 Occurrence of severe CDC Stage B or Stage C disease or death (cumulative after 3.5 years).

3.1.2.2 Occurrence of Grade 3 or 4 (clinical or laboratory) adverse events.

3.1.2.3 Time from randomisation to starting or needing to start continuous therapy.

3.1.2.4 Hospital admissions, time to first hospital admission, and duration of hospitalization.

3.1.2.5 Cumulative viral resistance mutations at the time of failure of first line therapy.

3.1.2.6 Time to death alone or death plus life threatening stage C events or HIV events associated with permanent end-organ damage. (Life-threatening Stage C events will be defined as a malignancy (lymphoma or Kaposi sarcoma), progressive multifocal leukoencephalopathy or intercurrent infection that requires admission to an intensive care unit). HIV events associated with permanent end-organ damage will be assessed at the end of the trial and will include HIV encephalopathy, chronic lung disease, cardiomyopathy and nephropathy where the parameters used for making the initial diagnosis have not reverted to normal.

4.0 SELECTION AND ENROLLMENT OF STUDY PARTICIPANTS

The following study eligibility criteria apply to all participants:

4.1 Inclusion Criteria

4.1.1 Children ≥6 weeks and ≤12 weeks of age at the time of randomisation.

4.1.2 HIV-1 infection identified by one positive DNA PCR or qualitative RNA PCR confirmed by viral load (plasma HIV-1 RNA PCR with a value >1,000 copies/mL) by 12 weeks of age. This requires two separate blood specimens from two different time points.

4.1.3 Antiretroviral naïve except for receipt of antiretroviral drugs used to prevent mother-to-child transmission.

4.1.4 The parent or legal guardian of the infant must be able and willing to provide written informed consent and comply with the study requirements.

4.1.5 A breastfed child can be included in the study.

4.2 Exclusion Criteria

4.2.1 Presence of any major congenital abnormalities that are life-threatening.

4.2.2 Documented laboratory values of Grade 3 or greater for AST, ALT, ANC, hemoglobin, electrolytes, creatinine or clinical toxicity at screening, as defined by age appropriate toxicity tables (Appendices II and III).

4.2.3 The parent or legal guardian is unable or unwilling to attend regularly scheduled study visits.

4.2.4 Any acute and clinically significant medical event at randomisation that would preclude his/her participation in the study (randomisation can take place as soon as the intercurrent illness has resolved if the child is still ≤12 weeks of age).

4.2.5 Use of the following medications:

- All investigational drugs.
- All prohibited medications listed in Section 5.6.

4.2.6 Inability to tolerate oral medication.

4.2.7 Infants with a birth weight <2 kilograms.

4.3 Screening Procedures and Identification of Potential Study Participants

4.3.1 Participant Recruitment

During antenatal visits, HIV-positive mothers receive counseling where issues of PMTCT, safe infant feeding, and the routine care and testing of the child are discussed. Mothers will receive information on how to access available resources and about possible participation in research. Following delivery these issues are reinforced before the mother is discharged home. At this point, the mothers will be informed of this study and will be given appointments to come to the sites from when the child is four weeks old.

There are further opportunities to inform the mothers of this study, for example, at the postnatal check up, which takes place between three days and two weeks after delivery.

In addition to recruiting participants at the study sites, healthcare workers at MTCT prevention programs and at other sources (i.e., private practitioners or hospital outpatient departments and wards) will be informed about the trial so that eligible subjects can be referred for possible participation in this study. We do not anticipate differences in participant recruitment procedures as these healthcare workers will be informed about the trial in the same way (IRB-approved letters and personal contact).

4.3.2 Eligibility

Mothers will be encouraged to return to the clinics four weeks after delivery for pre-screening and HIV-1 diagnosis of their child, but may return later if they choose. Randomisation will only be permitted up to the age of 12 weeks.

Information about **CIPRA ZA 004** will be given at this visit. **CIPRA ZA 004** is an open-label, comparative study that will evaluate the safety and immunogenicity of a 7-valent polysaccharide-protein conjugate vaccine (PncCV) and the immunogenicity of Haemophilus influenzae type b conjugate vaccine (HibCV) in a cohort of HIV-1 infected and HIV-1 uninfected children. The cohort of HIV-1 infected children will be participants enrolled in **CIPRA ZA 002.**

Mothers who return for their child's appointment should have their child seen for a Pre-screening Visit (Week –2). At the Pre-screening Visit they will receive more detailed information about the trial and pre-test counseling for the HIV-1 diagnosis of their child. The parents or legal guardians will be asked to sign a screening consent form (see Appendix XIIIA) and their child will be assigned a screening number. This will allow for a DNA PCR or RNA PCR and the collection of demographic and clinical data. Mothers will be encouraged to return one week after the Pre-screening Visit for their child's DNA PCR or RNA PCR results.

A positive HIV-1 PCR obtained through another research study or early infant HIV-infection identification program is acceptable as long as adequate source documentation is available.

Infants with a negative DNA PCR or RNA PCR will be referred to the Expanded Program for Immunization (EPI) where they will receive routine immunizations, infant feeding counseling and an HIV ELISA at the age of 12 months.

Infants with a positive DNA PCR or RNA PCR will be seen for a Screening Visit (Week –1). This will include another blood draw for a confirmatory quantitative RNA PCR (to check eligibility for enrollment), T-cell subsets (for randomisation) and other laboratory and clinical assessments. Permission will also be sought for additional blood specimens to be collected for **CIPRA ZA 00**5, a laboratory-based study investigating affordable tests for diagnosis and monitoring of HIV infection. Studies include the comparison of dried blood spots and anti-coagulated whole blood for diagnostic DNA and quantitative RNA PCR and the HIV antibody avidity index for early diagnosis of HIV-infection in infants. Parents/legal guardians will receive more information about the trial. They will be asked to return a week later for enrollment (Week 0) of their child into the study. Parents/legal guardians will not be asked to consent for the trial at the Screening Visit because they will have just received the positive HIV-1 DNA PCR or RNA PCR results for their child on this day.

At the Randomisation Visit (Week 0), the parent or legal guardian will be asked to sign and date a study consent form (see **Appendix XIII**). If the clinical and laboratory eligibility criteria have been met, the child will be enrolled and randomised into the study. Information about **CIPRA ZA 00**4 will be given at this visit as well. Immunizations will be offered to the infants at the study sites in order to minimize health care visits and facilitate enrollment in **CIPRA ZA 00**4.

A study database will record information on all children who have been assigned a screening number. The study database will record the name, date of birth (DOB), date of randomisation and the randomisation number, if applicable. If a participant is not eligible for randomisation, the reason for ineligibility will be recorded in the study database. The study database will be password-protected with access limited to study staff.

4.4 Study Enrolment Procedures

Prior to implementation of this protocol, sites must have the protocol and consent form(s) approved by their local Institutional Review Board (IRB)/Independent Ethics Committee (IEC) and approved by the Medicines Control Council (MCC). Sites must be registered with and approved by the Protocol Registration Office at the DAIDS through the Regulatory **Support** Center (**RSC**) (**via the website**

https://daidses.niaid.nih.gov/ProtocolRegistration/ via email: Protocol@tech.res.com or by telephone: 301-897-1707). Site registration must occur before the site can enrol any participants into this study.

Once a candidate for study entry has been identified, details will be carefully discussed with the participant's parent or legal guardian. The parent or legal guardian will be asked to read and sign the consent form that was approved by the Ethics Committees at the two

sites, the MCC and the **RSC** Protocol Registration Office. If the parent or legal guardian cannot read the consent form, it will be read to him/her. The consent form must be signed and dated by the parent or legal guardian prior to conducting any study procedures.

4.4.1 Randomisation/Registration

Randomisation will be performed through the PHRU SDMC (Statistical and Data Management Center) for both sites. A randomisation list will be prepared under the direction of the trial statistician. Participant randomisation numbers will be prepared separately for each site. The study coordinator or an investigator at each site will be responsible for submitting the request for randomisation to the PHRU SDMC. The trial statistician or a designee will return the randomisation information to the site via email or fax. Randomisation will not take place until after the parent/legal guardian has given informed consent and eligibility criteria are met. Parents/legal guardians will be told their child's allocation on the day of randomisation.

4.5 Co-enrolment Guidelines

The parent or legal guardian will be informed about CIPRA **CIPRA ZA 00**4 at the screening visits and the Randomisation Visit. Co-enrolment in CIPRA **ZA 00**4 will be permitted, but is not a requirement for inclusion into this study. Parents or legal guardians will be required to sign a separate informed consent to enrol their child into **CIPRA ZA 004**. The majority of study visits, duration of follow-up and the primary health care providers for both **CIPRA ZA 002 and 004** are the same. Therefore, participants will be required to see the same study doctor and will have the same clinical records for both studies, but there will be study-specific CRFs. Co-enrollment in other studies offering primary **or non-primary therapy can only be allowed once the participant failed first line therapy (reached primary endpoint) and would benefit from switching to second line therapy.**

For specific questions and approval, investigators should consult the protocol co-chair at the site. If necessary, the protocol co-chairs will discuss the issue. All decisions will be documented in the source documents and all communication between protocol co-chairs by email.

5.0 STUDY TREATMENT

Antiretroviral study drugs used in CIPRA **ZA 002**/CHER trial will be provided for the duration of the study (up to **330** weeks). Participants will become eligible for antiretroviral agents after the completion of the study through the South African National Treatment Plan.

5.1 Antiretroviral Regimens, Administration and Duration

5.1.1 First Line Regimen

The proposed first line regimen is AZT+3TC+LPV/r. Study participants will start the first line regimen according to the Arm to which they have been randomised. Participants who fail the first line therapy regimen may switch to the second line therapy regimen.

The dosages are based on established guidelines and available data from previous studies. Dosages, formulation, and toxicities are outlined in Appendix VIII. Appendix VIII serves as a treatment guideline for investigators, and gives background and dosing information for all the allowed medications, doses and regimens. All the information in this appendix should be used as a guideline for prescribing the participants' regimens. In general, dosage based on weight or body surface area will be recalculated at every physician visit. When the weight or body surface area (BSA) changes so that the dose (in mg) differs by 10% from the previously dispensed dose, the dose must be adjusted. Doses may be changed at less than 10% difference based on the site preference.

Single drug substitution is permitted for toxicity and is described under Toxicity Management (see Section 7.1). These individual drug substitutions are not considered a regimen failure.

Table 2. First Line Regimen and Dosing

Medication	**Dose**
Zidovudine (AZT)	240 mg/m^2 twice daily • Doses can be rounded upward to the nearest 0.5ml at the clinicians discretion **AZT capsules (100mg) /tablet (300mg)** • **8-13.9kg: 24 mg/kg/day: 1 capsule twice daily.** • **14-19.9kg: 18 mg/kg/day: 2 capsules (200mg) in the morning and 1 capsule (100mg) in the evening** • **20-29.9 kg: 400mg/day: 2 capsules (200mg) in the morning and 2 capsules (200mg) in the evening** • **≥30kg: 600 mg/day: 3 capsules or 1 tablet (300mg) twice daily**
Lamivudine (3TC)	4 mg/kg twice daily with a maximum of 300 mg daily Doses can be rounded upward to the nearest 0.5ml at the clinicians discretion **3TC scored tablets (150mg)** • **14–19.9kg: ½ tablet (75mg) twice daily** • **20–24.9kg: 1 tablet (150mg) in the morning**

	and ½ tablet (75mg) in the evening • **≥ 25kg: 1 tablet (150mg) twice daily**
Lopinavir/Ritonavir* (LPV/r)	• <6 months of age: 300 mg/m^2 LPV/75 mg/m^2 RTV twice daily • ≥6 months of age and <7 kg: 230 mg/m^2 LPV/57.5 mg/m^2 RTV twice daily with a maximum of 400 mg LPV/100 mg RTV twice daily • ≥6 months of age and ≥7 kg: 7 to 10 kg: 1.25 mL twice daily >10 to <15 kg: 1.75 mL twice daily 15 to 20 kg: 2.25 mL twice daily >20 to 25 kg: 2.75 mL twice daily >25 to 30 kg: 3.5 mL twice daily >30 to 35 kg: 4.0 mL twice daily >35 to 40 kg: 4.75 mL twice daily The maximum dose of LPV/r is 400 mg LPV/100 mg RTV twice daily Doses can be rounded upward to the nearest 0.1ml at the clinicians discretion
Lopinavir/Ritonavir* film coated tablets	**Number of 100/25mg tablets:** • **10-13.9kg: 2 tablets in the morning and 1 tablet in the evening** • **14-19.9kg: 2 tablets twice daily** • **20-24.9kg: 3 tablets in the morning and 2 tablets in the evening** • **25-34.9kg: 3 tablets twice daily** • **>35kg: Adult dosing, 4 tablets twice daily** **Number of 200/50mg tablets:** • **25-34.9kg: 2 tablets in the morning and 1 tablet in the evening** • **>35kg: Adult dosing, 2 tablets twice daily** *** Tablets must not be chewed, broken or crushed.**

* Therapeutic drug monitoring (TDM) may be undertaken where possible and clinically appropriate in infants below 24 months of age. Trough levels will be drawn. Dosages will be adjusted accordingly. In such case the Team will be informed of results of TDM and proposed dosage adjustments. Team approval is required.

5.1.2 Second Line Regimen

Second line therapy will be made available to children who fail first line therapy. Refer to Section 7.3.3 for guidelines for switching from first line to second line therapy.

The proposed second line regimen is ddI+ ABC+ EFV or NVP. The choice of EFV or NVP will be determined by the participant's age and weight.

However, this regimen does not constitute the only option. If a child requires an individual substitution while on first line therapy (e.g., ddI for 3TC), the most appropriate nucleoside backbone combination will be used for the second line regimen. The individual drug substitutions will be made according to Section 7.1.

The information in Appendix VIII will be used as a guideline for prescribing the participant's second line regimen.

As children become older, solid formulations become more acceptable and are usually much easier to transport and store. As soon as children are able to swallow tablets, they will be prescribed solid formulations at the recommended dosing for solid formulations.

Table 3. Second Line Regimen and Dosing

Medication	**Dose**
Didanosine (ddI)	• 2 weeks to 8 months of age: 100 mg/m^2 every 12 hours • After 8 months of age: 120 mg/m^2 every 12 hours • **Ages 6-18 years and body weight >20kg: capsules** **20 to <25kg: 200 mg once daily** **25 to <60kg: 250 mg once daily** **>60kg: 400 mg once daily**
*Abacavir (ABC)	8 mg/kg twice daily with a maximum of 300mg twice daily *Prior to initiating ABC, subjects will be screened for HLA Human Leukocyte Antigen-B*5701. If present, the subject will not receive Abacavir. Consult with the protocol team for alternative choices. **Abacavir tablets (300mg):** **≥ 25kg: 1 tablet (300mg) twice daily**
Efavirenz (EFV) (for infants >3 years of age and >10 kg)	Administered once daily: 10 to <15 kg: 200 mg 15 to <20 kg: 250 mg 20 to <25 kg: 300 mg 25 to <32.5 kg: 350 mg 32.5 to <40 kg: 400 mg

	≥40 kg: 600 mg
Nevirapine (NVP) (for infants <3 years of age or <10 kg)	• 150-200 mg/m^2 twice daily. Note: Initiate therapy with 150 mg/m^2 administered once daily for 14 days (maximum 200 mg once daily). Increase to full dose (150-200 mg/m^2) administered twice daily if no rash or other untoward effects (maximum 200 mg twice daily). (Lead-in period should not exceed 28 days. If untoward effect persists at 28 days, switch to alternative medication. NVP should NOT be initiated if AST/ALT ≥ Grade 3 or child is Child-Pugh stage B or C)

5.1.3 Additional Drugs for Single Exchange in First Line Regimen

Table 4. Additional Drugs and Dosages for Single Drug Exchange in First Line Regimen if Intolerance or Toxicity or if Treating TB

Medication	**Dose**
Stavudine (d4T)	**≤30 kg: 1mg/kg twice daily** **>30 kg – 60 kg: 30mg twice daily** **≥60 kg: 40mg twice daily**
Nelfinavir **(**)** (NFV)	• <12 months of age: 75 mg/kg twice daily • 12-24 months of age: 60 mg/kg twice daily • >2 years of age: 55 mg/kg twice daily with a maximum of 1250 mg twice daily
Ritonavir (*) (**) (RTV)	• >1 month of age: 350 to 400 mg/m^2 of body surface area twice daily with a maximum dose of 600 mg twice daily. Initiate treatment at 250 mg/m^2 twice daily and stepwise increase the dose by 50 mg/m^2 every 2-3 days as tolerated. As rifampicin decreases the RTV levels, a dosage up to 450mg/m^2 will be permitted if also on rifampicin (***).

* Some clinicians in South Africa choose to minimise stepwise increase in order to avoid potential confusion to the caregiver. In such cases, notice will be sent to the protocol team.

**TDM may be undertaken where possible and clinically appropriate. Trough levels will be drawn. Dosages will be adjusted accordingly. In such case the team will be informed of results of TDM and proposed dosage adjustments will need team approval.

*** Updated guidelines for the use of rifamycins for the treatment of tuberculosis among HIV-infected patients taking protease inhibitors or non-nucleoside reverse transcriptase inhibitors, 2004. (Accessed 11th March 2004 at http://www.cdc.gov/nchstp/tb/TB_HIV_Drugs/TOC.htm).

5.2 Drug Formulation

Registered drug or FDA approved generics will be used.

5.2.1 First Line Regimen and Formulation

Table 5. First Line Regimen, Formulation, and Storage

Medication	Formulation	Storage
Zidovudine (AZT)	Syrup: 10 mg/mL Capsules: 100 mg Tablets: 300 mg	Store at 2º-30ºC (36º-86ºF) and protect from light.
Lamivudine (3TC)	Oral solution: 10 mg/mL Tablets: 150 mg	Store at 2º-25ºC (36º-77ºF) and protect from light.
Lopinavir/Ritonavir (LPV/r)	Oral solution: 80 mg LPV and 20 mg RTV per mL	Store at 2º-8ºC (36º-46ºF). Avoid exposure to excessive heat. Instruct caregivers to store LPV/r under refrigeration if possible. Once brought to room temperature (up to 25ºC or 77ºF), use LPV/r oral solution within 2 months.
Lopinavir/Ritonavir (LPV/r)	**Film-coated tablets 200mg/50mg, 100mg/25mg**	**No special storage conditions are required**

5.2.2 Second Line Regimen and Formulation

Table 6. Second Line Regimen, Formulation, and Storage

Medication	Formulation	Storage
Didanosine (ddI)	• Pediatric powder for oral solution when reconstituted as solution containing antacid: 10 mg/mL • Chewable tablets with buffers: 50, 100, 150, 200	Store at room temperature 15º -25ºC (59-77ºF). Once the didanosine powder has been constituted with water and antacid, it must be stored between 2º-8ºC and

	mg • **Enteric coated tablets: 125 mg, 200 mg, 250 mg and 400 mg**	will only remain stable for 30 days.
*Abacavir (ABC)	• Oral solution: 20 mg/mL • Tablets: 300 mg	Store at room temperature 15º -30ºC (59º-86ºF) away from heat and direct light. Do not freeze solution.
Efavirenz (EFV)	• Capsules: 50 and 200 mg	Capsules are stored at room temperature below 30ºC.
Nevirapine (NVP)	• Oral solution: 10 mg/mL • Tablets: 200 mg	Viramune® tablets packaged in plastic bottles: store below 25ºC. Viramune® oral solution: store below 30ºC. Do not refrigerate. Shake bottle gently before use.

5.2.3 Alternative Antiretrovirals to be Used in Case of Intolerance or Single Drug Substitution for Toxicity

Table 7. Alternative Drugs, Formulation and Storage

Medication	**Formulation**	**Storage**
Stavudine (d4T)	• Oral solution: 1mg/mL • Capsules: **15,** 20 **and** 30 mg	Zerit® capsules and powder for solution should be stored at room temperature 15º-30ºC (59º-86ºF), in tightly sealed containers, protected from heat and moisture. After constitution, store Zerit® solution in tightly sealed bottles under refrigeration (2º-8ºC) for up to 30 days.
Nelfinavir (NFV)	• Powder for oral solution: 50 mg/g • Tablets: 250 mg	Store at room temperature 15º -30ºC (59º-86ºF) away from heat and direct light in original containers. Nelfinavir powder should be constituted for every dose, not in the original container. After constitution, Nelfinavir solution is only stable for up to 6 hours.
Ritonavir (RTV)	• Oral solution: 80 mg/mL	Ritonavir solution should be stored at room temperature (20º-25ºC) and not

		refrigerated. Shake well before use and use within 30 days of dispensing. The solution should be stored in its original container and protected from excessive heat and freezing.

5.3 Product Supply, Distribution, and Pharmacy

5.3.1 Study Product Supply and Distribution

The study drugs for this study will be acquired in the form of innovator drugs **or FDA approved generics** from the Department of Health Provincial ARV Depots (Gauteng Depot for PHRU pharmacy and Western Cape Depot for KID-CRU pharmacy) and/or pharmaceutical companies (AZT, 3TC and ABC will be received from GlaxoSmithKline Pty). Where the innovator forms of drug cannot be supplied by the Department of Health Depots, the innovator forms of drug will be supplied by International Health Distributors Pty Ltd, Kinesis Logistics or Motswedi Pharmaceuticals as noted in the Manual of Procedures.

The site pharmacists at the PHRU Pharmacy (Chris Hani Baragwanath Hospital in Soweto) and the KID-CRU Pharmacy (Tygerberg Children's Hospital in Cape Town) will obtain the study agents for this protocol by following the instructions in the Pharmacy section of the Manual of Procedures.

5.3.2 Study Drug Accountability

Drug accountability will be maintained throughout the study according to site SOPs, in accordance with the pharmacy plans for each site.

Three levels of drug accountability will be required for the management of this protocol: (1) Site Pharmacy at the PHRU and the KID-CRU; (2) Drug cupboard storage at the primary care sites. These drug cupboards will be inside the PHRU and KID-CRU Pharmacies; and (3) Subject Drug Accountability.

5.3.2.1 Site Pharmacy

Each of the sites will be required to complete a site pharmacy plan, according to the site establishment requirements of DAIDS. The responsibilities of drug accountability, storage and labelling will be maintained by the site pharmacist. The site pharmacist will be responsible for recording the number of participants on each treatment regimen and projecting the monthly requirements for drug. Accountability will be maintained of all shipments received.

With a request from the site, in the form of a prescription, the site pharmacist will label drug on a per-participant basis. Labelling of the drug includes:

- Participant study initials
- Participant identification number
- Drug name and strength
- Quantity of tablets/solutions/suspensions
- Expiration date - only required if dispensed out of original container
- Directions for administration
- Prescribing site
- Dispenser contact details (site pharmacist)
- Dispensing date
- Visit number

Accountability records will be maintained for all of the treatment prescriptions filled or provided to the participant on drug accountability records.

5.3.2.2 Primary Care Site Drug Cupboards

Study drug at PHRU and KID-CRU is placed into a CIPRA-specific lockable, temperature controlled drug cupboard within the pharmacy.

A member of the primary care team, after establishing enrollment of the participant, will provide an initial prescription to the site pharmacist. Thereafter any changes in dosage or regimen will require a new prescription.

All study drug dispensing done at PHRU and KID-CRU pharmacies will be done based on a participant-specific prescription.

The site pharmacist will provide the parent/legal guardian with the pre-labeled study drug for a particular visit. The site pharmacist is responsible for drug accountability of the CIPRA-specific drug cupboard. The site pharmacist will be responsible for explaining the details of the treatment prescription, warnings and advisories, dosing schedule and adherence to the parents/legal guardians.

LPV/r solution will require refrigeration at the primary care site until it is dispensed.

5.3.3 Subject Drug Accountability

Subject drug accountability will be conducted by the site pharmacist. Drug returns will be weighed or measured by the site pharmacist. Calculations of

accountability will be part of the Manual of Procedures. The site pharmacist will record the Calculated "Percentage Adherence" as well as the weight or amount of drug dispensed and returned in the source documents. Only the adherence percentage will be recorded in the CRF. When containers are not returned, the site pharmacist will be required to record, in the source document, the reasons for the participant not returning unused drugs. Drug accountability that cannot be calculated due to theft of containers for example, will be recorded as unknown.

The procedures to be followed will adhere to the Good Clinical Practices (GCP) guidelines on drug accountability. Drug accountability will be monitored by the Core A Quality Control Associate (CIPRA QCA**).**

5.3.4 Study Drug Destruction

The site pharmacist will receive all study drug directly from the participants' parents/legal guardians, with weighing/measuring and accountability completed by the site pharmacist, at the site pharmacy. Following the site's SOP for drug destruction, the site pharmacist will submit the returns of dispensed medicines to the relevant Medical Supply Disposals Company for destruction. A certificate of destruction obtained from the destruction company, confirming the specific investigational products destroyed, will be placed into the site file. Compliance with the site SOP for drug destruction will be documented and monitored.

5.4 Concomitant Medications

All participants will start cotrimoxazole prophylaxis from age 4 to 6 weeks per the standard of care, but this is not considered study therapy.

To avoid adverse events caused by drug interactions, the sites must refer to the most recent package inserts of study drugs and concomitant agents whenever a concomitant medication is initiated or a dose is changed.

Parents/legal guardians will be encouraged to seek advice prior to giving any other medication to their child. Participants should not receive antiretroviral therapy outside the trial.

5.5 Allowed Medications

Any medication not listed as disallowed may be prescribed by participant's clinician.

Children receiving chronic steroids will be allowed to continue in the study.

5.6 Disallowed Medications

Package inserts of antiretroviral and concomitant agents should be referred to whenever a concomitant medication is initiated or the dose is changed to avoid drug interaction adverse events. Please refer to the following website for the most current list of drug interactions: http://www.hivinsite.com/InSite.jsp?page=ar-00-02. The following medications are disallowed:

- Disallowed with NNRTIs
  - Antihistamines (with EFV)
    - Astemizole
    - Terfenadine
  - Anti-infectives
    - Ketoconazole
  - GI Motility
    - Cisapride
  - Sedative/hypnotic
    - Midazolam (can give a single dose only if adequate monitoring)
    - Triazolam
  - Ergot alkaloids and derivatives
  - Calcium Channel Blockers
    - Bepridil
  - Neuroleptic
    - Pimozide
- Disallowed with PIs
  - Antiarrhythmics
    - Amiodarone
    - Flecainide
    - Propafenone
    - Quinidine
  - Anticonvulsants
    - Phenytoin
  - Antihistamines
    - Astemizole
    - Terfenadine
  - Anti-infectives
    - Systemic Itraconazole
  - GI motility
    - Cisapride
  - HMG-CoA Reductase Inhibitors
    - Lovastatin
    - Simvastatin
  - Psychiatric medication
    - Pimozide
  - Sedative/hypnotic
    - Midazolam (Oral midazolam is contraindicated. If parenteral midazolam is co-administered in subjects on Lopinavir/ritonavir, close monitoring for respiratory depression and/or prolonged sedation should be exercised and dosage adjustment should be considered.)
    - Triazolam
  - Ergot alkaloids and derivatives

5.7 Precautionary Medications

Package inserts of antiretroviral and concomitant agents should be referred to whenever a concomitant medication is initiated or the dose is changed to avoid drug interaction adverse events. Use of the agents listed below while on study may require additional monitoring of drug levels or for adverse events. These precautionary medications include, but may not be limited to:

- Precautionary with NNRTIs
  - Anticonvulsants
    - Carbamazepine
    - Phenytoin
    - Phenobarbitol
  - Anti-infectives
    - Atovaquone
    - Clarithromycin
    - Dapsone
    - Fluconazole
    - Systemic Ketoconazole
    - Proguanil
    - Rifampicin and Rifabutin
    - Quinupristin/Dalfopristin
    - Voriconazole
    - Itraconazole
  - HMG-CoA Reductase Inhibitors
    - Lovastatin
    - Simvastatin
  - Hormonal agents
    - Glucocorticoids
  - Sedative /hypnotic
    - All benzodiazepines
  - Other
    - Warfarin
    - Sildenafil
    - Antacids/H2-blockers/Proton Pump inhibitors
    - Diltiazem
    - Felodipine, Nicardipine, Nifedipine, Verapamil
- Precautionary with PIs
  - Analgesics
    - Codeine and other opiates
    - Propoxyphene
  - Antiarrhythmics
    - Bepridil
    - **Digoxin**
    - Disopyramide
    - Lidocaine
    - Mexiletine
  - Anticonvulsants

- Carbamazepine
- Phenobarbitol
- Phenytoin

- Anti-infectives
  - Atovaquone
  - Clidamycin
  - Clarithromycin
  - Dapsone
  - Fluconazole
  - Miconazole
  - Rifabutin and Rifampicin
- Beta-blockers
  - Atenolol
  - Carvedilol
  - Esmolol
  - Labetolol
  - Propranolol
- Calcium channel blockers
  - Nifedipine
  - Verapamil
- HMG-CoA Reductase Inhibitors
  - Atorvastatin
  - Fluvastatin
- Hormonal agents
  - Glucocorticoids
- Psychiatric Medications
  - All SSRI (selective serotonin re-uptake inhibitors) antidepressants
  - Tricyclic antidepressants
- Sedative/hypnotics
  - All benzodiazepines
- Other
  - Sildenafil
  - Warfarin
  - Theophylline

6.0 CLINICAL AND LABORATORY EVALUATIONS

6.1 Timing of Evaluations

All clinical and laboratory evaluations are outlined in the Schedule of Events in Appendices IA, IB and IC. Appendix IA is the Schedule of Events for all participants in Steps 1a, 1b and 1c. Appendix IB is the Schedule of Events for all participants in Step 2 who start/restart first line therapy. Appendix IC is the Schedule of Events for all participants in Step 3 who start second line therapy.

All participants in Arm 1 (Step 1a) will follow the schedule outlined in Appendix IA until first line therapy is required. When therapy is required, they will progress to Step 2 and follow the Schedule of Events in Appendix IB. All participants in Arms 2 and 3 (Steps 1b and 1c) will follow the schedule outlined in Appendix IA until the restart of first line therapy is required or planned interruption cannot occur. When restart of first line therapy is required, they will progress to Step 2 and follow the schedule outlined in Appendix IB. All participants progressing to Step 3 will follow the schedule outlined in Appendix 1C.

Participant status and progression will be tracked through a "Step" approach as follows (see Table 8 below and Figure 2 on page 32):

Table 8. Step assignation for tracking status of study participants

Step	ART	PART	ARM
1a*	Delay until needed	A	1
1b**	Up to week 40 Up to week 96	A	2 3
1c**	Interrupted phase	A	2 & 3
2	1st line	A, B	1, 2 & 3 (post interruption)
3	2nd line	A, B	1, 2, 3

* Part 1 is closed to accrual
**Part B subjects on Step 1b and 1c will be reassigned to Step 2

6.1.1 Pre-screening Visit

Pre and Post Test Counseling will be conducted as per site SOP. Written consent must be given by the participant's parent or legal guardian prior to start of any screening evaluations.

The Pre-screening Visit occurs when the child is between 4 and 12 weeks of age.

6.1.2 Screening Visit

All screening evaluations must occur prior to the participant starting any study medications, treatments or interventions.

The Screening Visit will occur prior to randomisation. The time between the Screening Visit and the Randomisation Visit should ideally be within one week and not exceed two weeks. A maximum of 2 days beyond 2 weeks will be permitted with permission of the site PI.

6.1.3 Entry Evaluations (Baseline Visit/Week 0)

Eligibility for enrolment will be confirmed. Participants who meet all the eligibility criteria, with no contraindications to starting ART, and who are ≤12 weeks of age will be randomised. Participants with CD4 percentage ≥25% will be randomised in Part A to Arms 2 or 3. Participants with CD4 percentage <25% will be assigned to Part B (continuous therapy). Accrual to Arm 1 has stopped.

All participants will start cotrimoxazole prophylaxis from age 4 to 6 weeks per the standard of care, but this is not considered study therapy.

All participants randomised to Arms 2 and 3 will start ART. Participants in Arms 2 and 3 will continue on ART until planned interruption at Weeks 40 and 96, respectively. If a participant does not meet the criteria for planned interruption, if clinical, **virological** or immunological disease progression as defined in Section 3.1 occurs, ART will continue.

All participants will receive routine immunizations (see Appendix XI).

6.1.4 Post-Entry Evaluations

Study visits must be scheduled according to the Schedule of Events. Initially, the participants in all three arms will have the same schedule of follow-up visits according to the Schedule of Events in Appendix IA. A window period of ± 7 days will be allowed for post-entry evaluations.

6.2 Overview of Management of Participants

6.2.1 Participants Randomised to Arm 1

On-study evaluations will be performed during regular monitoring visits after completing the screening and randomisation visits. Participants randomised to Arm 1 (deferred ART) will have follow-up visits at Weeks 2, 4, 8, 12, 16, 20**,** 24, 32, 40 and 48, then every 12 weeks until the end of the study, or premature study discontinuation.

Parents/legal guardians will be required to attend monthly dispensing visits from randomisation until Week 40.

Once ART starts, scheduled study visits will be at Weeks 2, 4, and 12, with a possible interim visit at Week 8 at the study doctor's discretion. Participants receiving therapy will come to the clinic for monthly dispensing visits and be seen by the study doctor at least every 12 weeks, according to the Schedule of Events outlined in Appendix IB. Arm 1 is closed to accrual.

6.2.2 Participants Randomised to Arm 2

On-study evaluations will be performed during regular monitoring visits after completing the screening and randomisation visits. Participants randomised to

Arm 2 (ART until Week 40) will have follow-up visits at Weeks 2, 4, 8, 12, 16, 20, 24, 32, 40 and 48, then every 12 weeks until the end of the study, or premature study discontinuation.

Parents/legal guardians will be required to attend monthly dispensing visits while their child is receiving study medication.

The study doctor will assess the participant for eligibility to interrupt therapy at Week 40. He/she will record this decision in the participant's source documents and CRFs. Interruption can occur if the participant has not reached an endpoint before or on Week 40 (planned interruption), otherwise the participant will not be eligible for treatment interruption and ART will continue.

Participants who are eligible for treatment interruption will be evaluated at Week 40 by the study doctor. If the participant meets the eligibility criteria for planned interruption, ART will be stopped at this visit. At Week 48, a study visit will be scheduled to ascertain whether any adverse events occurred following interruption.

When ART is restarted, scheduled study visits will be at Weeks 2, 4, and 12, with a possible interim visit at Week 8 at the study doctor's discretion. Participants receiving therapy will come to the clinic for monthly dispensing visits and be seen by the study doctor at least every 12 weeks, according to the Schedule of Events outlined in Appendix IB.

6.2.3 Participants Randomised to Arm 3

On-study evaluations will be performed during regular monitoring visits after completing the screening and randomisation visits. Participants randomised to Arm 3 (ART until Week 96) will have follow-up visits at Weeks 2, 4, 8, 12, 16, 20, 24, 32, 40 and 48, then every 12 weeks until the end of the study, or premature study discontinuation.

Parents/legal guardians will be required to attend monthly dispensing visits while their child is receiving study medication.

The study doctor will assess the participant for eligibility to interrupt therapy at Week 96. He/she will record this decision in the participant's source documents and CRFs. Interruption can occur if the participant has not reached an endpoint by Week 96. If a participant has reached an endpoint before or on Week 96 (planned interruption), the participant will not be eligible for treatment interruption and ART will continue.

Participants who are eligible for treatment interruption will be evaluated at Week 96 by the study doctor. If the participant meets the eligibility criteria for planned interruption, ART will be stopped at this visit. At Week 104, a study visit will be scheduled to ascertain whether any adverse events occurred following interruption.

When ART is restarted, scheduled study visits will be at Weeks 2, 4, and 12, with a possible interim visit at Week 8 at the study doctor's discretion. Participants receiving therapy will come to the clinic for monthly dispensing visits and be seen by the study doctor at least every 12 weeks, according to the Schedule of Events outlined in Appendix IB.

6.3 Unscheduled Visits

If a participant has an intercurrent illness or an adverse event, the parent/legal guardian will be encouraged to attend the clinic during regular hours to have the child evaluated by a study team member for medical care. Laboratory evaluations will be performed as indicated.

The parent/legal guardian will be provided with a 24-hour emergency contact number of a member of the study team for visits outside the regular clinic hours. The "on-call" study doctor will evaluate the participant and make arrangements for further medical care, as required.

If a participant is seen at a centre other than the trial clinic, the parent/legal guardian will be instructed to inform the doctor/clinic sister of their child's participation in the trial. The parent/legal guardian will have a letter with details regarding the child's participation in the trial, details of the trial itself and the contact details of the trial physician. This letter will also request that the doctor/clinic sister notify the trial physician by telephone through the 24-hour emergency contact number and provide a written report as soon as possible (see Appendix XII).

6.4 Premature/Permanent Discontinuation Evaluations

Parents/legal guardians who do not wish for their child to be followed and desire permanent study discontinuation will be asked to allow the study doctor to complete the final study evaluations on the child's last visit. The study staff will document the reason for premature discontinuation.

6.5 Study Completion Evaluations

Study completion evaluations will include a history and physical examination, vital signs, laboratory assessments (CD4+ count, hematology, chemistry, lipid profile, storage and urinalysis [if possible]), assessment of adverse events, medication dispensing as appropriate, and adherence determination.

6.6 Adherence Assessment

The following strategies will be implemented to monitor adherence: Caregivers will be instructed to return all medication bottles. The site pharmacist will note the quantity (volume or weight) dispensed and returned at the subsequent visit. At each dispensing visit, the counselor or nurse will complete an Adherence Questionnaire with the caregivers.

6.7 Definition of Evaluations and Special Instructions

All clinical information within this protocol must be recorded on the CRFs and entered into the database unless otherwise stated. Laboratory data will be captured in the laboratory database. Sites must refer to the Source Document Guidelines Standard Operating Procedure (SOP).

6.7.1 Informed Consent

Written informed consent must be obtained from the parent or legal guardian before any screening procedures are initiated for all participants.

