INFORMATION TO USERS

HUMAN IMMUNODEFICIENCY VIRUS AND WEIGHT

OUTCOMES OF INFANTS IN KISUMU, KENYA

by

Cara Lea Frankenfeld

A Thesis Submitted to the Faculty of the

DEPARTMENT OF NUTRITIONAL SCIENCES

In Partial Fulfillment of the Requirements
For the Degree of

MASTER OF SCIENCE

In the Graduate College

THE UNIVERSITY OF ARIZONA

2000

STATEMENT BY AUTHOR

This thesis has been submitted in partial fulfillment of requirements for an advanced degree at the University of Arizona and is deposited in the University Library to be made available to borrowers under rules of the Library.

Brief quotations from this thesis are allowable without special permission, provided that accurate acknowledgment of source is made. Requests for permission for extended quotation from or reproduction of this manuscript in whole or in part may be granted by the head of the major department or the Dean of the Graduate College when in his or her judgment the proposed use of the material is in the interests of scholarship. In all other instances, however, permission must be obtained from the author.

SIGNED: _____

APPROVAL BY THESIS DIRECTOR

This thesis has been approved on the date shown below:

_____   _____
Douglas Taren, Ph.D.   Date
Associate Professor of Nutritional Sciences

ACKNOWLEDGEMENTS

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

ABSTRACT

Almost 600,000 infants acquire human immunodeficiency virus (HIV) infection from their mothers each year, with the majority of these infants living in developing countries. Knowledge regarding the impact of maternal and infant HIV infections upon birthweight is controversial. Little is known regarding the presence of HIV infection upon concurrent growth in developing countries. Data from a cohort of HIV-positive and HIV-negative women and their infants in Kisumu, Kenya was analyzed to assess maternal and infant HIV status with birthweight, growth and mortality of the infants. Three hundred and seventy-nine infants were assessed for health at four week intervals for the first year of life. The results of the analyses suggest that although differences in birthweight by HIV status alone are not present, HIV-infected infants subsequently gain less weight in the first year of life. Lower weight gain and positive HIV-status were independent predictors of mortality.

INTRODUCTION

Statement of the Problem

It has been estimated that 5.6 million people were newly infected with the human immunodeficiency virus (HIV) in 1999, bringing the estimated total number of people living with HIV or AIDS to 33.6 million (1). Approximately 95% of the people infected with HIV live in developing countries. Sub-Saharan Africa is home to the majority of HIV cases, with an estimated 23.3 million adults and children living with HIV. This translates to approximately seventy percent of total cases living in a region in which only about ten percent of the world's population lives. Among the people newly infected with HIV in 1999, an estimated 570,000 were children under the age of fifteen. Approximately ninety percent of these children were infants who acquired the infection from their mothers; in utero, intrapartum or postpartum through breastmilk. Additionally, ninety percent of the children newly infected with HIV live in Sub-Saharan Africa (1).

Due to the increasing numbers of infants infected with HIV, it is important to understand the impact of HIV upon other health outcomes, such as growth. A limited number of longitudinal growth studies of infants examining HIV status have been conducted in developing countries. In many of the earlier studies, antibody testing was utilized to determine HIV status. Since maternal HIV antibodies are present in infants into the second year of life, antibody testing cannot be used to determine timing of HIV transmission in HIV-positive breastfeeding populations. This is an important consideration because it is not known whether HIV infection impairs growth of infants

who were previously not growth impaired. This study will look at patterns of growth in infants with consideration for the timing of their infection in the first year of life.

Review of the Literature

Birthweight

Birthweight is an important component of infant morbidity and mortality. Infants with low birthweight (LBW), weighing less than 2500 grams at birth, are at increased risk of death during infancy, particularly in the neonatal period (2). This increased risk has been shown to persist after controlling for birth order, maternal human immunodeficiency virus (HIV) and syphilis infections, maternal education, and socioeconomic status (SES) (3). The relationship of birthweight with morbidity is less well characterized (2). Related to HIV infection, LBW was documented to be associated with intrauterine transmission of HIV to infants in New York City (4). The LBW infants had a four times greater risk of intrauterine transmission, after adjusting for AZT use during pregnancy, maternal CD4+ counts, and duration of membrane rupture (4). This underscores the importance of understanding whether and how a relationship exists between HIV and birthweight.

There are some important considerations for interpreting studies of birthweight in HIV-infected populations. The outcome of interest is usually whether maternal HIV status or infant HIV status influences birthweight. When considering maternal status, it is important to control for other variables known to influence birthweight, such as smoking, concurrent infections, or socioeconomic status (SES). Determining the impact

of infant HIV status and birthweight is more difficult. With studies that utilized antibody testing for the infant in breastfeeding populations, it is not possible to determine the actual timing of HIV transmission for the infants who became HIV-positive. Viral culture or polymerase chain reaction (PCR) tests can be performed on infant blood earlier than 18 months to accurately determine infant HIV status. Recently, the importance of distinguishing between intrauterine and intrapartum transmission has been established, because risk factors may differ between these two types of vertical transmission. To make this distinction, an early PCR test must be administered for the infant. It is recommended to test infant blood within 48 hours following birth (5). Since a rapid increase in sensitivity of PCR occurs in the first week of life, the further from birth the measurement is taken, the less likely a distinction between intrauterine and intrapartum transmission can be made (6). However, umbilical cord blood is not a reliable assessment of infant HIV status, because of potential contamination with maternal blood.