6.7.2 Documentation of HIV-1 Infection

HIV-1 infection will be documented by one positive DNA PCR or qualitative RNA PCR confirmed by viral load (with a plasma HIV-1 RNA with a value >1000 copies/mL) by 12 weeks of age. This requires two separate blood specimens from two different time points.

The documentation of HIV-1 infection is required before study entry and can be conducted during the screening process. Guidelines for voluntary counseling and testing need to be followed.

6.7.3 Medical History

A medical history must be present in the source documents. The medical history must include all diagnoses. Any allergies to medications and their formulations must be documented. The medical history will be obtained from the parent/legal guardian.

6.7.4 Medication History

A medication history must be present in the source document. A complete history of any prescription/non-prescription medications taken, including HIV treatment, should be recorded with actual or estimated start and stop dates.

6.7.5 Concomitant Medications

Participants should take as few as possible concomitant medications as medically feasible. All medications, including over the counter and herbal preparations and blood products administered during the study must be recorded on the case report form (CRF). For each concomitant medication, the generic name of the drug, start and stop dates and the route given will be recorded on the concomitant medication page of the CRF.

6.7.6 Study Medication

Study medication will be recorded on the source documents and on the study medication page of the CRF. The name of the drug, dose and start and stop dates of medication will be recorded.

6.7.7 Study Treatment Modifications

Record all study drug modifications, including initial doses, participant-initiated and/or protocol-mandated modifications, inadvertent and deliberate interruptions, and permanent discontinuation of treatment at each visit in the source documents and on the CRFs.

6.7.8 Clinical Assessments

Complete Physical Exam

A complete physical examination is required at screening, baseline, Weeks 2, 4, 8, 12, 16, 20, 24, 32, 40 and 48, and every 12 weeks until the end of the study or premature study discontinuation. It includes, at a minimum, an examination of the skin, head, mouth, and neck; auscultation of the chest; cardiac exam; abdominal exam; and neurological exam. The complete physical exam will also include signs and symptoms, diagnoses, and vital signs. **Prior to 2 years of age blood pressure will be measured when necessary; from 2 years of age blood pressure will be routinely measured.**

Targeted Physical Exam

A targeted physical examination includes vital signs (temperature and pulse [respiration rate is optional]) and is to be driven by any previously identified or new signs or symptoms, including diagnoses that the participant has experienced either since birth or the last visit.

Height, weight, head circumference and body surface area

Refer to Appendix VII for procedures.

Signs and Symptoms

At entry, all grades must be recorded according to the Division of AIDS Toxicity Table for Grading Pediatric Adverse Experiences (April 1994), which can be found on the Regulatory **Support** Center (**RSC**) Web site: **http://rsc.tech-res.com/.** All post-entry, abnormal signs and symptoms must be recorded. Record all signs and symptoms that lead to a change in treatment, regardless of grade.

Adverse Event Assessment

At each scheduled visit, a study physician will conduct an evaluation of intercurrent illnesses, and a review of systems and any available laboratory results.

Adverse Event Check

Record any intercurrent illnesses.

Diagnoses

Record all diagnoses, both confirmed and probable, in the source document. Diagnoses will be reported on the CRFs as per CRF guidelines.

6.7.9 Laboratory Evaluations

At screening, all pre-entry and entry laboratory values must be recorded. For post-entry assessments, record all laboratory values. All laboratory toxicities that lead to a change in treatment, regardless of Grade, must be recorded. Sites must refer to the Division of AIDS Toxicity Table for Grading Severity of Pediatric Adverse Experiences (April 1994). (See Appendices II and III).

Hematology

Hematology samples will be analysed for hemoglobin, hematocrit, mean corpuscular volume, white blood cells, differential (lymphocytes, neutrophils, monocytes, eosinophils) and platelets.

Blood chemistry

Chemistry samples will be analyzed at screening for full liver function tests (bilirubin, AST, ALT, alkaline phosphatase and gamma GT [GGT]) and urea and electrolytes (urea, creatinine, chloride, potassium, and sodium). AST and ALT will be analyzed for all visits following screening and urea and electrolytes will be analyzed every six months.

Lipid Profile

Cholesterol and triglyceride samples will be collected at screening and every 6 months thereafter. This test will be performed in a non-fasting state. However, if results for cholesterol are >300 and triglycerides are >750 (see Appendix IIIB: Supplemental Table for Grading of Cholesterol and Triglycerides), a complete fasting-state lipid profile must be drawn. Participants <2 years of age must fast for ≥4 hours, and participants ≥2 years must fast for ≥8 hours. Samples will be analyzed for HDL, LDL, triglycerides and total cholesterol.

Urinalysis

Urinalysis will be analyzed at baseline and will be attempted at least every six months. If urine cannot be collected at a specific time point, it can be deferred sequentially to each next visit until a sample is obtained. Analysis will include urine dipsticks for protein and glucose (leucocytes, nitrites and blood if indicated). If a participant has ≥2+ proteinuria, in absence of UTI, collect an early morning urine sample for the protein/creatinine ratio to exclude nephropathy.

Immunologic Studies

CD4+/CD8+/CD3+ count and percentages will be obtained from a certified laboratory.

Stored specimens

Plasma/serum and PBMCs or cell pellets will be stored for viral load and resistance testing. If a participant's parent/legal guardian declines permission to store specimens, their child may still participate in the study. Arrangements for long-term storage of samples in a repository have not yet been arranged. Sites should store these samples on site until further instructions for storage, testing and/or shipping are provided. See CIPRA ZA 002 Laboratory Instructions Manual.

Viral load

A quantitative PCR will be done at screening and every three months **after the baseline visit of each step** for participants on continuous ART.

7.0 PARTICIPANT MANAGEMENT

7.1 General Toxicity Management

General Information

CHER will follow the Division of AIDS Toxicity Table for Grading Severity of Pediatric Adverse Experiences (April 1994) (Appendices II and III) as a guideline for grading and managing clinical and laboratory toxicities. The Supplemental Toxicity Table for Grading Severity of Adult and Pediatric Cutaneous/Skin Rash/Dermatitis Adverse Experiences (Appendix IV) will be used to grade skin manifestations.

Toxicities may be due to non-study drugs, such as cotrimoxazole, antibiotics or anti-tuberculosis medications, either alone or as a result of interaction with study medication. Toxicities may be due to non-drug etiologies and as a general rule for all Grade 3 and 4 toxicities, an evaluation is recommended to exclude other causes.

For all Grade 3 and 4 laboratory and clinical abnormalities, every effort should be made to have the study participant come to the site for repeat clinical observation, and/or laboratory test and exclusion of non-drug related causes within 72 hours. If unable to do this, document the process followed to locate the participant. Re-evaluation of the study participant should also be done at that time.

The general principle is to re-introduce study medications once toxicity has resolved to $\leq$ Grade 2, unless there is a specific contraindication (e.g. following hypersensitivity; see

sections 7.2.2. for NVP or EFV and 7.2.9 for ABC) or implication of a specific study drug, as noted below. For certain conditions an immediate substitution is possible.

If toxicities do not resolve within three weeks of the study medication being withheld, a consultation with the protocol co-chair is necessary.

Single drug substitution is allowed in this trial and does not constitute a regimen failure. Specific information on single drug substitution can be found in Table 3 in Section 5.1.2.

All discussions with the protocol co-chair regarding toxicity management should be documented in the source documents and should be sent by email to the designated officer whenever possible.

For hematological toxicity, rash, pancreatitis, increase in values for AST and ALT, and lactic acidosis, follow course of action and toxicity management outlined under 7.2.1, 7.2.2, 7.2.3, 7.2.4 and 7.2.8, respectively.

Due to the fact that many participants in this study may also be enrolled in CIPRA ZA 004, if a systemic toxicity occurs, it may not be possible to distinguish whether the toxicity is related to the ART being received through this study or to the study vaccines being administered in CIPRA ZA 004. Where it is not possible to attribute the toxicity to a specific cause, it will be considered a study related AE in both studies.

Severity Grading

Grade 1:

Continue study drugs; routine monitoring.

Grade 2:

Continue study drugs. If relevant, monitor closely and consider more frequent clinical and laboratory assessments.

Grade 3:

The clinician has the option of immediately stopping the study drugs if a confirmatory laboratory test cannot be performed within 72 hours, or if he/she determines that the continuation of study drugs is unsafe while awaiting test results. However, the clinician may decide that for certain Grade 3 laboratory abnormalities, study participants should continue taking study drugs pending receipt of repeat laboratory tests.

For all confirmed suspected drug-related toxicities, stop all study drugs until the toxicity resolves to ≤ Grade 2.

- For toxicities not directly attributed to a study drug, re-start therapy (all study medications). If toxicity recurs to ≥ Grade 3 soon after reintroduction and is not attributable to only one drug, the current ARV regimen must be revised to second line therapy.

- For all toxicities clearly attributable to a specific drug (NRTI, PI or NNRTI):

    - The implicated drug may be switched with re-introduction of the remaining two ARVs once toxicity resolves to ≤ Grade 2.

    OR

    - The study drugs may be restarted at the discretion of the clinician, unless there is a specific contraindication (e.g. following hypersensitivity; see sections 7.2.2. for NVP or EFV and 7.2.9 for ABC), but if the toxicity recurs, consideration should be given to permanently discontinuing the implicated drug.

The new replacement drug will usually be selected while the child is off study medications. Clinicians must discuss the drug options with the protocol co-chair. This change in therapy due to toxicity will not constitute a "switch" from first-to second-line therapy. If toxicity recurs to Grade 3 or greater soon after reintroduction, consider another replacement drug or a switch to second line therapy.

If the clinical circumstances support that the adverse event is unrelated to study medication or is believed to be clinically insignificant, the clinician has the option of continuing study medication after notification of the protocol co-chairs. For a persistent abnormality at the same grade, consult the protocol co-chair.

Grade 4:

Stop all medications pending confirmation of adverse experience and assessment of management strategy.

For all suspected drug-related toxicities hold all study drugs until the toxicity resolves to ≤ Grade 2, then re-start therapy (all study medications) unless there is a specific contraindication (e.g. following hypersensitivity; see sections 7.2.2. for NVP or EFV and 7.2.9 for ABC). If toxicity recurs soon after reintroduction and is not attributable to only one drug, the current ARV regimen must be revised to second-line therapy.

For all toxicities that can be clearly attributed to a specific drug (NRTI, PI or NNRTI), hold all study drugs until the toxicity resolves to ≤ Grade 2. Then, restart therapy unless there is a specific contraindication (e.g. following hypersensitivity; see sections 7.2.2. for NVP or EFV and 7.2.9 for ABC), but strongly consider discontinuing the implicated drug permanently and continue all other medications. The selection of the new drug for replacement will be done while the child is off study medications. Clinicians must discuss the drug options with the protocol co-chairs. This change in therapy due to toxicity will not constitute a "switch" from first-to-second line therapy. If the toxicity recurs to a Grade 3 or greater after a specific switch within a class, the current ARV regimen must be revised to second-line therapy.

To avoid a delay in diagnosis and minimise the risk of a life-threatening hypersensitivity reaction (see section 7.2.9), ABC should be permanently discontinued if hypersensitivity

cannot be ruled out, even when other diagnoses are possible (respiratory diseases, flu-like illness, gastroenteritis or reactions to other medications).

If the clinical circumstances support that the adverse event is unrelated to study medication or is believed to be clinically insignificant, the clinician has the option of continuing study medication after notification of the protocol co-chairs. For a persistent abnormality at the same grade, consult the protocol co-chair.

If there is any doubt about participant management, discuss with the protocol co-chairs who, in turn, may consult the protocol team.

7.2 Protocol Specific Toxicity Management

The management of specific hematological, dermatological and hepatic toxicities, suspected pancreatitis and lactic acidosis is described below.

7.2.1 Hematological Toxicity

The use of G-CSF/GM-CSF and erythropoietin is not standard of care in South Africa and therefore will not be used for the management of hematological toxicity. Although weekly checks are preferable, checks may be ordered every second week depending on study participant circumstances and after discussion with the protocol co-chair.

7.2.1.1 Anemia

Grade 1:
- Continue study drugs.
- Check that correct doses of ARVs and cotrimoxazole have been given.
- Look for reversible causes.

Grade 2:
- Continue study drugs.
- Check that correct doses of ARVs and cotrimoxazole have been given.
- Consider more frequent laboratory assessments.

Grade 3:
- Check that correct doses of ARVs and cotrimoxazole have been given. If dosage has been too high, give correct dosages and repeat after a week.
- If dosage was correct, stop cotrimoxazole and repeat hemoglobin in 2 weeks.
    - If recovery to ≤ Grade 2, then change to 3X weekly cotrimoxazole and repeat hemoglobin in 2 weeks.
        - If ≤ Grade 2, continue 3X weekly (and ascribe toxicity to cotrimoxazole daily dosage).
        - If Grade 3 toxicity recurs, change to alternative pneumocystis prophylaxis.

- If Grade 3 persists, change to alternative pneumocystis prophylaxis.

- If patient not receiving cotrimoxazole or if toxicity persists after altering cotrimoxazole dosing, consider either interrupting ARVs or switching AZT to d4T. Consider alternative etiologies for anemia.
  - Interrupt ARVs.
    - Check weekly until ≤ Grade 2 and then reintroduce previous regimen.
    - If toxicity recurs to Grade 3 or greater, switch AZT to d4T and manage as below.

  OR

  - Drug substitution.
    - Switch from AZT to d4T.
    - Check weekly until ≤ Grade 2.
    - If recovery to ≤ Grade 2, ascribe toxicity to AZT.
    - If no recovery, stop all ARVs, await recovery to ≤ Grade 2 and revise regimen to second line therapy after discussion with protocol co-chair.

Grade 4:

- Consider a blood transfusion.
- Stop ARVs and cotrimoxazole.
  - Check that correct doses of ARVs and cotrimoxazole have been given.
- Once recovery to ≤ Grade 2:
  - If dosage had been too high, restart ARVs and cotrimoxazole at correct doses and repeat after a week.
  - If correct doses had been given consider either:
    - Re-introduction of ARVs.
      - If toxicity recurs to Grade 3 or greater, await recovery to ≤ Grade 2 and switch from AZT to d4T.
      - If toxicity recurs to Grade 3 or greater after the switch to d4T, stop all ARVs, await recovery to ≤ Grade 2 and revise regimen to second line therapy after discussion with protocol co-chair.

    OR

    - Drug substitution.
      - Switch from AZT to d4T
      - If toxicity recurs to Grade 3 or greater, stop all ARVs, await recovery to ≤ Grade 2 and revise regimen to second line therapy after discussion with protocol co-chair.
  - When ARV therapy is stabilized re-introduce cotrimoxazole.

7.2.1.2 Management of Neutropenia

Grade 1:

- Continue study drugs.
- Check that correct doses of ARVs and cotrimoxazole have been given.
- Look for reversible causes.

Grade 2:

- Continue study drugs.
- Check that correct doses of ARVs and cotrimoxazole have been given.
- Consider more frequent laboratory assessments.

Grade 3:

- Check that correct doses of ARVs and cotrimoxazole have been given and consider alternative etiologies. If dosage has been too high, give correct dosages and repeat after a week.
- If correct doses and on cotrimoxazole:
  - Stop cotrimoxazole (and continue on ARVs)
  - If recovery to ≤ Grade 2
    - Change to 3X weekly cotrimoxazole.
    - Check weekly.
      - If remains ≤ Grade 2, continue 3X weekly (and ascribe toxicity to cotrimoxazole daily dosage).
      - If Grade 3 returns, stop cotrimoxazole and change to alternative pneumocystis prophylaxis.
      - If no recovery, change to alternative pneumocystis prophylaxis.
- If Grade 3, not attributable to cotrimoxazole
  - Switch from AZT to d4T
    - Check weekly
    - If recovery to ≤ Grade 2, continue current ARV regimen, and ascribe toxicity to AZT.
    - If no improvement after 2 weeks, stop all ARVs, check weekly until ≤ Grade 2 and then start second line regimen after discussion with protocol co-chair.

Grade 4:

- Stop ARVs and cotrimoxazole.
  - Check that correct doses of ARVs and cotrimoxazole had been given.
- Once recovery to ≤ Grade 2:
  - If dosage had been too high, restart drugs with correct doses and repeat after a week.
  - If correct doses had been given, re-introduce ARVs but switch from AZT to d4T.
  - Monitor weekly
    - If no recurrence of Grade 3 or greater toxicity, continue therapy and re-introduce daily cotrimoxazole.

- Monitor weekly X2 and then every second week X2.
- If relapse to Grade 3 or greater toxicity, consider toxicity due to cotrimoxazole and change to alternative pneumocystis prophylaxis.

▪ If Grade 3 or greater toxicity recurs, stop all ARVs, check weekly until ≤ Grade 2 and then start second line regimen after discussion with protocol co-chair.

7.2.2 **Rash**

7.2.2.1 Management of Skin rash in Participants Not Receiving NVP, EFV or ABC Containing Regimens (Participants on First Line Therapy)

Grade the skin rash using Appendix IV (the Supplemental Toxicity Table for Grading Severity of Adult and Pediatric Cutaneous/Skin Rash/Dermatitis Adverse Experiences). Drugs in first line therapy most likely to be associated with rash are 3TC and LPV/r, although rash occurs uncommonly with these drugs. The most likely agent concomitant medication to cause rash is cotrimoxazole.

Grade 1 Rash

For Grade 1 rash (erythema with or without pruritis), study drug(s) should be continued. If rash does not resolve within 14 days of onset, the protocol co-chair should be contacted.

Grade 2 Rash

For Grade 2 rash, treatment should continue without interruption but should be monitored closely by the site investigator. Participants may be treated symptomatically with antipyretics, antihistamines and/or non-steroidal anti-inflammatory medications.

If participant is receiving cotrimoxazole, the drug may be discontinued, at the discretion of the clinician or at the request of the family. If withdrawn, cotrimoxazole should not be re-introduced. Rather proceed with Dapsone.

Grade 3 Rash

For Grade 3 rash, participants should discontinue all study drugs. If the Grade 3 rash is considered most likely to be due to concomitant illness or medications, standard management, including discontinuation of other likely causal agents should be undertaken. Upon resolution to < Grade 1, the clinician should discuss with the protocol co-chair whether re-introduction of study drugs is appropriate. If rash remains at ≥ Grade 3, all study drugs should be permanently discontinued. The participant has then

met the end-point of regimen-limiting toxicity and should change to second line therapy.

Grade 4 Rash

For Grade 4 rashes (e.g., exfoliation, mucosal involvement or target lesions [erythema multiforme]) or any evidence of Stevens-Johnson syndrome (SJS)), participants should discontinue all study drugs. No re-introduction with the causative agent is permitted.

7.2.2.2 Management of Skin rash in Participants Receiving NVP, EFV or ABC Containing Regimens (Participants on Second Line Therapy)

Any rash that occurs in the setting of EFV or NVP or abacavir (ABC) use should prompt an investigation for non-nucleoside reverse transcriptase inhibitor (NNRTI) or ABC hypersensitivity and include clinical evaluation for systemic symptoms and laboratory evaluation of liver transaminases, complete blood count with differential and creatinine. Clinical symptoms and supporting laboratory values consistent with NNRTI hypersensitivity include fever, rash, mucosal involvement or conjunctivitis, allergic symptoms (urticaria, wheezing), elevated transaminases (>2 x ULN), peripheral eosinophilia, elevated serum creatinine (>Grade 2), arthralgia or myalgia. Clinical symptoms and supporting laboratory values consistent with ABC hypersensitivity most commonly include fever, rash and gastrointestinal symptoms. Some subjects are initially thought to have a flu-like illness or respiratory disease. Apart from elevated transaminases, creatinine and creatine phosphokinase may also be elevated. Lymphopenia also occurs.

In participants experiencing hypersensitivity reactions (HSR) to NVP, EFV or ABC, the drugs should be permanently discontinued regardless of the grade of the rash. Isolated rash in the setting of NVP, EFV or ABC therapy does not constitute an HSR.

In addition participants should be advised that the rash (rash of any grade) may worsen and to seek medical care or contact the study site clinicians as soon as possible if there is any worsening of the rash, or if systemic signs or symptoms develop that could be compatible with an HSR. It is important to monitor the participant, especially if EFV or NVP has not been interrupted because of rash, to ensure that symptoms do not worsen or that signs or symptoms consistent with clinical hepatitis or with hypersensitivity to NNRTI do not develop.

Pruritis or rash with pruritis and minor accompanying symptoms may be managed with antihistamines, antipyretics and/ or non-steroidal anti-inflammatory (NSAI) medications.

Participants who experience rash during the first 2 weeks of the NVP lead in period should not have their NVP dose increased until the rash resolves. If rash persists to 28 days of NVP lead-in period, NVP should be permanently discontinued.

Grade 1 Rash

For participants who develop a Grade 1 rash (erythema with or without pruritis), but have no abnormalities consistent with an HSR (clinical and laboratory abnormalities listed above) and no evidence of clinical hepatitis, study drug(s) should be continued with very close follow up. If rash does not resolve within 14 days of onset, the protocol co-chair should be contacted.

Subjects on ABC for less than 6 weeks who develop a Grade 1 rash will be instructed to contact the investigators as soon as possible. The subject will be carefully evaluated for ABC HSR. Should the rash continue for more than 7 days without alternative explanation, ABC will be discontinued at the discretion of the site PI.

Grade 2A Rash

Grade 2A rashes, are diffuse erythematous macular or maculopapular rash or dry desquamation with or without pruritis, but without constitutional findings or target lesions without blister/vesicle or ulceration in lesions. If there is no abnormality consistent with an HSR (clinical and laboratory abnormalities listed above) and no evidence of clinical hepatitis, study drugs and cotrimoxazole should be continued with very close observation. If the rash does not resolve within 14 days of onset, the protocol co-chair should be contacted.

Subjects on ABC for less than 6 weeks who develop a Grade 2A rash will be instructed to contact the investigators as soon as possible. The subject will be carefully evaluated for ABC HSR. Should the rash continue for more than 7 days without alternative explanation, ABC will be discontinued at the discretion of the site PI.

Grade 2B (urticaria)

For participants who have mild to moderate urticaria, without constitutional symptoms, without increase above baseline of AST/ALT, and without evidence of clinical hepatitis or HSR, EFV or NVP may be continued with close follow up and may be treated symptomatically with antihistamines; systemic steroids should not be used. However, if the patient's family or the site investigator decides to discontinue either of these drugs, they may not be reintroduced; they are permanently discontinued. The protocol co-chairs should be notified.

If participant is receiving cotrimoxazole, the drug may be discontinued, at the discretion of the clinician or at the request of the family. If withdrawn, cotrimoxazole should not be re-introduced. Rather proceed with Dapsone.

For subjects on ABC for less than 6 weeks developing urticaria, ABC HSR will be considered most likely and ABC will be discontinued unless there is convincing evidence of an alternative cause.

Grade 3

All study drugs should be permanently discontinued.

Grade 4

All study drugs should be permanently discontinued.

7.2.3 Pancreatitis

If a participant develops nausea, vomiting, or abdominal pain of any grade associated with elevation of serum lipase (> Grade 1), or develops a clinical syndrome that, in the opinion of the participant's clinician, is classified as pancreatitis, current ARVs should be stopped. The participant should be monitored closely until toxicity resolves.

Other causes of lipase elevation need to be excluded. These include bowel perforation and infarction, pancreatic pseudocyst, peritonitis and drugs (bethanechol, cholinergic medications, codeine, indomethacin, meperidine, methacholine, heparin, and morphine). Excessive hemolysis causes a "false elevation" due to interference in the assay (Tietz, 1995). Elevation of lipase ≥ Grade 2 will be regarded as evidence of pancreatitis.

If symptoms of pancreatitis develop without elevated lipase, the clinician has the option to continue treatment. If the participant is receiving ABC, then the clinician should evaluate for abacavir hypersensitivity (see section 7.2.9). Consider performing an abdominal ultrasound or CT scan to exclude a pancreatic pseudocyst and gall stones. However, treatment may be continued. The lipase level should be repeated in two weeks.

For a diagnosis of pancreatitis**,** all study medication and concomitant medication should be discontinued. Hold all study drugs until the lipase has normalised.

- Restart ARVs. Do not use ddI or d4T.
- If recurrence of elevation of lipase ≥ Grade 2, consider switch to new regimen.

7.2.4 Increase in Values for AST and ALT

An increase in AST and ALT can either be asymptomatic or be associated with clinical evidence of hepatitis. In the first 8 to 10 weeks, elevations are often drug-

related. Lactic acidosis becomes more likely at approximately 6 months. Elevated AST and ALT levels may also be associated with ABC hypersensitivity, see section 7.2.9.

For any increase in AST or ALT in the presence of clinical evidence of hepatitis, study drug should be discontinued until resolution of clinical symptoms and AST and ALT levels ≤ Grade 1.

Consider the impact of concomitant medications such as cotrimoxazole and anti-tuberculosis medication in both symptomatic and asymptomatic increase in AST or ALT.

7.2.4.1 Asymptomatic increase in AST and ALT

Grade 1:

Monitor carefully and continue ARVs.

Grade 2:

All study drugs, excluding NNRTIs may be continued at the discretion of the protocol co-chair if there is an obvious aetiology, i.e., concomitant illness and concomitant medication.

Grade 3:

If the participant is receiving cotrimoxazole, this alone may initially be withdrawn, but with careful re-evaluation after a week. Otherwise, study drugs should be held.

Generally, medications once discontinued should be withheld until levels are ≤ Grade 1, at which time therapy may be reintroduced. All medications may be restarted if the laboratory abnormalities were thought secondary to a concomitant illness. If the participant was receiving an NNRTI (EFV or NVP), either of these medications should be considered the most likely cause of the elevations and permanently discontinued. If recurrence of Grade 3 toxicity, after consultation with the protocol co-chair, consider "within class" drug substitutions or commence second line therapy. If the decision is made to switch to NVP, AST and ALT must have reverted to Grade 2 or less.

Grade 4:

All current and concomitant ART should be discontinued until levels are ≤ Grade1. If alternative aetiology is identified, resume ART. If the participant was receiving an NNRTI (EFV or NVP), either of these medications should be considered the most likely cause of the elevations and permanently discontinued.

If the study participant had been on cotrimoxazole, withhold cotrimoxazole and recheck after 1 to 2 weeks. If the participant had not been on cotrimoxazole, switch ARVs. Consider substitution within class. Consult with the protocol co-chair.

7.2.4.2 Drug-related hepatitis

Symptoms of clinical hepatitis include the following: fatigue, malaise, anorexia, nausea, acholic stools, bilirubinuria, jaundice, liver tenderness, or hepatomegaly. Clinical hepatitis should be considered in participants who have signs and symptoms of hepatitis, even if LFTs are normal or alternative diagnoses are possible. Study drugs, in particular NVP, should be interrupted as soon as possible in participants who have symptoms consistent with clinical hepatitis. NVP should be permanently discontinued. Other study drugs may be restarted on resolution of symptoms after consideration of nucleoside induced hepatic steatosis/lactic acidosis (see Section 7.2.8) or ABC hypersensitivity (see section 7.2.9) as a cause of the symptoms and review of management with protocol co-chairs.

7.2.5 Raised AST and ALT in Study Participants on Anti-tuberculosis Therapy and ART

Tuberculosis is endemic in Johannesburg and Cape Town. The prevalence of HIV in children with culture-confirmed tuberculosis varies from 13 to 70% (Lucas, 1996; Mukadi, 1997; Madhi, 2000; Jeena, 2002).

There may be overlapping toxicity for study participants on ART and anti-tuberculosis therapy (Burman, 2001). The most common overlapping profiles include skin rash, nausea and vomiting, and hepatitis.

In the event of toxicity, anti-tuberculosis therapy takes precedence over ART. This implies that anti-tuberculosis therapy may be continued instead of ART, when managing toxicity.

7.2.6 Hyperglycemia/Glycosuria

If the urinary chemistry is >2+ positive, a random blood glucose level should be obtained. If the random blood glucose is >11 mmol/L, refer to an endocrinologist regarding possible new onset diabetes. If the random glucose level is between 8.25 and 11 mmol/L, perform a fasting blood glucose. If the fasting blood glucose level is >8.25 mmol, refer to an endocrinologist for further assessment.

7.2.7 Elevated Cholesterol or Triglycerides

Initiation of HAART, with or without PIs and/or NNRTIs, has been associated with elevated cholesterol and triglyceride levels. A non-fasting lipid profile will be done on all participants at screening and every 6 months thereafter. If elevated, it should be managed as follows:

- For cholesterol levels >300, do a fasting lipid profile.
- For triglyceride levels >750, do a fasting lipid profile.

Cholesterol

If Grade 2 or greater toxicity is recorded, sites should contact the protocol co-chair to discuss further management.

Triglycerides

If Grade 2 or greater toxicity is recorded, sites should contact the protocol co-chair to discuss further management.

Please refer to Appendix IIIB: Supplemental Table for Grading Cholesterol and Triglycerides.

7.2.8 Lactic Acidosis

The relevance of asymptomatic lactic acid elevations is unclear, and lactates are not part of the routine safety evaluations for this study. Routine lactate monitoring is not currently recommended. No baseline lactic acid levels will be obtained.

A sometimes-fatal syndrome of lactic acidosis, often associated with evidence of hepatic steatosis, is a recognized but rare complication of NRTI therapy. This syndrome is felt to be secondary to mitochondrial toxicity induced by the inhibitory effect of NRTIs on DNA polymerase gamma, a key enzyme needed for mitochondrial DNA synthesis. Current knowledge regarding this syndrome is incomplete. Obesity, gender and prolonged NRTI exposure may be risk factors. It has been reported in children. Symptoms of lactic acidosis frequently involve non-specific symptoms, such as fatigue, weakness and fever, but in the majority of cases also involves symptoms suggestive of hepatic dysfunction such as nausea, vomiting, abdominal or epigastric discomfort, abdominal distension, hepatomegaly and new onset elevated liver enzymes. (If the participant is receiving ABC, then the clinician should evaluate for ABC hypersensitivity (see section 7.2.9)). A high index of suspicion may be required to diagnose this condition.

Alternatively, it is possible that unwarranted concern may be raised by over-interpretation of lactic acid levels. NRTI toxicity is only one cause of lactic acidosis. Lactic acid elevations are also seen in the context of diabetes mellitus, uremia, liver disease, infections, malignancies, alkaloses and drug and toxin ingestion of such substances as ethanol, methanol, ethylene glycol, and salicylates.

For participants with persistent ALT and AST values of ≥5 times ULN (≥ Grade 2 toxicity), with no easily discernible etiology (e.g., acute hepatitis A, B, C, or chronic hepatitis B or C), or participants with clinical acidosis, a serum bicarbonate and electrolytes will be performed and an anion gap ($[A^-]$) calculated.

$$[A^-] = \{[Na^+]+ [K^+]\} – \{[HCO_3^-] +[Cl^-]\}$$

A value of <12 mEq/liter will be accepted as normal (Behrman, 2003). If the [A^-] is increased, ARVs should be withheld and an urgent serum lactate performed. Urinalysis should be performed to look for ketones (to consider ketoacidosis) and a blood glucose test should be performed. If lactic acidosis is suspected, serum lactate should be measured regardless of the anion gap, which may be normal in the presence of elevated lactate.

- A serum lactate of <2 mmol/L is considered normal (Ravel, 1994). The ARV regimen should then be restarted, provided there are no other contraindications.
- An elevated serum lactate between 2 to 5 mmol/L may be indicative of lactic acidosis. Study drugs should be withheld at the discretion of the protocol co-chair and may be cautiously restarted when the serum lactate level is <2 mmol/L.
- A serum lactate >5 mmol/L will be considered as indicative of lactic acidosis. All study drugs should be withheld; contact the protocol co-chair. This will usually be regarded as regimen-limiting toxicity unless a highly associated and non-recurring contributory factor is identified. The protocol team will work with the participant's clinician to determine the best course of action for participants in whom NRTI associated liver dysfunction syndrome and lactic acidosis are confirmed. Supportive care should be given to the participant. If a contributing factor is identified, consideration may be given to cautiously restarting the same regimen.

7.2.9 Abacavir Hypersensitivity

In clinical studies, approximately 8% of subjects receiving an ABC-containing product developed a hypersensitivity reaction, which in rare cases has proved fatal.

7.2.9.1 Description of the Hypersensitivity Reaction

The ABC hypersensitivity reaction is characterised by the appearance of symptoms indicating multi-organ involvement. The majority of patients have fever and/or rash as part of the syndrome; however reactions have occurred without rash or fever.

Symptoms can occur at any time during treatment with ABC, but usually appear within the first six weeks of initiation of treatment (median time to onset 11 days). The symptoms worsen with continued therapy and can be life threatening. These symptoms usually resolve shortly after discontinuation of ABC.

Frequently observed signs and symptoms include fever, rash, malaise or fatigue, gastrointestinal symptoms such as nausea, vomiting, diarrhoea, or abdominal pain and respiratory symptoms such as dyspnoea, sore throat, or cough. Other signs

and symptoms include myalgia, arthralgia, oedema, pharyngitis, headache, paresthesia or myolysis.

Physical findings may include rash (usually maculopapular or urticarial), lymphadenopathy or mucous membrane lesions (conjunctivitis, mouth ulceration). Abnormal chest x-ray findings may also be present (predominantly infiltrates, which can be localised).

Laboratory abnormalities may include elevated liver function tests (such as hepatic transaminases), increased creatine phosphokinase or creatinine levels, and lymphopenia.

Anaphylaxis, hypotension, liver failure, renal failure, adult respiratory distress syndrome or respiratory failure may occur.

Some patients with hypersensitivity were initially thought to have respiratory disease (pneumonia, bronchitis, pharyngitis), a flu-like illness, gastroenteritis or reactions to other medications. This delay in diagnosis of hypersensitivity has resulted in ABC being continued or re-introduced, leading to a more severe hypersensitivity reaction or death. Therefore, the diagnosis of hypersensitivity reaction should be carefully considered for patients presenting with symptoms of these diseases. If hypersensitivity reaction can not be ruled out, no medicinal product containing ABC (Ziagen®, Trizivir®, Epzico® or Kivex®) should be restarted.

Patients who develop a hypersensitivity reaction must discontinue ABC and must never be rechallenged with any medicinal product that contains ABC (Ziagen®, Trizivir®, Epzicom® or Kivexa®). Restarting any ABC-containing product following a hypersensitivity reaction results in a prompt return of symptoms within hours. This recurrence of the hypersensitivity reaction may be more severe than on initial presentation, and may include life-threatening hypotension and death.

There have been infrequent reports of hypersensitivity reactions following reintroduction of ABC, where the interruption was preceded by a single key symptom of hypersensitivity (rash, fever, malaise/fatigue, gastrointestinal or a respiratory symptom).

On very rare occasions hypersensitivity reactions have been reported in patients who have re-started therapy, and who had no preceding symptoms of a hypersensitivity reaction.

7.2.9.2 Management of Hypersensitivity Reactions

Patients developing signs or symptoms of hypersensitivity MUST contact their doctor immediately for advice.

If a hypersensitivity reaction is diagnosed the ABC-containing product MUST be discontinued immediately. The patient should be asked to return all unused supplies of the ABC-containing product for disposal to prevent an accidental re-challenge.

An ABC containing medicinal product (Ziagen®, Trizivir®, Epzicom® or Kivexa®), MUST NEVER be administered following a hypersensitivity reaction, as more severe symptoms will recur within hours and may include life-threatening hypotension and death.

To avoid a delay in diagnosis and minimise the risk of a life-threatening hypersensitivity reaction, the ABC-containing product should be permanently discontinued if hypersensitivity cannot be ruled out, even when other diagnoses are possible (respiratory diseases, flu-like illness, gastroenteritis or reactions to other medications).

Symptomatic support for ABC hypersensitivity may be indicated. This should include, for example, administration of intravenous fluids to patients who develop hypotension. Antihistamines or corticosteroids have been used in cases of ABC hypersensitivity, however there are no clinical data demonstrating the benefit of these in the management of the reaction.

Laboratory and other investigations which may be useful in the evaluation and treatment of ABC hypersensitivity include, but may not be limited to, measurement of ALT, AST, creatine phosphokinase, serum creatinine and white blood cell differential count and chest x-ray, if respiratory symptoms are present.

7.2.9.3 Special Considerations Following an Interruption of ABC Therapy

If therapy with ABC has been discontinued and restarting therapy is under consideration, the reason for discontinuation should be evaluated to ensure that the patient did not have symptoms of a hypersensitivity reaction. If hypersensitivity reaction cannot be ruled out, no medicinal product containing ABC (Ziagen®, Trizivir®, Epzicom® or Kivexa®) should be restarted.

There have been infrequent reports of hypersensitivity reaction following reintroduction of an ABC-containing product where the interruption was preceded by a single key symptom of hypersensitivity (rash, fever, malaise/fatigue, gastrointestinal symptoms or a respiratory symptom). If a decision is made to restart any ABC-containing product in these patients, this should be done only under direct medical supervision.

On very rare occasions hypersensitivity reactions have been reported in patients who have re-started therapy, and who had no preceding symptoms of a hypersensitivity reaction. If a decision is made to re-start an ABC-containing product, this must be done only if medical care can be accessed readily by the patient or others.

7.2.9.4 Essential Patient Information

Investigators must ensure that patients are fully informed regarding the following information on the hypersensitivity reaction:

- Patients must be made aware of the possibility of a hypersensitivity reaction to ABC that may result in a life threatening reaction or death.

- Patients developing signs or symptoms possibly linked with a hypersensitivity reaction MUST CONTACT their doctor IMMEDIATELY.

- Patients who are hypersensitive to ABC should be reminded that they must never take any ABC containing medicinal product (Ziagen®, Trizivir®, Epzicom® or Kivexa®) again.

- In order to avoid restarting the ABC-containing product, patients who have experienced a hypersensitivity reaction should be asked to return the remaining tablets or oral solution to the pharmacy.

- Patients who have stopped an ABC -containing product for any reason, and particularly due to possible adverse reactions or illness, must be advised to contact their doctor before restarting.

- Each patient should be reminded to read the Package Leaflet included in the pack.

- Patients should be reminded of the importance of removing the Alert Card included in the pack, and keeping it with them at all times.

7.2.9.5 Reporting of Hypersensitivity Reactions

All cases of potential ABC hypersensitivity should be reported using the Non-GSK ABC HSR CRF Module provided; these should be completed and faxed to the CIPRA-SA Safety Desk within one week of the onset of the hypersensitivity reaction and recorded on an AE CRF.

7.2.9.6 Stevens Johnson Syndrome, Toxic Epidermal Necrolysis or Erythema Multiforme

Serious skin reactions such as Stevens Johnson Syndrome, Toxic Epidermal Necrolysis or Erythema Multiforme have been reported very rarely in patients taking ABC-containing products. These patients generally do not have the cluster of additional symptoms (e.g., gastrointestinal and respiratory) that characterize the ABC hypersensitivity reaction, but they do have features typical of these serious skin reactions.

If a serious skin reaction develops, the ABC -containing product should be discontinued, and the patient should not be rechallanged with any ABC -containing medicinal product (Ziagen®, Trizivir®, Epzicom® or Kivexa®).

As many products other than ABC also cause these serious skin reactions, all other medicinal products that the patient is receiving should also be reviewed and discontinued as appropriate.

7.3 Management of Study Regimen

7.3.1 Criteria for Proceeding with Therapy Interruption (Step 1c)

For participants in the following situations:

- CD4% ≥20% at previous visit;
- Absence of severe CDC Stage B or Stage C disease (see Appendices V and VI);
- Absence of regimen limiting toxicity.

Interruption can be delayed for up to 6-8 weeks (while the participant is being evaluated for a specific event) after which the decision may be referred to the protocol team for resolution

7.3.2 Criteria for Proceeding with Continuous Therapy, First Line Regimen (Start or Restart) (Step 2)

For participants in the following situations:

- The CD4% falls below 25% or the CD4 count is less than 1000 cells/mm^3 regardless of CD4% for children if <12 months of age;
- The CD4% falls below 20% for children 12-**59** months of age*
- **The CD4% falls below 20% or the CD4 count <350 cells/mm^3 for children ≥ 60 months of age**
- Or severe CDC Stage B or Stage C disease (see Appendices V and VI);
- In addition, any severe condition felt to be HIV-related can lead to restarting ART after discussion and approval **by the study team**
- There should always be < Grade 3 AST, ALT, ANC, hemoglobin, electrolytes, creatinine or clinical toxicity, as defined by age appropriate toxicity tables (see Appendices II and III). However, one may discuss individual cases with the protocol co-chair.
- All Part B participants.
- Participants randomized to Arm 1 and who do not meet study defined clinical and/or immunologic criteria for proceeding to continuous treatment can nevertheless receive first line continuous ARV treatment at

the discretion of the treating doctor if he/she deems necessary according to the DSMB recommendation; such decision will be recorded in the study database.

* If there is discordance between CD4 % and count the investigator may use an absolute CD4 count to reintroduce ART: 12-35 months CD4% <20% or CD4 count <750 cells/ mm^3; **36-59 months** CD4% <20% or CD4 count <500 cells/ mm^3

7.3.3 Criteria for Proceeding with Continuous Therapy, Second Line Regimen (Step 3)

Participants may be switched to the second line regimen if they fail the first line regimen, as defined by:

- CD4% <20% on two separate occasions, within 4 weeks.
- Severe CDC Stage B or Stage C disease (see Appendices V and VI);
- Regimen-limiting toxicities.

Additional criteria that must be met in order to start the second line regimen include:

- Rash, if present, resolved to ≤ Grade 1;
- AST/ALT if abnormal, resolved to ≤ Grade 1;
- All other toxicities resolved to ≤ Grade 2;
- If nevirapine is prescribed, absence of active hepatitis;
- If efavirenz is prescribed, participant must be >3 years of age and >10 kg.
- For ABC, subject must be negative for HLA B*5701. (This applies to subjects newly started on ABC once the assay has been available. Subjects already on stable ABC without hypersensitivity do not require the HLA B*5701 test and can continue ABC).

Before deciding to switch therapy regimens, the following should be considered:

- Clinicians will be encouraged not to switch the first line therapy within 12 weeks of starting or restarting ART, even if the participant has reached an endpoint.
- Clinicians will also be encouraged to make every attempt to ensure that participants are adherent to ART before a switch to the second line therapy regimen is considered.
- The clinician will consult with the protocol co-chair prior to switching therapy.

- In the event of toxicity, the participant should not be switched to the second line therapy until the toxicity resolves.

7.3.4 Criteria for Failure of Second Line Therapy

In the opinion of the investigator, clinical, **virological** and immunological progression may warrant a change in therapy. Laboratory toxicity requires change or discontinuation of regimen if specified in the Toxicity Management section of the protocol. Participants who fail the second line regimen will receive standard of care, and the parent/legal guardian will be asked to continue the child on study until completion of the study visits.

7.4 Management of Children with Tuberculosis

If a study participant develops tuberculosis (TB) on first line antiretroviral therapy, boost LPV/r with additional RTV so that equal dosage of LPV and RTV are given (Ren, 2008). This translates into giving an additional 0.75ml of RTV for each ml of LPV/r being given. **Under these circumstances, care must be taken where subjects have pre-existing cardiac disorders because of prolonged PR interval on an electrocardiogram (EKG).**

For subjects requiring antituberculosis therapy while on NFV, review the indication(s) for switching from the first line PI (LPV/r).

i) If it was due to taste intolerance of LPV/r, an attempt will be made to switch the patient back to LPV/r. In this case TB treatment will be co-administered together with ritonavir (RTV) as outlined above.
ii) If LPV/r was discontinued because of toxicity, for example non-resolving Grade 4 or Grade 3 toxicity, LPV/r or RTV may not be used again. Patients who are clinically stable will interrupt antiretroviral (ARV) therapy until they finish TB treatment and then resume their revised regimen. If the patient's CD4 is low and concurrent treatment for HIV and TB is required then the patient should be switched to the most appropriate second line therapy. As no further switches are allowed in the first line regimen the patient would have reached an endpoint.

If the participant is on second line ART and develops TB, continue with EFV and TB therapy. If efavirenz (EFV) cannot be used as part of second line therapy, ART may need to be interrupted for the duration of the anti-tuberculosis therapy (6 to 9 months) as TB therapy takes precedence over ARV therapy.
For patients on rifampicin and either efavirenz or ritonavir, trough levels for either ARV will be drawn, if possible, for therapeutic drug monitoring and dosage of EFV or RTV will be modified accordingly once approval is obtained from the Protocol Team.

7.5 Management of Immune Reconstitution Inflammatory Syndrome (IRIS)

Definition: Paradoxical clinical deterioration after starting HAART, due to the improving immune system interacting with organisms colonizing the body during the early stages of HIV infection.

Causes: A wide range of pathogens may induce IRIS, including *Mycobacterium tuberculosis* (MTB), *Mycobacterium avium complex, Cryptococcus neoformans, Aspergillus fumigatus, Candida albicans, Pneumocystis jiroveci*, CMV, JC virus, Human Herpes viruses, Human Papilloma virus and hepatitis B and C viruses (HBV, HCV).

Presentation: IRIS usually presents during the first 2 weeks until approximately 6 months of starting HAART. Clinical presentations vary and depend on the causative organism and the organ system that is colonised. For example, disease caused by MTB may present with high fever, lymphadenopathy, worsening of the original tuberculous lesion, and/or deteriorating chest radiographic manifestations, including the development of a military pattern or pleural effusion.

Management plan:

- Continue antiretroviral treatment unless the subject is receiving ABC and hypersensitivity cannot be ruled out (see section 7.2.9).
- Confirm diagnosis of the opportunistic infection (OI).
- Continue or initiate specific therapy for the OI.
- Evaluate the participant clinically to exclude a new infectious process if the participant was already under therapy for the OI.
- Initiate anti-inflammatory agents, initially non-steroidals or, if needed, corticosteroids at the discretion of the clinician.
- Consult with the protocol co-chair.

7.6 Treatment of Acute Retroviral Syndrome

Management plan:

- After interruption of therapy, the parent/legal guardian will be instructed to report any constitutional symptoms or signs to the study doctor.
- The parent/legal guardian will be instructed to bring the study participant for an unscheduled visit if they are concerned.
- There is a scheduled visit eight weeks after interruption where the parent/legal guardian will be questioned about feeding habits, constitutional symptoms, such as fever, and intercurrent events since interruption. A physical examination will be performed.
- In cases of fever, new onset of oral candidiasis or constitutional symptoms, the study participant will be investigated for intercurrent infection. Although lymphocyte subsets are scheduled for measurement eight weeks after interruption, they will be measured before that time for any serious adverse event.

- ART will be resumed if the participant **shows signs of:**
  - clinical disease progression or
  - immunological **disease progression**
    - CD4% falls below 25% or CD4 count <1000 cells/mm^3 regardless of CD4% if <12 months of age;
    - below 20% if 12-**59** months of age*;
    - **below 20% or CD4 count <350 cells/mm^3 if ≥ 60 months,**
  - or severe CDC Stage B or Stage C disease).

Otherwise, there will be no specific treatment.

* If there is discordance between CD4 % and count the investigator may use an absolute CD4 count to reintroduce ART: 12-35 months CD4% <20% or CD4 count <750 cells/ mm^3; **36-59 months** CD4% <20% or CD4 count <500 cells/ mm^3

7.7 Mothers that Breastfeed

Breastfed children are eligible for the study. In Soweto and the Western Cape/Tygerberg area, the state provides infant formula at no charge for the first six months. The mother's choice on infant feeding will be supported. The mode of infant feeding, breast or formula will be noted at each visit. If the mother is breastfeeding and receiving ART, her regimen will be noted.

7.8 Prophylaxis Against Opportunistic Infections:

Decisions on prophylaxis against opportunistic infections will follow the South African Department of Health Guidelines for the treatment of children with HIV in South Africa. Cotrimoxazole prophylaxis will be given to all children for approximately the first year of life, irrespective of CD4+ count. After the first year of life, cotrimoxazole prophylaxis is recommended for all HIV infected children who are not on ART.

7.9 Immunizations

Immunizations will be given according to the EPI program in South Africa. Children co-enrolled in Project 4 will receive additional immunizations as part of that study. Study visits for Project 2 have been planned to coincide with the immunization schedule for Project 4.

8.0 CRITERIA FOR DISCONTINUATION

8.1 Criteria for Discontinuation from the Study

8.1.1 Participants will be withdrawn from the study if the child's parent or legal guardian withdraws consent. Under these circumstances, ART will not continue to be supplied from the trial medication. The participant will still have access to the standard of care, as delineated by the guidelines of the South African Department of Health.

8.1.2 The study will be discontinued at the discretion of CIPRA-**SA**, South African Medicines Control Council, the Food and Drug Administration (FDA), the NIAID, the CIPRA Program Principal Investigator, the IRB/IECs, the pharmaceutical supporter or as recommended by the DSMB.

8.2 Criteria for Discontinuation of Antiretroviral Therapy

Treatment will be permanently discontinued in all participants who meet one of the following criteria:

8.2.1 The child experiences an adverse event that, in the clinician's opinion, requires the discontinuation of the ART; if no alternative ARTs are available, or if there are insufficient choices to maintain HAART, the child should continue on study, but off treatment.

8.2.2 The child's parent or legal guardian requests discontinuation of ARTs, but wishes the child to remain on study.

8.2.3 Clinical reasons believed life-threatening by the study physician, even if not addressed in the toxicity management of the protocol.

8.2.4 Participant judged by the protocol co-chair to be at significant risk of failing to comply with the provisions of the protocol as to cause harm.

9.0 STATISTICAL CONSIDERATIONS

9.1 General Design Issues

- CHER compares three strategies for giving ART to infants aged ≥6 weeks. Eligible infants with CD4 percentage ≥25% are randomly allocated to either deferred therapy (Arm 1, Step 1a), a short course of primary treatment **(PTX)** given for 40 weeks (approximately until the first birthday) (Arm 2, Step 1b), or a longer course of primary treatment given for 96 weeks (approximately until the second birthday) (Arm 3, Step 1b). Infants with CD4 percentage <25% at entry are to be randomly allocated to either Arm 2 or Arm 3 and form a separate stratum (Part B of the study). Long term treatment (LTX) taken during (Step 2) is to be started if CD4 percentage drops to **< 25% or CD4 count <1000 cells/mm^3 regardless of CD4% if <12 months of age; below 20% if 12-59** months of age*; **below 20% or CD4 count <350 cells/mm^3 if ≥ 60 months, o**r the child develops a severe CDC Stage B or Stage C disease at any time after randomisation in Arm 1, and at any time after beginning treatment interruption (Step 1c) in Arms 2 and 3; LTX will consist of the same regimen used in primary treatment PTX. First line ART will be continued until the child reaches the primary endpoint. Any child who reaches the primary endpoint will be considered for switching to second line therapy, but this will not be mandatory.

* If there is discordance between CD4 % and count the investigator may use an absolute CD4 count to reintroduce ART: 12-35 months CD4% <20% or CD4 count <750 cells/ mm^3; **36-59 months** CD4% <20% or CD4 count <500 cells/ mm^3

9.2 Primary Outcome Measure

The primary outcome measure is time from randomisation to death or failure of the first line regimen. The first line regimen is the regimen used at the initiation of Step 2 in Arm 1 and Step 1b and Step 2 in Arms 2 and 3, allowing for substitution of drugs within the same class because of toxicity. Failure of the first line regimen is defined as either:

Immunological failure, defined as failure to achieve CD4% ≥20% by 24 weeks after the start (or restart) of the regimen, or CD4% drop to a level <20% on two occasions within 4 weeks, at any time after 24 weeks of therapy (start or restart).

OR

Clinical failure, defined as the development of a severe CDC Stage B or Stage C disease (as defined in Appendices V and VI).

OR

Failure because of regimen limiting toxicity requiring the addition of or switch to a new drug class or requiring a permanent treatment discontinuation.

OR

Virological failure defined as HIV-1 RNA value ≥10,000 copies per/ml recorded on two consecutive separate occasions after 24 weeks of treatment (initial therapy or restart). Confirmatory HIV-RNA value should be obtained at least 1 week and up to 4 weeks from the initial value unless otherwise approved by the protocol team.

9.3 Method of Randomisation

Randomisation will be stratified by clinical site. The trial statistician will prepare all four randomisation schedules (Parts A and B for each of two sites). Randomised blocks of variable sizes will be used to achieve balance with respect to the number assigned to each treatment Arm. Randomisation assignments will be performed by PHRU for both participating sites.

9.4 Sample Size and Accrual

A total of approximately **451** infants (4**11** in Part A plus 40 in Part B) will be followed for a minimum of 182 weeks (3.5 years). Calculation of the sample required to achieve the primary objective is limited to the number needed for inclusion in Part A of the study and is based on the distribution of the following random durations.

Arm 1:
A_1 = time from randomisation to death while untreated; assumed to have a piecewise exponential distribution with different rates before and after one year.
B_1 = time from randomisation to starting LTX (i.e. duration of Step 1a); assumed to have a piecewise exponential distribution with different rates before and after one year.

C_1 = time from starting LTX to regimen failure or death; assumed to have an exponential distribution after 26 weeks conditional of no regimen failure in the first 26 weeks

Arm 2:
X_2 = time from randomisation to PTX regimen failure or death during Step 1b, assumed to have an exponential distribution after 26 weeks conditional of no regimen failure in the first 26 weeks.

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A_2 = time from stopping PTX to death while off therapy; assumed to have an exponential distribution with rate the same as the post-year-1 rate of A_1.

B_2 = time from stopping PTX (after 40 weeks from randomisation) to starting LTX (i.e. duration of Step 1a); assumed to have an exponential distribution.

C_2 = time from starting LTX to regimen failure or death; assumed to have an exponential distribution after 26 weeks conditional of no regimen failure in the first 26 weeks.

Arm 3:
X_3 = time from randomisation to PTX regimen failure or death during Step 1b assumed to have an exponential distribution after 26 weeks conditional of no regimen failure in the first 26 weeks.

A_3 = time from stopping PTX to death while off therapy; assumed to have an exponential distribution with rate the same as the post-year-1 rate of A_1.

B_3 = time from stopping PTX (after 96 weeks from randomisation) to starting LTX (i.e. duration of Step 1a); assumed to have an exponential distribution.

C_3 = time from starting LTX to regimen failure or death; assumed to have an exponential distribution after 26 weeks conditional of no regimen failure in the first 26 weeks.

Sample size calculation was implemented using the procedure described in Barthel, Babiker, Royston and Parmar 2006 with software described in Royston and Babiker, 2002. This requires the assumed distribution of time, T, from randomisation to the primary end-point in the three Arms. Event rates were based on the information provided below. The distribution of T was generated empirically by drawing 100,000 replicates of (X_2, X_3, A_i, B_i, C_i) for i=1, 2, 3, and calculating T as follows:

Arm 1: $T = A_1$, if $A_1 \leq B_1$, otherwise $T = B_1 + C_1$.

Arm 2: $T = X_2$ if $X_2 \leq 40/52$;
$= 40/52 + A_2$, if $X_2 > 40/52$ and $A_2 \leq B_2$;
$= 40/52 + B_2 + C_2$; otherwise.

Arm 3: $T = X_3$, if $X_3 \leq 96/52$;
$= 96/52 + A_3$, if $X_3 > 96/52$ and $A_3 \leq B_3$;
$= 96/52 + B_3 + C_3$; otherwise.

Based on longitudinal data on more than 464 untreated children with CD4+ percentages greater than 25% at age between 6 and 12 weeks – a subset of children studied by the HIV Pediatric Prognostic Markers Collaborative Study Group (HPPMCS 2003), the cumulative probabilities of CD4% falling below 20% (below 25% if under one year of age) at 1, 2 and 3 years were estimated as 38%, 53% and 58%, respectively. The median time to CD4% drop to < 20% (<25% if under 1 year) was 1.86 years and the average death rate during the first year was 8.3 per 100 person years (PY) during the first year and 7.5 per 100 PY during the second and third year of follow-up.

Data from the PENTA 5 trial of response to HAART in previously untreated children (Pediatric European Network for Treatment of AIDS, 2002) indicate that:

- In children with baseline CD4% <20% but >10% (Stratum 1), ART failure rates were 1.5% due to AIDS or death during the first 6 months and 14% due to failure to achieve CD4% >20% by 6 months. Cumulative progression rate to CDC Stage C disease or death by 2 years was 6% and cumulative immunological failure by 2 years was 40%.

- In children with baseline CD4% >20% (Stratum 2), the regimen failure rate was 0.8% due to clinical progression during the first 6 months. Cumulative immunological or clinical progression was 6% by 2 years.

We shall apply Stratum 2 rates for PTX regimen failure during the pre-interruption phase (Step 1b) in Arms 2 and 3. This is likely to be a conservative assumption since PTX is started at randomisation when all children have CD4% > 25%. LTX regimen failure rates are assumed to be a bit lower than in stratum 2 of PENTA 5 because a few children would start LTX at a CD4% between 20 and 25% if under one year of age.

Based on the above data, we make the following assumptions:

- The Death rate during the first year of follow-up but before starting therapy in Arm 1 is 8.3 per 100PY.

- The Death rate after the first year of follow-up but before starting therapy in Arm 1 is 7.5 per 100PY, a 10% reduction relative to the rate in year 1.

- The cumulative probability of starting therapy (LTX) by 1 and 2 years after randomisation in Arm 1 is 0.38 and 0.53 respectively; median time to starting therapy (Step 2) is 1.86 years.

- The probability of regimen failure or death by 26 weeks after starting LTX in Arm 1 is 0.10.

- The cumulative probability of regimen failure or death by two years after starting LTX in Arm 1 is 0.35.

- The probability of PTX regimen failure or death by 26 weeks from randomisation in Arms 2 and 3 is 0.01.

- The cumulative probability of PTX regimen failure or death by 2 years in Arms 2 and 3 is 0.06.

We further assume:

- The death rate in Arms 2 and 3 off therapy during step 1c is 7.5 per 100PY, the same as in Arm 1 after the first year.

- The probability of LTX regimen failure or death during the first 26 weeks after starting LTX in Arms 2 and 3 is 0.10 (same as in Arm 1).

- The cumulative probability of LTX regimen failure by two years in Arms 2 and 3 is 0.35 (same as in Arm 1).

- The median time from stopping PTX to starting LTX in Arms 2 and 3 is longer than the median time from randomisation to starting LTX in Arm 1 by 0.25 and 0.5 years respectively, a relative increase of 27% in Arm 3 and 13.5% in Arm 2.

- The total duration of the study is **6** years, with an enrollment period of 1.5 years (minimum follow-up is 3.5 years).

- 10 % loss to follow-up by **6** years.

Under the above assumptions, the average reduction in the hazard for the primary endpoint is about 23% in Arm 2 and 43% in Arm 3, relative to Arm 1 in Part A, and a total of 396 children (132 per Arm) with 138 primary events would be required to detect a difference in the distribution of the primary outcome between the three Arms, with power 80% and alpha of 0.05.

Based on unpublished data from Chris Hani Baragwanath Hospital, Johannesburg (A. Violari, personal communication, 2004), 10% of HIV-1 infected infants tested at 6 weeks of age had a CD4% less than 25%. It is therefore estimated that no more than 40 such infants will be included in Part B of this study.

9.5 Monitoring Plan and Interim Analysis

An independent Data Safety Monitoring Board (DSMB), established by NIAID, will monitor all aspects of the trial and make recommendations to NIAID and the study team. The DSMB will consider findings from any other relevant studies and review trial data on enrollment, safety, adherence to randomised strategies and efficacy, in strict confidence, at least yearly. The DSMB will report to the protocol co-chairs, the CIPRA Program Principal Investigator and the NIAID Program Officer if, in their view, the data provide proof, beyond reasonable doubt, that one of the allocated strategies is better (or worse) than its comparators in terms of a difference of clinically significant magnitude in mortality or in the primary outcome. The guiding statistical criteria for "proof beyond

reasonable doubt" is a Haybittle-Peto type rule, based on a difference of at least 3 standard deviations in the log relative hazard (or $p < 0.001$) in any interim analysis.

The frequency and timing of interim reviews by the DSMB will be determined by the DSMB but there should be at least one full review every year. Unscheduled reviews may be requested by the study data center, which will monitor mortality pooled across the three arms monthly. An unscheduled review will be triggered if the observed cumulative mortality by age 24 weeks is in excess of 0.15 by more than 0.67 standard errors; i.e. the lower one-sided 75% confidence bound > 0.15.

A subcommittee of the DSMB was provided with the following interim analyses every 3 months until 26 March 2008. Subsequently, the DSMB requests 6-monthly review.

- Number of deaths tabulated by treatment Arm separately for Parts A and B
- Kaplan-Meier plots of time to death by treatment Arm separately for Parts A and B
- Global test of significance of the difference in survival between the 3 Arms in Part A over all available follow-up and also restricted to the first year of follow-up
- Pair - wise comparisons of survival time separately for Parts A and B

The subcommittee could call for an unscheduled review by the full board if the interim pairwise comparison between any two Arms of the study provided sufficient evidence that there was a relative reduction in the hazard of death of at least 20% in one Arm compared to the other. i.e. the adjusted 95% upper confidence bound for the hazard ratio is no more than 0.8. Because of multiple testing in several planned interim analyses, it is important to control the probability of type 1 error and we therefore propose using a confidence interval of nominal significance 99.5% which would approximately correspond to the adjusted 95% confidence interval.

The meta-analysis by Marie-Louise Newell (Newell, 2004) reported 20% cumulative probability of death by 6 months of age which is equivalent to a mortality rate of 45 per 100 PY. We would expect a lower mortality rate in Arm 1 of Part A even before starting ART because of the careful safety monitoring of individual children in the study compared to what would happen outside a clinical trial. If the early mortality rate in Arm 1 is in fact 30 per 100PY (equivalent to 14% cumulative probability of death by 6 months) then a 20% relative reduction in the hazard of death would reduce the mortality rate to 22.5 per 100 PY equivalent to 10% cumulative probability of death by 6 months (an absolute reduction of 4%)

9.6 Analysis Plan

All comparisons involving the 3 arms will use data from Part A only. Analysis of Part B will be descriptive.

9.6.1 Primary Analysis

The three Arms in Part A will be compared, as randomised (intent-to-treat principle), in terms of the primary outcome measure (time from randomisation to death or failure of the first line ART regimen) using Kaplan-Meier plots and the log-rank test (stratified by site). The null hypothesis is that the three strategies lead to similar outcome in terms of the primary endpoint. This will be tested by a log-rank test (with 2 degrees of freedom) against the global alternative that the three Arms are not equivalent. Cox proportional hazards modelling will be used to compare the time to death or treatment failure in Arm 2 and in Arm 3, relative to Arm 1.

9.6.2 Secondary Analysis

9.6.2.1 The three Arms will be compared in terms of the cumulative probability of the development of a new severe CDC Stage B or Stage C disease or death at 182 weeks (3.5 years). Kaplan-Meier methods will be used to estimate the cumulative probability of death or the development of severe CDC Stage B or Stage C diseases. A log-rank (Wald) test of no difference (against the global alternative) between the three Arms in terms of this cumulative probability will be performed. If this provides evidence of a difference between the Arms, pair-wise comparisons will be performed.

9.6.2.2 The three Arms will be compared in terms of: a) the incidence rate of Grade 3 or 4 clinical or laboratory adverse events using Poisson regression with robust estimates of the covariance matrix to allow for extra-Poisson variation due to multiplicity of events within participants; and b) time to first Grade 3 or 4 adverse event using a log-rank test. In both cases, pair-wise comparisons will be undertaken if the test against the global alternative provides evidence of a difference between the Study Arms.

9.6.2.3 The three Arms will be compared in terms of time to starting continuous therapy (Step 2) or the need to start continuous therapy, defined as:

- a CD4% < 25% or CD4 count <1000 cells/mm^3 regardless of CD4% if <12 months of age;
- below 20% if 12-**59** months of age*;
- **below 20% or CD4 count <350 cells/mm^3 if ≥ 60 months, or**
- the development of severe CDC Stage B or Stage C disease.

A log-rank test will be conducted to assess the global difference between the three Arms. Cox proportional hazards modeling will be used for pair-wise comparison between the three Arms. These comparisons must be interpreted in the light of the fact that, by design, Arms 2 and 3 are not at risk for this outcome during the first 40 weeks and 96 weeks, respectively. To aid interpretation, the gain in median time from randomisation to Step 2 in Arms 2 and 3, relative to Arm 1, will be estimated using Kaplan-Meier methods.

If there is discordance between CD4 % and count the investigator may use an absolute CD4 count to reintroduce ART: 12-35 months CD4% <20% or CD4 count <750 cells/ mm^3; **36-59 months** CD4% <20% or CD4 count <500 cells/ mm^3

9.6.2.4 The incidence rate of hospital admissions and time to first hospital admission will be compared among the three Arms, as described in the analysis of Grade 3 or 4 adverse events (Section 9.6.2.2), using Poisson regression and a global log-rank test. In addition, the distribution of total duration of hospital admissions, up to 3.5 years, will be described and compared among the three Arms using quantile regression.

9.6.2.5 The proportion of children with primary viral resistance mutations (indicative of resistance to one or more drugs in the first line regimen) at the time of treatment failure will be described.

9.6.2.6 The three Arms in Part A will be compared in terms of time from randomisation to life threatening CDC stage C disease or death using Kaplan-Meier plots and log-rank test . The null hypothesis of no difference between the three arms will first be tested by a log-rank test (with 2 degrees of freedom) against the global alternative that the incidence of life threatening CDC stage C disease or death is not the same in three Arms. Cox proportional hazards modelling will be used to estimate the relative hazard in Arm 2 and in Arm 3, compared to Arm 1.

10.0 DATA COLLECTION AND MONITORING

10.1 Record Keeping

Case report forms (CRFs) will be provided for each participant. Participants will not be identified by name on any CRFs. Participants will be identified by a screening number at pre-screening and a unique study number will be provided by the PHRU Statistical and Data Management Center (SDMC) upon randomisation.

10.2 Role of Central Data Management

Data Management for the CIPRA ZA 002 sites will be centralized within the PHRU SDMC. Central data management responsibilities are specified in the PHRU Standard Operating Procedures. These will include: coordinating the CRF development process; formatting the finalized CRFs; designing and validating the study database; validating the DataFax system; developing the Data Quality Management Plan; coordinating centralized randomisation; data capturing of CRFs; implementing central data quality management (i.e., centralized data quality control and data quality assurance procedures); quality control reporting to the sites; expedited reporting; other reporting to the sites (such as participant accrual, CRF generation metrics, QC resolution time); data coding; reconciliation of adverse events with the safety desk; exporting data from DataFax for database locks (for interim and final analyses, safety reporting, and other expedited reporting); coordinating laboratory data transfers and merging with the clinical databases; providing final databases in specified format to designated investigators and sponsors; training site personnel on the sites' data management responsibilities; and providing other project-related assistance and support to site data management, as necessary.

10.3 Type of Data Collection

Sources of information will include history, physical exam and specimens obtained from study participants. Baseline evaluation of all participants will include demographic data, medical and surgical history, current and concomitant medication history, and previous antiretroviral therapy, which will be obtained from the child's parent/ legal guardian. Follow-up evaluation will be determined per protocol specifications.

Drug compliance, self-report and pharmacy records will be used for assessing drug compliance in this study. The site pharmacists are required to maintain complete records of all study medication dispensed and returned. The procedures to be followed will be sent directly to the site pharmacists and will adhere to the Good Clinical Practices (GCP) guidelines on drug accountability.

10.4 Data Collection Plan

DataFax is a clinical trials data management system that integrates fax and computer technologies for processing CRFs. The clinics/sites retain the original CRF hard copy and transmit an electronic image to the PHRU SDMC. Electronic transmission is accomplished by a standard fax machine via phone lines or via the Internet by either an Internet-ready fax machine or by scanning the CRFs into a local computer. All transmissions are stored as electronic images at the SDMC.

The clinics/sites will fax completed forms to the SDMC as soon as possible after completion, and will respond promptly to SDMC requests for clarifications and corrections. Early detection of recurrent problems provides an opportunity to reduce errors and improve data quality through re-training and/or written clarification.

Each DataFax CRF is identified by a barcode denoting the study number (protocol) and type of form. Pages do not need to be faxed in sequence. DataFax processes images by separating a fax into individual pages, adjusting each page to correct for proper alignment and rotation, and identifying each page based on the barcode information. DataFax uses intelligent character recognition (ICR) to extract data from checkboxes and enter numbers into numerical fields. SDMC staff reviews each CRF, comparing the data entered by the ICR process with the actual data image and correcting any discrepancies. Data in text and comment fields are manually entered. Data validation will be completed by at least two data management staff members to reduce errors in the data entry process.

Data fields requiring clarification or correction, e.g., missing data or out-of-range values, are flagged with quality control (QC) notes that are included on QC Reports regularly faxed or emailed to the clinics/sites for review. Corrections or clarifications are made on the original CRF and re-faxed to the SDMC. DataFax stores each image of a CRF that has been received and tracks all versions of each CRF, along with all associated QC notes.

10.5 Data Storage

CRFs will be kept in participant specific CRF binders at each clinic/site. The binders will be stored by each clinic in a secure and hazard free data storage environment, with double lock. Access will be restricted to study personnel authorized to handle study CRF documents.

Study databases will reside within the DataFax system on a UNIX® server at the SDMC. Study databases or subsets of study data can be exported from DataFax into any required format. Laboratory data will be accessed in electronic format from laboratory service providers at each site and stored on the server at the SDMC. Backup and archiving of all study data will adhere to the PHRU standard operating procedure for data backups, which specifies that backups will be made across several media at regular intervals.

10.6 Data Quality Control

At the clinics/sites, each CRF will be reviewed for accuracy, completeness and legibility by a trained staff clinician before faxing to the SDMC. Clinic/site staff will ensure that PHRU standard operating procedures for CRF completion and GCP are adhered to.

The protocol Data Manager at SDMC will incorporate quality control (QC) measures into the database application, such as variable type, length, legal range, and coding parameters. Additionally, the Data Manager will incorporate QC measures into the data capture system, including double data entry and in-process edit checks. Additional programming will be incorporated to ensure the quality of data, such as database queries to identify missing or duplicate data, and out of range values or discrepant data.

Quality Control Reports are generated by the SDMC on a bi-weekly basis. These reports are compiled automatically by DataFax, and include any missed visits, as well as any problems identified during routine QC procedures. QC Reports are emailed to the Study Coordinator (or designee) at each clinic/site. The Study Coordinator or designee will correct any problems identified on the QC Report (in consultation with the clinician, where appropriate) and refax the corrected form pages to the SDMC. SDMC staff will incorporate corrected information from re-faxed images into the study database. For audit trail purposes, all versions of CRF images faxed to the SDMC are archived with time and date stamps for the duration of the protocol.

10.7 Internal Site Monitoring

The CIPRA-SA project will employ internal quality control associates for the studies. Their main responsibility is to oversee the progress of the trial, assess protocol compliance, and ensure that the protocol is conducted in compliance with ICH GCP guidelines and applicable regulatory requirements. The internal quality control associates will conduct a record review of a specified number of CRFs and source documents, as outlined in the Quality Management Plan. They will ensure that the data is accurately and completely recorded and reported, and confirm that informed consent is being obtained and recorded for all participants prior to their participation in the trial. The Quality Management Plan documents the internal monitoring activities.

The monitors will interact with the respective study coordinators and protocol co-chairs for each site to discuss their findings from the visit, to identify the strengths and

weaknesses of the site, and to identify any corrective action that needs to be taken. This will be documented in a written report to the CIPRA Quality Manager and a formal letter to the site.

10.8 External Site Monitoring

10.8.1 Clinical Site Monitoring and Record Availability

Site monitors under contract to the National Institute of Allergy and Infectious Diseases (NIAID), will visit participating clinical research sites to review the individual participant records, including consent forms, CRFs, supporting data, laboratory specimen records, and medical records (physicians' progress notes, nurses' notes, individuals' hospital charts) to ensure protection of study participants, compliance with the protocol, and accuracy and completeness of records. The monitors will also inspect the sites' regulatory files, to ensure that regulatory requirements are being followed, and site pharmacies, to review product storage, management, and accountability.

10.9 Access to Source Documents

The protocol co-chairs will make study documents (e.g., consent forms, drug distribution forms, CRFs) and pertinent hospital or clinic records readily available for inspection by the local regulatory authorities and IECs, the site monitors, the FDA, the NIAID, the Office for Human Research Protections (OHRP), and the pharmaceutical supporter(s) or their designee for confirmation of the study data.

11.0 SAFETY MONITORING AND REPORTING

The site investigators are responsible for the detection, documentation and reporting of events which are classified as Adverse Events/Expedited/Serious Adverse Events. These events will be reported to the DAIDS through the Regulatory **Support** Center (**RSC**), the South African Medicines Control Council and the relevant Institutional Review Boards/Independent Ethics Committees for each participating site. Furthermore, all adverse events determined to require expedited reporting, must be reported to all other co-sponsors according to stipulations in the relevant contracts. Reports generated to the regulatory entities will be based on the individual criteria required by each entity, i.e., the reportable events and the required timeframe for reporting.

Requirements, definitions and methods for expedited reporting of Adverse Events (AEs) are outlined in Version 2.0, dated January 2010 of the DAIDS EAE Manual, which is available on the RSC website at http://rsc.tech-res.com/safetyandpharmacovigilance/.

The DAERS internet-based reporting system must be used for expedited AE reporting to DAIDS. In the event of system outages or technical difficulties, expedited AEs may be submitted via the CIPRA Safety Desk on the DAIDS EAE Form available at http://rsc.tech-res.com/. For questions about DAERS, please

contact DAIDS-ES at DAIDS-ESSupport@niaid.nih.gov or from within the DAERS application itself.

11.1 Adverse Event Reporting to DAIDS

- **For the period of the study, all Serious Adverse Events (SAEs), as defined by ICH guidelines regardless of relationship to the study agent(s) must be reported in an expedited fashion to DAIDS Regulatory Support Center (RSC) Safety Office as defined in the DAIDS EAE Manual version 2.0.**
- **The SUSAR (Suspected, Unexpected Serious Adverse Reaction) Reporting Category will be used post study. A SAE with onset after exposure to study agent will be reported if the SAE is related to study drug and unexpected.**
- **Assessment of AEs is based on the following characteristics: seriousness, relationship of the AE to the study agent(s), expectedness of the AE, and severity (intensity) of the AE. Assessment of the expectedness of an AE with study agent(s) is performed only for the SUSAR Reporting category.**

  A Serious Adverse Event (SAE) is any untoward medical occurrence that at any dose:
  - **Results in death,**
  - **Is life-threatening,**
  - **Requires inpatient hospitalization or prolongation of existing hospitalization[1],**
  - **Results in persistent or significant disability/incapacity, or**
  - **Is a congenital anomaly/birth defect**

Important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the patient or may require intervention to prevent one of the outcomes listed in the definition above must also be reported in an expedited timeframe to DAIDS. Such determination may be made through medical or scientific judgment [ICH E2A].

For all SAE's submitted to RSC, sites must file an updated SAE report to RSC with the final or stable outcome unless the initial SAE reported already had a final or stable outcome.

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[1] Per ICH SAE definition, hospitalization itself is not an adverse event, but is an outcome of the event. Thus, hospitalization in the absence of an adverse event is not regarded as an AE, and is not subject to expedited reporting.

The following are examples of hospitalization that are not considered to be AEs:

- Protocol-specified admission (e.g. for procedure required by study protocol)
- Admission for treatment of target disease of the study, or for pre-existing condition (unless it is a worsening or increase in frequency of hospital admissions as judged by the clinical investigator)
- Diagnostic admission (e.g. for a work-up of an existing condition such as persistent pretreatment lab abnormality)
- Administrative admission (e.g. for annual physical)
- Social admission (e.g. placement for lack of place to sleep)
- Elective admission (e.g. for elective surgery)
-

The study agents for which relationship assessments are required are:

- **Zidovidine (AZT)**
- **Lamividine (3TC)**
- **Lopinavir/Ritonavir (LPV/r)**
- **Abacavir (ABC)**
- **Didanosine (ddI)**
- **Nevirapine (NVP)**
- **Efavirenz (EFV)**
- **Nelfinavir (NFV)**
- **Stavudine (d4T)**
- **Ritonavir (RTV)**

SAE reporting will not be required for AEs occurring in subjects who have never been exposed to study agents.