Inconsistent results have been observed regarding the significance of maternal HIV infection with birth size between developed and developing countries. Most studies conducted in developed countries observed no effect of maternal HIV infection on birthweight. Mean birthweight was not observed to be significantly different between HIV-positive and HIV-negative women in five studies conducted in developed countries (7-11). LBW was observed more frequently in HIV-positive women compared with HIV-negative women in one study conducted in New York (12), but not in two others (7, 13) conducted in Maryland and Scotland. Twenty-nine percent of the HIV-positive women in the New York study gave birth to an infant with LBW, compared with

approximately nine percent of HIV-negative women (12). After adjustment for illicit drug and cigarette use, SES and prenatal care, the HIV-positive women had a two-fold higher risk (odd ratio: 2.09, 95% confidence interval: 1.54-2.69) of delivering a LBW infant.

Significant effects of HIV infection in pregnant women on birthweight have been observed more frequently in developing countries. HIV-positive women in the Democratic Republic of Congo, Rwanda, and Haiti were observed to give birth to infants with lower mean birthweight than infants born to HIV-negative women (14-16). The difference in mean birthweights between infants born to HIV-negative and HIV-positive women ranged from 119 grams (14) to 170 grams (15) lower for infants of HIV-positive women. LBW has also been observed to be significantly lower for infants born to HIV-positive women compared with HIV-negative controls (14, 16-19). HIV-positive women consistently have a greater incidence of LBW infants compared with HIV-negative women. These differences in LBW between women by HIV status have ranged from 7.2% to 13.1%, with one study reporting a 28% rate of LBW incidence in HIV-positive women (14, 16, 18). The increased risk of giving birth to an infant with LBW persisted after adjustment for variables known to influence birthweight. After adjustment for sociodemographic factors and sexual history variables, the observed odds were 1.5 (95% CI: 1.0-2.4) for LBW with maternal HIV infection (14). After adjustment for SES and concurrent infections, the observed odds ratio for LBW with maternal HIV infection was 5.7 (95% CI: 2.5-12.8) (18).

Additionally, a meta-analysis was conducted utilizing seventeen prospective cohort studies from both developing and developed countries (20). An overall odds ratio for LBW of 2.04 (95% CI: 1.86-2.35) was predicted for HIV-positive women. Not surprising, the authors noted significant heterogeneity between the studies (20). Not all studies conducted adjusted for potential confounders, nor could they adjust for the same confounders. This does indicate the need for careful interpretation of the results.

Different results have also been observed for the significance of infant HIV infection upon birthweight. Many earlier studies conducted in developing countries cannot be interpreted for this result due to antibody testing in breastfeeding populations. In non-breastfeeding populations of HIV-positive women, antibody testing can distinguish whether the infection was present at birth. In five such studies in non-breastfeeding populations, no significant difference was observed for mean birthweight and HIV-positive infants, compared with uninfected infants born to HIV-positive women (seroreverters) (21-25). Distinction between intrauterine and intrapartum transmission cannot be made when HIV infection is not assessed within the first week of life. This distinction is important because intrauterine transmission may be causative for lower birthweight, whereas lower birthweight may be causative for intrapartum transmission. Two known studies that utilized an early PCR test observed different results regarding the association of LBW with presumed intrauterine and intrapartum transmissions (5, 26). The number of subjects and the covariates considered were similar between the two studies. Mock et al. studied a cohort of 218 formula-fed infants in Thailand. Twelve of the infants had presumed intrauterine transmission and thirty-seven infants had presumed

intrapartum transmission. Kuhn et al. studied a cohort of 276 infants of HIV-positive women in New York. In this study, 12 infants had presumed intrauterine transmission and 36 had presumed intrapartum transmission. Both studies controlled for obstetric factors, maternal immunological parameters at birth, and length of gestation. Mock et al. observed that LBW was associated with presumed intrauterine transmission. The adjusted odds ratio for this outcome was 5.2 (1.3-18.9) (26). Kuhn et al. observed that LBW was associated with presumed intrapartum transmission, with an adjusted odds ratio of 4.26 (1.57-11.56) (4). However, they also observed an association of small-for-gestational age and presumed intrauterine transmission (4). The different results noted may reflect different proportion of infants with LBW and presumed intrapartum transmission present in the studies. Only two of the thirty-seven infants with presumed intrapartum transmission were observed to have LBW in Thailand, but nineteen were observed to have LBW in New York. Further research is needed to elucidate an understanding of the association of birth size and presumed intrauterine and intrapartum transmissions.