Requirements, definitions and methods for expedited reporting of Adverse Events (AEs) are outlined in Version 2.0 dated January 2010 of the DAIDS EAE Manual, which is available on the RSC website at http://rcc.tech-res.com/safetyandpharmacovigilance/.

11.2 Timeframe for Expedited Reporting of Adverse Events

The expedited adverse event reporting will apply for the entire study duration for an individual subject (from study enrolment until study completion or discontinuation of the subject from study participation for any reason).

After the end of the protocol-defined AE reporting period unless otherwise noted, only SUSARs as defined in Version 2.0 of the EAE Manual will be reported if the study site staff becomes aware of the event on a passive basis, i.e., from publicly available information.

Table 10. AE Reporting

	Initial Reports (using DAERS reporting system)	Follow-up Reports	Send to
CIPRA-SA Safety Desk	Within 3 working days	Submit Follow-up Reports when the AE has resolved or when new or more information becomes available that may affect the safety of	Fax: 0866845378 Email: ciprasafety@phru.co.za
DAIDS **RSC** Safety Office	Within 3 working days of site awareness, by no later than 5 p.m. US EST on the third day		Fax:(800) 275-7619 (301) 897-1710 Tel: (301) 897-1709 E-mail:

		participants of the conduct of the trial.	**DAIDSRSCSafetyOffice@tech-res.com**
South Africa Medicines Control Council	15 Days (7 days if fatal or life-threatening)		Tel: (012) 312-0295 Fax: (012) 312-1076
WITS HREC	15 Days (7 days if fatal or life-threatening)		Tel: (011) 274-9200 Fax: (011) 274-9257
US CPT	21 Days (48 Hrs if life-threatening)		Tel: (021) 938-9075 Fax: (021) 931-3352

11.3 Procedures for Expedited Reporting of Adverse Events

Sites using the DAERS internet-based reporting system for submission of SAEs to DAIDS will follow DAERS processes as outlined in DAERS training information. For questions about DAERS, please contact DAIDS-ES at DAIDS-ESSupport@niaid.nih.gov or from within the DAERS application itself. The sites must also notify the CIPRA-SA Safety Desk as soon as possible but within 3 working days of site awareness of the event. The event form can be faxed to 0866845378 or emailed to ciprasafety@phru.co.za. The event will then be reported to the relevant regulatory agencies and the Institutional Review Board/Independent Ethics Committee at the participating sites as per their reporting guidelines.

If the site cannot use DAERS to report an AE on an expedited basis, the AE must be documented on the DAIDS Expedited Adverse Event Reporting Form (EAE Reporting Form) available on the RSC website: http://rsc.tech-res.com/. This report must then be submitted directly to the CIPRA-SA Safety Desk who will report the event as required.

Follow-up reports will be entered on the DAERS reporting system and copied to the CIPRA-SA Safety Desk. These reports will then be forwarded to the relevant regulatory authorities and local IRB/IECs by the CIPRA-SA Safety Desk.

11.4 Additional Adverse Event Reporting Requirements

South African Medicines Control Council

Sites must also adhere to the local regulatory agency, Medicines Control Council (MCC), requirements for adverse event reporting. Reporting to the local regulatory agency will be performed according to the required guidelines, as set out in the 'Reporting of Adverse Drug Reactions in South Africa Guidelines (2.11 ADR Reporting May 03 v1)'. According to these guidelines, Serious Adverse Events are defined as in the ICH E6 GCP guidelines

(defined above). All unexpected Serious Adverse Events, related to study drug that are not fatal or life-threatening, should be reported within 15 days of the site becoming aware of the event (7 days for fatal or life-threatening events).

Institutional Review Boards/Independent Ethics Committees

Sites will also report all reportable events to their relevant local Institutional Review Boards/Independent Ethics Committee. For this study, the Johannesburg site will report AEs to the University of the Witwatersrand (WITS) Human Research Ethics Committee and the Cape Town site will report AEs to the University of Stellenbosch Committee for Pharmaceutical Trials (US CPT).

The Human Research Ethics Committee (HREC) of the University of the Witwatersrand (WITS) Reporting guidelines and requirements for the WITS HREC are in line with the MCC 'Reporting of Adverse Drug Reactions in South Africa Guidelines (2.11 ADR Reporting May03 v1)'. Serious Adverse Events are defined as in the ICH E6 GCP guidelines (defined above) and all unexpected Serious Adverse Events, related to study drug, should be reported within 15 days of the site becoming aware of the event. WITS HREC also requires all deaths to be reported, regardless of relationship.

The University of Stellenbosch Committee for Pharmaceutical Trials (US CPT): Reporting guidelines and requirements for the reporting of Serious Adverse Events are outlined in the updated CPT SOP dated May 2010. According to these guidelines, all significant adverse events occurring at the investigator's site must be reported by the investigator within a maximum of 21 days. However any event which in the opinion of a reasonable and competent investigator, could have serious negative consequences for research participants, research team members, the project as a whole, or the university, should be reported within 48 hours of the investigator becoming aware of the event. Serious adverse drug events are defined as in the FDA Title 21 Part 312.32 and ICH E6 GCP guidelines.

Additionally, all suspected cases of ABC HSR will be collected and reported using the 'Non-GSK ABC HSR CRF Module' within one week of first becoming aware of the event.

11.5 Periodic Summary Adverse Experience Safety Reviews

Monitoring Committee Reviews

Interim monitoring reports will be generated by the study statisticians and data managers for review by the Data Safety Monitoring Board (DSMB). Detailed descriptions of the report contents will be included in the overall Study Monitoring Plan. A full interim review will be performed at least annually.

The DSMB will monitor the data from CIPRA-**SA**. The data to be reviewed will include:

- Screening data
- Baseline data
- Efficacy data
- Safety data
- Quality assurance data
- Accrual status, including projections
- Total number of case report forms that are in-house
- Total number of case report forms that have been quality assured
- Time to milestones
- Safety data from other sources, as it becomes available
- Other data that will help in the assessment of the effectiveness of the clinical trial

In addition, each protocol co-chair will present an overall progress statement for the annual meeting. This written statement will be very brief. This statement should contain an assurance that each protocol co-chair considered the clinical trial's progress and that there is no evidence of toxicity or safety issues which should be addressed by the DSMB.

DSMB members will receive the statistical report for review by overnight mail, three weeks in advance of scheduled meetings. The data in the report will include all data reported to the data analysis center, up to and including 30 days in advance of scheduled meetings. Any deaths and serious and unexpected adverse drug experiences that occur in this 30 day interval will be reported by the protocol co-chairs and investigators directly to the DSMB during the open session. For ad hoc meetings, due to the time constraint, the board may be naïve to the data under review.

Individuals involved in the conduct of the clinical trial (i.e., clinical and medical monitors, statisticians, programmers, data coordinators, clinical investigators and their study staff, participants) will be excluded from the closed session of the annual and ad hoc meetings and will not be apprised of the interim analysis.

After each DSMB meeting, the Chairperson will issue a written report describing all recommendations, including additional safety steps. This report will be

submitted directly to the protocol co-chairs, the CIPRA Program Principal Investigator and the NIAID Program Officer. The FDA adverse drug experience reporting timelines will be utilized to disseminate feedback from the DSMB to the CIPRA Program Principal Investigator, protocol co-chairs and sub-investigators. That is, three days for acute circumstances and ten days for non-acute circumstances. The DSMB's Chairperson, through the PHRU Program Principal Investigator, will issue these directives in writing.

At a minimum, the DSMB will meet on an annual basis. The annual meeting will be held in-person. Any additional meetings may be assembled as an in-person meeting or conducted via teleconference or mail correspondence.

12.0 HUMAN SUBJECTS PROTECTION

All the procedures described here have been reviewed and approved by:

University of the Witwatersrand, Human Research Ethics Committee, Johannesburg, South Africa (Federal Assurance # SF, IORG 000862, IRB 00001223); and

University of Stellenbosch, Committee for Pharmaceutical Trials, Stellenbosch, South Africa (Federal Assurance # SF, IORG 00001263, IRB 00001681).

12.1 Institutional Review Board (IRB)/Independent Ethics Committee (IEC) Review

This protocol and the informed consent documents (Appendix XIII) and any subsequent modifications will be reviewed and approved by the IRB or IEC responsible for oversight of the study. This research will be conducted with human subjects' oversight from the Independent Ethics Committees of the University of the Witwatersrand, Johannesburg and the University of Stellenbosch, Cape Town. Our proposal is developed by local researchers and responds to the need of providing adequate care to children. Research conducted on children living with HIV is a priority of this study team to address all aspects of the HIV epidemic in resource poor settings.

12.2 Protocol Registration

Prior to implementation of this protocol, and any subsequent full version amendments, each site must have the protocol and the protocol informed consent form(s) approved, as appropriate, by their local institutional review board (IRB)/ethics committee (EC) and any other applicable regulatory entity (RE). Upon receiving final approval, sites will submit all required protocol registration documents to the DAIDS Protocol Registration Office (DAIDS PRO) at the Regulatory Support Center (RSC). The DAIDS PRO will review the submitted protocol registration packet to ensure that all of the required documents have been received.

Site-specific informed consent forms (ICFs) WILL be reviewed and approved by the DAIDS PRO and sites will receive an Initial Registration Notification from the DAIDS PRO that indicates successful completion of the protocol registration process. A copy of the Initial Registration Notification should be retained in the site's regulatory files.

Upon receiving final IRB/EC and any other applicable RE approval(s) for an amendment, sites should implement the amendment immediately. Sites are required to submit an amendment registration packet to the DAIDS PRO at the RSC. The DAIDS PRO will review the submitted protocol registration packet to ensure that all the required documents have been received. Site-specific ICF(s) *WILL NOT* be reviewed and approved by the DAIDS PRO and sites will receive an Amendment Registration Notification when the DAIDS PRO receives a complete registration packet. A copy of the Amendment Registration Notification should be retained in the site's regulatory files.

For additional information on the protocol registration process and specific documents required for initial and amendment registrations, refer to the current version of the DAIDS Protocol Registration Manual.

12.3 Informed Consent

A signed consent form will be obtained from at least one parent or legal guardian as the participants are infants and cannot provide consent themselves. Eligible participants will be identified from mothers already diagnosed as HIV-positive. A copy of the consent form will be given to the parent or legal guardian, and this fact will be documented in the participant's record.

The consent form will describe the purpose of the study, the study procedures, potential risks, the right to withdraw consent, compliance information, and a description of the treatment strategy, provision of ART and of post-trial treatment. Before signing, the parent or legal guardian will be questioned for comprehension of the consent form, and will be given the opportunity to ask questions to confirm understanding of the study risks and time commitment involved with participation. Documents will be written in English, Afrikaans, Zulu, Xhosa and Sotho. All translations will undergo an independent translation to verify content.

12.4 Subject Confidentiality

All laboratory specimens, evaluation forms, reports and other records that leave the site will be identified by coded number only to maintain subject confidentiality. Personal identifiers will not be stored in the data set. All records, including study documentation, will be kept locked in a secure location. Study numbers will be stored separately in locked cabinets. All study staff will follow these procedures. Staff involved in cohort retention will not disclose the identity or the nature of participation to the household or the community. Strict security procedures will be followed to ensure confidentiality and security of data. All computer entry and networking programs will be executed with coded numbers only and all computers will be protected by anti-virus software. All data will be backed up on compact discs (CDs) at the end of each day, and these CDs will be kept in locked storage cabinets in the study office. Access to the security password will be given only to the protocol co-chair and appropriate personnel.

Clinical information will not be released without written permission of the participant's parent/legal guardian, except as necessary for monitoring by the IRB/IECs, the MCC, the FDA, the NIAID, the OHRP, or the pharmaceutical supporter(s) or the sponsor's designee.

12.5 Potential Risks and Precautions to Minimize Risks

Potential risks identified include the development of ART complications, such as drug intolerance during continuous therapy or drug resistance during periods of drug interruption or reintroduction, vaccination in the presence of HAART, blood draw associated complications and breach of confidentiality. Both long term and short term side effects of antiretroviral therapy will be described to the participant's parents/legal guardian and monitored by qualified personnel for reporting purposes. There is no evidence that immunization in the presence of HAART is harmful. Laboratory safety blood sampling will include regular hematology and biochemistry testing. Regular clinic visits with study physicians assessing the participants will be undertaken with frequent monitoring for identified study risks. Each child's parent or legal guardian will also be aware of complications and asked to contact the study team, outside the study schedule, should any untoward reactions occur. Participants may also feel discomfort during blood draws or experience bruising at the needle site.

Confidentiality will be protected by restricting participant identifiers at the clinical site and assigning a code to study documents and samples when shipped. Access to computerized personal information will be restricted by password. Key personnel will sign confidentiality agreements and the penalties of disclosure will be stressed. All contact information provided at entry by the parent or legal guardian will be reviewed and consent to contact and preferred method of contact (e.g., confirmation of appointments, etc.) will be noted. All identifiers and records will be kept in locked files within the clinic, in accordance with strict confidentiality guidelines at the clinical site.

12.6 Risk-Benefit Ratio

All participants will benefit from treatment with a potent combination ARV therapy regimen. Participation by study participants will also benefit the South African and international therapy programs by exploring new strategies for treatment that are specifically suited for resource poor settings.

One of the constraints of treatment introduction is the concern about the ability to guarantee life-long access to ART on an individual basis. The provision of five years of therapy in this study is consistent with anticipated benefits of proposed NGO or governmental therapy programs in South Africa. The South African government has made known that the provision of treatment for this program will be for the duration of the study and continued after the study. Alternative local clinical research programs in the area (HPTN 052, ACTG 5175 and MTCT Plus) currently are unable to secure life-long treatment access. The post-trial treatment access will be monitored at the government pilot sites.

12.7 Inclusion of Women and Children

This protocol is targeted to enrolling infants, both male and female, with perinatally acquired HIV infection, between the ages of six and 12 weeks. The majority of children will be recruited through active, well established MTCT prevention programs that identify HIV-positive pregnant women at each of the sites. Participants referred from other sources, such as private practitioners or hospital outpatient departments and wards, will also be eligible for screening for participation. Although the parent/ legal guardian will not receive antiretroviral therapy through this trial, as part of the screening process, mothers will receive information on how to access available resources and about possible participation in other research studies.

12.8 Inclusion of Minority Populations

Minority populations in South Africa include Caucasian and Asian populations. No limitation on the recruitment of these populations has been planned. However, the populations served by the study sites are predominantly black and mixed race. It is anticipated that over 90% of the recruitment will be from black and mixed race populations.

12.9 Study Discontinuation

The study may be discontinued at any time by the IRB/IECs, the FDA, the NIAID, the pharmaceutical supporter(s), the MCC or other government agencies, as part of their duties to ensure that research participants are protected.

13.0 PUBLICATIONS OF RESEARCH FINDINGS

Publication of the results of this trial will be governed by CIPRA-**SA** and DAIDS policies. Any presentation, abstract, or manuscript will be made available for review by the pharmaceutical supporter(s) prior to submission.

14.0 BIOHAZARD CONTAINMENT

As the transmission of HIV and other blood-borne pathogens can occur through contact with contaminated needles, blood and blood products, appropriate blood and secretion precautions will be employed by all personnel in the drawing of blood and shipping and handling of all specimens for this study, as currently recommended by the Centers for Disease Control and Prevention and the National Institutes of Health.

All infectious specimens will be transported using packaging mandated in the Code of Federal Regulations, CDC 42 CFR Part 72. Sites must refer to individual carrier guidelines, e.g., Federal Express, Airborne Express, for specific instructions.

15.0 REFERENCES

Abrams EJ, Wiener J, Carter R, *et al.* Maternal health factors and early pediatric antiretroviral therapy influence the rate of perinatal HIV-1 disease progression in children. AIDS. 2003; 17(6): 867-77.

Actuarial Society of South Africa. AIDS Model. Accessed July 20, 2004. Available URL: www.assa.org.za.

Adderson EE. Antibody repertoires in infants and adults: effects of T-independent and T-dependent immunizations. Springer Semin Immunopathol. 2001; 23: 387-403.

Altfeld M, Rosenberg ES. The role of CD4(+) T helper cells in the cytotoxic T lymphocyte response to HIV-1. Curr Opin Immunol. 2000; 12(4): 375-80.

Altfeld M, Allen TM, Yu XG, *et al.* HIV-1 superinfection despite broad CD8+ T-cell responses containing replication of the primary virus. Nature. 2002; 420(6914): 434-9.

Baba TW, Jeong YS, Pennick D, *et al.* Pathogenicity of live, attenuated SIV after mucosal infection of neonatal macaques. Science. 1995; 267(5205): 1820-5.

Behrman RE, Kliegman RM, Jenson HB. Nelson's Textbook of Pediatrics. 17th ed: Elsevier Science. 2003.

Berrey MM, Schacker T, Collier AC, *et al.* Treatment of primary human immunodeficiency virus type 1 infection with potent antiretroviral therapy reduces frequency of rapid progression to AIDS. J Infect Dis. 2001; 183: 1466-75.

Blanco F, Garcia-Benayas T, Jose de la Cruz J, *et al.* First line therapy and mitochondrial damage: different nucleosides, different findings. HIV Clin Trials. 2003; 4(1): 11-9.

Bobat R, Coovadia H, Coutsoudis A, *et al.* Determinants of mother-to-child transmission of HIV-1 infection in a cohort from Durban, South Africa. Pediatr Infect Dis J. 1996; 15: 604-10.

Borrow P, Lewicki H, Hahn BH, *et al.* Virus-specific CD8+ cytotoxic lymphocyte activity associated with control of viremia during primary human immunodeficiency virus type 1 infection. J Virol. 1994; 68: 6103-10.

Bryson YJ, Luzuriaga K, Sullivan JL, *et al.* Proposed definitions for in utero versus intrapartum transmission of HIV-1. N Engl J Med. 1992; 327(17): 1246-7.

Burman WJ, Jones BE. Treatment of HIV-related tuberculosis in the era of effective antiretroviral therapy. Am J Crit Care Med. 2001; 164: 7-12.

Cao J, McNevin J, Holte S, *et al.* Comprehensive analysis of human immunodeficiency virus type 1 (HIV-1)-specific gamma interferon-secreting CD8+ T cells in primary HIV-1 infection. J Virol. 2003; 77: 6867-78.

CDC (Centers for Disease Control and Prevention). Updated guidelines for the use of rifamycins for the treatment of tuberculosis among HIV-infected patients taking protease inhibitors or non-nucleoside reverse transcriptase inhibitors. 2004. Available URL: http://www.cdc.gov/nchstp/tb/tb_hiv_drugs/toc.htm.

Chadwick EG, Caparelli EV, Yogev R, *et al.* Pharmacokinetics, effacy and safety Lopinavir/Ritonavir for HIV-1-infected Infants less than 6 months of age: 24 week results. AIDS 2008; 22: 249-55

Chaisson RE, Sterling TR, Gallant J. General physical manifestations of human immunodeficiency virus infection. In: Mandell GL, Bennett JE, Dolin R, eds. Principles and practice of infectious diseases. Philadelphia: Churchill Livingstone. 2000: 1404-5.

Compagnucci A, Saidi Y, Chaix ML, *et al* for the PENTA 7 Executive Committee. Difficulties in achieving suppression of viral replication in vertically HIV-1 infected infants early treated with d4T+ddI+NFV: The PENTA 7 Study. 9th Conference on Retroviruses and Opportunistic Infections. February 24-28, 2002. Seattle Poster Number 809.

Cooper D, Gold J, Maclean P, *et al.* Acute AIDS retrovirus infection: definition of a clinical illness associated with acute seroconversion. Lancet. 1985; 1: 537-40.

Cooper ER, Charurat M, Mofenson LM, *et al.* Combination antiretroviral strategies for the treatment of pregnant HIV-1-infected women and prevention of perinatal HIV-1 transmission. J Acquir Immune Defic Syndr. 2002; 29(5): 484-94.

Coovadia HM, Rollins NC, Bland RM, *et al.* Mother-to-child transmission of HIV-1 infection during exclusive breastfeeding in the first 6 months of life: an intervention cohort study. Lancet 2007; 369(9567):1107-16.

Cotton MF, Schaaf HS, Willemsen E, *et al.* The burden of mother to child transmission of HIV-1 disease in a "low" prevalence region, a five year study of hospitalized children. S Afr J Epidemiol Infect. 1998; 13: 46-9.

Coutsoudis AA, Coovadia HB, Pillay K, Kuhn LB. Are HIV-infected women who breastfeed at increased risk of mortality? AIDS. 2001; 15:653-5.

Crespo M, Paradineiro JC, Ribera E, *et al.* A Case of Multiorgan Failure Following Interruption of Antiretroviral Treatment. Eur J Clin Microbiol Infect Dis. 2003.

Dabis F, Leroy V, Bequet L, *et al.* Effectiveness of a short course of zidovudine + nevirapine to prevent mother-to-child transmission (PMTCT) of HIV-1: The Ditrame Plus ANRS 1201 Project in Abidjan, Cote d'Ivoire. In: 14th International AIDS Conference. July 7-12, 2002. Barcelona, Spain: International AIDS Society.

de Martino M, Tovo PA, Galli L, *et al.* Prognostic significance of immunologic changes in 675 infants perinatally exposed to human immunodeficiency virus. The Italian Register for Human Immunodeficiency Virus Infection in Children. J Pediatr. 1991; 119(5): 702-9.

de Martino MD, Tovo PA, Balducci M, *et al.* Reduction in mortality with availability of antiretroviral therapy for children with perinatal HIV-1 infection. JAMA. 2000; 284: 190-7.

de Rossi A. Primary HIV infection in infants: impact of highly active antiretroviral therapy on the natural course. J Biol Regul Homeost Agents. 2002; 16: 53-7.
Department of Health. National HIV seroprevalence survey of women attending public antenatal clinics in South Africa. 2000. Available URL: http://www.doh.gov.za.

Desai N, Mathur M, Weedon J. Lactate levels in children with HIV/AIDS on highly active antiretroviral therapy. AIDS. 2003; 17(10): 1565-8.

Dickover RE, Dillon M, Leung KM, *et al.* Early prognostic indicators in primary perinatal human immunodeficiency virus type 1 infection: importance of viral RNA and the timing of transmission on long-term outcome. J Infect Dis. 1998; 178(2): 375-87.

Eshleman SH, Mracna M, Guay LA, *et al.* Selection and fading of resistance mutations in women and infants receiving nevirapine to prevent HIV-1 vertical transmission (HIVNET 012). AIDS. 2001; 15(15): 1951-7.

Faye A, Bertone C, Teglas JP, *et al.* Early multitherapy including a protease inhibitor for human immunodeficiency virus type 1-infected infants. Pediatr Infect Dis J. 2002; 21: 518-25.

Gibb DM, Dunn DT, Duong T, *et al.* Immunological changes during unplanned treatment interruptions in children with HIV-1 infection. Paediatric Infectious Disease Journal. 2004; May.

Gortmaker SL, Hughes M, Cervia J, *et al.* Effect of combination therapy including protease inhibitors on mortality among children and adolescents infected with HIV-1. N Engl J Med. 2001; 345(21): 1522-8.

Guay LA, Musoke P, Fleming T, *et al.* Intrapartum and neonatal single-dose nevirapine compared with zidovudine for prevention of mother-to-child transmission of HIV-1 in Kampala, Uganda: HIVNET 012 randomised trial. Lancet. 1999; 354(9181): 795-802.

Hel Z, Venzon D, Poudyal M, *et al.* Viremia control following antiretroviral treatment and therapeutic immunization during primary SIV251 infection of macaques. Nat Med. 2000; 6(10): 1140-6.

HIV Paediatric Prognostic Markers Collaborative Study Group (HPPMCS). Short-term risk of disease progression in HIV-1-infected children receiving no antiretroviral therapy or zidovudine monotherapy: a meta-analysis. Lancet. 2003; 362(9396): 1605-11.

Holt PG. Postnatal maturation of immune competence during infancy and childhood. Pediatr Allergy Immunol. 1995; 6: 59-70.

Hussey GD, Reijnhart RM, Sebens AM, *et al.* Survival of children in Cape Town known to be vertically infected with HIV-1. S Afr Med J. 1998; 88: 554-8.

Jeena PM, Pillay P, Pillay T, *et al.* Impact of HIV-1 co-infection on presentation and hospital-related mortality in children with culture proven pulmonary tuberculosis in Durban, South Africa. Int J Tuberc Lung Dis. 2002; 6(8): 672-8.

Kaufmann GR, Cunningham A, Kelleher AD, *et al.* Patterns of viral dynamics during primary human immunodeficiency virus type 1 infection. J Infect Dis. 1998; 178: 1812-5.

Keoshkerian E, Ashton LJ, Smith DG, *et al.* Effector HIV-specific cytotoxic T-lymphocyte activity in long-term nonprogressors: associations with viral replication and progression. J Med Virol. 2003; 71(4): 483-91.

Lallemant M, Jourdain G, Le Coeur S, *et al.* Single-dose perinatal nevirapine plus standard zidovudine to prevent mother-to-child transmission of HIV-1 in Thailand. N Engl J Med. 2004 Jul 15; 351(3):217-28.

La Porte CJL, Colbers EPH, Bertz R, *et al*. Pharmacokinetics of adjusted-dose lopinavir-ritonavir combined with rifampin in healthy volunteers. Antimicrob Agents Chemother 2004; 48:1553–60.

Leonard EG, McComsey GA. Metabolic complications of antiretroviral therapy in children. Pediatr Infect Dis J. 2003; 22(1): 77-84.

Litalien C, Faye A, Compagnucci A, *et al.* Pharmacokinetics of nelfinavir and its active metabolite, hydroxy-tert-butylamide, in infants perinatally infected with human immunodeficiency virus type 1. Pediatr Infect Dis J. 2003; 22(1): 48-55.

Lucas SB, Peacock CS, Hounnou A, *et al.* Disease in children infected with HIV in Abidjan, Côte d'Ivoire. Br Med J. 1996; 312: 335-8.

Luzuriaga K, Sullivan JL. Viral and immunopathogenesis of vertical HIV-1 infection. Pediatr Clin North Am. 2000a; 47: 65-78.

Luzuriaga K, McManus K, Catalina M, *et al.* Early therapy of vertical human immunodeficiency virus type 1 (HIV-1) infection: control of viral replication and absence of persistent HIV-1-specific immune responses. J Virol. 2000b; 74: 6984-91.

Luzuriaga K, McManus M, Mofenson LM, *et al.* A trial of three antiretroviral regimens in HIV-1-infected children. N Engl J Med. 2004; 350(24): 2471-80.

Madhi SA, Huebner RE, Doedens L, *et al.* HIV-1 co-infection in children hospitalised with tuberculosis in South Africa. Int J Tuberc Lung Dis. 2000; 4(5): 448-54.

McIntosh K, Shevitz A, Zaknun D, *et al.* Age- and time-related changes in extracellular viral loads in children vertically infected by human immunodeficiency virus. Pediatr Infec Dis. J 1996; 15: 1087-91.

Meyers TM, Pettifor JM, Gray GE, *et al.* Pediatric admissions with human immunodeficiency virus infection at a regional hospital in Soweto, South Africa. J Trop Pediatr. 2000; Aug 46(4): 224-30.

Mofenson LM, Korelitz J, Meyer III WA, *et al.* The relationship between serum human immunodeficiency virus type 1 (HIV-1) RNA level, CD4 lymphocyte percent, and long-term mortality risk in HIV-1-infected children. J Infect Dis J. 1997; 175: 1029-38.
Mofenson LM, Munderi P. Safety of antiretroviral prophylaxis of perinatal transmission for HIV-infected pregnant women and their infants. J Acquir Immune Defic Syndr. 2002; 30(2): 200-15.

Moodley D, Moodley J, Coovadia H, *et al.* A Multicenter Randomised Controlled Trial of Nevirapine Versus a Combination of Zidovudine and Lamivudine to Reduce Intrapartum and Early Postpartum Mother-to-Child Transmission of Human Immunodeficiency Virus Type 1. J Infect Dis J. 2003; 187: 725-35.

Mukadi YD, Wiktor SZ, Coulibaly IM, *et al.* Impact of HIV infection on the development, clinical presentation, and outcome of tuberculosis among children in Abidjan, Cote d'Ivoire. AIDS. 1997; 11: 1151-8.

Musey L, Hughes J, Schacker T, *et al.* Cytotoxic T cell responses, viral load and disease progression in early human immunodeficiency virus type 1 infection. N Eng J Med. 1997; 337: 1267-74.

National Institutes of Health. Guidelines for the use of antiretroviral agents in pediatric HIV infection. 26 October, 2006. Available URL: http://AIDSinfo.nih.gov.

Nduati R, Richardson BA, John G, *et al.* Effect of breastfeeding on mortality among HIV-1-infected women: a randomised trial. Lancet. 2001; 357: 1651-5.

Newell ML, Peckham C, Dunn DT, *et al.* Natural history of vertically acquired human immunodeficiency virus-1 infection. The European Collaborative Study. Pediatrics. 1994; 94: 815-9.

Newell, M. L. Coovadia, H. Cortina-Borja, *et al*. Mortality of infected and uninfected infants born to HIV-infected mothers in Africa: a pooled analysis Lancet 2004; 364: 1236-43.

Palumbo P, Raskino C, Fiscus S, *et al.* Disease progression in HIV-infected infants and children: predictive value of quantitative plasma HIV RNA and CD4 lymphocyte count. JAMA. 1998; 279: 756-61.

Pediatric European Network for Treatment of AIDS (PENTA). Comparison of dual nucleoside-analogue reverse transcriptase inhibitor regimens with and without nelfinavir in children with HIV-1 who have not previously been treated: the PENTA 5 randomised trial. Lancet. 2002; 359(9308): 733-740.

Pediatric European Network for Treatment of AIDS (PENTA). Accessed September 5, 2004. Available URL: http://www.ctu.mrc.ac.uk/penta/home.htm.

PETRA study team (Perinatal Transmission). The efficacy of three short-course regimens of zidovudine and lamivudine in preventing early and late transmission of HIV-1 from mother to

child in Tanzania, South Africa, and Uganda (PETRA study): a randomised, double-blind, placebo-controlled trial. Lancet. 2002; 359(9308): 1178-86.

Ravel R. Clinical Laboratory Medicine. 6th ed. San Diego: Elsevier; 1994.

Ren Y, Nuttall JJ, Egbers C, *et al*. Effect of Rifampicin on Lopinavir Pharmacokinetics in HIV-Infected Children With Tuberculosis. *J Acquir Immune Defic Syndr* 2008 (e-pub. www.jaids.com/pt/re/jaids/paptoc.htm - 14 Feb 2008).

Richardson BA, Mbori-Ngacha D, Lavreys L, *et al.* Comparison of human immunodeficiency virus type 1 viral loads in Kenyan women, men, and infants during primary and early infection. J Virol. 2003; 77: 7120-3.

Robins G, Shaffer R, Smeaton L, *et al.* Antiretroviral strategies in naive HIV-infected subjects: comparisons of sequential 3-drug regimens (ACTG 384). In: XIV International AIDS Conference. July 7-14, 2002; Barcelona, Spain.

Rosenberg ES, Altfeld M, Poon SH, *et al.* Immune control of HIV-1 after early treatment of acute infection. Nature. 2000; 407(6803): 523-6.

Rosenberg ES, LaRosa L, Flynn T, *et al.* Characterization of HIV-1-specific T-helper cells in acute and chronic infection. Immunol Lett. 1999; 66(1-3): 89-93.

Rosenwirth B, ten Haaft P, Bogers WM, *et al.* Antiretroviral therapy during primary immunodeficiency virus infection can induce persistent suppression of virus load and protection from heterologous challenge in rhesus macaques. J Virol. 2000; 74(4): 1704-11.

Rouet F, Elenga N, Msellati P, *et al.* Primary HIV-1 infection in African children infected through breastfeeding. AIDS. 2002; 16(17): 2303-9.

Roux P, Henley L, Cotton MF, Eley B. Burden and cost for inpatient care for HIV-positive paediatric patients - status in the Cape Town Metropole during the second week of March 1999. S Afr Med J. 2000; 90: 1008-11.

Royston P, Babiker A. A menu-driven facility for complex sample size calculation in randomised controlled trials with a survival or a binary outcome. Stata J. 2002;2:151-163.

Saez-Llorens X, Violari A, Deetz CO, *et al.* Forty-eight-week evaluation of lopinavir/ritonavir, a new protease inhibitor, in human immunodeficiency virus-infected children. Pediatr Infect Dis J. 2003; 22(3): 216-24.

Schacker TW, Hughes JP, Shea T, *et al.* Biological and virologic characteristics of primary HIV infection. Ann Intern Med. 1998; 128: 613-20.

Shearer WT, Quinn TC, LaRussa P, *et al.* Viral load and disease progression in infants infected with human immunodeficiency virus type 1. N Engl J Med. 1997; 336: 1337-42.

Shapiro RL, Holland DT, Capparelli E, *et al.* Antiretroviral concentrations in breast-feeding infants of women in Botswana receiving antiretroviral treatment. J Infect Dis. 2005 Sep 1; 192(5):720-7.

Shapiro RL, Ndung'u T, Lockman S, *et al.* Highly active antiretroviral therapy started during pregnancy or postpartum suppresses HIV-1 RNA, but not DNA, in breast milk. J Infect Dis. 2005 Sep 1; 192(5):713-9.

Stanford University. Stanford HIV drug resistance database [Online database]. 2003. Available URL: http://www.hivdb.stanford.edu.

Staprans SI, Dailey PJ, Rosenthal A, *et al.* Simian immunodeficiency virus disease course is predicted by the extent of virus replication during primary infection. J Virol. 1999; 73(6): 4829-39.

Sterling TR, Vlahov D, Astemborski J, *et al.* Initial plasma HIV-1 RNA levels and progression to AIDS in women and men. N Eng J Med. 2001; 344: 720-5.

Taha TE, Kumwenda NI, Hoover DR, *et al.* Association of HIV-1 load and CD4 lymphocyte count with mortality among untreated African children over one year of age. AIDS. 2000; 14(4): 453-9.

Thior I, Lockman S, Smeaton LM, *et al.* Breastfeeding plus infant zidovudine prophylaxis for 6 months vs formula feeding plus infant zidovudine for 1 month to reduce mother-to-child HIV transmission in Botswana: a randomised trial: the Mashi Study. Jama 2006; 296(7):794-805.

Tietz NW (Ed.). Clinical Guide to Laboratory Tests. 3rd Ed. Philadelphia USA: WB Saunders. 1995.

Tovo PA, de Martino M, Gabiano C, *et al.* Prognostic factors and survival in children with perinatal HIV-1 infection. The Italian Register for HIV Infections in Children. Lancet. 1992; 339(8804): 1249-53.

van Rossum AM, Fraaij PL, de Groot R. Efficacy of highly active antiretroviral therapy in HIV-1 infected children. Lancet Infect Dis. 2002; 2(2): 93-102.

Wilfert CM, Wilson C, Luzuriaga K, *et al.* Pathogenesis of pediatric human immunodeficiency virus type 1 infection. J Infect Dis. 1994; 170(2): 286-92.

Wilson CB, Penix L, Weaver WM, *et al.* Ontogeny of T lymphocyte function in the neonate. Am J Reprod Immunol. 1992; 28(3-4): 132-5.

Wiznia A, Stanley K, Krogstad P, *et al.* Combination nucleoside analog reverse transcriptase inhibitor(s) plus nevirapine, nelfinavir, or ritonavir in stable antiretroviral therapy-experienced HIV-infected children: week 24 results of a randomised controlled trial--PACTG 377. Pediatric AIDS Clinical Trials Group 377 Study Team. AIDS Res Hum Retroviruses. 2000; 16(12): 1113-21.

World Health Organization. Scaling up antiretroviral therapy in resource limited settings: guidelines for a public health approach. 2002. Available URL: http://emro.who.int/ASD/ rapid/ScalingUpAntiretroviralTherapy.pdf.

Worthington MG, Ross JJ. Aseptic meningitis and acute HIV syndrome after interruption of antiretroviral therapy: implications for structured treatment interruptions. 2003. AIDS 17; 2145-2146.

Yogev R, Lee S, Wiznia A, *et al.* Stavudine, nevirapine and ritonavir in stable antiretroviral therapy-experienced children with human immunodeficiency virus infection. Pediatr Infect Dis J. 2002; 21(2): 119-25.

Zwi K, Pettifor JM, Soderlund N. Paediatric hospital admissions at a South Africa urban regional hospital: the impact of HIV, 1992-1997. Ann Trop Paediatr. 1999; Jun 19(2): 135-42.

Zwi K, Pettifor JM, Soderland N, *et al.* HIV infection and in-hospital mortality at an academic hospital in South Africa. Arch Dis Child. 2000; 83(3): 227-30.