Growth

Growth is an important predictor of morbidity and mortality for HIV-infected infants. Growth impairment has been observed to be associated with the occurrence of persistent diarrhea (27) and bacterial lung infections in HIV-infected infants (28). The presence of these illnesses also increased the risk for mortality (19, 27, 29). Most studies indicate that, among HIV-infected children, impaired weight growth is predictive for

survival (30-33). However, the impact of impaired length growth is less well defined. Carey et al. noted that both weight and height velocity provided significant information of the risk of death (32), but McKinney et al. observed no predictive value of initial height-for-age z-score and height growth rates for survival (33). The results of these studies suggest that maintaining adequate growth, particularly weight, of HIV-infected infants could be important for decreasing morbidity and increasing survival time.

Although some studies indicate that infants born to HIV-positive women exhibit similarities in birth size, regardless of the infant's HIV status, the growth patterns of infected infants differ from those of seroreverted infants. Lower length-for-age and weight-for-age values are commonly observed for HIV-infected infants, but decreased weight-for-length is not as frequently observed. However, significant differences in growth between uninfected infants born to HIV-positive mothers and infants born to HIV-negative mothers have not been observed. When seroreverted infants are observed to be smaller at birth, they appear to exhibit a catch-up growth that does not occur in the HIV-infected infants. After this catch-up growth, seroreverted infants exhibit growth similar to infants born to HIV-negative women.

Limited studies are available assessing longitudinal growth of children born to HIV-positive women, particularly in developing countries (15, 30, 34, 35). Three of these studies were conducted utilizing antibody testing for HIV determination in children (30, 34, 35). Since these are populations in which essentially all women breastfeed, actual timing of transmission could not be known due to antibody testing. The timing of

transmission is an important consideration, because differences in growth may exist prior to and following seroconversion.

Length

Impairment of linear growth has been frequently observed in children infected with HIV infection. Many of the studies conducted in developed countries assessed the difference in linear growth between HIV-positive infants and seroreverted infants. Four of six such studies observed significantly lower linear growth in the infected infants and children (21, 23, 25, 36). The observed age of which HIV-infected infants begin to exhibit lower length-for-age compared to seroreverted infants varies from birth to fifteen months. In Europe, HIV-positive children followed from birth to forty-eight months of age were noted to be 0.8 cm shorter by three months of age (21). They remained significantly shorter than did seroreverted infants throughout the study. From birth to eighteen months, mean length-for-age z-scores were observed to be significantly lower in HIV-infected infants (23). The HIV-infected infants were calculated, after adjustment for gender and maternal characteristics, to be 2.25 cm shorter than seroreverted infants by eighteen months of age. HIV-infected children followed from birth to seventy months of age did not exhibit significantly lower height than seroreverted children until after fifteen months of age (25). In retrospective analysis of HIV-infected and seroreverted infants, seroreverted infants were observed to be longer from four to twenty-four months of age (36). Additionally, thirty-three percent of HIV-infected children, aged three months to fifteen years, exhibited severe height growth failure in a multi-center United States

clinical trial (32). This height growth failure was independent of examining center, parity, race, sex, gestational age, and caregiver. Two studies did not observe differences in linear growth between infected and seroreverted infants (24, 37). These studies contained smaller sample sizes than the other studies, which may have limited the power to detect differences in growth. Compared with seroreverters, lower mean length values for HIV-infected infants were noted only at months three and six, in a study conducted in New York City in which children were followed for their first 18 months of life (24). This study contained a sample of sixty-four infants. Peters et al. only observed stunting in symptomatic children receiving nutritional support (37). In contrast to other studies of HIV and growth, very sick children were included.

In developing countries, the presence of HIV infection has been observed to be associated with stunting in cross-sectional and longitudinal analyses. HIV-infected children aged seventeen to forty-one months in Uganda had approximately a six and one half times greater odds of being stunted compared to HIV-negative children in a cross-sectional analysis (38). Of four longitudinal studies conducted in developing countries, all four observed lower linear growth in HIV-infected infants (15, 30, 34, 35). Infants followed for twenty-five months were noted to have lower length-for-age growth curves than both seroreverted infants and infants born to HIV-women (30). This difference in growth persisted after the exclusion of children with early mortality. Two studies observed lower growth in HIV-infected infants from birth (34, 35). In Rwanda, infants were followed from birth to forty-eight months of age (34). HIV-infected infants in this study generally had significantly lower length-for-age values than infants born to HIV-

negative mothers from birth to thirty-six months of age. Throughout a twenty-four month study in Malawi, linear growth was significantly lower, after adjustment for vitamin A status, in HIV-infected infants compared with seroreverted infants (35). Length values were not noted to be significantly different at birth in infants studied in the Democratic Republic of Congo (15). HIV-infected infants in this group were significantly shorter by three months of age, and remained shorter than seroreverted infants and infants born to HIV-negative women throughout the twenty month study. HIV infection and associated illnesses were independent predictors of growth impairment in this population. Additionally, the odds of falling below –2 standard deviations of mean reference curves for length for age by twenty months of age was 2.10 (95% CI: 1.30-3.39) for HIV-positive infants compared with uninfected infants, after controlling for maternal stature (15).

Few studies have compared seroreverted children with children born to HIV-negative women. In Italy, a study comparing growth of seroreverted infants and infants born to HIV-negative women did not observe lower length-for-age z-scores until eighteen months of age for seroreverted infants (39). The effect of being a seroreverter persisted after adjustment for SES, sex, and maternal characteristics. However, growth between seroreverted infants and infants born to HIV-negative women has been observed to be similar in developing countries. Utilizing control groups of infants born to HIV-negative women and comparing growth with seroreverted infants in the Democratic Republic of Congo and Rwanda, no differences in linear growth were observed between these two groups (15, 34).

Weight

Impairment of weight has also been observed in HIV-infected children, and exhibits a similar pattern to differences in length. The same four studies that observed differences in length between HIV-infected infants and seroreverted infants also observed differences in weight between these two groups (21, 23, 25, 36). Three months after birth, HIV-positive infants were observed to be an average of 400 grams lighter than were the seroreverted infants (21). The HIV-infected infants became significantly lighter at three months of age and remained lighter for the 48-month study. Mean weight-for-age z-scores were lower in HIV-infected infants measured from birth to eighteen months of age (23). The HIV-infected infants were calculated to be 710 grams lighter than seroreverted infants were at eighteen months, after adjustment for gender and maternal characteristics (23). Compared to observations that length differed between HIV-infected and seroreverted children after fifteen months of age, significant differences in weight were not observed until after thirty-six months of age (25). This is an interesting finding because it is contrary to the usual development of malnutrition, in which significant weight impairment is noted before significant length impairment. Weight was generally lower in HIV-infected infants in retrospective analysis of infants less than twenty-five and one half months of age (36). Additionally, 20% of HIV-infected children in a United States multi-center trial exhibited severe weight growth failure, independent of sociodemographic variables (32). The two studies in which significant differences in

weight between HIV-infected and seroreverted children were not observed contained

smaller sample sizes (24, 37) and very sick children (37).

In developing countries, HIV-infected infants were observed to be significantly

lighter in four studies (15, 30, 34, 35). HIV-infected infants were observed to have

significantly lower weight-for-age growth curves than seroreverted infants and infants

born to HIV-negative women in the first twenty-five months of life (30). Lower growth

curves remained significant after exclusion of infants with early mortality. In Rwanda

(34) and Malawi (35), HIV-infected infants were noted to be lighter compared with

controls at birth and remained lighter throughout the studies, which were conducted for

forty-eight and twenty-four months, respectively. HIV-infected infants in the Democratic

Republic of Congo were significantly lighter by three months of age compared with

seroreverted infants and infants born to HIV-negative women (15). These infected

infants remained significantly lighter throughout the remainder of the 20-month study.

The HIV-positive infants in this sample had an observed odds ratio of 2.84 (95% CI:

1.58-5.11) for falling below –2 standard deviations of mean references curves for weight

for age compared with uninfected infants, after control for maternal stature.

In general, seroreverted infants have not exhibited lower weight growth compared

to infants born to HIV-negative women in both developed and developing countries.

Differences in weight-for-age z-scores were not observed between seroreverted infants

and infants born to HIV-negative women in Italy followed for two years after birth (38).

Similarly, differences in weight growth patterns were not observed between seroreverted

infants and infants born to HIV-negative women in the Democratic Republic of Congo and in Rwanda followed for twenty months and forty-eight months, respectively (15, 34)

Weight-for-length

Inconsistent results regarding differences in weight-for-length between infected and uninfected infants have been observed. In the United States, HIV-infected infants exhibited lower mean weight-for-length z-scores than uninfected infants did during the first eighteen months of life (23). HIV infection was the only independent predictor of weight-for-length at eighteen months of age, when assessed in multivariate regression with gender, prenatal smoking, prenatal alcohol, prenatal drugs, maternal education, and prenatal T-lympocyte CD4+ percentage as covariates in the model. No differences in weight-for-length between infected and uninfected children less than 25 1/2 months of age were observed in the United States (36). HIV-infected children in the Democratic Republic of Congo became significantly wasted after 12 months of age and remained wasted throughout the remaining eight months of the study (15). Weight-for-length, measured at three month intervals in the first year of life and at six month intervals thereafter for forty-eight months, was generally not observed to be lower in HIV-infected infants (34). It was also observed that, for HIV-infected infants, weight-for-length z-scores remained near or above reference medians (36). This finding may not reflect growth impairment in HIV-infected children if both length and weight are proportionally decreased.

Mechanisms of Impaired Growth for HIV-Infected Infants

The specific etiology for growth impairment in HIV-infected infants has not yet been elucidated, but likely involves a combination of mechanisms. Linear growth of infants with HIV has been observed to be correlated with viral load, but the mechanisms by which this correlation occurs are unknown (24). Malnutrition and subsequent growth impairment can also occur as a result of decreased intake, increased energy demands or both. Decreased intake may be secondary to decreased oral intake or decreased absorption. Increased demands can occur as a result of altered metabolic or endocrine functions. The presence of HIV infection may put infants at greater risk for such alterations. Abnormalities in absorption, metabolism, and endocrine are frequently observed in HIV-infected adults with wasting, but relatively few pediatric studies have been conducted (40). It is also important to consider the potential impact of other infections upon growth. HIV infection may increase the risk for infections, such as diarrhea and respiratory illnesses.