- **APPENDICES**

APPENDIX I: SCHEDULES OF EVENTS

APPENDIX IA: SCHEDULE OF EVENTS for Step 1a (delayed therapy), Step 1b (early therapy for 40/96 weeks) and Step 1c (interruption)

☐ Study visit occurs within specified Step ▭ Study visit does **not** occur within specified Step

Weeks on Study	**-2 Pre-Scr**	**-1 Scr**	**0[20] Bsl**	**2[21]**	**4**	**8**	**12**	**16/20**	**24**	**28**	**32**	**36**	**40[22]**	**44**	**48**	**52/56**	**60**	**64/68**	**72**	**76/80**	**84**	**88/92**	**96[22]**	**104**	**Q12 Thereafter**
Arm 1 Step 1a																									**Q12 Until Step 2**
Arm 2 Step 1b																									
Step 1c													**Bsl 0**		**8**		**20**		**32**		**44**		**56**		**Q12 Until Step 2**
Arm 3 Step 1b																									
Step 1c																							**Bsl 0**	**8**	**Q12 Until Step 2**
Screening Informed Consent	x																								
Study Informed Consent/information			x																						
Demographics[1]	x																								
Past Medical History [2]		x																							
Incl/Exclu Criteria		x	x																						
HIV-1 DNA/RNA PCR		x																							
Results of PCR & Counselling [3]		x																							
Quantitative PCR [4]		x					**x**		**x**			**x**			**x**		**x**		**x**		**x**		**x**		**x**
Randomisation [5]			x																						
History & Physical Examination		x	x	x	x	x	x	x	x		x		x		x		x		x		x		x	x	x
Vital Signs [6]		x	x	x	x	x	x		x		x		x		x		x		x		x		x	x	x
T-cell subsets		x	x		x		x		x		x		x		x		x		x		x		x	x	x
Haematology [7]		x	x		x		x		x		x		x		x		x		x		x		x	x	x
Chemistry: LFTs [8]		x	x		x		x		x		x		x		x		x		x		x		x	x	x
Chemistry: urea and electrolytes[9]		x							x						x				x				x		
Weeks on Study	**-2 Pre-Scr**	**-1 Scr**	**0[20] Bsl**	**2[21]**	**4**	**8**	**12**	**16/20**	**24**	**28**	**32**	**36**	**40[22]**	**44**	**48**	**52/56**	**60**	**64/68**	**72**	**76/80**	**84**	**88/92**	**96[22]**	**104**	**Q12 Thereafter**

Arm 1	**Step 1a**																								**Q12 Until Step 2**
Arm 2	**Step 1b**																								
	Step 1c												**Bsl 0**		**8**		**20**		**32**		**44**		**56**		**Q12 Until Step 2**
Arm 3	**Step 1b**																								
	Step 1c																						**Bsl 0**	**8**	**Q12 Until Step 2**
Lipid profile [10]			x						x						x				x				x		
Storage[11]			x	x		x		x		x		x		x		x		x		x		x		x	x
Urinalysis [12]				x					x						x				x				x		**Q24 from wk 96**
Adherence determination [13]					x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x		x
Trial medication dispensing [14]				x		x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x		x
AE check [15]									x		x		x		x		x		x		x				
AE assessment [16]					x	x	x	x		x		x		x		x		x		x		x		x	x
Concomitant therapy				x	x	x	x	x	x	x		x		x		x		x		x		x			x
Stop ARV													ARM 2										ARM 3		
DBS DNA PCR [17]			**x**																						
DBS RNA PCR [18]			x	x		(x)		(x)		(x)		(x)		(x)											
Avidity Index			**x**					x		x		x		x											
Conclusion of study[19]																									Wk 182/ Wk 260
Total blood volumes (mLs)		3.5	9.5	7.5		7.5		8.5		**10**.5		8.5		8.5		**9**.0		7.0		**9**.0		7.0		**9**.0	7.0	**9**.0

(1) When possible, and particularly if there is doubt about the place of residence of a participant, a study nurse or field worker will accompany the participant home and draw a map indicating the place of residence.

(2) Medical history to detect intercurrent illness, HIV disease progression and adverse events.

(3) If the PCR results are positive, the screening consent form allows for safety bloods, a confirmatory PCR and T-cell subsets to be done at this visit. Informed consent for the study will only be taken at the Randomisation Visit.

(4) A quantitative PCR will be used to confirm the diagnosis **and monitored every 12 weeks thereafter for participants on treatment. Confirmatory HIV-RNA value should be obtained at least 1 week and up to 4 weeks from the initial value unless otherwise approved by the protocol team.**

(5) Randomisation can occur from ≥6 weeks to ≤12 weeks of age.

(6) This includes height and weight determinations and body surface area (see Appendix VII).

(7) Haematology samples will be analysed for haemoglobin, hematocrit, mean corpuscular volume, white blood cells, differential (lymphocytes, neutrophils, monocytes, eosinophils) and platelets.

(8) Chemistry samples will be analysed at screening for full liver function tests (LFTs). AST and ALT will be analyzed at all scheduled visits following screening.

(9) Chemistry samples will be analysed at screening and every 6 months thereafter for urea and electrolytes (U&E).

(10) Cholesterol and triglyceride samples will be collected at screening and every 6 months thereafter. This test will be performed in a non-fasting state. However, if results for cholesterol are >300 and triglycerides are >750, a complete fasting-state lipid profile must be drawn. Participants <2 years of age must fast for ≥4 hours, and participants ≥2 years of age must fast for ≥8 hours. Samples will be analyzed for HDL, LDL, triglycerides and total cholesterol.

(11) Recommended schedule for plasma and PBMC or cell pellet storage.

(12) Urinalysis will be analyzed at randomisation and every 6 months thereafter. If urine cannot be collected at a specific time point, it can be deferred sequentially to each next visit until a sample is obtained. Analysis must include urine dipsticks for protein and glucose (leucocytes, nitrites and blood if indicated). If a participant has ≥2+ proteinuria, in absence of UTI, collect an early morning urine sample to test the protein/creatinine ratio to exclude nephropathy.

(13) Adherence will be determined by an Adherence Questionnaire (see Appendix IX) administered by either the study nurse or a counsellor.

(14) Trial medication will be dispensed to Arms 2 and 3 until planned interruption time. For Arm 1, no trial medication until participants develop severe CDC Stage B or Stage C or CD4% falls to <25% or CD4 count <1000 cells/mm^3 regardless of CD4% if <12 months of age; below 20% if 12-**59** months of age*; **below 20% or CD4 count <350cells/mm^3 if ≥ 60 months,** (see Section 3.0).

(15) A study staff member will ask the parent/legal guardian at dispensing visits about the child's health.

(16) The doctor will document the severity and likely relationship of the adverse events to ARVs.

(17) Dried blood spot (DBS) DNA PCR will be collected at screening for **CIPRA ZA 00**5. One hundred (100) HIV-positive and fifty (50) HIV-negative specimens are

needed. Once that number is reached **CIPRA ZA 00**5 staff will notify **CIPRA ZA 002** staff to stop further collection.

(18) Dried blood spot (DBS) RNA PCR will be collected from participants who are not on ARVs, when possible. A total of 375 specimens is needed and once that target is reached **CIPRA ZA 00**5 staff will notify **CIPRA ZA 002** staff to stop collection.

(19) The final visit will occur once the last participant enrolled has been on study for at least 3.5 years (Week 182). The study doctor will see the participants for the final visit.

(20) The time between the Screening Visit (Week -1) and Randomisation (Week 0) should preferably be within 1 week and not exceed 2 weeks. If >2 weeks have passed and the child is still eligible, he/she should be re-screened.

(21) Caregivers will return for further adherence counseling if the participant is on therapy. Caregivers of participants not on therapy will receive general HIV counseling.

(22) The doctor will assess participants at these visits to determine if planned interruption of ART for Arms 2 and 3 can occur.

* If there is discordance between CD4 % and count the investigator may use an absolute CD4 count to reintroduce ART: 12-35 months CD4% <20% or CD4 count <750 cells/ mm^3; **36-59 months** CD4% <20% or CD4 count <500 cells/ mm^3

APPENDIX IB: SCHEDULE OF EVENTS for Step 2 (start or restart first line therapy)

Weeks	**Baseline 0**	**2**	**4**	**8**	**12**	**Monthly After Wk 12**	**Q12 After Wk 12**
History & Physical Examination	x		x		x		x
Vital Signs [(1)]	x		x		x		x
T-cell subsets	x				x		x
Haematology [(2)]	x		x		x		x
Chemistry: LFTs [(3)]	x		x		x		x
Chemistry: Urea and electrolytes [(4)]	x						
Lipid profile [(5)]	x						
Storage [(6)]	x		x		x		x
Urinalysis [(7)]							
Quantitative PCR[(8)]	**x**				**x**		**x**
Adherence determination [(9)]	x	x	x	x	x	x	x
Trial medication dispensing	x		x	x	x	x	x
AE check [(10)]				x		x	
AE assessment [(11)]	x	x	x		x		x
Concomitant therapy	x	x	x		x		x
Conclusion of study [(12)]							
Total blood volumes (mLs)	7.0		7.0		7.0		**9**.0

APPENDIX IB (cont)

(1) This includes height and weight determinations and body surface area (see Appendix VII).
(2) Haematology samples will be analysed for haemoglobin, hematocrit, mean corpuscular volume, white blood cells, differential (lymphocytes, neutrophils, monocytes, eosinophils) and platelets.
(3) Chemistry samples will be analysed at baseline for full liver function tests (LFTs). AST and ALT will be analyzed at all scheduled visits following screening.
(4) Chemistry samples will be analysed at baseline and every 6 months thereafter for urea and electrolytes (U&E).
(5) Cholesterol and triglyceride samples will be collected at baseline and every 6 months thereafter. This test will be performed in a non-fasting state. However, if results for cholesterol are >300 and triglycerides are >750, a complete fasting-state lipid profile must be drawn. Participants <2 years of age must fast for ≥4 hours, and participants ≥2 years of age must fast for ≥8 hours. Samples will be analyzed for HDL, LDL, triglycerides and total cholesterol.
(6) Recommended schedule for plasma and PBMC or cell pellet storage.
(7) Urinalysis will be analyzed at randomisation and every 6 months thereafter. If urine cannot be collected at a specific time point, it can be deferred sequentially to each next visit until a sample is obtained. Analysis will include urine dipsticks for protein and glucose (leucocytes, nitrites and blood if indicated). If a participant has proteinuria of ≥2+, in absence of UTI, collect early morning urine for protein/creatinine ratio to exclude nephropathy.
(8) A quantitative PCR will be taken every 12 weeks from baseline step 2. Confirmatory HIV-RNA value should be obtained at least 1 week and up to 4 weeks from the initial value unless otherwise approved by the protocol team.
(9) Adherence will be determined by an Adherence Questionnaire (see Appendix IX) administered by either the study nurse or a counsellor.
(10) A study staff member will ask the parent/legal guardian at dispensing visits about the child's health.
(11) The doctor will document the severity and likely relationship of the adverse events to ARVs.
(12) The final visit will occur once the last participant enrolled has been on study for at least 3.5 years (Week 182). The study doctor will see the participants for the final visit.

APPENDIX IC: SCHEDULE OF EVENTS for Step 3 (start second line therapy)

Weeks	**Baseline 0**	**2**	**4**	**8**	**12**	**Monthly After Wk 12**	**Q12After Wk 12**
History & Physical Examination	x		x		x		x
Vital Signs [(1)]	x		x		x		x
T-cell subsets	x		x		x		x
Haematology [(2)]	x		x		x		x
Chemistry: LFTs [(3)]	x		x		x		x
Chemistry: Urea and electrolytes[(4)]	x						
Lipid profile [(5)]	x						
Storage[(6)]	x		x		x		x
Urinalysis [(7)]							
Quantitative PCR[(8)]	**x**				**x**		**x**
Adherence determination [(9)]	x	x	x	x	x	x	x
Trial medication dispensing	x		x	x	x	x	x
AE check [(10)]						x	
AE assessment [(11)]	x	x	x	x	x		x
Concomitant therapy	x	x	x	x	x		x
Conclusion of study [(12)]							
Total blood volumes (mLs)	7.0	0.0	7.0		7.0		**9**.0

(1) This includes height and weight determinations and body surface area (see Appendix VII).

(2) Haematology samples will be analysed for haemoglobin, hematocrit, mean corpuscular volume, white blood cells, differential (lymphocytes, neutrophils, monocytes, eosinophils) and platelets.

(3) Chemistry samples will be analysed at screening for full liver function tests (LFTs). AST and ALT will be analyzed at all scheduled visits following baseline.

(4) Chemistry samples will be analysed at baseline and every 6 months thereafter for urea and electrolytes (U&E).

(5) Cholesterol and triglyceride samples will be collected at baseline and every 6 months thereafter. This test will be performed in a non-fasting state. However, if results for cholesterol are >300 and triglycerides are >750, a complete fasting-state lipid profile must be drawn. Participants <2 years of age must fast for ≥4 hours, and participants ≥2 years of age must fast for ≥8 hours. Samples will be analyzed for HDL, LDL, triglycerides and total cholesterol.

(6) Recommended schedule for plasma and PBMC or cell pellet storage.

(7) Urinalysis will be analyzed at randomisation and every 6 months thereafter. If urine cannot be collected at a specific time point, it can be deferred sequentially to each next visit until a sample is obtained. Analysis will include urine dipsticks for protein and glucose (leucocytes, nitrites and blood if indicated). If a participant has proteinuria of ≥2+, in absence of UTI, collect early morning urine for protein/creatinine ratio to exclude nephropathy.

(8) A quantitative PCR will be taken every 12 weeks from baseline step 2. Confirmatory HIV-RNA value should be obtained at least 1 week and up to 4 weeks from the initial value unless otherwise approved by the protocol team.

(9) Adherence will be determined by an Adherence Questionnaire (see Appendix IX) administered by either the study nurse or a counselor.

(10) A study staff member will ask the parent/legal guardian at dispensing visits about the child's health.

(11) The doctor will document the severity and likely relationship of the adverse events to ARVs.

(12) The final visit will occur once the last participant enrolled has been on study for at least 3.5 years (Week 182). The study doctor will see the participants for the final visit.

APPENDIX II: DIVISION OF AIDS TOXICITY TABLE: AGE < 3 MONTHS

DIVISION OF AIDS (DAIDS) TABLE FOR GRADING SEVERITY OF PEDIATRIC (≤ 3 MONTHS OF AGE) ADVERSE EXPERIENCES, APRIL 1994

For other findings, the toxicity table for children > 3 months of age (April, 1994; Appendix III) is applicable. All values are for term newborns. Preterm infants should be judged by a comparison of local normal ranges and the newborn ranges identified here.

PARAMETER	GRADE 1	GRADE 2	GRADE 3	GRADE 4
HEMATOLOGY				
Hemoglobin				
1-7 days old	13.0-14.0	12.0-12.9	<12	Cardiac Failure secondary to anemia
8-21 days old	12.0-13.0	10.0-11.9	<10.0	Cardiac Failure secondary to anemia
22-35 days old	9.5-10.5	8.0-9.4	<8.0	Cardiac Failure secondary to anemia
36-56 days old	8.5-9.4	7.0-8.4	<7.0	Cardiac Failure secondary to anemia
57-90 days old	9.0-9.9	7.0-8.9	<7.0	Cardiac Failure secondary to anemia
Absolute Neutrophil Count				
1 day old	5000-7000	3000-4999	1500-2999	<1500
2-7 days old	1750-2500	1250-1749	750-1249	<750
8-56 days old	1200-1800	900-1199	500-899	<500
57-90 days old	750-1200	400-749	250-399	<250
Bilirubin				
<7 days old		20-25	26-30	>30
7-60 days old	1.0-1.9xN	2.0-2.9xN	3.0-7.5xN	>7.5xN
61-90 days old	1.0-1.9xN	2.0-2.9xN	3.0-7.5xN	>7.5xN
Creatinine				
<7 days old	1.0-1.7	1.8-2.4	2.5-3.0	>3.0
7-60 days old	0.5-0.9	1.0-1.4	1.5-2.0	>2.0
61-90 days old	0.6-0.8	0.9-1.1	1.2-1.5	>1.5
Creatinine Clearance				
<7 days old	35-40	30-34	25-29	<25
7-60 days old	45-50	40-44	35-39	<35
61-90 days old	60-75	50-59	35-49	<35
Low Calcium				
<7 days old	6.5-6.9	6.0-6.4	5.5-5.9	<5.5
7-60 days old	7.6-8.0	7.0-7.5	6.0-6.9	<6.0
61-90 days old	7.8-8.4	7.0-7.7	6.0-6.9	<6.0
High Calcium				
<7 days old	12.0-12.4	12.5-12.9	13.0-13.5	>13.5
7-60 days old	10.5-11.2	11.3-11.9	12.0-13.0	>13.0
61-90 days old	10.5-11.2	11.3-11.9	12.0-12.9	≥ 13.0

APPENDIX III: DIVISION OF AIDS TOXICITY TABLE: AGE > 3 MONTHS

DIVISION OF AIDS (DAIDS) TABLE FOR GRADING SEVERITY OF PEDIATRIC (> 3 MONTHS OF AGE) ADVERSE EXPERIENCES, APRIL 1994

PARAMETER	GRADE 1	GRADE 2	GRADE 3	GRADE 4
HEMATOLOGY				
Hemoglobin > 3 mo.- < 2 y.o.	9.0-9.9 g/dL	7.0-8.9 g/dL	<7.0 g/dL	Cardiac Failure secondary to anemia
Hemoglobin ≥ 2 y.o.	10-10.9 g/dL	7.0-9.9 g/dL	<7.0 g/dL	Cardiac Failure secondary to anemia
Abs Neutrophil Ct/µL	750-1200	400-749	250-399	<250
Platelets/µL		50,000-75,000	25,000-49,999	<25,000 or bleeding
PT (seconds)	1.1-1.25 x N	1.26-1.5 x N	1.51-3.0 x N	>3 x N
PTT (seconds)	1.1-1.66 x N	1.67-2.33 x N	2.34-3.0 x N	>3 x N
GASTROINTESTINAL				
Bilirubin	1.1-1.9 x N mg/dL	2.0-2.9 x N mg/dL	3.0-7.5 x N mg/dL	>7.5 x N mg/dL
AST (SGOT)	1.1-4.9 x N U/L	5.0-9.9 x N U/L	10.0-15.0 x N U/L	>15.0 x N U/L
ALT (SGPT)	1.1-4.9 x N U/L	5.0-9.9 x N U/L	10.0-15.0 x N U/L	>15.0 x N U/L
GGT	1.1-4.9 x N U/L	5.0-9.9 x N U/L	10.0-15.0 x N U/L	>15.0 x N U/L
Pancreatic Amylase	1.1-1.4 x N U/L	1.5-1.9 x N U/L	2.0-3.0 x N U/L	>3.0 x N U/L
Total Amylase + Lipase*	1.1-1.4 x N U/L	1.5-2.4 x N U/L	2.5-5.0 x N U/L	>5.0 x N U/L
Uric Acid	7.5-9.9 mg/dL	10-12.4 mg/dL	12.5-15.0 mg/dL	>15.0 or Gout mg/dL
CPK	See Neuromuscular Toxicity			
Abdominal Pain	Mild	Moderate- No Rx Needed	Moderate- Rx Needed	Severe- Hospital and Rx
Diarrhea	Soft stools	Liquid stools	Liquid Stools & Mild Dehydration Bloody stools	Dehydration requiring IV therapy or Hypotensive Shock
Constipation	Mild	Moderate	Severe	Distention and Vomiting
Nausea	Mild	Moderate- Decreased po intake	Severe-Little po intake	Unable to ingest food or fluid for >24 hours
Vomiting	<1 episode/day	1-3 episodes/day or duration >3d	>3 episodes/day or duration >7d	Intractable Vomiting

Comments:

*Both amylase and lipase must be elevated to the same Grade or higher (i.e. if total amylase is Grade 4, but lipase is only Grade 1, the Toxicity Grade is 1. In pediatric HIV patients, the most common source of serum amylase is the salivary glands. Salivary amylase elevations are generally not clinically significant. When amylase is released from damaged pancreatic cells, it can be a marker of pancreatitis. In most cases of clinical pancreatitis, lipase will also be elevated. However, lipase is also a non-specific marker. Combined elevation of amylase and lipase (each >5 x normal) often indicates pancreatic disease and requires evaluation. However, in the absence of pancreatic disease, drug can be resumed even at Grade 3 and 4 toxicities.

APPENDIX III (cont.)

PARAMETER	GRADE 1	GRADE 2	GRADE 3	GRADE 4
RENAL AND ELECTROLYTES				
CREATININE*				
2 Month-2 Years	0.6-0.8 mg/dL	0.9-1.1 mg/dL	1.2-1.5 mg/dL	>1.5 mg/dL
2 Years-Adolescent	0.7-1.0 mg/dL	1.1-1.6 mg/dL	1.7-2.0 mg/dL	>2.0 mg/dL
Adolescents	1.0-1.7 mg/dL	1.8-2.4 mg/dL	2.5-3.5 mg/dL	>3.5 mg/dL
Creatinine Clearance	60-75 cc/min/1.73 m^2	50-59 cc/min/1.73 m^2	35-49 cc/min/1.73 m^2	<35 cc/min/1.73 m^2
ELECTROLYTES				
High Sodium (mmo/L)	145-149		150-155	>155 or mental status changes
Low Sodium (mmo/L)	130-135		124-129	<124 or mental status changes
High Potassium (mmo/L)	5.0-5.9	6.0-6.4	6.5-7.0	>7.0 or Cardiac arrhythmias
Low Potassium (mmo/L)	3.0-3.5	2.5-2.9	2.0-2.4	<2.0
High Calcium	10.5-11.2 mg/dL	11.3-11.9 mg/dL	12.0-12.9 mg/dL	≥13.0 mg/dL
Low Calcium	7.8-8.4 mg/dL	7.0-7.7 mg/dL	6.0-6.9 mg/dL	<6.0 mg/dL
Low Magnesium (mg/dL)	1.2-1.4	0.9-1.1	0.6-0.8	<0.6 or Cardiac arrhythmias
Hypoglycemia (mg/dL)	55-65	40-54	30-39	<30 or Mental status changes
Hyperglycemia (mg/dL)	116-159	160-249	250-400	>400 or Ketoacidosis
Proteinuria (mg/dL)	Tr-1+ <150 mg/day	2+ 150-499 mg/day	3+ 500-1000 mg/day	4+, or nephrotic syndrome >1000 mg/day
Hematuria	Microscopic <25 cells/hpf	Microscopic ≥25 cells/hpf	Gross	Obstruction or Transfusion requirement
Comments: Calcium values are corrected for albumin concentration. CrCl values do not apply to infants <2 months old.				
OTHER				
Allergy	Pruritis without Rash	Pruritic Rash	Mild Urticaria	Severe Urticaria Anaphylaxis, Angiedema
Drug Fever (Rectal)		38.5-40 °C	>40 °C	Sustained Fever: >40 °C, >5 days
Cutaneous		Diffuse maculo-papular rash, dry desquamation	Vesiculation, ulcers	Exfoliative dermatitis, Stevens-Johnson or Erythema multiforme, Moist desquamation
Stomatitis	Mild discomfort	Painful, difficulty swallowing, but able to eat and drink	Painful: unable to swallow solids	Painful: requires IV fluids

***Conversion factor for Creatinine mg/dL X 76.3 = μmol/L**

APPENDIX III (cont.)

SYMPTOM	GRADE 1	GRADE 2	GRADE 3	GRADE 4
CENTRAL NERVOUS SYSTEM				
Seizures	None	1 Uncomplicated seizure +/- Temp Elevation	1 seizure /Month for ≥2 Consecutive Months Or 3 seizure over 6 Months; No Temp Elevation	>1 seizure /Month; No Temp Elevation; No Decrease in seizure Frequency Despite dose reduction
Seizures are a ubiquitous symptom of numerous systemic or CNS disturbances; alternative explanations should be vigorously sought and eliminated. Status epilepticus represents a severe end of the seizure spectrum, but should be considered as a single seizure event. The need for chronic or acute anticonvulsant medication should be made on a clinical basis. Seizures as a manifestation of drug toxicity are usually primarily generalised. Focal (partial onset) seizures are suggestive of focal central nervous system pathology and should be appropriately investigated, although they may be a manifestation of drug toxicity. Beware of focal seizures, which secondarily generalise; these should be approached diagnostically as partial onset seizures. Children with underlying epileptic conditions who experience persistent breakthrough seizures despite maximal anticonvulsant therapy coincident with beginning the trial medication should be considered Grade 4.				
Headache	≤1/Month <2 Hrs duration Mild	>1/Month >2 Hrs Duration Moderate to Severe Responds to non-narcotic analgesia or prophylaxis	>2/Month >2 Hrs Duration Moderate to Severe Responds to narcotic analgesia, or does not respond to prophylaxis	>4/Month; >2 Hrs Duration; Moderate to Severe; Non-Responsive to narcotic Analgesia; or persistently Recurrent despite prophylaxis No decrease in frequency or Severity despite dose reduction
Headache is a non-specific symptom, but may be a symptom of CNS/intracranial pathology. Appropriate diagnostic measures should be pursued. Duration refers to the waxing and peak phases, not to the resolution/waning phases of the headache. Mild refers to a grade of headache pain, which does not affect function or activity. Moderate to severe refers to a grade of headache, which affects function or activity.				
Mental Status And Behaviour	Changes which do not Affect Function	Changes requiring pharmacological or other therapy; or mild lethargy, sedation or somnolence which resolves with rest	Changes not improved by drugs or other therapies; or onset of confusion, memory impairment, lethargy, sedation, or somnolence which does not respond to rest	Onset of delirium, obtundation, coma, or psychosis, or Grade 3 toxicity which does not respond to dose reduction
Behaviour refers to the development of attention deficits with or without hyperactivity, depression, mania, agitation, sleep disorders, phobias, obsessive-compulsive behaviours, or anxiety. Mental status refers to the level of consciousness, memory function, language and analytical operations, and non-dominant hemisphere functioning. Alternative explanations should be sought.				
Balance & Posture	None	None	Ataxia, dizziness, vertigo, tremor, impaired postural balance	Onset of movement disorder; or Grade 3 toxicity which does not respond to dosage adjustment
"Ataxia" can be mistakenly diagnosed in the face of central weakness or peripheral neuropathy, which should not be considered a drug toxicity of this category. Movement disorders refer to tardive or other dyskinesias, dystonias, chorea, or ballismus. Alternative explanations should be sought.				

APPENDIX III (cont.)

SYMPTOM	GRADE 1	GRADE 2	GRADE 3	GRADE 4
Visual	None	Blurring, diplopia, or horizontal nystagmus of < 1 hour duration, with spontaneous resolution	> = 1 episode of Grade 2 symptoms per week, or an episode of Grade 2 seizure lasting 1 hour with spontaneous resolution by 4 hours or vertical nystagmus	Decrease in visual acuity, visual field deficit, or oculogyric crisis, or Grade 3 seizure which persist after dose reduction
Many of the symptoms in this category can be the result of CNS pathology, or alternatively can be an external (i.e., non-CNS) neuro-ophthalmologic disorder. Appropriate diagnostic investigations should be pursued.				
Myelopathy	None	None	None	Myelopathic/spinal cord symptoms, such as: Pyramidal tract weakness and disinhibition, sensory level, loss of proprioception, bladder/bowel dysfunction
HIV can cause spinal cord syndromes rarely in children. Other infectious agents can cause myelopathies as well. Alternative explanations should be sought.				
PERIPHERAL NERVOUS SYSTEM				
Neuropathy/ Lower Motor Neuronopathy	None	Mild transient Paresthesia only	Persistent or progressive paresthesias, burning sensation in feet, or mild dysesthesia; no weakness; mild to moderate deep tendon reflex changes; no sensory loss	Onset of significant weakness, decrease or loss of DTRs, sensory loss in "stocking glove" distribution, radicular sensory loss, multiple cranial nerve involvement; bladder or bowel dysfunction, fasciculations, respiratory embarrassment from chest wall weakness. Grade 3 symptoms which do not resolve with dose reduction
Infectious agents other than HIV can precipitate a neuropathy and should be considered, especially CMV. Neuropathies, which do not resolve after dose reduction or discontinuation, should be pursued for alternative infectious or non-infectious etiologies, since drug-related neuropathies will usually resolve after dose reduction or drug discontinuation. It should be borne in mind that many participants will worsen for up to one month after drug discontinuation prior to improvement ("coasting"). Abnormalities should be confirmed by nerve conduction studies (NCS) +/- electromyographic studies (EMG).				

APPENDIX III (cont.)

SYMPTOM	GRADE 1	GRADE 2	GRADE 3	GRADE 4
Myopathy or Neuromuscular Junction Impairment	Normal or mild (<2 x N) CPK elevation	Mild proximal weakness and/or atrophy not affecting gross motor function. Mild myalgias, +/- mild CPK elevation (<2 x N)	Proximal muscle weakness and/or atrophy affecting motor function +/- CPK elevation; or severe myalgias with CPK >2 x N; Consider confirmatory EMG and/or muscle bx	Onset of myasthenia-like symptoms (fatigable weakness with external, variable ophthalmoplegia and/or ptosis), or neuromuscular junction blockade (acute paralysis) symptoms (confirm with EMG); or Grade 3 symptoms which do not resolve on dose adjustment; confirm with muscle bx
HIV can produce a myopathy, and should be differentiated. Drug-induced myopathy can be accompanied by normal CPK levels. On occasion, neuropathic or central weakness can mimic myopathic weakness.				
Clinical symptoms *not otherwise specified* in this table	No therapy; monitor condition	May require minimal intervention and monitoring	Requires medical care and possible hospitalization	Requires active medical intervention, hospitalization, or hospice care
Laboratory values *not otherwise specified* in this table	Abnormal, but requiring no immediate intervention; follow	Sufficiently abnormal to require evaluation as to causality and perhaps mild therapeutic intervention, but not of sufficient severity to warrant immediate changes in study drug	Sufficiently severe to require evaluation and treatment, including at least temporary suspension of study drug	Life-threatening severity. Requires immediate evaluation, treatment, and usually hospitalization. Study drug must be stopped immediately and should not be restarted until the abnormality is clearly felt to be caused by some other mechanism that study drug.

APPENDIX IIIA: UNIT OF MEASUREMENT CONVERSION FOR BLOOD GLUCOSE

The units of measurement for blood glucose will be reported in mmol/L in South Africa. The following table provides the conversion to mmol/L.

PARAMETER	GRADE 1	GRADE 2	GRADE 3	GRADE 4
Hypoglycemia	3.0-3.6 mmol/L	2.2-2.97 mmol/L	1.65-2.15 mmol/L	
Hyperglycemia	6.38-8.7 mmol/L	8.8-13.6 mmol/L	13.7-22 mmol/L	>22 mmol/L or Ketoacidosis

Conversion Factor: mg/dL X 0.055 = mmol/L

References:

Tietz N, *et al.* Textbook of Clinical Chemistry. 2nd ed. WB Saunders. 1994.

APPENDIX IIIB: SUPPLEMENTAL TABLE FOR GRADING CHOLESTEROL AND TRIGLYCERIDES

PARAMETER	GRADE 1	GRADE 2	GRADE 3	GRADE 4
Cholesterol (fasting)	200-239 mg/dL 5.18-6.19 mmol/L	240-300 mg/dL >6.19-7.77 mmol/L	301-400 mg/dL >7.77-10.36 mmol/L	NA
LDL (fasting)	130-<160 mg/dL 3.37-<4.14 mmol/L	160-<190 mg/dL 4.14-<4.92 mmol/L	>190 mg/dL 4.92 mmol/L	NA
Triglycerides (fasting)	–	500-750 mg/dL 5.65-8.48 mmol/L	751-1200 mg/dL 8.49-13.56 mmol/L	>1200 mg/dL >13.56 mmol/L

The units of measurement for cholesterol and blood triglycerides in South Africa will be reported in mmol/L. The following formula should be used for converting mmol/L to mg/dL:

Conversion factor for cholesterol: mmol X 38.67 = mg/dL.

Conversion factor for triglycerides: mmol X 88.57 = mg/dL.

APPENDIX IV: SUPPLEMENTAL TOXICITY TABLE FOR GRADING SEVERITY OF ADULT AND PEDIATRIC CUTANEOUS/SKIN RASH/ DERMATITIS ADVERSE EXPERIENCES

GRADE 1	GRADE 2	GRADE 3	GRADE 4
CUTANEOUS/SKIN RASH/DERMATITIS			
Erythaema, with or without pruritis	A. Diffuse erythaematous macular or maculopapular cutaneous eruption or dry desquamation with or without pruritis (without the presence of any additional constitutional findings as described for Grade 3); OR typical target lesions without blistering, vesicles, or ulcerations in the lesions. B. Urticaria	A. Diffuse erythaematous macular or maculopapular cutaneous eruption or moist desquamation with or without pruritis together with any of the following constitutional findings considered related to study drug: 1. 5 x ULN AST, ALT or 2 x baseline if baseline > ULN. 2. fever, >39^{o}C 3. blistering and/or vesiculation of cutaneous eruptions 4. any site of mucosal lesions; OR B. angioedema; OR C. exfoliative dermatitis defined as severe widespread erythaema and dry scaling of the skin, with generalised superficial lymphadenopathy, and with other constitutional findings such as fever, weight loss, hypoproteinemia possibly related to study drug; OR D. diffuse rash and serum sickness-like reactions defined as a clinical symptom complex manifested as fever, lymphadenopathy, edema, myalgia, and/or arthralgia; OR E. diffuse cutaneous eruptions, usually starting on the face, trunk or back, often with prodromal symptoms plus one of the following: 1. cutaneous bullae, sometimes confluent with widespread sheet-like detachment of skin (<10% body surface area), (Nikolski's sign)(Stevens Johnson Syndrome, SJS) 2. two or more anatomically distinct sites of mucosal erosion or ulceration not due to another cause.	Diffuse cutaneous eruptions, usually starting on the face, trunk or back, often with prodromal symptoms plus cutaneous bullae with widespread sheet-like detachment of skin (>10% of body surface area), (Nikolski's sign), (SJS/Toxic Epidermal Necrolysis (TEN) overlap syndrome; TEN)

APPENDIX V: CDC PEDIATRIC HIV CLASSIFICATION

CDC 1994 REVISED CLASSIFICATION SYSTEM FOR HIV-INFECTION IN CHILDREN LESS THAN 13 YEARS OF AGE. 1994

TABLE 1. Pediatric human immunodeficiency virus (HIV) classification*

	Clinical Categories			
Immunological Categories	N: No signs/ symptoms	A: Mild signs/ Symptoms	B:** Moderate signs/symptoms	C:** Severe signs/symptoms
1: No evidence of suppression	N1	A1	B1	C1
2: Evidence of moderate suppression	N2	A2	B2	C2
3: Evidence of severe suppression	N3	A3	B3	C3

* Children whose HIV infection status is not confirmed are classified by using the above grid with a letter E (for perinatally exposed) placed before the appropriate classification code (e.g., EN2).

** Both Category C and lymphoid interstitial pneumonitis in Category B are reportable to state and local health departments as acquired immunodeficiency syndrome.

TABLE 2. Immunological categories based on age-specific CD4+ T-lymphocyte Counts (L) and percent of total lymphocytes

	Age of Child		
Immunological	< 12 months	1-5 years	6-12 years
Category	L (%)	L (%)	L (%)
1: No evidence of suppression	≥ 1,500 (≥ 25)	≥ 1,000 (≥ 25)	≥ 500 (≥ 25)
2: Evidence of moderate suppression	750-1,499 (15-24)	500-999 (15-24)	200-499 (15-24)
3: Evidence of severe suppression	< 750 (< 15)	< 500 (< 15)	< 200 (< 15)

APPENDIX V (cont.)

Box 2. Clinical Categories for Children with Human Immunodeficiency Virus (HIV) Infection

CATEGORY N: NOT SYMPTOMATIC

Children who have no signs or symptoms considered to be the result of HIV infection or who have only one of the conditions listed in Category A.

CATEGORY A: MILDLY SYMPTOMATIC

Children with two or more of the conditions listed below but none of the conditions listed in Categories B and C.

- Lymphadenopathy ($\geq$ 0.5 cm at more than two sites; bilateral = one site)
- Hepatomegaly
- Splenomegaly
- Dermatitis
- Parotitis
- Recurrent or persistent upper respiratory infection, sinusitis, or otitis media

CATEGORY B: MODERATELY SYMPTOMATIC

Children who have symptomatic conditions, other than those listed for Category A or C that are attributed to HIV infection. Examples of conditions in clinical Category B include, but are not limited to:

- Anemia (< 8 gm/dl), neutropenia (<1,000/mm3) or thrombocytopenia (<100,000/mm3) persisting $\geq$30 days
- Bacterial meningitis, pneumonia, or sepsis (single episode)
- Candidiasis, oropharyngeal (thrush), persisting (>2 months) in children >6 months of age
- Cardiomyopathy
- Cytomegalovirus infection, with onset before 1 month of age
- Diarrhea, recurrent or chronic
- Hepatitis
- Herpes simplex virus (HSV) stomatitis, recurrent (more than two episodes within 1 year)
- HSV bronchitis, pneumonitis, or esophagitis with onset before 1 month of age
- Herpes zoster (shingles) involving at least two distinct episodes or more than one dermatome

- Leiomyosarcoma
- Lymphoid interstitial pneumonia (LIP) or pulmonary lymphoid hyperplasia complex
- Nephropathy
- Nocardiosis
- Persistent fever (lasting >1 month)
- Toxoplasmosis, onset before 1 month of age
- Varicella, disseminated (complicated chickenpox)

CATEGORY C: SEVERELY SYMPTOMATIC

Children who have any condition listed in the 1987 surveillance case definition for acquired immunodeficiency syndrome, with the exception of LIP (Box 3).