Decreased oral intake obviously can result in subsequent growth impairment. This has not been directly observed in HIV-infected children. HIV-infected children studied in the United States had intakes meeting the United States Recommended Daily Allowances (41). However, intake did predict weight, and children responded positively to increased oral intake. This suggests that other factors beyond decreased oral intake influence growth in HIV-infected children.

Decreased intake can also be secondary to decreases in gastrointestinal absorption. The presence of HIV may increase the risk of infection with enteric

pathogens. These pathogens may act directly to decrease absorption by causing diarrhea, gastrointestinal bleeding and intestinal dysfunction (40). Malabsorption may also occur indirectly due to HIV infection of the epithelium, but this has not been directly documented in HIV-infected children. Malabsorption is a frequent finding in children with HIV infection (42, 43). However, it is not likely to be the sole etiology of growth impairment in children. Growth failure in children has been observed in the absence of clinical symptoms indicating malabsorption (42).

Increased demands can occur as a result of altered endocrine or metabolic function. Increases in some cytokines, such as tumor necrosis factor (TNF), can induce a catabolic state in humans. Elevated TNF has been observed in HIV-infected children (44), but an association with growth impairment has not been established. Endocrine function was assessed in fourteen HIV-infected children, age six months to ten years (45). Overt abnormalities in endocrine function were not observed. Overall, the children were clinically and biochemically euthyroid, but the authors noted a state of compensated hypothyroidism in a subset of five of the children (45). A similar trend, but not significant, of hypothyroidism was also observed in the assessment of nine HIV-infected children (46). Whether decreased thyroid function impairs growth in children is not known.

Studies are limited which examine the effect of HIV seroconversion after birth on infant growth patterns. Assessing the impact of infection after birth and whether growth decreases as a result would be difficult. The majority of infants who are infected postpartum are infected within the first few months of age. This limits the number of

growth measurements available prior to seroconversion. Determining the exact timing of

HIV transmission is also difficult. However, the effect of HIV infection upon

immunologic factors indicates that associations may exist between seroconversion and

growth. Cytokine production, particularly interleukin-6 (IL-6), may have osteotrophic

effects. In mice, treatment with mouse IL-6 and soluble mouse IL-6 receptor resulted in

osteoclast-like multinucleated cell formation (47). Results of this study suggested that

the soluble IL-6 receptor induces osteoclast formation in the presence of IL-6. Elevated

levels of IL-6 and the IL-6 receptor were present in cells of adult donors infected with

HIV (48). These observed elevated levels of IL-6, compared with HIV-negative controls,

were irrespective of the diagnosis for the subject as positive without AIDS Related

Complex (ARC) or AIDS, as having ARC, or as having AIDS. Similar metabolic

responses to cytokine production might also occur in children with HIV, who have

exhibited elevated levels of IL-6 compared with controls (49).

It is also important to consider co-morbidity as a factor in growth impairment of

HIV-infected infants. Diarrheal disease is associated with increases in growth

impairment of children (50). The etiology for this is likely multifactorial, involving

mucosal changes, metabolic responses, and decreased energy consumption. Energy

consumption from non-breastmilk foods, but not breastmilk, has been observed to

decrease in infants with diarrhea or fever (51). Diarrhea also increases the transit time of

nutrients through the gastrointestinal tract. HIV-infected infants are more susceptible to

concurrent infection than uninfected infants. HIV-infected children in Zaire were noted

to have significantly more episodes of acute and persistent diarrhea than did the

uninfected children (26). However, it is difficult to measure the relative contributions of concurrent infections and HIV infection upon infant growth, and no studies are known to have separated the effects of co-morbidity and HIV infection upon infant growth.

Objectives

Three questions formed the foundation for the analyses. First, do differences in birthweight exist between infants by maternal or infant HIV status in a developing country setting? Second, do differences in growth exist between infants by infant HIV status? Third, what are the contributions of growth and HIV status upon infant mortality? From these questions, seven objectives were created.

1. Create a suitable data set for analysis from the original data set

2. Examine demographic characteristics of HIV-positive and HIV-negative women and their infants

3. Determine rates of vertical transmission in this sample

4. Examine differences in and factors influencing birthweight in infected and uninfected infants born to HIV-positive women and infants born to HIV-negative women

5. Examine differences in and factors influencing weight at the end of the first year of life in HIV-infected and uninfected infants born to HIV-positive women and infants born to HIV-negative women using stagewise regression

6. Examine differences in and factors influencing weight throughout the first year of life in HIV-infected and uninfected infants born to HIV-positive women and infants born to HIV-negative women

7. Examine the contributions of growth and HIV status upon mortality of the infants

Hypotheses

Based on the review of literature, three hypotheses for the analysis were developed.