Box 3. Conditions Included in Clinical Category C for Children Infected with Human Immunodeficiency Virus (HIV)

CATEGORY C: SEVERELY SYMPTOMATIC*

Serious bacterial infections, multiple or recurrent (i.e., any combination of at least two culture-confirmed infections within a 2-year period), of the following types:

Septicemia, pneumonia, meningitis, bone or joint infection, or abscess of an internal organ or body cavity (excluding otitis media, superficial skin or mucosal abscesses, and indwelling catheter-related infections)

- Candidiasis, esophageal or pulmonary (bronchi, trachea, lungs)
- Coccidiomycosis, disseminated (at site other than or in addition to lungs or cervical or hilar lymph nodes)
- Cryptococcosis, extrapulmonary
- Cryptosporidiosis or isosporiasis with diarrhea persisting >1 month
- Cytomegalovirus disease with onset of symptoms at age >1 month (at a site other than liver, spleen, or lymph nodes)
- Encephalopathy (at least one of the following progressive findings present for at least 2 months in the absence of a concurrent illness other than HIV infection that could explain the findings:
  - A) failure to attain or loss of developmental milestones or loss of intellectual ability, verified by standard developmental scale or neuropsychological tests;
  - B) impaired brain growth or acquired microcephaly demonstrated by head circumference measurements or brain atrophy demonstrated by computerised tomography or magnetic resonance imaging (serial imaging is required for children < 2 years of age);
  - C) acquired symmetric motor deficit manifested by two or more of the following: Paresis, pathologic reflexes, ataxia, or gait disturbance

- Herpes simplex virus infection causing a mucocutaneous ulcer that persists for >1 month; or bronchitis, pneumonitis, or esophagitis for any duration affecting a child >1 month of age
- Histoplasmosis, disseminated (at a site other than or in addition to cervical or hilar lymph nodes)
- Kaposi's sarcoma
- Lymphoma, primary, in brain
- Lymphoma, small, noncleaved cell (Burkitt's), or immunoblastic or large cell Lymphoma of B-cell or unknown immunological phenotype
- *Mycobacterium tuberculosis*, disseminated or extrapulmonary
- *Mycobacterium*, other species or unidentified species, disseminated (at a site other than or in addition to lungs, skin, or cervical or hilar lymph nodes)
- *Mycobacterium avium* complex or Mycobacterium kansasii, disseminated (at site other than or in addition to lungs, skin, or cervical or hilar lymph nodes)
- *Pneumocystis carinii* pneumonia
- Progressive multifocal leukoencephalopathy
- Salmonella (nontyphoid) septicemia, recurrent
- Toxoplasmosis of the brain with onset at >1 month of age
- Wasting syndrome in the absence of a concurrent illness other than HIV infection that could explain the following findings:

  A) persistent weight loss >10% of baseline

  OR

  B) downward crossing of at least two of the following percentile lines on the Weight-for-age chart (e.g., 95th, 75th, 50th, 25th, 5th) in a child ≥1 year of age

  OR

  C) <5th percentile on weight-for-height chart on two consecutive measurements, ≥30 days apart **PLUS**

  a) chronic diarrhea (i.e., at least two loose stools per day for ≥30 days)

  OR

  b) documented fever (for ≥30 days, intermittent or constant)

Immunological Categories

The three immunological categories (Table 2) were established to categorize children by the severity of immunosuppression attributable to HIV infection. CD4+ T-lymphocyte depletion is a major consequence of HIV infection and is responsible for many of the severe manifestations of HIV infection in adults. For this reason, CD4+ counts are used in the adult HIV classification system. However, several findings complicate the use of CD4+ counts for assessing immunosuppression resulting from HIV infection in children. Normal CD4+ counts are higher in infants and young children than in adults and decline over the first few years of life. In addition, children may develop opportunistic infections

APPENDIX V (cont.)

at higher CD4+ levels than adults. Although insufficient data exist to correlate CD4+ levels with disease progression at all age groups, low age-specific CD4+ counts appear to correlate with conditions associated with immunosuppression in children. Therefore, despite these complications, classification based on age-specific CD4+ levels appears to be useful for describing the immunological status of HIV-infected children.

Fewer data are available on age-specific values for CD4+ T-lymphocyte percent of total lymphocytes than for absolute counts. However, the CD4+ T-lymphocyte percent has less measurement variability than the absolute count. To establish the age-specific values of CD4+ percent that correlate with the CD4+ count thresholds, CDC compiled data from selected clinical projects in the United States and Europe. The data included >9,000 CD4+ counts, with the corresponding CD4+ percent determinations, from both HIV-infected and uninfected children <13 years of age. Nonparametric regression modelling was used to establish the CD4+ percent boundaries that best correlated with the CD4+ count boundaries in the classification system.

The immunological category classification (Table 2) is based on either the CD4+ T-lymphocyte count or percent of total lymphocytes. If the CD4+ count and the CD4+ percent indicate different classification categories, the child should be classified into the more severe category. Repeated or follow-up CD4+ values that result in a change in classification should be confirmed by a second determination. Values thought to be in error should not be used. A child should not be reclassified to a less severe category regardless of subsequent CD4+ determinations.

Clinical Categories

Children infected with HIV or perinatally exposed to HIV may be classified into one of four mutually exclusive clinical categories based on signs, symptoms, or diagnoses related to HIV infection (Box 2). As with the immunological categories, the clinical categories have been defined to provide a staging classification (e.g., the prognosis for children in the second category would be less favourable than for those in the first category).

Category N, not symptomatic, includes children with no signs or symptoms considered to be the result of HIV infection or with only one of the conditions listed in Category A, mildly symptomatic. Category N was separated from Category A partly because of the substantial amount of time that can elapse before a child manifests the signs or symptoms defined in Category B, moderately symptomatic. Also, more staging information can be obtained during this early stage of disease by separating Categories N and A. In addition, for children who have uncertain HIV-infection status (prefix E), Categories N and A may help to distinguish those children who are more likely to be infected with HIV (i.e., children in Category EA may be more likely to be infected than children in Category EN).

Category B includes all children with signs and symptoms thought to be caused by HIV infection but not specifically outlined under Category A or Category C (severely

symptomatic). The conditions listed in Box 2 are examples only; any other HIV-related condition not included in Category A or C should be included in Category B. Anemia, thrombocytopenia, and lymphopenia have defined thresholds in the new classification system.

Category C includes all AIDS-defining conditions except lymphoid interstitial pneumonitis (LIP) (Box 3). Several reports indicate that the prognosis for children with LIP is substantially better than that for children who have other AIDS-defining conditions. Thus, LIP has been separated from the other AIDS-defining conditions in Category C and placed in Category B.

Signs and symptoms related to causes other than HIV infection (e.g., inflammatory or drug-related causes) should not be used to classify children. For example, a child with drug-related hepatitis or anemia should not be classified in Category B solely because these conditions may be associated with HIV infection. In contrast, a child with anemia or hepatitis should be classified in Category B when the condition is related to HIV infection. The criteria for diagnosing some conditions and determining whether a child's signs, symptoms, or diagnoses are related to HIV infection may not be clear in all cases, and therefore, may require judgment of the clinicians and researchers using the classification system.

(Source: CDC. *Revised classification system for HIV infection in children less than 13 years of age. MMWR.* 1994; 43(RR-12): 2-6.)

APPENDIX VI: DEFINITIONS OF SEVERE CDC STAGE B DISEASES

NEPHROPATHY

The diagnostic criteria for nephropathy is early morning urine protein/creatinine ratio of >200mg/mmol in the absence of a urinary tract infection and an absence of an axillary temperature of ≥38.0°C (Rao TK, 2001; Webb NJA, 2003; Yoshimoto M, 1990).

If the urinalysis shows ≥2+ proteinuria, in the absence of a urinary tract infection or febrile illness with temperature ≥38.0°C, measure the urine protein/creatinine ratio. If the participant has a febrile illness where the temperature is ≥38.0°C, repeat urinalysis 2 to 3 weeks after the illness/temperature has subsided. If the urinalysis is still ≥2+ proteinuria, measure the urine/creatinine ratio.

CHRONIC HIV-ASSOCIATED LUNG DISEASE (INCLUDING BRONCHIECTASIS) AND LYMPHOCYTIC INTERSTITIAL PNEUMONIA (LIP)

The definition of chronic HIV-associated lung disease (including bronchiectasis) is chronic irreversible dilatation of the bronchi due to chronic infection (Coovadia HM, 2001).
Symptoms and signs include a history of productive cough of copious amounts of purulent sputum, with or without clubbing, halitosis, and clinical signs on chest auscultation of widespread crackles and wheezes and a chest radiograph showing lobar/ diffuse honey-comb appearance (small cysts) and/or persistent areas of opacification and/or widespread lung destruction, with fibrosis and loss of volume. The chest radiograph will be interpreted by a radiologist or pediatric pulmonologist.

A CT chest may be used to confirm the diagnosis, but is not mandatory.

The definition of lymphocytic interstitial pneumonia is: chest X-ray with bilateral reticulonodular interstitial pulmonary infiltrates present for more than two months with no response to antibiotic treatment and no other pathogen found.

CARDIOMYOPATHY (CMO)

The diagnostic criteria for cardiomyopathy require diagnostic confirmation and exclusion of other causes of congestive cardiac failure/large heart. The left ventricle and right ventricle are enlarged. The end-diastolic and end-systolic dimensions of the left or right ventricle are increased (≥2 SD from the mean for body surface area), with a reduced fractional shortening and ejection fraction (≥2 SD from the mean) (Park MK, 2002; Appendix, 2001).

All children with suspected cardiac dysfunction will be investigated for cardiomyopathy (Coovadia HM, 2001).

TO START OR RESTART ART:

The conditions listed below are applicable when evaluating the need to start or restart ART.

FAILURE TO THRIVE (FTT)

Documented loss of body weight OR body weight crossing two major centile lines (50^{th}, 25^{th}, 10^{th} or 5^{th}) and failure to gain weight on standard management within 4 weeks and no other cause identified during investigation OR for those already below the 5^{th} centile, if weight gain fails to parallel the 5^{th} centile (no chronic diarrhea or fever for >30 days required) (WHO, 2006).

SEVERE ORAL CANDIDIASIS

Severe oral candidiasis beyond the neonatal period not resolving on adequately administered topical therapy and that recurs within 6 months.

RECURENT SEVERE BACTERIAL PNEUMONIA

A current episode plus one in the previous 6 months. Acute onset (<2 weeks) of severe symptoms (such as fever, cough, dyspnoea and chest pain) PLUS new consolidation on clinical examination or chest radiograph PLUS response to antibiotics (unless it is a terminal event).

REFERENCES

1. Rao TK. Human Immunodeficiency virus infection and renal failure. Infect Dis Clin North Am. 2001; Sept 15(3): 883-850.

2. Webb NJA, Postlethwaite RJ. Clinical Paediatric Nephrology. 3rd Ed. Pendlebury, UK: Oxford University Press. 2003. Abnormal urinanalysis, haematuria, proteinuria, by Milford DV & Robson AM: Normal values for proteinuria and Urinary prot/creat ratios, p16-17.

3. Yoshimoto M, Tsukahara H, Saito M, Hayashi S, Haruki S, Fujisawa S, Sudo M. Evaluation of variability of proteinuria indices. Paediatric Nephrology. 1990; 4:136-139.

4. Coovadia HM, Wittenberg DF. Paediatrics and Child Health, A Manual for Health Professionals in the Third World. 4th ed. Cape Town, SA: Oxford University Press. 2001. Part 7, System-based disorders; Chapter 25, Respiratory disorders by Gie RP: Suppurative lung disease, p464-465.

5. Coovadia HM, Wittenberg DF. Paediatrics and Child Health: A Manual for Health Professionals in the Third World, 4th Ed, Copyright © 2001 Oxford University Press, Cape Town, SA: Part 7, System-based disorders; Chapter 28, Cardiac disorders by Levin SE: Cardiomyopathy, p553-555.

6. Park MK, George R, Troxler MPH. Paediatric Cardiology for Practitioners. 4th Ed. St Louis, USA: Mosby, Inc. 2002. Part 5: Acquired Heart Disease, Chapter 18: Primary Myocardial Disease: Dilated or Congestive Cardiomyopathy, p274.

7. Pediatric AIDS Clinical Trials Group. Appendix 40: Pediatric/maternal diagnoses. Cardiopulmonary Disorders, p 51.Available URL: http://pactg.s-3.com.

8. World Health Organization Case Definitions of HIV for Surveillance and Revised Clinical Staging and Immunological Classification of HIV-related Disease in Adults and Children. 2006; annex 1: 20.

APPENDIX VII: ANTHROPOMETRIC MEASUREMENTS

MEASUREMENT OF HEIGHT, LENGTH, WEIGHT, HEAD CIRCUMFERENCE and BODY SURFACE AREA (PROCEDURES TO BE FOLLOWED BY CHER STUDY)

1. Height

   - Once a participant is greater than 24 months of age and can stand upright, stature should be measured using a calibrated, wall-mounted stadiometer.

   - For best results, the participant is measured wearing a gown that allows the measurer to visualise the participant's body position. The participant stands with bare feet close together, body and legs straight, arms at sides, relaxed shoulders, and head, back, buttocks, and heels against the wall or shaft of the stadiometer.

   - Instruct the participant to look straight ahead and stand tall, keeping heels on the ground.

   - Bring the headboard down to the top of the participant's head while at eye-to-eye level with the participant and record the height to the nearest 0.1 cm.

2. Length

   - Measure participant's recumbent length up to 24 months of age and, for those unable to stand, up to 36 months of age on a calibrated length board with a stable headboard and a sliding footboard.

   - Two people are required to perform an accurate length measurement. One person holds the participant's head in place while the other slides the footboard and takes the reading. The participant's foot should be flat against the footboard with toes pointing straight upward and legs straight at the time of measurement.

     Participants who are longer than standard length boards but cannot stand should be measured on a hospital bed as accurately as possible. Two measurers must be involved. One holds the participant's head in place while the other marks the bed paper where the top of the head meets the bed. The feet need to be flexed with toes pointing upward. Legs and torso need to be straight during the time of marking the bed at the heel. Using a ruler, draw a straight line from the marks to the edge of the bed paper. Measure between the lines without the participant moving.

3. Weight

   - Use an electronic or beam scale with non-detachable weights.

   - Zero the scale prior to each measure. Use a calibrated scale.
   - Infants and young toddlers should be weighed lying down wearing only a diaper during the measurement.

- Weigh children who can stand on a beam scale, preferably one with "handle bars" for support. Calm children down and reduce their movement as much as possible for accurate measurements. Take a child's weight while he or she is barefoot and wearing only a gown.

- For children who have disabilities that prevent them from standing on a beam scale but are too large for an infant scale, use a bed scale. Alternatively, an adult may stand on the scale with and without the child. The weight of the adult is subtracted from the combined adult-child weight to determine the child's weight.

4. Head Circumference

   The tape is placed across the forehead with the lower border of the tape just above the eyebrows, around the head, above the ears and over the occipital prominence at the back of the head. Pull tape firmly to compress the hair and underlying soft tissues. Positioning of the tape over the forehead and occiput should be done to yield the maximum head circumference. Record head circumference in centimetres to the nearest 0.1 cm.

5. Body surface Area

   The body surface area is calculated using the following formula:

$$BSA(m2) = \sqrt{\text{Weight in kilograms x height in centimeters}/3600}$$

APPENDIX VIII: ANTIRETROVIRAL THERAPY BACKGROUND INFORMATION AND POTENTIAL DRUG INTERACTION

This appendix is divided into 5 sections for each drug

1. Current preparations
2. Dosing recommendations
3. Side effects
4. Drug interactions
5. Special instructions

A. NUCLEOSIDE REVERSE TRANSCRIPTASE INHIBITORS

Increased lactic acid in the blood (lactic acidosis) and an enlarged fatty liver may result in liver failure. Other complications and death have been reported with these medicines alone or in combination with other anti-HIV medicines. Some non-specific symptoms that might indicate lactic acidosis include: unexplained weight loss, stomach discomfort, nausea, vomiting, fatigue, weakness, and shortness of breath.

1. ABACAVIR (ABC)

Preparations:

- Oral solution: 20 mg/mL
- Tablets: 300 mg

Dosage Recommendations:

- 8 mg/kg twice daily with a maximum of 300 mg twice daily

ABC tablets (300mg):

- **≥ 25kg: 1 tablet (300mg) twice daily**

Abacavir is currently approved for infants and children 3 months of age and older by the US Food and Drug Administration at a dose of 8 mg/kg twice daily. It is highly unlikely that children less than 3 months of age would receive abacavir but if it was needed, the study would use the same dose as approved for other children.
Prior to use, subjects must be screened for HLA B*5071, which, if present, is a containdication to using ABC. Subjects who commenced ABC prior to availability of the HLA test and who are tolerating ABC, can continue the drug and do not require the assay.

Side Effects:

The following side effect has been associated with use of abacavir:
An allergic reaction which may include many different symptoms, such as fever, rash, feeling tired, upset stomach, vomiting, loose or watery stools, abdominal pain, cough, sore throat, shortness of breath, achiness, or a general feeling of illness.

These symptoms usually appear within the first six weeks after starting this drug but can occur at any time during treatment. This reaction can be severe and could even lead to

death if abacavir is not stopped. The severe form of allergic reaction can also recur if abacavir is restarted after it has been stopped and can even lead to death.

The frequency and clinical presentation of the allergic reaction to abacavir in children is similar to that seen in adults. In addition to those symptoms listed for adults, this syndrome may be seen as weakness or sluggishness, poor appetite, difficulty breathing, or a clinical picture similar to symptoms seen with infection of the blood.

If you think your child might be developing a reaction to abacavir, do not give your child any more doses and contact the doctor at the site immediately.

Note: Severe or fatal allergic-type reactions can occur within hours after abacavir is restarted in participants who have interrupted abacavir therapy. Allergic-type reactions to abacavir can occur in participants who have had no prior identified history or whose symptoms were previously unrecognized. If you interrupt abacavir for any reason, immediately contact the medical staff at the site. If the study doctor decides to restart abacavir, your child may need to be monitored more closely in the clinic or in the hospital.

Other side effects include:

- Upset stomach
- Vomiting
- Vague overall feeling of discomfort
- Feeling tired
- Decrease in appetite
- Loose or watery stools
- Inflammation or swelling of the pancreas with abdominal pain
- Headache
- Increased triglycerides

Drug Interactions:

- No significant interactions between ABC, AZT and 3TC.
- ABC neither inhibits nor is metabolized by hepatic cytochrome P450 enzymes. Thus, it should not cause changes in drug levels of PIs and NNRTIs metabolized through these pathways.

Special Instructions:

- Can be given without regard to food.
- Parents and caregivers must be cautioned about the risk of serious hypersensitivity.
- Participants' therapy should not be interrupted without consulting with their study physician.
- As the participant is enrolled into the study, the site must ensure that:
    1. The parent/caregiver receives the ABC warning card.
    2. The designated health care provider reviews the signs and symptoms of hypersensitivity reaction with the parent/caregiver.

3. The parent/caregiver verbalizes an understanding of the steps to take in the event of a suspected hypersensitivity reaction, including when and how to contact the study site.

2. DIDANOSINE (ddI)

Preparations:

- Pediatric powder for oral solution when reconstituted as solution containing antacid: 10 mg/mL
- Chewable tablets with buffers: **50, 100, 150, 200 mg**
- **Enteric coated tablets: 125 mg, 200 mg, 250mg and 400 mg**

Dosage:

- 2 weeks to 8 months of age: 100 mg/m^2 every 12 hours
- After 8 months of age: 120mg/m^2 every 12 hours
- **Ages 6-18 years and body weight >20kg: capsules**
  20 to <25kg: 200 mg once daily
  25 to <60kg: 250 mg once daily
  >60kg: 400 mg once daily

Didanosine is approved for HIV infected infants and children 2 weeks of age and older by the US Food and Drug Administration. Infants less than 8 months of age requiring therapy with ddI will be treated at a dose of 100 mg/m^2/dose, twice daily.

Side Effects:

The following side effects have been associated with use of didanosine:

- Inflammation of the pancreas, which may cause abdominal pain and may be fatal
- Changes in the eye that may lead to visual changes
- Loose or watery stools
- Numbness, tingling, and pain in your hands and feet
- Upset stomach and vomiting
- Headache
- Abnormal pancreatic function tests
- Increase in uric acid in the bloodstream
- Abnormal liver function tests
- **Non-cirrhotic portal hypertension.**

Non cirrhotic portal hypertension occurs when the blood flow in the major vein in the liver slows down which can lead to the development of severely enlarged esophageal veins (varices) in the gastrointestinal system. Because esophageal varices are thin and portal hypertension increases the pressure of blood flow in these veins, esophageal varices can break open, resulting in serious bleeding and in some cases death.

When didanosine is used with other medicines with similar side effects, these side effects may be seen more often and may be more severe than when didanosine is used alone. Participants who take didanosine together with stavudine (d4T), with or without hydroxyurea, may be at greater risk for abnormal liver function tests and for inflammation of the pancreas, which may be fatal. Death from liver problems has occurred in participants receiving the combination of stavudine, didanosine and hydroxyurea. Participants who take didanosine with stavudine may also have a greater risk for experiencing numbness, tingling, and pain in the hands or feet.

Drug Interactions:

- Ketoconazole and itraconazole: Possible decrease in absorption. Administer at least two hours before or two hours after ddI.
- Dapsone: Possible decrease in absorption. Administer at least two hours before or two hours after ddI.
- Tetracycline: Should be given one hour before or four hours after ddI.
- Iron salts: Should be given one hour before or four hours after ddI.
- Fluoroquinolone antibiotic: Absorption significantly decreased (chelation of drug by antacid in pediatric powder and tablets); administer two hours before or four to eight hours after ddI (varies with fluoroquinolone antibiotic).
- Administration with PIs:
    - Indinavir should be administered at least one hour before ddI on an empty stomach.
    - Ritonavir should be administered at least two hours before or one hour after ddI.
    - Nelvinavir should be administered at least one hour after ddI.

Special Instructions:

- ddI formulation contains buffering agents or antacids. Food decreases absorption; administer ddI on an empty stomach (30 minutes before or two hours after a meal).
- When administering chewable tablets, at least two tablets should be administered to ensure adequate buffering capacity (i.e., if the child's dose is 50 mg, administer two 25 mg tablets and not one 50 mg tablet).
- Decreased dosage should be used for participants with impaired renal function.

3. LAMIVUDINE (3TC)

Preparations:

- Oral solution: 10 mg/mL
- Tablets: 150 mg

Dosage:

- 4 mg/kg twice daily with a maximum of 300 mg daily

3TC scored tablets (150mg)

- **14–19.9kg:½ tablet (75mg) twice daily**
- **20–24.9kg:1 tablet (150mg) in the morning and ½ tablet (75mg) in the**

evening

- **≥ 25kg: 1 tablet (150mg) twice daily**

3TC is approved as part of combination therapy in adults, adolescents and children older than 3 months of age. Recommended doses of 3TC in this study follow approved dosing recommendations for this age group. For infants younger than 3 months, the same dose is proposed. This dosing has previously been studied in a number of studies including Pediatric ACTG 345 - a Phase I/II Study of Ritonavir Therapy in HIV-1 Infected Infants and Children, as well as Pediatric ACTG 356 - Early Intensive Antiretroviral Combination Therapy in HIV-1-Infected Infants and Children (Luzuriaga, 2004). Preliminary findings have not demonstrated any untoward toxicities. Additionally, this is the currently recommended dosing for this age group by the US Guidelines for the Use of Antiretroviral Agents in Pediatric HIV Infection (Working Group on Antiretroviral Therapy and Medical Management of HIV-Infected Children, 2004).

Side Effects:

The following side effects have been associated with use of lamivudine:

- Headache
- Feeling of vague overall discomfort
- Feeling tired
- Dizziness
- Depression
- Upset stomach
- Vomiting
- Loose or watery stools
- Decrease in appetite
- Abdominal cramps
- Sleeplessness
- Rash
- Numbness, tingling, and pain in the hands or feet
- Decrease in the number of white blood cells that help fight infection
- An increase in a substance in the blood (a type of a pancreatic enzyme) which could mean a problem with the pancreas
- Increased liver function tests, which could mean liver damage
- Inflammation of the pancreas with abdominal pain

The following side effect has been seen in children:

- Inflammation of the pancreas with abdominal pain

Participants who are infected with both Hepatitis B and HIV should be warned that their liver function tests may increase, and symptoms associated with hepatitis (an acute inflammation of the liver) may worsen after lamivudine has been stopped. Although most of these cases have resolved without treatment, some deaths have been reported.

Drug Interactions:

- Cotrimoxazole increases 3TC blood levels (possibly competes for renal tubular secretion); unknown significance.
- When used with AZT may prevent emergence of AZT resistance, and for AZT-resistant virus, revision to phenotypic AZT sensitivity may be observed.

Special Instructions:

- Can be administered with food.
- Decrease dosage in participants with impaired renal function.

4. STAVUDINE (d4T)

Preparations:

- Oral solution: 1 mg/mL
- Capsules: **15,** 20 **and** 30 mg

Dosage:

- **≤ 30 kg:** 1mg/kg twice daily with a maximum of 30 mg twice daily
- **>30 kg – 60 kg: 30mg twice daily**
  ≥ 60 kg: 40mg twice daily

Stavudine is currently approved for children 6 months of age and older at a dose of 1mg/kg every twelve hours. It is unlikely, although possible, that some infants less than 6 months of age would require treatment in this study. For those children, this study proposes to use stavudine at the same dosing amount and interval as in older children. This is the recommended dose in the US for pediatric HIV infection treatment. Preliminary pharmacokinetic and safety data using this dose in infants has recently been presented (Wade, 2003).

Side Effects:

The following side effects have been associated with use of stavudine:

- Numbness, tingling, and pain in your hands or feet
- Rash
- Upset stomach and vomiting
- Abdominal pain
- Muscle aches and pain
- Abnormal liver function tests
- Abnormal pancreatic function tests
- Inflammation or swelling of the pancreas with abdominal pain
- Rare cases of muscle weakness, which may progress to paralysis and inability to breath. This may be associated with elevation of lactic acid in the blood.

When stavudine is used with other medicines with similar side effects, these side effects may be seen more often, and may be more severe, than when stavudine is used alone. Participants who take stavudine together with didanosine (ddI), with or without hydroxyurea, may be at greater risk for abnormal liver function tests and for inflammation of the pancreas, which can be fatal. Death from liver problems has occurred in participants receiving the combination of stavudine, didanosine and hydroxyurea.

Participants who take stavudine with didanosine may also have a greater risk for having numbness, tingling, and pain in their hands or feet.

Drug Interactions:

1. Drugs that decrease renal function could decrease clearance.
2. Should not be administered in combination with AZT (poor antiretroviral effect).
3. Do not give in combination with ddI.

Special Instructions:

- Can be administered with food.
- Need to decrease dose in participants with renal impairment.

5. ZIDOVUDINE (AZT)

Preparations:

- Syrup: 10 mg/mL
- Capsules: 100 mg
- Tablets: 300 mg

Dosage:

- 240 mg/m^2 twice daily

AZT capsules (100mg)/ tablet (300mg)

- **8-13.9kg: 24 mg/kg/day: 1 capsule twice daily.**
- **14-19.9kg: 18 mg/kg/day: 2 capsules (200mg) in morning and 1 capsule (100mg) in the evening**
- **20-29.9 kg: 400mg/day: 2 capsules (200mg) in the morning and 2 capsules (200mg) in the evening**
- **≥30kg: 600 mg/day: 3 capsules or 1 tablet (300mg) twice daily**

Although the recommended pediatric schedule for AZT in the product package insert is as a thrice daily regimen for a total dose of 480 mg/m^2, AZT is used routinely in older populations twice daily. AZT will be administered to children in this study in a twice daily regimen (240 mg/m^2 twice daily), as well. The twice daily dosing recommendation is supported by the recommendations of the South African National Antiretroviral Treatment Guidelines (National Department of Health South Africa, 2004), PENTA guidelines (Pediatric European Network for Treatment of AIDS, 2004) and a study by Saez-Llorens and colleagues (Saez-Llorens, 2001).

Side Effects:

The following side effects have been associated with use of zidovudine:

- Decrease in the number of white blood cells that help fight infection
- Decrease in the number of red blood cells that may cause weakness, dizziness, and fatigue
- Muscle aches, weakness and wasting
- Headache
- Upset stomach

- Decrease in appetite
- Vague overall feeling of discomfort
- Lack of energy
- Feeling tired
- Sleeplessness
- Heartburn

Drug Interactions:

1) Increased toxicity may be observed with concomitant administration of the following drugs (therefore, more intensive toxicity monitoring maybe warranted):
    a. Ganciclovir
    b. Interferon-alpha
    c. TMP/SMX
    d. Acyclovir
    e. Other drugs that can be associated with bone marrow suppression

2) The following drugs may increase AZT concentrations (and therefore potential toxicity):
    a. Probenecid
    b. Atovaquone
    c. Methadone
    d. Valproic acid
    e. Fluconazole

3) Cimetidine: Decreased renal clearance may be observed with co-administration of cimetidine (may be significant in participants with renal impairment).

4) Rifampin and rifabutin: AZT metabolism may be increased with co-administration of rifampicin and rifabutin (clinical significance unknown).

5) Clarithromycin may decrease concentrations of AZT, probably by interfering with absorption (preferably administer four hours apart).

6) Ribavirin decreases the intracellular phosphorylation of AZT (conversion to active metabolite).

7) Phenytoin concentrations may increase or decrease.

8) d4T should not be administered in combination (poor antiretroviral effect).

Special Instructions:

- Can be administered with food (although the manufacturer recommends administration 30 minutes before or one hour after a meal).
- Decrease dosage in participants with severe renal impairment.

- Substantial granulocytopenia or anemia may necessitate interruption of therapy until marrow recovery is observed; use of erythropoietin or filgrastim is not standard of care in this South Africa population.
- Reduced dosage may be indicated in participants with substantial hepatic dysfunction.

B. NON-NUCLEOSIDE REVERSE TRANSCRIPTASE INHIBITORS

The use of potent antiretroviral drug combinations (which commonly include a protease inhibitor) may be associated with an abnormal placement of body fat and wasting. Some of the body changes include:

- Increase in fat around the waist and stomach area
- Increase in fat on the back of the neck
- Thinning of the face, legs, and arms
- Breast enlargement

6. NEVIRAPINE (NVP)

Preparations:

- Oral solution: 10 mg/mL
- Tablets: 200 mg

Dosage: for infants <10 kg or <3 years of age

- 150-200 mg/m^2 twice daily. Note: Initiate therapy with 150 mg/m^2 administered once daily for 14 days (maximum 200 mg once daily). Increase to full dose (150-200 mg/m^2) administered twice daily if no rash or other untoward effects (maximum 200 mg twice daily).

This is not the dose recommended by the United States and South Africa regulatory agencies for children 2 months of age and older (4 mg/kg once daily for 14 days and if no sign of rash, increase to 7 mg/kg twice daily). However, the proposed dosing for this study is widely used in clinical trials, and has been specifically studied in infants in Pediatric ACTG 356 - Early Intensive Antiretroviral Combination Therapy in HIV-1-Infected Infants and Children (Luzuriaga, 2004). Preliminary findings in that study have not demonstrated any untoward toxicities. Additionally, this is a currently recommended dosing strategy for this age group by the US Guideline for the Use of Antiretroviral Agents in Pediatric HIV Infection (Working Group on Antiretroviral Therapy and Medical Management of HIV-Infected Children, 2004) and is being incorporated in the European pediatric treatment guidelines, as well. Using mg/m^2 dosing guidelines provides more reliable scaling of an appropriate dose for children than using just weight for this drug.

Side Effects: (continuous dosing, not single dose regimens)
The following serious side effects have been associated with use of nevirapine:

Severe liver damage that can cause death may occur. Participants with higher CD4+ cell counts are at increased risk for developing liver damage, which is often associated with a rash. Women with CD4 cell counts greater than 250, including pregnant women receiving chronic nevirapine therapy, are at even higher risk for developing liver damage. Participants who have abnormal liver function tests before starting nevirapine and participants with active Hepatitis B or C infection are also at higher risk for liver damage. NVP should NOT be initiated if AST/ALT ≥ Grade 3 or child is Child-Pugh Stage B or C for liver dysfunction.

If a participant is developing liver damage, one or more of the following may result:

- Tiredness
- General feeling of illness
- Loss of appetite
- Nausea
- Pale stools
- Dark urine
- Yellowing of the skin or whites of the eyes
- Liver tenderness or abnormal liver function tests

Rash is the most common side effect associated with nevirapine. Rash occurs more often in women. Most rashes occur early during treatment. The rash may be severe and rarely, may cause death. One of the risk factors for developing serious skin reactions includes failure to take nevirapine properly during the first 14 days of treatment.

Hypersensitivity reactions (HSR), which can rarely be fatal, may occur. Symptoms associated with an HSR include rash, fever, fatigue, muscle or joint aches, blisters, mouth sores, facial swelling, red eyes and irritation of the eyes, general feeling of discomfort, hepatitis, kidney problems, and/or changes in white blood cell levels.

The risk of participants developing any of the serious side effects listed above is greatest during the first few months of treatment, but these side effects also can occur later. If your child develops any of the side effects listed above, no matter how long he/she has been receiving nevirapine, you must contact the study doctor right away and have your child seen by the study staff before the next dose. If you and your doctors then decide to stop the treatment because of symptomatic hepatitis, hypersensitivity or severe skin reactions, your child should never take nevirapine again.

Other side effects include:

- Fever
- Headache
- Upset stomach

Drug Interactions:

There could potentially be multiple drug interactions. Before administration, the participant's medication profile should be carefully reviewed for potential drug interactions.

- Not recommended for concurrent use: See Sections 5.6 and 5.7.

- Drug interactions requiring careful monitoring if co-administered:

    1) Administration with PIs: Indinavir (IDV) concentrations are decreased significantly (approximately 25%-30%) when administered with NVP. The adult guidelines recommend that IDV doses be increased by 20% when administered in combination with NVP; standard doses of nelvinavir (NFV) and ritonavir (RTV) are recommended when these drugs are used in conjunction with NVP. However, LPV/r lopinavir/ritonavir doses need to be increased by 30% when used in combination with NVP.

    2) Antifungals: NVP significantly reduces ketoconazole concentrations and these drugs should not be use concomitantly. If indicated, an alternate antifungal agent should be used.

    3) Rifabutin: Rifabutin has a lesser effect on NVP concentration than rifampicin, which is contra-indicated.

    4) Anticonvulsants and psychotropics: There are no data on the extent of drug interactions with the anticonvulsants phenobarbital, phenytoin, and carbamazepine. Serum concentrations of these agents should be monitored. Many of the psychotropics are metabolized by similar metabolic pathways as NVP and may interact; participants should be monitored carefully when these medications are used concomitantly.

Special Instructions:

- Can be administered with food.
- May be administered concurrently with ddI. Be aware of hypersensitivity reaction.

7. EFAVIRENZ (EFV)

Preparations:

- Capsules: 50 and 200 mg

Dosage: for infants >3 years of age and >10 kg

Administered once daily:

- 10 to <15 kg: 200 mg
- 15 to <20 kg: 250 mg
- 20 to <25 kg: 300 mg
- 25 to <32.5 kg: 350 mg
- 32.5 to <40 kg: 400 mg
- ≥40 kg: 600 mg

Efavirenz is approved for use in combination antiretroviral regimens in adults, adolescents, and in children 3 years of age and older. Efavirenz will be dispensed in this protocol in the approved age groups, at the recommended doses.

Side Effects:

Effects on mental function include:

- Dizziness
- Trouble sleeping, such as inability to sleep, abnormal dreams, and drowsiness
- Confusion
- Difficulty concentrating
- Hallucinations
- A feeling of strangeness and losing touch with reality
- An exaggerated feeling of well-being
- Agitation or anxiety

Other risks include:

- Rash
- Upset stomach
- Loose or watery stools
- Headache

Increases in substance in the blood which can mean problems with the pancreas, such as inflammation or swelling of the pancreas with abdominal pain:

- Increase in cholesterol
- Increase in triglycerides
- Abnormal liver function tests and inflammation of the liver (hepatitis)
- Abnormal vision
- Fever
- An abnormal or unusual distribution of body fat

Drug Interactions:

Mixed inducer/inhibitor of cytochrome P450 3A4 enzymes; concentrations of concomitant drugs can be increased or decreased depending on specific enzyme pathway involved.

- Not recommended for concurrent use: See Sections 5.6 and 5.7.
- Drug interactions require careful monitoring if co-administered with:
    1. Warfarin: (levels potentially increased or decreased).
    2. Rifampicin and rifabutin: Enzyme inducers, such as rifabutin, may decrease EFV concentrations; the clinical significance is unknown.
    3. Phenobarbital and phenytoin: May decrease EFV concentrations; the clinical significance is unknown.
    4. EFV is highly plasma protein bound and has the potential for drug interactions with other highly protein bound drugs (i.e., phenobarbital and phenytoin).

5. Clarithromycin: Levels are decreased while the levels of its metabolite are increased; alternatives to clarithromycin, such as azithromycin, should be considered.
6. Other macrolide antibiotics have not been studied in combination with EFV.
7. Protease inhibitors: Numerous drug interactions are possible when EFV is administered in combination with protease inhibitors; dose adjustments are required for LPV/r, as well as IDV, and increased monitoring is needed when combined with RTV.

Special Instructions:

- Efavirenz can be taken with and without food.
- The relative bioavailability of EFV was increased by 50% (range 11-126%) following a high fat meal. Because there is no information on safety of EFV when given above the recommended dose, administration with a high fat meal should be avoided due to the potential for increased absorption.
- Capsules may be opened and added to liquids or foods (EFV has a peppery taste); grape jelly has been used to disguise the taste.
- Bedtime dosing is recommended, particularly during the first two to four weeks of therapy, to improve tolerability of central nervous system side effects.

C. PROTEASE INHIBITORS

The use of potent combinations of anti-HIV medicines (which commonly include a protease inhibitor) may be associated with an abnormal placement of body fat and wasting. Some of the body changes include:

- Increase in fat around the waist and stomach area
- Increase in fat on the back of the neck
- Thinning of the face, legs, and arms
- Breast enlargement

The use of protease inhibitors may also be associated with altered fat metabolism, including elevated triglycerides and/or elevated cholesterol and with the development of or the worsening of increased blood sugar and diabetes.

There have been reports of increased bleeding in HIV-infected persons with hemophilia who were treated with protease inhibitors, although it is not known if protease inhibitors were the cause of these bleeding episodes.

8. LOPINAVIR/RITONAVIR (LPV/r)

Preparations:

- Oral solution: 80 mg LPV and 20 mg RTV per mL
- For children with a body surface area of less than 1.3 m^2: Kaletra® oral solution is recommended.

Dosage:

LPV/r Oral Solution Dosage

- <6 months of age: 300 mg/m^2 LPV/75 mg/m^2 RTV twice daily
- ≥6months of age and <7 kg: 230 mg/m^2 LPV/57.5 mg/m^2 RTV twice daily with a maximum of 400 mg LPV/100 mg RTV twice daily
- ≥6 months of age and ≥7 kg:

  7 to 10 kg: 1.25 mL twice daily
  >10 to <15 kg: 1.75 mL twice daily
  15 to 20 kg: 2.25 mL twice daily
  >20 to 25 kg: 2.75 mL twice daily
  >25 to 30 kg: 3.5 mL twice daily
  >30 to 35 kg: 4.0 mL twice daily
  >35 to 40 kg: 4.75 mL twice daily

The maximum dose of LPV/r is 400 mg LPV/100 mg RTV twice daily

LPV/r Tablet Dosage

Number of 100/25mg tablets.

- **10-13.9kg: 2 tablets (100/25mg) in the morning and 1 tablet (100/25mg) in the evening**
- **14-19.9kg: 2 tablets twice daily**
- **20-24.9kg: 3 tablets in the morning and 2 tablets in the evening**
- **25-34.9kg: 3 tablets twice daily**
- **>35kg: Adult dosing**

Number of 200/50mg tablets.

- **25-34.9kg: 2 tablets in the morning and 1 tablet in the evening**
- **>35kg: Adult dosing**

*** Tablets must not be chewed, broken or crushed**.

Lopinavir/ritonavir is approved for the treatment of adults, adolescents and children 6 months of age and older with HIV infection. In this study, lopinavir/ritonavir is being used in the initial regimen for children. Pharmacokinetic and safety data has been obtained in South African children 6 months of age and older and supports the current dosages for children (Saez-Llorens, 2003). For children less than 6 months of age, the dose of 300 mg/m^2 LPV/75 mg/m^2 RTV twice daily is proposed by this study. The dose has been established in Pediatric ACTG 1030 – a Phase I/II study of lopinavir/ritonavir in HIV-1 infected infants <6 months of age. (Chadwick, 2008).

Side Effects:

The following side effects have been associated with use of lopinavir/ritonavir:

- Gastrointestinal disturbances, including abdominal pain, upset stomach, vomiting, abnormal bowel movements (or stools), and loose or watery stools
- Large increases in the amount of fatty acid in the blood
- Large increases in cholesterol in the blood
- Abnormal liver function tests (increased liver enzymes) and worsening liver disease, possibly leading to death

- Elevation of pancreatic enzymes and inflammation of the pancreas (pancreatitis) which may result in death
- Feeling weak/tired
- Headache
- Rash (seen in children)

Drug Interactions:

LPV/r is extensively metabolized by hepatic cytochrome P450 3A (CYP3A). Drugs metabolized by the hepatic cytochrome P450 enzyme system have the potential for significant interactions with multiple drugs, some of which may be life-threatening. Before administration, the participant's medication profile should be carefully reviewed for potential drug interactions.

- LPV/r is not recommended for concurrent use with: See Sections 5.6 and 5.7.
- Special consideration should be given when administering LPV/r in combination with the following:
  1. St. John's Wort.
  2. EFV and NVP: Induction of the metabolism of LPV/r and decrease in plasma concentrations. A dose increase of LPV/r is recommended (see dosage section).
  3. Anticonvulsant drugs: Carbamazepine and phenobarbital increase CYP3A activity, leading to increased clearance and lower levels of LPV/r; should be used with caution.
  4. Dexamethasone: Decreases LPV/r serum concentrations. Use with caution.
  5. HMG-CoA reductase inhibitors: LPV/r increases serum concentrations of some HMG-CoA reductase inhibitors. Pravastatin is the preferred agent.
  6. Clarithromycin: LPV/r increases serum concentration and clarithromycin dose adjustment is recommended in participants with impaired renal function (CrCl 30-60 mL/min.: decrease clarithromycin dose by 50%; CrCl <30 mL/min.: decrease clarithromycin dose by 75%).
  7. Rifabutin: LPV/r increases metabolite concentrations; dose reduction of rifabutin by at least 75% of the usual dose is recommended.
  8. Sildenafil (Viagara): Increases serum concentrations. Reduce dose of sildenafil and monitor toxicity.
  9. Antiarrhythmics: Increases serum concentrations of amiodarone, bepridil, lidocaine (systemic) and quinidine. Monitoring of antiarrhythmic serum concentrations is recommended.
  10. Immunosuppressant agents: May increase serum concentrations of cyclosporine, tacrolimus, and rapamycin. Monitor serum concentrations of these agents when co-administered.
  11. Dihydropyridine calcium channel blockers (i.e., felodipine, nifedipine, nicardipine): Increases serum concentrations; clinical monitoring is recommended.
  12. Ketoconazole: Serum concentration of this drug is increased by LPV/r. High doses of this agent (>200mg/day) is not recommended.
  13. Atovaquone: Decreases concentrations. The clinical significance is unknown.

14. Other PIs: Appropriate doses of LPV/r have not been established.
15. Metronidazole: LPV/r oral solution contains 42.4% alcohol and can cause a disulfiram-like reaction when co-administered.
16. **Lopinavir/ritonavir should be used with caution and with clinical monitoring in patients who are also using other drugs that prolong the PR interval, such as atazanavir, digoxin or beta blockers**

Special Instructions:

- Administer with food or formula. High fat meal increases absorption, especially of the liquid preparation.
- If co-administered with ddI, ddI should be given one hour before or two hours after LPV/r.

9. NELFINAVIR (NFV)

Preparations:

- Powder for oral solution: 50 mg/g
- Tablets: 250 mg tablet

Dosage:

- <12 months of age: 75 mg/kg twice daily
- 12-24 months of age: 60 mg/kg twice daily
- >2 years of age: 55 mg/kg twice daily with a maximum of 1250 mg twice daily

The US Food and Drug Administration has approved nelfinavir for children 2 years of age and older. The doses recommended by the study for that age group are consistent with the US approved dose. Determining appropriate dosing for young children and infants has proven quite difficult. Pediatric investigators in Europe and the US have conducted studies to better guide treatment guidelines. The proposed dosing for infants up to 2 years of age is 60-75 mg/kg twice daily and is based on recently published data from PENTA 7 (Aboulker, 2004) that indicated considerably higher doses, on a weight adjusted basis, are needed in infants to achieve comparable nelfinavir exposure seen in adults (Litalien C, 2003).

Side Effects:

The following side effects have been associated with use of nelfinavir:

- Loose or watery stools
- Upset stomach
- Gas
- Abdominal pain
- Rash
- Decrease in the number of white blood cells that help fight infection
- Abnormal liver enzyme test
- Allergic reactions, including bronchospasm, fevers and edema
- Jaundice and elevated bilirubin

Note: Participants with phenylketonuria should be aware that nelfinavir oral powder. (Viracept Oral Powder) contains 11.2 mg phenylalanine per gram of powder.

Drug Interactions:
NFV is in part metabolized by cytochrome P450 3A4 (CYP3A4). There could potentially be multiple drug interactions. NFV decreases the metabolism of certain drugs, resulting in increased drug levels and potential toxicity. Before administration, the participant's medication profile should be carefully reviewed for potential drug interactions.

- NFV is not recommended for concurrent use with: See Sections 5.6 and 5.7.
- Special consideration in the following cases:
  1. Rifabutin: Causes less decline in NFV concentrations than rifampicin, if coadministered with NFV, rifabutin should be reduced to one half the usual dose.
  2. NVP: There are no data on co-administration with NVP, but some experts use higher doses of NFV if used in combination with NVP.
  3. Administration with other PIs:
     - Co-administration with IDV increases concentration of both drugs.
     - Co-administration with RTV increases concentration of NFV, without change in RTV concentration.

Special Instructions:
- Administer with meal or light snack.
- Administered two hours before or one hour after ddI.
- Do not mix with any acidic food or juice because of resulting poor taste.
- Do not add water to bottles of oral powder; a special scoop is provided with oral powder for measuring purposes.
- Tablets readily dissolve in water and produce a dispersion that can be mixed with milk or chocolate milk; tablets also can be crushed and administered with pudding.

10. RITONAVIR (RTV)

Preparations:
- Oral solution: 80 mg/mL

Dosage:
>1 month of age: 350 to 400 mg/m^2 of body surface area twice daily by mouth and should not exceed 600mg twice daily. Ritonavir should be started at 250 mg/m^2 and increased at 2 to 3 day intervals by 50 mg/m^2 twice daily. If patients do not tolerate 400 mg/m^2 twice daily due to adverse events, the highest tolerated dose may be used for maintenance therapy in combination with other antiretroviral agents, however, alternative therapy should be considered. When possible, dose should be administered using a calibrated dosing syringe.

Ritonavir is approved by the US Food and Drug Administration for children one month of age and older (http://www.fda.gov/oashi/aids/listserve/ October 6th, 2005). No children under 1 month of age will be treated with ritonavir on this study.

Side Effects:

The following side effects have been associated with use of ritonavir:

- Stomach and bowel problems including abdominal pain, upset stomach, vomiting, abnormal stools, and loose or watery stools
- An increase in triglycerides
- Numbness and tingling in the arms, legs and around the mouth
- Rash
- Abnormal liver function tests
- Fever
- A change in the sense of taste

Drug Interactions:

RTV is extensively metabolized by hepatic cytochrome P450 3A (CYP3A). There could potentially be multiple drug interactions. Before administration, the participant's medication profile should be carefully reviewed for potential drug interactions.

- RTV is not recommended for concurrent use with: See Sections 5.6 and 5.7.
- Special consideration in the following cases:
  1. Theophylline: RTV increases metabolism, levels should be monitored, and dose may need to be increased.
  2. Clarithromycin: RTV increases levels, dose adjustment may be necessary in participants with impaired renal function.
  3. Desipramine: Dose adjustment may be necessary.
  4. Warfarin: Monitoring of anticoagulant effect is necessary.
  5. Digoxin: May increase or decrease levels; monitoring of levels is recommended.
  6. Anticonvulsants including carbamazepine and phenobarbital: Levels should be monitored because RTV can affect the metabolism of these drugs.
  7. Dexamethasone: Leads to increased clearance of RTV.
  8. **Rifampicin: For subjects on PI's such as lopinavir, co-formulated with RTV in small doses for therapeutic levels, RTV must be increased to equivalent dose in mg once rifampicin is needed for anti-mycobacterium therapy. Under these circumstances, if there is pre-existing heart disease, there may be a prolongation of PR interval.**

Special Instructions:

- Administration with food increases absorption and helps decrease gastrointestinal side effects.
- If RTV is prescribed with ddI, there should be two hours between taking each of the drugs.

- To minimize nausea, therapy should be initiated at a low dose and increased to full dose over five days, as tolerated.
- **For RTV alone and if in full dosage with lopinavir, use with caution for preexisting structural heart disease, conduction system abnormalities, or other cardiac diseases.**
- Techniques to increase tolerance in children:
  1. Mixing oral solution with milk, chocolate milk, vanilla or chocolate pudding, or ice cream.
  2. Dulling the taste buds before administration by chewing ice, giving popsicles or spoonfuls of partially frozen orange or grape juice concentrates.
  3. Coating the mouth by giving peanut butter to eat before the dose; or administration of strong-tasting foods, such as syrup, cheese, or strong-flavored chewing gum immediately after dose.

REFERENCES

Aboulker JP, Babiker A, Chaix ML, *et al.* Paediatric European Network for Treatment of AIDS. AIDS. 2004;18(2):237-245.

Chadwick EG, Caparelli EV, Yogev R, et al. Pharmacokinetics, effacy and safety Lopinavir/Ritonavir for HIV-1-infected Infants less than 6 months of age: 24 week results. AIDS 2008;22:249-55

Litalien C, Faye A, Compagnucci A, *et al.* Pharmacokinetics of nelfinavir and its active metabolite, hydroxy-tert-butylamide, in infants perinatally infected with human immunodeficiency virus type 1. Pediatr Infect Dis J. 2003; 22(1): 48-55.

Luzuriaga K, McManus M, Mofenson L, *et al.* A trial of three antiretroviral regimens in HIV-1-infected children. N Engl J Med. 2004;350(24):2471-2480.

National Department of Health South Africa. National Antiretroviral Treatment Guidelines. HIV and AIDS Policy Guideline, First Edition, Minuteman Press. Available URL: http://www.hepo-hiv.co.za/DH16_ART_Guidelines.pdf.

Pediatric European Network for Treatment of AIDS (PENTA). PENTA guidelines for the use of antiretroviral therapy in paediatric HIV infection. 2004. Available URL: http://www.ctu.mrc.ac.uk/penta/guidelines.htm.

Saez-Llorens X, Nelson RP, Jr, Emmanuel P, *et al.* A randomised, double-blind study of triple nucleoside therapy of abacavir, lamivudine, and zidovudine versus lamivudine and zidovudine in previously treated human immunodeficiency virus type 1-infected children. The CNAA3006 Study Team. Pediatrics 2001; 107(1):E4.

Saez-Llorens X, Violari A, Deetz CO, *et al.* Forty-eight-week evaluation of lopinavir/ritonavir, a new protease inhibitor, in human immunodeficiency virus-infected children. Pediatr Infect Dis J. 2003; 22(3): 216-24.

Wade N, Unadkat J, Huang S, *et al.* Pharmacokinetics and Safety of d4T (Stavudine) and 3TC (Lamivudine) in HIV-infected pregnant women and their infants (PACTG). 2003.

Working Group on Antiretroviral Therapy and Medical Management of HIV-Infected Children. Guidelines for the Use of Antiretroviral Agents in Pediatric HIV Infection, November 30, 2004. (**http://aidsinfo.nih.gov**)

APPENDIX IX: DRUG ADHERENCE CALCULATION WORKSHEET

(To be completed by the site pharmacist)

		Drug			
		A	**B**	**C**	**D**
1. Units dispensed:	1				
2. Units returned:	2				
3. Units administered:	3				
4. Units that should have been used:	4				
5. Compliance (%): [(#3/#4) x 100]	5				

Units = mL OR mg OR number of tablets

Site Pharmacist signature: _____ Date:_____

APPENDIX X: MEASURES TO RETAIN CHILDREN IN FOLLOW-UP

1. Demographics
    a. A careful note of the contact address is to be made. If necessary, a field worker will do a home visit to check the address.
    b. If possible, a second contact address is to be obtained (e.g., the address of a neighbour or a family member or friend).
    c. Contact telephone numbers should be obtained wherever possible – either of the caregiver, a family member, a neighbour, or a friend.
    d. All contact details will be checked at every visit to confirm that no changes have occurred since the last visit.

2. Visit schedule
    a. At the start of a study, the year's planned visit schedule will be given to the caregiver.
    b. At each visit, the next visit date will be checked with the caregiver.

3. Follow-up visits are to coincide with any other planned visits for either the caregiver or her child (e.g., visits for mother if on Project 1, visits for the childhood immunizations if on Project 4), wherever possible.

4. Missed visit – a clinic list of children expected for the week will be generated each week. Should a caregiver and her child not attend on a particular day, contact tracing will be initiated within 24 hours of the missed visit. (e.g., a telephone call if appropriate, a letter to be sent to the family, a field worker to be sent to the home).

5. 24 hour contact details of a member of the study team will be given to the caregiver. Should the child experience any problems or illnesses, the caregiver will be encouraged to either bring the child to the clinic for review or to contact the on-call number.

6. Meetings with support groups to be arranged for the caregivers – to coincide with clinic visits.

7. The caregivers/mothers of the children to be encouraged to have regular medical review to assess their clinical disease status and to have referral for ART, as appropriate.

8. Transportation money will be provided for study visits.

9. Assist the caregiver to access social services, if appropriate.

10. Ongoing information to be given to the caregivers about the trial, about the clinical status of the children or any other issues that may affect the caregiver's decision to continue the trial.

11. Ongoing adherence counselling – to explain the importance of strict adherence, which includes study visit follow-up.

12. Side-effects – potential side-effects of the ARVs, the likely duration of these side-effects, and basic management of these side effects to be discussed with the caregivers, prior to initiating ARVs, so that they are fully informed.

13. If a participant is "lost to follow-up"**,** all attempts will be made to ascertain the participant's whereabouts and the reason for the clinic non-attendance. If this child returns to the clinic at a later date, he/she will once again be started on the study.

14. Ongoing encouragement and assessment of the caregivers to ensure they are coping with providing the study medication.

APPENDIX XI: IMMUNIZATION SCHEDULE IN SOUTH AFRICA

Age	BCG	OPV	DPT	Hep B	Hib	Measles	DT
Birth	BCG	OPV 0					
6 weeks		OPV 1	DPT 1	Hep B 1	Hib 1		
10 weeks		OPV 2	DPT 2	Hep B 2	Hib 2		
14 weeks		OPV 3	DPT 3	Hep B 3	Hib 3		
9 months						Measles 1	
18 months		OPV 4	DPT 4			Measles 2	
5 years		OPV 5					DT

BCG Vaccine against tuberculosis
OPV Oral polio vaccine (OPV 0 refers to dose at birth with subsequent doses numbered accordingly)
DPT Diphtheria, pertussis and tetanus vaccine
Hep B Hepatitis B vaccine
Hib Hemophilus influenzae type b vaccine
DT Diphtheria and tetanus vaccine

APPENDIX XII: LETTER TO MEDICAL OFFICER/ CLINIC SISTER

LETTER TO MEDICAL OFFICER and/or CLINIC NURSE

Dear Dr/Sr _____

(Name of child) _____ is participating in the CHER research study at _____ (name of site). This research study is comparing three antiretroviral treatment strategies in infants with perinatally acquired HIV-1 infection.

All children in the study are being followed by the study physicians for a minimum of 3.5 years. They are currently receiving the following study drugs under the care of the study physicians:

____ Zidovudine (AZT)
____ Lamivudine (3TC)
____ Kaletra (LPV/r)
____ Didanosine (ddI)
____ Abacavir (ABC)
____ Nevirapine (NVP)
____ Efavirenz (NFV)
____ Nelfinavir (NFV)
____ Ritonavir (RTV)
____ Stavudine (d4T)

Concomitant medication:

____ Cotrimoxazole

The contact details for the study physicians are listed below. They request that you notify them of this visit by telephone and provide a written report as soon as possible. You are welcome to call if you would like a more detailed explanation of the trial or have any comments or any problems. Please contact the study physicians for dosage of ARVs.

Dr. Avy Violari
Perinatal HIV Research Unit
University of the Witwatersrand
New Nurses Home
Chris Hani Baragwanath Hospital
Old Potch Road
Soweto, 2013
Johannesburg, South Africa
Tel: (011) 989 9707 or (082) 857-4126

Prof. Mark Cotton
Children's Infectious Diseases Clinical Research Unit (KID-CRU), Ward J8
University of Stellenbosch
Tygerberg Children's Hospital
Francie van Zijl Avenue
Parow, 7505
Cape Town, South Africa
(021) 938-4219 or (083) 379-5283

24-hr contact numbers: (082) 456-8554 for Johannesburg and (084) 641-0735 (a/h) for Cape Town.

Thank you for your cooperation.

APPENDIX XIII: SAMPLE STUDY INFORMED CONSENT FORM

PATIENT INFORMATION LEAFLET AND STUDY INFORMED CONSENT FORM

Title of the study: A Phase III, Randomised, Open-label Trial to Evaluate Strategies for Providing Antiretroviral Therapy to Infants Shortly after Primary Infection in a Resource Poor Setting

Short title of the study: Children with HIV Early Antiretroviral Therapy
CHER Trial

Sponsored by: The National Institute of Allergy and Infectious Disease
Division of AIDS (DAIDS)
6700B Rockledge Drive
Bethesda, Maryland 20852
United States

Protocol co-chairs:

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Dr. Mark Cotton
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University of Stellenbosch
Tygerberg Children's Hospital
Francie van Zijl Avenue
Parow, 7505
Cape Town, South Africa
(021) 938-4219 or (083) 379-5283

This information is provided to help you decide whether to take part in this clinical research study. Please read this form carefully and ask the study doctor or the study staff to explain any words or procedures you do not understand.

Dear Parent
Good day, my name is _____ and I am a _____ at _____ I would like to invite your child to take part in the research study called "CHER", because he/she is infected with the human immunodeficiency virus (HIV-1). Before agreeing to participate in this study, it is important that you fully understand what is involved. If you have any questions, which are not fully explained in this leaflet and consent form, do not hesitate to ask the doctor in charge of the study or the study staff at any time.

If you allow your child to take part in this study, you will be asked to sign and date this consent form. You will get a copy to keep. You should only sign the consent form if all details of the study are completely clear to you, if you are willing to let your child to be part of the study and if you completely understand your child's rights as a participant in this study.

This study will be conducted in accordance to the Declaration of Helsinki.

WHY IS THIS STUDY BEING DONE?
HIV-1 infection in children is a major problem in South Africa. Even with projects to prevent the spread of HIV from mother to child, there are still children born every day who are infected with HIV.

HIV-1 infection in young children is usually more severe than in older children and adults, because their immune system is weaker. That means that many children who are infected with HIV-1 may get sick even before they are one year old and some of them may even die. Therefore, we need to find a way that will lead to more HIV-infected children staying well longer.

The CHER trial compares three treatment strategies in infants who were infected with HIV-1 at birth. Infants with stronger immune systems were randomly assigned to deferred therapy (Arm 1), receiving therapy until the first birthday (Arm 2), or therapy until the second birthday (Arm 3). We want to see if stopping this treatment will be useful and will change the course of disease in children. Infants with weaker immune systems will receive continuous therapy as they are already sicker than the other infants. Arm 1 is closed to accrual.

When infants in Arm 1 become sick (the CD4% drops to <25% or CD4 count <1000 cells/mm^3 regardless of CD4% if ≤12 months of age or <20% if 12 months of age and older or if your child develops a severe CDC Stage B or Stage C disease), they will be treated with antiretroviral therapy (ART).
If your child has interrupted treatment, we will restart therapy if:

- the CD4% drops to < 25% or CD4 count <1000 cells/mm^3 regardless of CD4% if <12 months of age;
- below 20% if 12-**59** months of age;
- **below 20% or CD4 count <350cells/mm^3 if ≥ 60 months,** or
- your child develops a severe CDC Stage B or Stage C disease).

They will be given the same ART that they took earlier (before therapy was stopped) to ensure that they do not become sicker. If the first regimen of ART stops working, the study doctors will consider giving the infants a second regimen of ART. The study doctors will consider giving your child a second regimen of ART if your child's CD4% drops to <20%, if your child develops a severe CDC Stage B or Stage C disease**, if your child's viral load goes up** or if your child experiences severe side effects which prevent continued use of that specific drug regimen.

HOW MANY CHILDREN WILL TAKE PART AND HOW LONG WILL MY CHILD BE IN THIS STUDY?
A total of approximately **451** children from Cape Town and Johannesburg will participate in the study. The duration of the study will be approximately **6** years. The first child who enrols will be followed up for **6** years and the last child enrolled for 3.5 years. Your child may be in the study between 3.5 years to **6** years (182 weeks to **330** weeks) depending on the time of enrolment.

WHAT IS THE HISTORY OF THIS STUDY?
The study commenced in July 2005. 377 infants were put into one of three arms by randomization (like tossing a coin). In "Arm 1" children were closely checked for immune weakness or disease and started anti-HIV treatment (treatment that keeps the amount of HIV-1 virus low in the blood) when they needed it.

In “Arm 2”, children were given anti-HIV medicines until their first birthday and in “Arm 3” until their second birthday. Afterwards, they are carefully followed up to see how soon they will need anti-HIV medicines again. The medicines will be restarted if they do need them again. This would occur if the immune system shows signs of weakening or if they start becoming sick. The medicines will be given to the child for as long as he/she derives benefit from them (until he/she fails that regimen).

On June 20th 2007, the Data Safety Monitoring Board (a group of experts who continually check what happens to children on the trial to see if one arm better or worse than the others), found there were fewer deaths in infants on Arms 2 and 3 (early therapy) than arm 1 (later therapy). Ten of 252 **infants** (4%) died in Arms 2 and 3 combined and 20 of 125 **infants** (16%) in Arm 1. For this reason, no more participants will be enrolled on Arm 1.

WHAT DOES MY CHILD HAVE TO DO IF HE/SHE IS IN THIS STUDY?

The study has different treatment “Arms” and your child will be allocated to one of these Arms by chance, like flipping a coin. In “Arm 1” children were closely checked for immune weakness or disease and started anti-HIV treatment (treatment that keeps the amount of HIV-1 virus low in the blood) when they need it. Recruitment to Arm 1 is closed.

In “Arm 2”, children will be given anti-HIV medicines until their first birthday and in “Arm 3” until the second birthday. Afterwards, they will be carefully followed up to see how soon they will need anti-HIV medicines again. The medicines will be restarted if they do need them again. This would occur if the immune system shows signs of weakening or if they start becoming sick. The medicines will be given to the child for as long as he/she derives benefit from them (until he/she fails that regimen).

Children whose immune systems are already weakened will be referred to continuous therapy. The treatment your child will receive will include prevention of lung infections and/or anti-HIV treatment. Your child must be 6 to 12 weeks old at enrolment and must be infected with HIV in order to take part in this study.

All participants will receive cotrimoxazole prophylaxis from the Randomisation Visit until Week 40, regardless of treatment arm, as standard of care.

STUDY PROCEDURES

Randomisation Visit (Week 0-Baseline)

Your child will need to come to the clinic for an entry or enrolment visit. If you decide to let your child take part in this study, you will need to sign and date this study consent form. After signing and dating the consent form, your child will be randomised to one of the study arms. Randomisation is a process similar to flipping a coin. Your child will have an equal chance of being randomised to any one of the study arms. You cannot choose which arm you wish your child to be in. If your child is randomised to Arm 1, they will follow the same tests and procedures as Arm 2 and 3, except the anti-HIV treatment will only be applicable once your child has started therapy:

If your child is randomised to Arms 2 or 3, this will be the day that your child starts the anti-HIV treatment (study treatment). The following tests and procedures will be done:

- The study doctor will look at your child's blood results that were taken at the screening visit to see if it is safe for your child to start the anti-HIV treatment
- You will be asked about your child's previous illnesses and hospitalisation.
- A physical examination.
- Your child's weight, height, heart rate and temperature (breathing rate and blood pressure if necessary) will be measured
- Your child's body surface area will be worked out (if applicable).
- Approximately 10 mL of blood (2 teaspoons) will be taken to check your child's FBC (test of the number of blood cells), LFTs (tests of liver and kidney function), and CD4+ (test to see how many immune/fighter cells are in the blood).
- A small amount of blood will be stored and tested for drug levels, drug resistant virus and viral load (the amount of virus in blood) at a later stage. Your child's blood will not be used for genetic research without your permission.
- If your child's weight is ≥3 kg and you give special permission a few drops of this blood will be put on thick paper (filter paper) to see if the RNA PCR test on this filter paper will be able to show how much virus is in your child's blood.
- Urine will be collected to test for any abnormalities.
- Go to the pharmacy where you will be given a month's supply of treatment drugs for your child (if applicable).
- The site pharmacist and/or study doctor will give you full instructions about how to give the drugs to your child and you will also have a chance to give the treatment drugs to your child in the pharmacy to show that you know how to give the drugs correctly (if applicable).

The Randomisation Visit may take up to 2 hours of your time to perform all the evaluations. The results of the enrolment tests will be given to you at the Week 2 Visit. You will be given a list of warning signs. If you notice any of these signs in your child, please see a doctor straight away as your child may be very sick and need urgent medical care. Please also contact your study doctor immediately.

WEEK 2 VISIT

Children in Arm 1

You will be asked to come back after 2 weeks for a brief review of your child's health.

Children in Arm 2 or 3

You will be asked to come back after 2 weeks of giving your child the treatment drugs. We want to make sure that you don't experience any problems in giving the treatment drugs and that your child does not experience any bad reactions to the treatment drugs.

STUDY WEEK 4, 8, 12, 16, 20, 24, 32, 40, 48 THEN EVERY 12 WEEKS TO END OF THE TRIAL

Children in Arm 1, 2 or 3

Your child will have follow-up visits with a study doctor at **Study** Week 4, 8, 12, 16, 20, 24, 32, 40, 48, and then every 12 weeks until the end of the study or premature study discontinuation. At these visits, the following tests and procedures will be done:

- A physical examination.
- Your child's weight, height, heart rate, breathing rate (optional) and temperature will be measured. Your child's blood pressure will be measured, if necessary, during visits prior to 104 weeks of age (2 years); beginning with Week 104, until the end of the trial, your child's blood pressure will be routinely measured.
- Your child's body surface area will be worked out at some visits (if applicable).
- You will be asked about any illnesses and/or reactions to the study drugs, hospitalisation or change in medication given to your child since the last visit.
- Approximately 10 mL of blood (2 teaspoons) will be taken check your child's FBC (test of the number of blood cells), LFTs (tests of liver and kidney function), and CD4+ (test to see how many immune/fighter cells are in the blood). **A viral load (the amount of virus in your blood) will be measured every 3 months after the baseline visit if your child is on ART.** Blood will be drawn at all these visits except at Week 8, 16 and 20. Urea, electrolytes, lipids and viral load will be checked at Week 24 and every six months thereafter.
- **We may need to take extra blood (1 teaspoon) for therapeutic blood monitoring to determine the amount of drug that is in your child's blood. This will help us confirm that your child is getting the correct amount of drug.**
- If your child is on anti-tuberculosis medication, additional blood may be drawn to test for drug levels.
- A small amount of blood will be stored and tested for drug levels, drug resistant virus and viral load (the amount of virus in blood) at a later stage. Your child's blood will not be used for genetic research without your permission.
- If your child's weight is ≥3 kg and you give special permission, less than half a teaspoon of blood will be tested to see if the strength of the antibody test can help find out if a child is HIV-infected. This will only be done at four of the visits in the first year of the study.
- Urine will be collected every 2 or 3 months to test for any abnormalities. In the event that urine is not obtained for urinalysis on a particular visit, and if abnormalities are not suspected, (the urine collection maybe deferred until the next visit).
- Go to the pharmacy where you will be given a month's supply of treatment drugs for your child **(if applicable)**.

CHILDREN IN ARMS 2 AND 3 WHERE THE CD4 PERCENTAGE AT THE START OF THE TRIAL WAS BELOW 25 PERCENT

As a result of new information in adults of increased sickness off antiretrovirals, we now recommend that your child does not stop these medicines at 40 or 96 weeks as originally planned, but rather to continue the treatment without interruption. Should your infant have already entered an interruption phase related to the trial, we recommend that your child restarts antiretroviral therapy.

ALL MONTHLY SCHEDULED DISPENSING VISITS

<u>Children in Arm 1 (deferred therapy)</u>

At each scheduled dispensing visit, you will need to go to the pharmacy monthly until Week 40.

Children in Arm 2, 3 and Arm 1 (on treatment)

At each scheduled dispensing visit, you will need to bring all the unused treatment drugs you have received for your child at the previous visit. The following procedures will be done:

- Bring study drug returns.
- Complete the Drug Adherence Calculation Worksheet.
- Discuss any questions or problems you have about giving the drugs.
- The site pharmacist will then measure the unused treatment drugs to make sure that your child is getting the correct amount of each treatment drug.
- You will receive the next 4 weeks supply of treatment drugs.

INFORMATION ABOUT THE TREATMENT FOR HIV-1 INFECTED CHILDREN

The initial treatment your child will receive while he/she is on the study will be zidovudine (AZT) + lamivudine (3TC) + lopinavir/ritonavir (**LPV/r**). If your child has a bad reaction to any drug, we will change his/her treatment to a regimen that might include one or more of the following drugs: didanosine (ddI), abacavir (ABC), stavudine (STV), efavirenz (EFV), nevirapine (NVP), nelfinavir (NFV) or ritonavir (RTV). If your child gets tuberculosis, one of the drugs may be replaced by ritonavir (RTV) or efavirenz (EFV)

Your study doctor will tell you exactly how to give the treatment to your child. Each symptom, side effect or injury your child might experience during the study should be reported to your study doctor, even if you think it may not be important.

WHAT ARE THE RISKS TO MY CHILD WHEN HE/SHE PARTICIPATES IN THE STUDY?

All of the medicines used in this study are approved by the US Food and Drug Administration (FDA) and the Medicines Control Council (MCC). However, the dosages of some of the medicines have not yet been approved for very young children. That means that some of the anti-HIV medicines are being used in an investigational manner. This includes Lopinavir/ritonavir (LPV/r) in children <6 months of age and prescribing Ritonavir at the full dose instead of increasing in a step wise manner. Higher doses of Ritonavir will be prescribed should your child require therapy against Tuberculosis.

The drugs used in this study may have side effects, some of which are listed below. Please note that these lists do not include all the side effects seen with these drugs. These lists include the more serious or common side effects with a known or possible relationship. If you have questions concerning additional study drug side effects, please ask the medical staff at your site.

NUCLEOSIDE ANALOGUES (NRTIs)

Lactic acidosis **(elevated lactic acid levels in the blood)** and severe hepatomegaly (enlarged liver) with steatosis (fatty liver) that may result in liver failure, other complications and death have been reported with the use of antiretroviral nucleoside analogues alone or in combination. The liver complication and death have been seen more often in women on these drug regimens. Some non-specific symptoms that might indicate lactic acidosis include: unexplained weight

loss, stomach discomfort, nausea, vomiting, fatigue, **cramps, muscle pain**, weakness and shortness of breath.

Zidovudine (AZT) Side Effects

The following side effects have been associated with use of zidovudine:

- Decrease in the number of white blood cells that help fight infection
- Decrease in the number of red blood cells that may cause weakness, dizziness, and fatigue
- Muscle aches, weakness and wasting
- Headache
- Upset stomach
- Vomiting
- Decrease in appetite
- Vague overall feeling of discomfort
- Lack of energy
- Feeling tired
- Sleeplessness
- Heartburn

Lamivudine (3TC) Side Effects

The following side effects have been associated with use of lamivudine:

- Headache
- Feeling tired
- Dizziness
- Numbness, tingling, and pain in the hands or feet
- Depression
- Trouble sleeping
- Rash
- Upset stomach, vomiting, nausea, loose or watery stools
- Pancreatitis (inflammation of the pancreas), which may cause death. If you develop pancreatitis, you may have one or more of the following: stomach pain, nausea, and vomiting
- Abnormal pancreatic and liver function blood tests

Participants who are infected with both Hepatitis B and HIV should be warned that their liver function tests may increase, and symptoms associated with hepatitis (an acute inflammation of the liver) may worsen after lamivudine has been stopped. Although most of these cases have resolved without treatment, some deaths have been reported.

Didanosine (ddI) Side Effects

The following side effects have been associated with use of didanosine:

- Pancreatitis (inflammation of the pancreas), which may cause death. If you develop pancreatitis, your child may have one or more of the following: stomach pain, nausea, and vomiting
- Deaths from liver failure have been reported in pregnant women receiving the combination of didanosine and stavudine with other anti-HIV drugs.
- Numbness, tingling, and pain in your hands and feet

- Abnormal vision changes
- Upset stomach, vomiting and loose or watery stools
- Headache
- Abnormal pancreatic function blood tests or abnormal liver function blood tests
- Increase in uric acid in the bloodstream
- **New information includes**
    - **Deaths from lactic acidosis have been reported in pregnant women receiving the combination of didanosine and stavudine.**
    - **Serious liver problems have happened in some people (including pregnant women) who take didanosine. These problems, which include liver failure and high blood pressure in the large vein of the liver, may lead to death.**

If you are developing liver problems, you may have one or more of the following:

- **Yellowing of the skin or whites of your eyes**
- **Dark urine**
- **Pain on the right side of your stomach**
- **Easy bruising or bleeding**
- **Loss of appetite**
- **Upset stomach or vomiting**
- **Vomiting blood or dark colored stools**

When didanosine is used with other medicines with similar side effects, these side effects may be seen more often and may be more severe than when didanosine is used alone. Participants, who take didanosine together with stavudine (d4T), with or without hydroxyurea, may be at greater risk for pancreatitis or liver problems or both. These conditions may result in death.

Abacavir (ABC) Side Effects

The following side effects have been associated with use of abacavir:
Participants taking abacavir may have a serious allergic reaction that involves several organs of the body. **This reaction can be severe or may rarely cause death. Your risk of this allergic reaction is much higher if you have a certain type of gene called HLA-B*5701. Your doctor can determine if you have this by doing a blood test. If you have tested positive for this blood test before, do not take abacavir and let your doctor know right away. If you have two or more of the following while taking abacavir, stop taking the abacavir and call your doctor right away:**

- Fever
- Rash
- Upset stomach, vomiting, loose or watery stools, abdominal pain
- General feeling of illness, extreme tiredness, achiness
- Cough, sore throat, shortness of breath.

This serious reaction usually appears within the first six weeks after starting this drug but can occur at any time during treatment. This reaction can be severe and can lead to death especially if abacavir is not stopped. IF YOU STOP GIVING ABACAVIR BECAUSE OF AN ALLERGIC REACTION, YOU SHOULD NEVER GIVE YOUR CHILD ABACAVIR OR ANY OTHER ABACAVIR-CONTAINING MEDICINE AGAIN.

This serious allergic reaction is also seen in children and the side effects are similar to those seen in adults. In addition to the side effects listed for adults, children may experience one or more of the following: weakness or sluggishness, poor appetite, difficulty breathing.

Note: A more severe or fatal allergic-type reactions can occur within hours after abacavir is restarted in participants who have interrupted abacavir therapy for any reason. If the abacavir is interrupted for any reason other than an allergic reaction, immediately contact the medical staff at the site. If the study doctor decides to restart abacavir, your child may need to be monitored more closely in the clinic or in the hospital.

IF YOU THINK YOUR CHILD MIGHT BE DEVELOPING A REACTION TO ABACAVIR, DO NOT GIVE YOUR CHILD ANY MORE DOSES AND CONTACT THE DOCTOR AT THE SITE IMMEDIATELY.