1. Differences in birthweight will be observed between infants born to HIV-positive and HIV-negative women; no differences will be observed between uninfected and infected infants born to HIV-positive mothers

2. Growth will be lower for infected infants compared to uninfected infants and infants born to HIV-negative women; growth of uninfected infants born to HIV-positive women will be similar to infants born to HIV-negative women

3. Growth impairment and positive HIV status of the infant will be predictors for infant mortality

METHODS

Study Design and Location

This analysis of growth is a secondary analysis of data from a larger study on the interaction of placental malaria and vertical HIV transmission being conducted in Kisumu, Kenya. Kisumu is located next to Lake Victoria in the Nyanza province of Kenya. The larger study is being conducted jointly by the Kenya Medical Research Institute (KEMRI) and the United States Centers for Disease Control and Prevention (CDC). This study is a prospective cohort of women identified as being HIV-positive or HIV-negative after 32 weeks gestation and prior to birth. Women are asked to deliver their baby at the hospital and to bring their infant to well baby clinics every four weeks for follow-up. At follow-up visits, women are interviewed regarding feeding behaviors and infant health. Infants have their physical growth and health evaluated.

Subjects

The prevalence rate of HIV in pregnant women in urban areas of Kenya is approximately 16% (52). Women were recruited after 32 weeks of gestation at prenatal clinics at the Nyanza Provincial Hospital in Kisumu. Women had to be willing to deliver at the provincial hospital and have an uncomplicated singleton birth for inclusion into the study. Additionally, women could not exhibit symptoms of opportunistic infections characteristic for AIDS. Following birth, women had to bring their infant to well baby clinics every four weeks. Infants included in the analysis of growth were born between

August, 1996 and February, 1999. Within this population, essentially all women

breastfeed their infants, and colostrum is not routinely withheld from their infants.

Informed Consent, Approval and Human Subjects

Informed consent was obtained from the women. The study to evaluate malaria

and HIV transmission was approved by the Institutional Review Boards of KEMRI and

the CDC. As a secondary data analysis, the analysis was exempt from Human Subjects

review at the University of Arizona.

Data Collection

Knowledge regarding many specific details of the original data collection is

limited. At recruitment, maternal demographic characteristics were assessed. These

included age, education, marital status, husband's education, presence of electricity, and

gravida. Women were tested for syphilis and HIV. Women who tested positive for

syphilis were provided with treatment. Women were required to deliver their infant at

the Nyanza Provincial Hospital. At birth, blood was drawn for CD4+ count analysis and

infant weight was measured.

Women were instructed to bring their infants to well baby clinics every four

weeks. At each visit, infant blood was tested for HIV, hemoglobin, and malaria.

Polymerase chain reaction (PCR) testing was used to test for HIV in infants. Infants were

considered to be positive for HIV if two consecutive PCR tests were positive, with the

infant documented as HIV-positive from the date of the first test. Clinic staff took infant

weight, height, arm circumference, and head circumference measurements. Mothers were interviewed about breastfeeding behaviors, infant health, and other potential sources of HIV exposure for their infants.

Study Variables

Weight, birthweight, and mortality were outcomes of interest in the different analyses performed. Potential covariates considered included maternal and infant health characteristics, and SES indicators. Maternal health characteristics measured and included in this analysis were HIV status, age, gravida, CD4+ counts at birth, and syphilis status at recruitment. Infant health characteristics included were HIV status, estimated age of infection, sex, gestational age, introduction of mixed feeding, and presence of diarrheal illness. Birthweight was also included as a covariate in the analysis of longitudinal growth. Marital status, maternal educational level, husband's educational level, and presence of electricity were included as indicators of SES. A summary of how the variables were utilized for each analysis is provided in TABLE 1.

Data Analysis

Data Management

All data management and statistical analyses were performed utilizing STATA 6.0. (Stata Corporation, College Station, Texas, USA) A base data set was created from the original data from Kenya. It was from this base data set that data sets specific for the separate analyses were created.

TABLE 1: Summary of study variables

Variables	Birthweight Analysis	Stagewise Regression	4-Week Weight Analysis	Mortality Analysis
Birthweight	outcome	initial dependent variable; no transformation	no transformation	no transformation
CD4+ counts	quartiles plus category for missing	not included	not included	not included
Diarrhea 4 weeks prior	not included	not included	0=no, 1=yes, 2=indeterminate	0=no, 1=yes, 2=indeterminate
Diarrhea 4-8 weeks prior	not included	not included	0=no, 1=yes, 2=indeterminate	0=no, 1=yes, 2=indeterminate
Electricity	0=no, 1=yes	0=no, 1=yes	0=no, 1=yes	0=no, 1=yes
Gestation	no transformation	not included	not included	not included
Gravida	categorical: 0=>2, 1=≤2	categorical: 0=>2, 1=2, 2=1	categorical: 0=>2, 1=≤2	categorical: 0=>2, 1=≤2
Husband Education	categorical: 0=≥12 years, 1=<12 years	categorical: 0=≥12 years, 1=<12 years	categorical: 0=≥12 years, 1=<12 years	categorical: 0=≥12 years, 1=<12 years
Infant age of HIV infection	not included	quartiles	used to characterize infants at that age	used to characterize infants at that age
Infant HIV	0=not positive mother, 1=seroreverted, 2=postpartum, 3=presumed birth infected	0=not positive mother, 1=positive	0=not positive mother, 1=infant not positive at this age, 2=infant positive at this age	0=not positive mother, 1=infant not positive at this age, 2=infant positive at this age
Infant sex	0=female, 1=male	0=female, 1=male	0=female, 1=male	0=female, 1=male
Infant weight	not included	final weight as outcome of initial regression	predicted weight as outcome	predicted weight
Marital status	0=no, 1=yes	0=no, 1=yes	0=no, 1=yes	0=no, 1=yes
Maternal age	Box-Cox transformed	Box-Cox transformed	Box-Cox transformed	Box-Cox transformed
Maternal Education	categorical: 0=>8 years, 1=≤8 years	categorical: 0=>8 years, 1=≤8 years	categorical: 0=>8 years, 1=≤8 years	categorical: 0=>8 years, 1=≤8 years
Maternal HIV	0=no, 1=yes	0=no, 1=yes	0=no, 1=yes	0=no, 1=yes
Mixed Feeding Introduction	not included	quartiles	categorical at this age: 0=no, 1=yes	categorical at this age: 0=no, 1=yes
Mortality	not included	not included	not included	outcome
Syphilis result	0=no, 1=yes, 9=indeterminate	not included	not included	not included

The initial data cleaning process consisted of identifying infants with important information unavailable, assessing weight of each child, and checking for outliers in other variables. Infants were dropped from the analysis if no follow-up occurred after recruitment or if maternal HIV status was indeterminate or missing. Weight versus age graphs were examined for each child. Weight gains or losses that seemed physiologically improbable, as defined by an isolated large gain or loss, were noted. These weights were re-checked in Kenya for possible entry error. Those with entry error were corrected and those that remained questionable were changed to missing. Ranges for all other variables were examined for potential outliers.

Four distinct analyses were performed. The first analysis was to assess birthweight. The second and third analyses assessed infant growth. The last analysis assessed HIV, growth and infant mortality. The analyses utilized analysis of covariance (ANCOVA), linear and logistic regression models with appropriate regression diagnostics were performed for each regression. For analyses utilizing ANCOVA, adjusted mean weights by significant covariates for HIV status were calculated post-regression.

Demographic Characteristics

Comparisons of demographics and potential covariates were made by maternal HIV status and infant HIV status. Comparisons by maternal HIV status were made by t-test if variables were normally distributed, by chi-square for bivariate outcome variables, and Wilcoxon rank sum for continuous or integral variables that were not normally

distributed. Comparisons by infant HIV status were made by ANOVA for normally

distributed variables, by logistic regression for bivariate outcome variables, and Kruskal-

Wallis test for variables not normally distributed.

PCR Testing and Transmission Rates

Infants were tested for HIV utilizing PCR. Blood was collected at each visit from

a finger prick. Peripheral blood mononuclear cells were isolated by centrifugation, and

subsequently lysed. A nested PCR was utilized as decribed by Janini et al. (53) Negative

and positive controls were included in each experiment, and PCR reactions were done in

duplicate. Infants were considered to be HIV-positive if two consecutive PCR tests were

positive. Infants were noted to be infected at the date of the earlier positive PCR test. To

provide a better estimate of the timing of transmission, the midpoint between the of prior

negative test date and age of the first positive test date was taken to be estimated age of

infection. Mean age of infection and percentiles were calculated using the estimated age

of infection.

Analysis of Birthweight

Mean birthweights were compared by maternal HIV status and infant HIV status.

Birthweight was normally distributed, so transformation was not necessary. Infant

birthweight between HIV-negative and HIV-positive women was compared by unpaired

t-test. Infants were divided into five groups for comparison: infants born to HIV-

negative women, uninfected infants born to HIV-positive women (PCR-negative), infants

born to HIV-positive women not infected at birth but infected later, infants born to HIV-positive women presumed infected at birth, and infants for whom it was not possible to reasonably presume whether they had been infected at birth or postpartum. Infants were considered to have been presumed infected at birth if they tested positive for HIV at their first scheduled visit. Infants were classified as indeterminate if they tested positive on the first visit they attended, but did not attend the first scheduled visit.

Mean birthweight was then assessed as the dependent variable in stepwise ANCOVA. Potential covariates included in the ANCOVA were maternal HIV status, infant HIV status, gestation, gravida, maternal education, husband's education, marital status, electricity, CD4+ counts, infant sex, maternal syphilis status, and maternal age. Maternal HIV status and infant HIV status were collinear, so it was not possible to include both variables in the same model. Since it was of interest to assess both maternal status and infant status, two separate ANCOVAs were performed. Variables that were not normally distributed were either Box-Cox transformed (FIGURE 1) or converted to categorical variables. Logical two-way interactions were assessed in univariate analysis, and were included in multivariate analysis if significant.

FIGURE 1: Equation for Box-Cox Transformation (X)

$$x(^\lambda) = \{x^\lambda - 1\} / \lambda \qquad \text{if } x > 0 \text{ or } x < 0$$

$$x = \ln(x) \qquad \qquad \text{if } x = 0$$

<u>Analysis of Growth in the First Year of Life</u>

Two different analyses were performed to assess the impact of infant HIV status upon growth in the first year of life. First, a stagewise, also known as residual, regression was performed. Final weight was regressed upon birthweight, and the residuals of this regression were saved. These residuals became the outcome variable for regression with infant HIV status and other covariates. Infants were excluded from the analysis if either birthweight or final weight was missing, or if death occurred prior to the eleventh visit. Variables that did not meet the assumption of linearity were Box-Cox transformed or converted to categorical variables. Univariate analyses was performed with each covariate and logical two-way interactions. Those significant in univariate analyses were included in multivariate analysis.