Other than a serious allergic reaction, additional side effects include:

- Upset stomach
- Vomiting
- Vague overall feeling of discomfort
- Feeling tired
- Decrease in appetite
- Loose or watery stools
- Pancreatitis (inflammation of the pancreas), with one or more of the following: stomach pain, nausea or vomiting
- Headache

Stavudine (STV) Side Effects

The following side effects have been associated with use of stavudine:

- Deaths from liver failure have been reported in pregnant women receiving the combination of stavudine and didanosine with other anti-HIV drugs.
- Participants who take stavudine together with didanosine, with or without hydroxyurea, may be at greater risk for pancreatitis or liver problems or both. These conditions may results in death.
- Numbness, tingling, and pain in your hands or feet
- Pancreatitis (inflammation of the pancreas), which may cause death. If your child develops pancreatitis, he/she may have one or more of the following: stomach pain, nausea, and vomiting.
- Rash
- Upset stomach, vomiting and loose or watery stools
- Abdominal pain
- Abnormal liver function blood tests or abnormal pancreatic function blood tests
- Rare cases of muscle weakness, which may progress to paralysis and inability to breathe. This may be associated with elevation of lactic acid in the blood.

When stavudine is used with other medicines with similar side effects, these side effects may be seen more often, and may be more severe, than when stavudine is used alone.

PROTEASE INHIBITORS (PIs)

The use of protease inhibitors may be associated with the following:

- Increases in the amount of triglycerides and/or cholesterol in the blood
- Development of diabetes or the worsening of high blood sugar

There have been reports of increased bleeding in HIV-infected persons with haemophilia who were treated with protease inhibitors, although it is not known if protease inhibitors were the cause of these bleeding episodes.

Lopinavir/ritonavir (LPV/r) Side Effects

The following side effects have been associated with use of lopinavir/ritonavir:

- Pancreatitis (inflammation of the pancreas), which may cause death. If your child develops pancreatitis, he/she may have one or more of the following: stomach pain, nausea, vomiting or abnormal pancreatic function blood tests
- Abnormal bowel movements (stools), including loose or watery stools, upset stomach and stomach pain
- Large increases in triglycerides and cholesterol in the blood
- Liver problems and worsening liver disease, possibly leading to death. People with these conditions may have abnormal liver function blood tests. **If you child develops liver problems, he/she may have one or more of the following: yellowing of the skin or whites of your eyes, dark urine, pain on the right side of your stomach, loss of appetite, upset stomach or vomiting, pale colored stools, itchy skin.**
- **Rash, which could blister, and may be severe or life-threatening. Contact your Healthcare Provider if your child develops a rash**
- Feeling weak/tired
- Headache

Additional safety information includes the following:

LPV/r may lead to changes in your child's heart rhythm and can lead to serious heart problems. The electrical activity of your child's heart may be seen on an EKG (electrocardiogram). LPV/r prolongs the PR interval in some patients (the time between the contracting and relaxing of the heart). Cases of atrioventricular block (a block to a valve in the heart) have been reported.

Your child's risk for these problems may be higher if he/she already has a history of abnormal heart rhythm or other types of heart disease or if your child takes other medicines that can affect his/her heart rhythm while taking LPV/r. Tell the doctor right away if your child has any of these symptoms while taking LPV/r: dizziness, lightheadedness, fainting and sensations of abnormal heartbeats.

Ritonavir (RTV) Side Effects

The following side effects **have been associated with use of ritonavir:**

- Feeling weak and tired
- Stomach and bowel problems including abdominal pain, upset stomach, vomiting, abnormal stools, and loose or watery stools
- Loss of appetite
- Headache
- Dizziness
- Abnormal increases in triglycerides and cholesterol in the blood
- Numbness and tingling in the arms, legs and around the mouth
- Rash
- Abnormal liver function blood tests which may be due to possible liver problems. Liver problems including cases of death have occurred in people taking ritonavir
- Fever
- A change in the sense of taste
- Pancreatitis (inflammation of the pancreas), which may cause death. If your child develops pancreatitis, he/she may have one or more of the following: stomach pain, nausea and vomiting.
- **Ritonavir prolongs the PR interval (the time between the contracting and relaxing of the heart) in some patients and should be used with caution in patients who have preexisting structural heart disease, conduction system abnormalities, or other cardiac diseases.**
- **Abnormal heart rhythm and electrocardiogram (EKG) changes. This may be felt as a "flutter" in the chest or an irregular pulse. If your child develops abnormal heart rhythm, he/she may experience light-headedness, fainting spells or an abnormal heart beat.**

Nelfinavir (NFV) Side Effects

The following side effects have been associated with use of nelfinavir:

- Loose or watery stools
- Upset stomach
- Gas
- Abdominal pain

- Rash
- Decrease in the number of white blood cells that help fight infection
- Abnormal liver enzyme test
- Allergic reactions, including bronchospasm, fevers and edema
- Jaundice and elevated bilirubin

Note: Participants with phenylketonuria should be aware that nelfinavir oral powder (Viracept Oral Powder) contains 11.2 mg phenylalanine per gram of powder.

Should your child need treatment for tuberculosis or need to go onto nelfinavir or ritonavir, we may take an extra half-teaspoonful of blood to check if a dose adjustment for the antiretrovirals is necessary

NON-NUCLEOSIDE REVERSE TRANSCRIPTASE INHIBITORS (NNRTIs)

Nevirapine (NVP) Side Effects

The following serious side effects have been associated with use of nevirapine:
Severe liver damage that can cause death may occur and is often associated with a rash. Being female or having a higher CD4 cell count, regardless of gender, increase the risk of developing liver damage.
Women with CD4 cell counts greater than 250, including pregnant women receiving chronic nevirapine therapy, are at even higher risk for developing liver damage. Men with CD4 cell counts greater than 400 are also at increased risk. However, these reactions can happen at any CD4 count in both men and women. Participants who have abnormal liver function tests before starting nevirapine and participants with active Hepatitis B or C infection are also at higher risk for liver damage.

If a participant is developing liver damage, one or more of the following may result:
- Tiredness
- General feeling of illness or flu-like feeling
- Loss of appetite
- Nausea
- Pale stools
- Dark urine
- Yellowing of the skin or whites of the eyes
- Liver tenderness or abnormal liver function tests

Hypersensitivity reactions ("allergic reaction") may occur. These reactions are rarely fatal. The symptoms that you may notice are: rash, fever, tiredness, muscle or joint aches, flu-like feeling, blisters, mouth sores, facial swelling, red eyes and irritation fo the eyes, general feeling of discomfort, and /or liver damage described above, kidney problems, and/or changes in white blood cell levels.

Muscle break down causing muscle aches or pain has been observed in some people experiencing skin and/or liver reactions associated with nevirapine.

Rash is the most common side effect associated with nevirapine. Rash occurs more often in women. Most rashes occur early during treatment. The rash may be severe and rarely may cause

death. One of the risk factors for developing serious skin reactions includes failure to take nevirapine properly during the first 14 days of treatment.

The risk of participants developing any of the serious side effects listed above is greatest during the first few months of treatment, but these side effects also can occur later. If your child develops any of the side effects listed above, no matter how long he/she has been receiving nevirapine, you must contact the study doctor right away and have your child seen by the study staff before the next dose. Your study doctor will instruct you on what to do next. If you and your doctors then decide to stop your child's treatment because of liver damage, hypersensitivity or severe skin reactions, your child should never take nevirapine again.

Other than the serious side effects listed above, additional side effects include:

- Fever
- Headache
- Upset stomach (nausea, vomiting)

Efavirenz (EFV) Side Effects

The following side effects have been associated with the use of efavirenz:

A small number of people may experience the following serious psychiatric problems:

- Depression, which may be severe
- Suicidal thoughts or attempts (rarely)
- Aggressive behaviour
- Psychosis-like symptoms, such as abnormal thinking, paranoia, and delusions

People with a history of psychiatric problems may be at greater risk for these serious psychiatric problems.

Side effects associated with the central nervous system may include the following:

- Dizziness
- Trouble sleeping
- Abnormal dreams
- Drowsiness
- Confusion
- Difficulty concentrating
- Hallucinations
- A feeling of strangeness and losing touch with reality
- An exaggerated feeling of well-being
- Agitation or anxiety

If alcohol or mind- or mood-altering drugs are used with efavirenz, it is possible that the central nervous system side effects could become worse.

Serious liver problems and worsening liver disease can occur. These problems can be life-threatening. People with these conditions may have abnormal liver function blood tests. If your child is developing liver problems, he/she may have one or more of the following:

yellowing of the skin or whites of your eyes, dark urine, pain on the right side of the stomach, loss of appetite, upset stomach or vomiting, pale colored stools, itchy skin.

Additional side effects include:

- Rash
- Upset stomach
- Loose or watery stools
- Headache
- Pancreatitis (inflammation of the pancreas), with one or more of the following: stomach pain, nausea or vomiting
- Hepatitis (inflammation of the liver)
- Abnormal increases in pancreatic and liver enzyme levels in the blood. Abnormal increases in the amount of triglycerides and cholesterol in the blood
- Abnormal vision
- Fever

Use of Combination Antiretroviral Drugs

The use of potent antiretroviral drug combinations may be associated with an abnormal placement of body fat and wasting. Some of the body changes include:

- Increase in fat around the waist and stomach area
- Increase in fat on the back of the neck
- Thinning of the face, legs, and arms
- Breast enlargement

Immune Reconstitution Syndrome: In some people with advanced HIV infection, signs and symptoms of inflammation from other infections may occur soon after anti-HIV treatment is started.

Risk of stopping medication (for patients in Arm 2 or 3):

Stopping anti-HIV drugs usually causes virus in your child's blood to go up and CD4 counts to go down, and your child's risk of becoming sick might increase.

Concomitant Medications Risks

There is the risk of serious and/or life-threatening side effects when non-study medications are taken with study drugs. For your child's safety, you must tell the study doctor or nurse about all the medications your child may be taking before your child starts the study and before your child starts taking any non-study medications while on the study. In addition, you should check with the study doctor or nurse before enrolling your child in any other clinical trials while your child is on the study.

Blood Drawing Risks

During the collection of blood samples, your child may experience light-headedness, pain, bleeding and/or bruising at the site on the arm where blood is taken. Blood clots may form and infections may occur, but this is unusual. Fainting may occur during or shortly after having blood drawn. If your child experiences faintness, he/she should lie down immediately to avoid possible injuries caused by falling and you should immediately notify the study staff.

What if your child develops a serious infection of the respiratory tract such as pneumonia

If your child develops a serious infection such as pneumonia, we will collect secretions to find the cause. This will be done immediately and will help us treat your child with the right medicines. Sometimes special tests are necessary to find the cause of the infection. We will therefore freeze some of the secretions for special tests that cannot be done right away to identify germs causing infection. In this way we will learn more about germs causing infections such as pneumonia. This test may be done years later and we will not be able to inform you of these results.

NEW FINDINGS

All new findings that become known during the study, and that might be important for you to decide if you want to keep your child in this study, will be made known to you as soon as possible. If you want the results of the study, let the study staff know.

WHAT ABOUT CONFIDENTIALITY?

Information about your HIV-status, your child's HIV status, your child's care in the clinic and his/her blood test results will be stored in locked cabinets. All medical information collected during the study will be treated as confidential and will be available only to staff members involved with the CHER trial who are directly involved in your child's care. Any information that is used for research or publication purposes will be kept confidential and will not have your or your child's name recorded on it.

It is possible that the regulatory authorities, such as the Medicines Control Council (MCC), the Ethics Committees, the US National Institutes of Health (NIH), monitors, the drug companies, and the Food and Drug Administration (FDA) may want to review the study documents (for example, the original blood result forms, and doctors notes) at a later stage in which case every effort will be made to protect your child's confidentiality.

If your child becomes ill and you choose to take him/her to another health care facility, we would advise you to inform the health care worker (HCW) that your child is participating in a clinical study. Please ask the HCW to contact your child's study doctor should they have any concerns. If it is necessary to access clinical information for the study, the study doctor may contact the health care facility where your child is receiving care for information about his/her condition and treatment.

YOUR CHILD'S PARTICIPATION IS VOLUNTARY

Your child's participation in the study is voluntary. You can refuse to let him/her participate or stop his/her participation at any time that you choose. Your child's withdrawal from the study will not affect his/her access to other medical care. If you wish to stop giving the trial medication to your child, please consult the doctor in charge of the study (Dr. Violari or Dr. Cotton). The study doctor will ask you if the following evaluations can be performed on your child's last visit:

- A physical examination.
- Your child's weight, height, heart rate, breathing rate (optional) and temperature will be measured. Prior to 2 years of age blood pressure will be measured when necessary; beginning at 2 years of age blood pressure will be routinely measured.
- You will be asked about any illnesses and/or reactions to the study drugs, hospitalisations or changes in medication given to your child since the last visit.

- Approximately 10 mL of blood (2 teaspoons) will be taken to check your child's FBC (test of the number of blood cells), LFTs (tests of liver and kidney function), and CD4+ (test to see how many immune/fighter cells are in the blood).
- If your child is on anti-tuberculosis medication, additional blood may be drawn to test for drug levels.
- A small amount of blood is stored and tested for drug levels, drug resistant virus and viral load (the amount of virus in blood) at a later stage. Your child's blood will not be used for genetic research without your permission.

You will also be asked why you wish to withdraw your child from the study. The doctor will supervise the discontinuation as your child's health is the first priority.

WHY WOULD THE DOCTOR TAKE MY CHILD OFF THIS STUDY EARLY?

The study doctor may need to take your child off the study early, without your permission, if:

- A Data Safety Monitoring Board (DSMB) recommends that the study be stopped or cancelled (A DSMB is an outside group of experts who monitor the safety of the study);
- The study is cancelled by the regulatory authorities, e.g., the Ethics Committees, the MCC, the US National Institutes of Health or the US Food and Drug Administration (FDA);
- You are unable to attend the study visits as required by the study; or
- The study doctor retains the right to withdraw your child from the study if it is considered to be in his/her best interest.

WHAT OTHER TREATMENT OPTIONS DOES MY CHILD HAVE BESIDES THIS STUDY?

The South African government is providing treatment with anti-HIV medicines at this site and across the country. That means that in the future your child will be able to get anti-HIV drugs that are sponsored by the government and will be available through many hospitals. Prescription anti-HIV drugs are also available for purchase. Please talk to your doctor about these and other options available to your child. Your doctor can explain the risks and benefits of these options.
At the conclusion of the trial, your child will be referred to the public service program through your district.

INSURANCE

Compensation for any injury caused by taking part in this study will be in accordance with the guidelines of the Association of the British Pharmaceutical Industry (ABPI). Broadly speaking the ABPI guidelines recommend that the PHRU at the Chris Hani Baragwanath Hospital or KID-CRU, Ward J8 at Tygerberg Children's Hospital, without legal commitment, should compensate you without you having to prove that it is at fault. This applies in cases where it is likely that such injury results from giving any new drug or any other procedure carried out in accordance with the protocol for the study. The PHRU at the Chris Hani Baragwanath Hospital or KID-CRU, Ward J8 at Tygerberg Children's Hospital, will not compensate you where such injury results from any procedure carried out which is not in accordance with the protocol for the study. Your right at law to claim compensation for injury where you can prove negligence is not affected. Copies of these guidelines are available upon request.

ARE THERE POTENTIAL BENEFITS FOR MY CHILD FOR BEING IN THE STUDY?

By letting your child participate in the study, his/her health may improve with the anti-HIV drugs. However, no direct benefit can be guaranteed. This trial will help us learn whether all infected children should be started on treatment early in life or if we can wait until the immune system weakens. It will also tell us if we can stop treatment for a while once the body is stronger.

WHAT HAPPENS IF MY CHILD IS INJURED?

A study-related injury or illness is one that occurs as a direct result of the administration of the study medicine or study-specific procedures. If your child is injured as a result of being in this study, the Perinatal HIV Research Unit at the Chris Hani Baragwanath Hospital or KID-CRU, Ward J8 Tygerberg Children's Hospital will give your child immediate necessary treatment for the injuries. There is no program for compensation through the US National Institutes of Health (NIH).

IF YOUR CHILD HAS TO PERMANENTLY STOP TAKING STUDY-PROVIDED MEDICATION (ANTIRETROVIRALS (ARVs)) OR ONCE YOUR CHILD LEAVES THE STUDY, HOW WOULD THE MEDICATION BE PROVIDED?

During the study:

If your child must permanently stop taking study-provided ARVs before your child's study participation is over, the study staff will discuss other options that may be of benefit to your child.

After the study:

After your child has completed his/her study participation, the study will not be able to continue to provide you with the ARVs your child received on the study. If continuing to take these or similar ARVs would be of benefit to your child, the study staff will discuss how you may be able to obtain them.

WHAT IF MY CHILD NEEDS EMERGENCY CARE AND HOSPITALISATION?

If your child needs emergency care, or if hospitalisation is required at any time during the study, or up to two months after taking the last dose of study medicine, please tell the treating doctor that your child is/was enrolled in this research study and that the study doctors must be informed.

WHAT ARE THE COSTS TO ME?

Neither you nor your child's medical scheme will be expected to pay for any study medication, study related visits or study procedures.

WILL I RECEIVE ANY PAYMENT FOR MY CHILD'S PARTICIPATION?

You will not receive payment for your child's participation in the study, although you will receive R150 for transport expenses to the clinic for each scheduled study visit.

WHAT DO I DO IF I HAVE QUESTIONS OR PROBLEMS?

If at any time between your visits, you feel that any of your child's symptoms are causing you any problems, or you have any questions during the study, please do not hesitate to contact the doctor in charge of the study or the study staff.

The 24-hour telephone number that you can reach the doctor in charge or another authorized person is (082) 456-8554 for Johannesburg or (084) 641-0735 (a/h) or (021) 938-4302 (o/h) for Cape Town.

If you want any information regarding your child's rights as a research participant, or have complaints regarding this research study, you may contact:

Johannesburg:
Prof. Cleaton-Jones, Chairperson
The University of the Witwatersrand
Human Research Ethics Committee
Tel: (011) 717-2229

Cape Town:
Prof. B. Warren, Acting Chairperson
University of Stellenbosch
Committee for Pharmaceutical Trials
Tel: (021) 938-9075

These independent committees are established to help protect the rights of research participants and gave written approval for the study protocol.

If you have questions about this trial you should first discuss them with your doctor or the Ethics Committee. If you have not been provided with answers to your satisfaction, you should write to the South African Medicines Control Council (MCC) at:
The Registrar
South African Medicines Control Council
Department of Health
Private Bag X828
Pretoria, 0001
Fax: (012) 323-4474
E-mail: labusa@health.gov.za

CHER Trial
STUDY INFORMED CONSENT SIGNATURE PAGE

Name of participant: _____

To parents/legal guardians:

Have you read the information sheet about this study, or has someone read it to you and to your satisfaction?	Yes ☐	No ☐
Have you had an opportunity to ask questions and discuss this study?	Yes ☐	No ☐
Have you received satisfactory answers to all your questions?	Yes ☐	No ☐
Have you received enough information about this study?	Yes ☐	No ☐
Do you understand that you are free to withdraw your child from this study at any time?	Yes ☐	No ☐
Do you agree to let your child take part in this study?	Yes ☐	No ☐
Do you consent to the storage of your child's blood for drug levels, virus resistance testing and viral load at a later stage? (Your child may participate in the study even if you decline permission to store blood.	Yes ☐	No ☐
Do you consent to your child's participation in the extra study that collects dried blood spots and tests strength of the antibody?	Yes ☐	No ☐
Do you consent to the storage of your child's respiratory secretions for testing at a later stage? (Your child may participate in the study even if you decline permission to store respiratory secretions).	Yes ☐	No ☐

Name of Parent/Legal guardian_____

Signature _____ Date_____
(Parent/Legal guardian)

Name _____ Signature _____
Witness (if applicable) Witness (if applicable)

Date _____

I am satisfied that the parent/legal guardian understands what the CHER study is about and that his/her questions have been answered.

Name _____ Signature_____
Person obtaining consent Person obtaining consent
Date _____ Time_____

Instruction to the site: This addendum to the Informed Consent can be used for participants who have signed previous addendums to informed consents for LoA 1 and 2. If not, then the full version 3.0 ICF must be signed.

ADDENDUM TO THE PARTICIPANT INFORMATION LEAFLET AND STUDY INFORMED CONSENT FORM

Title of the study: A Phase III, Randomised, Open-label Trial to Evaluate Strategies for Providing Antiretroviral Therapy to Infants Shortly after Primary Infection in a Resource Poor Setting

Short title of the study: Children with HIV Early Antiretroviral Therapy
CHER Trial

Sponsored by: The National Institute of Allergy and Infectious Disease
Division of AIDS (DAIDS)
6700B Rockledge Drive
Bethesda, Maryland 20852
United States

Protocol co-chairs:

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Perinatal HIV Research Unit
University of the Witwatersrand
New Nurses Home
Chris Hani Baragwanath Hospital
Old Potch Road
Soweto, 2013
Johannesburg, South Africa
Tel: (011) 989 9707 or (082) 857-4126

Prof. Mark Cotton
Children's Infectious Diseases Clinical Research Unit (KID-CRU), Ward J8
University of Stellenbosch
Tygerberg Children's Hospital
Francie van Zijl Avenue
Parow, 7505
Cape Town, South Africa
(021) 938-4219 or (083) 379-5283

Dear Parent
You have already signed informed consent giving permission for your child to be enrolled in this study. We are asking you to sign this addendum to the informed consent to notify you of the safety update to Didanosine (ddI) and additional criteria for restarting treatment. Information located in the main consent that is not mentioned in this addendum is still applicable to your child's participation.

The following side effects have been associated with the use of didanosine:

- You have already been informed of Pancreatitis (swelling of the pancreas), which may cause death. If your child develops pancreatitis, he/she may have one or more of the following: stomach pain, nausea, and vomiting.
- New information includes:
    - **Deaths from lactic acidosis have been reported in pregnant women receiving the combination of didanosine and stavudine.**

- **Serious liver problems have happened in some people (including pregnant women) who take didanosine. These problems, which include liver failure and high blood pressure in the large vein of the liver, may lead to death.**

If you are developing liver problems, you may have one or more of the following:

- **Yellowing of the skin or whites of your eyes**
- **Dark urine**
- **Pain on the right side of your stomach**
- **Easy bruising or bleeding**
- **Loss of appetite**
- **Upset stomach or vomiting**
- **Vomiting blood or dark colored stools**

You have also already been informed about additional side effects associated with the use of didanosine include:

- Numbness, tingling, and pain in the hands or feet
- Abnormal vision changes
- Upset stomach, vomiting and loose or watery stools
- Headache
- Abnormal pancreatic function blood tests or abnormal liver function blood test
- Increase in uric acid in the bloodstream

Remember that when didanosine is used with other medicines with similar side effects, these side effects may be seen more often and may be more severe than when didanosine is used alone. People who take didanosine together with stavudine, with or without hydroxyurea, may be at greater risk for pancreatitis or liver problems or both. These conditions may result in death.

RESTARTING TREATMENT

If your child has interrupted treatment, we will restart therapy if the CD4% drops to < 25% or CD4 count <1000cells/mm^3 regardless of CD4% if ≤12 months of age; below 20% if 12-**59** months of age; **below 20% or CD4 count <350cells/mm^3 if ≥ 60 months,** or if your child develops a severe CDC Stage B or Stage C disease). They will be given the same ART that they took earlier (before therapy was stopped) to ensure that they do not become sicker. If the first regimen of ART stops working, the study doctors will consider giving the infants a second regimen of ART. The study doctors will consider giving your child a second regimen of ART if your child's CD4% drops to <20%, if your child develops a severe CDC Stage B or Stage C disease**, if your child's viral load goes up** or if your child experiences severe side effects which prevent continued use of that specific drug regimen.

HOW MANY CHILDREN WILL TAKE PART AND HOW LONG WILL MY CHILD BE IN THIS STUDY?

The final number of children on study is **451.** We initially informed you that the duration of the study will be approximately 5 years. However, in order to ensure that all the participants have at

least 3.5 years on study, we will be continuing with follow up visits for another year until this time point is reached.

ADDENDUM TO STUDY INFORMED CONSENT SIGNATURE PAGE

Name of participant: _____

To parents/legal guardians:

Have you read this addendum to the information sheet about this study, or has someone read it to you and to your satisfaction?	Yes ☐	No ☐
Have you had an opportunity to ask questions and discuss this addendum?	Yes ☐	No ☐
Have you received satisfactory answers to all your questions?	Yes ☐	No ☐

Parent/Legal Guardian:

_____	_____	_____	☐☐☐☐
Name	Signature	Date	Time

Witness (if applicable):

_____	_____	_____	☐☐☐☐
Name	Signature	Date	Time

Person conducting informed consent process:

I am satisfied that the parent/legal guardian understands what the addendum to the informed consent is about and that his/her questions have been answered.

_____	_____	_____	☐☐☐☐
Name	Signature	Date	Time

Appendix Table 1: All Grade 3 or 4 Events in 377 infants (ART-Def, ART-40W and ART-96W)

Variable	ART-Def	ART-40W	ART-96W
Total	**170**	**116**	**87**
Herpes	0	2	1
BCG lymphadenitis	7	2	0
CMV	5	0	0
Candida	4	2	3
Tuberculosis	3	2	1
Upper respiratory tract infection	5	7	0
Lower respiratory tract infection	40	27	31
Urinary Tract Infection	16	6	6
Fracture	1	1	1
Gastrointestinal tract	49	29	25
Anorexia	2	0	2
Hepatitis	2	1	0
Neurological abnormality	14	9	7
Cardiac	1	2	1
Skin Infestations	5	4	0
Dermatitis	2	5	2
Other*	2	3	0

*__Other__: Burns, Lipoatrophy, Hepatomegaly
Gastrointestinal Tract: Diarrhea, Acute Gastroenteritis, Dysentery, Gastroenteritis
Candida: Oral, Skin, Oesophageal
Neurological abnormality: AIDS encephalopathy, HIV Wasting syndrome, Brain disorder, Cerebral palsy, Hydrocephalus, Spastic diplegia, febrile seizures, focal seizures, seizures
Cardiac: Cardiac failure, Myocarditis, Cardiomyopathy
CMV: Viraemia, Colitis, Hepatitis, Pneumonia

Appendix Table 2: Baseline Characteristics of ART-40W and ART-96W after additional recruitment: Health Status

Variable	ART-40W		ART-96W		Total	
Number of participants enrolled	**143**		**143**		**286**	
Age at randomization						
Median (IQR)	7·6	(6·7 to 8·9)	7·6	(6·6 to 9·0)	7·6	(6·6 to 8·9)
Mother/Child receiving ART for PMTCT						
No Therapy	13 (9 %)	19 (13 %)	13 (9 %)	19 (13 %)	26 (9 %)	38 (13 %)
*NVP (%)	100 (70 %)	78 (55 %)	93 (65 %)	74 (52 %)	193 (67 %)	152 (53 %)
AZT (%)	7 (5 %)	3 (2 %)	3 (2 %)	3 (2 %)	10 (3 %)	6 (2 %)
AZT + NVP (%)	21 (15 %)	33 (23 %)	32 (22 %)	39 (27 %)	53 (19 %)	72 (25 %)
HAART (%)	2 (1 %)	0 (0%)	2 (1 %)	0 (0 %)	4 (1 %)	0 (0 %)
Birth weight (kg)						
< 2·5 (%)	19	(13·3 %)	23	(16·1 %)	42	(15 %)
[2·5 to 3.0) (%)	52	(36·4 %)	52	(36·4 %)	104	(36 %)
[3.0 to 3.5) (%)	52	(36·4 %)	56	(39·2 %)	108	(38 %)
> 3.5 (%)	20	(14·0 %)	12	(8·4 %)	32	(11 %)
Median (IQR)	3	(2·7 to 3·2)	2·9	(2·7 to 3·3)	3	(2·7 to 3·3)
Baseline Weight (kg)						
Median (IQR)	4·4	(4.0 to 4.9)	4·4	(3·8 to 5·0)	4·4	(4·0 to 4·9)
Weight Z - score						
Median (IQR)	-0·8	(-1·3 to -0·1)	-0·7	(-1·6 to 0·0)	-0·7	(-1·5 to -0·04)
CDC Classification						
Class N (%)	111	(77·6 %)	120	(83·9 %)	231	(81 %)
Class A (%)	26	(18·2 %)	14	(9·8 %)	40	(14 %)
Class B (%)	6	(4·2 %)	9	(6·3 %)	15	(5 %)
CD4 %						
Median (IQR)	35·2	(29·0 to 41·6)	34·3	(29·0 to 40·0)	35·0	(29·0 to 40·8)
CD4 Count (cells/mm^3)						
Median (IQR)	1985·5	(1476 to 2789)	2070	(1537 to 2763)	2012	(1518 to 2762)
Screening viral loads (X 1,000/ml)						
(10-100]	6	(4 %)	7	(5 %)	13	(5 %)
(100-500]	50	(35 %)	29	(20 %)	79	(28 %)
(500-750]	16	(11 %)	21	(15 %)	37	(13 %)
>750	71	(50 %)	86	(60 %)	157	(55 %)
Mean $\log_{10}$ viral load (std)	5·6	(0·3)	5·7	(0·4)	5·7	(0·3)

* Single dose nevirapine

Appendix Table 3: Primary endpoints including additional 34 children with baseline CD4 ≥25%

Variable	ART-40W	ART-96W
Number of Participants Enrolled	**143**	**143**
Total	**35 (24.5%)**	**30 (21·0%)**
Death	**11 (7·7%)**	**10 (7·0%)**
Immunological Failure on Therapy	**17(11·9%)**	**14(9·8%)**
Failure to reach a level > 20% by week 24	9(6·3%)	6(4·2%)
CD4% fell below 20% on two occasions after 24 weeks on therapy	8(5·6%)	8(5·6%)
Clinical Failure on Therapy	**6(4·2%)**	**5(3·5%)**
Severe Stage B Disease	1(0·7%)	2(1·4%)
CDC C Disease	5(3·5%)	3(2·1%)
Virological Failure on Therapy	**1(0·7%)**	**1(0·7%)**
Failure due to ART Limiting Toxicity	**0(0%)**	**0(0%)**

Pairwise comparison hazard ratio for ART-96W vs ART-40W: HR 0·843; 95% CI (0·518 to 1·373)

Appendix Table 4 (a): Clinical events for 2 early ART arms including additional 34 children with baseline CD4 ≥25%

Number of events	ART-40W (N=143)	ART-96W (N=143)	P-values
Deaths (Rate per 100 person years)	11 (2)	12 (2)	0·8
Severe Stage B Disease Events (Rate per 100 person years)	3 (0)	0 (0)	.
Other significant Events* (Rate per 100 person years)	15 (2)	13 (2)	0·7
CDC Stage C Events (Rate per 100 person years)	10 (2)	6 (1)	0·3
Total Clinical Events (Rate per 100 person years)	**39 (6)**	**31 (5)**	**0·4**
Total number of participants with clinical events	**23**	**18**	
Timing of events			
Primary Therapy (Rate per 100 person years)	15 (14)	27 (11)	
Interruption (Rate per 100 person years)	15 (8)	3 (2)	
Continuous Therapy (Rate per 100 person years)	9 (3)	1 (1)	
Severe Stage B Disease	**18**	**13**	
Cardiomyopathy	1	0	
Chronic Lung Disease	2	0	
Failure To Thrive	14	11	
Recurrent Pneumonia	1	2	
CDC Stage C Disease	**10**	**6**	
CMV Disease	0	0	
Disseminated Tuberculosis	0	3	
HIV Encephalopathy	6	2	
HIV Wasting Syndrome	4	1	
Other significant events			
Failure To Thrive	14	11	
Recurrent Pneumonia	1	2	
No. of hospitalizations (No. of children)	95(55)	86(56)	0·54
Total number of days hospitalized	558	533	
Hospitalization rates per 100 person years	16	14	
End of trial status			
Median CD4%	31	30	
Median CD4 Count	1092	959	
Viral Load > 1,000 copies/ml on ART	20	13	
Viral Load < 400 copies/ml on ART	130	133	
Median WAZ (IQR)	-0·33(-0·97 to -0·02)	-0·68 (-1·2 to -0·04)	
Median HAZ (IQR)	-0·7(-1·4 to -0·41)	-1·1 (-1·8 to -0·55)	
Median WHZ (IQR)	0·31(-0·49 to 0·98)	0·21(-0·29 to 0·59)	
Off ART	31	46	
Median time off ART in weeks (IQR)	211 (149 to 232)	153 (117 to 181)	

***** Other significant events: Failure to thrive and recurrent pneumonia

Appendix Table 4b: Adverse clinical events including additional 34 children with baseline CD4 ≥25%

Variable	ART-40W (N=143)	ART-96W (N=143)	P-value
Total Grade 3 or 4 Clinical Events	**124**	**95**	
Rate per 100 person years	**20**	**16**	**0·07**
Total Study Drug Related Grade 3 or 4 Clinical Events	**3**	**2**	
Acute Renal Failure[1]	1	0	
Gastroenteritis[1,2]	1	1	
Lipoatrophy[3]	1	0	
Myocarditis[1]	0	1	
Total Grade 3 or 4 Laboratory Events	**48**	**37**	
Rate per 100 person years	**8**	**6**	**0·3**
Total Study Drug Related Grade 3 or 4 Laboratory Events	**18**	**17**	**0·87**
Gamma Glutamyl Transpeptidase	0	2	
Alanine or Aspartate Aminotransferase	3	1	
Anemia	8	3	
Neutropenia	5	8	
Thrombocytopenia	1	3	
Creatinine	1	0	

[1]Probably not related, [2]Possibly related, [3]Probably related

Appendix Table 5: All Grade 3 or 4 Events in early ART arms including additional 34 children with baseline CD4 ≥25%

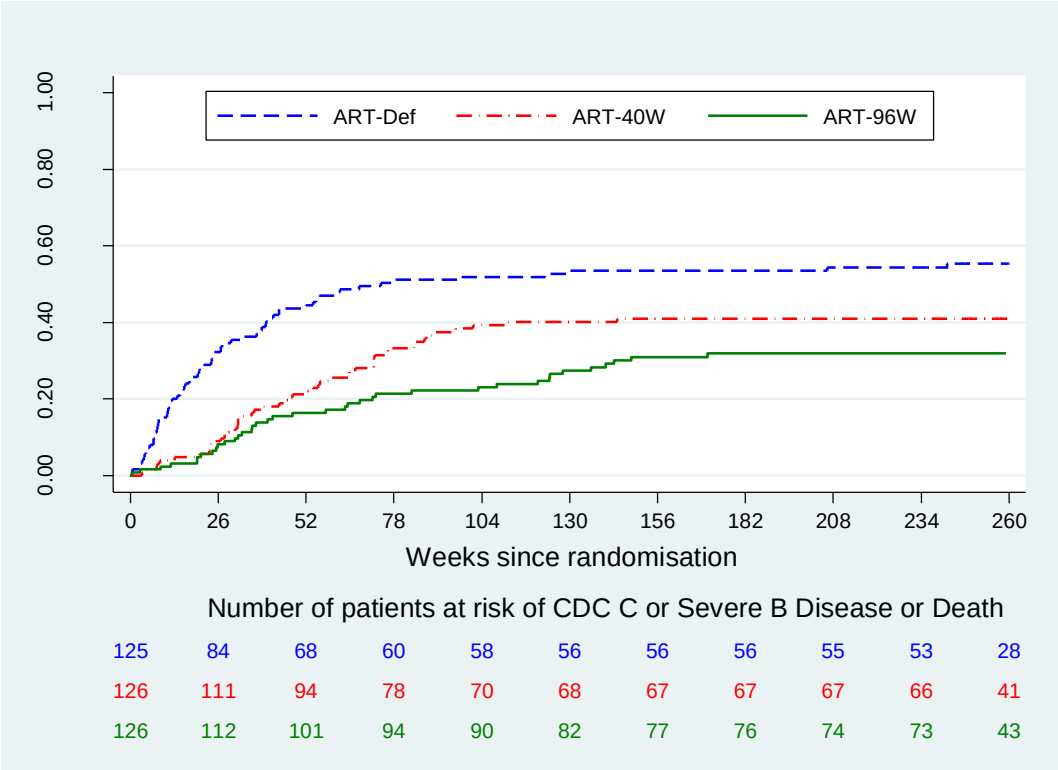
Variable	ART-40W	ART-96W
Total	**122**	**93**
Anorexia	0	2
BCG lymphadenitis	2	0
CMV	0	0
Candida	2	3
Cardiac	2	1
Dermatitis	5	2
Failure to thrive	15	10
Fracture	1	1
Gastrointestinal tract	31	27
Hepatitis	1	0
Herpes	2	1
Lower respiratory tract infection	28	31
Neurological abnormality	11	7
Skin infestations	4	0
Tuberculosis	2	1
Upper respiratory tract infection	7	0
Urinary tract infection	6	7
Other*	3	0

*__Other:__ Burns, Lipoatrophy
Gastrointestinal Tract: Diarrhea, Acute Gastroenteritis, Dysentery, Gastroenteritis
Candida: Oral, Skin, Oesophageal
Neurological abnormality: AIDS encephalopathy, HIV Wasting syndrome, Brain disorder, Cerebral palsy, Hydrocephalus, Spastic diplegia, febrile seizures, focal seizures, seizures
Cardiac: Cardiac failure, Myocarditis, Cardiomyopathy
CMV: Viraemia, Colitis, Hepatitis, Pneumonia

Appendix Figure 1: Time to primary endpoint (ART-40W, ART-96W) including additional 34 children with baseline CD4 ≥25%

Appendix Figure 2: Progression to CDC Stage C disease or Severe Stage B Disease or Death in 377 children with baseline CD4 ≥25%
(All site reported events)

Variable	Hazard Ratio	95% CI	P-Value
ART-96W – ART-40W	0·706	(0·462-1·079)	0·1081
ART-96W – ART-Def	0·424	(0·285-0·631)	<·0001
ART-40W – ART-Def	0·575	(0·398-0·831)	0·0032



Appendix Figure 3: Progression to CDC Stage C disease or Severe Stage B Disease or Death in 2 early ART arms including additional 34 children with baseline CD4 ≥25%
(Endpoint review committee accepted events)

Variable	Hazard Ratio	95% CI	P-Value
ART-96W – ART-40W	0·709	(0·372-1·350)	0·2951