The second analysis consisted of assessment of growth at the end of each four-week interval. Since each infant was not measured at exactly four-week intervals, weight at four-week intervals was predicted. Linear splines were fit for each infant with age as the independent variable and weight as the dependent variable. Knots for the splines were placed at the ages that the infant had weight measurements taken. If there were more than 16 weeks between visits, weight was not predicted between these ages. Sixteen weeks was chosen because linearity decreases as the time between visits increases. Stepwise ANCOVA was performed for each four-week time point with the predicted weight of infants as the outcome variable. Infants were divided into five groups for assessment of HIV status: infants born to HIV-negative women, infants born to HIV-positive women who did not become positive, infants presumed infected at birth,

infants known to be infected postpartum, and infants indeterminate for presumed or postpartum infection. Of the variables utilized in the ANCOVA models, those that did not meet the assumption of linearity were Box-Cox transformed or converted to categorical variables. Two-way interactions were assessed for the other covariates and infant HIV-status in univariate analysis. Significant interactions were included in multivariate analysis.

Analysis of Growth, HIV and Mortality

Infants had been classified in the data as still active, died or out of study. Covariates were also examined for differences between infants who remained in the study and those classified as out of the study. Infants classified as out of study were excluded from the analysis. Since the actual date of death for the infants is not known, logistic regression, as opposed to survival analysis, was utilized to examine the effects of growth and HIV status upon later mortality. Predicted weight and HIV status, along with other potential covariates, were assessed as predictors of later mortality by logistic regression at eight weeks of age. The outcome variable was whether the infant would still be active at the end of the first year of life or have died prior to the end of the first year. Logistic regression diagnostics were performed, and the models were assessed by Pearson chi-square goodness of fit tests and area under the receiver operating characteristic (ROC) curve.

RESULTS

Base Data Set

There were 414 infant-mother pairs in the initial data set. (FIGURE 2) Of the

mothers, 113 had tested HIV-negative, 283 had tested HIV-positive, 15 had indeterminate

HIV status and 3 had missing HIV status. Women with indeterminate or missing HIV

status were dropped from the analysis. Three infants born to HIV-negative mothers and

fourteen infants born to HIV-positive mothers had no additional follow-up and were

excluded from the study. A total of 35 infants were dropped from the study and 379 were

kept. Those that remained in the analysis included 110 infants born to HIV-negative

women, 73 infants born to HIV-positive women who would become HIV-positive within

the first year of life, and 196 uninfected children born to HIV-positive women.

FIGURE 2: Flowchart of inclusion and exclusion of infants from base data set
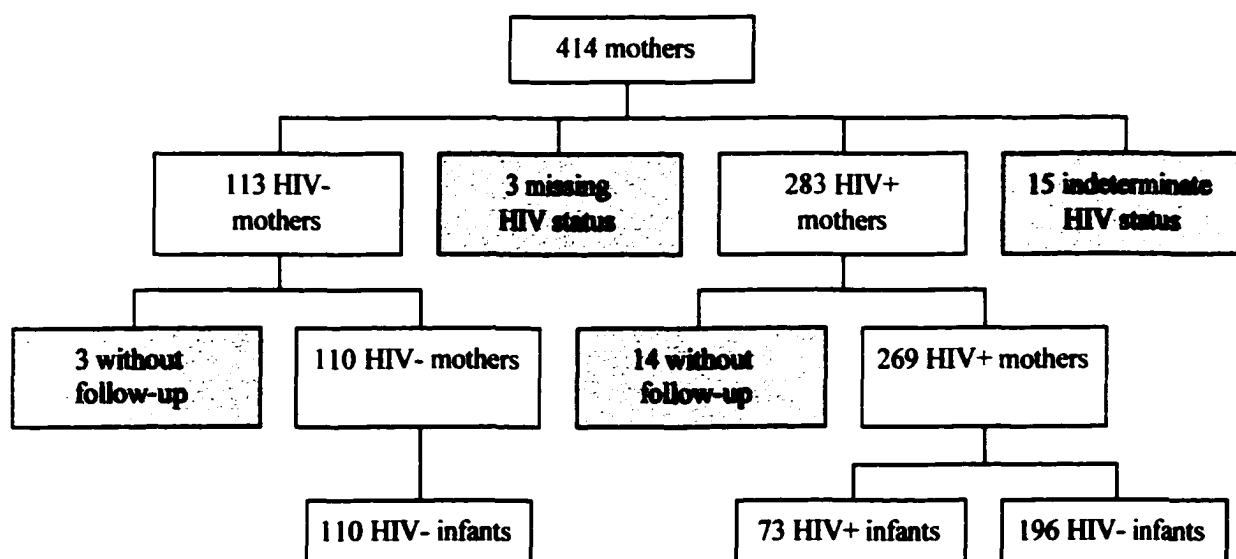
A total of 379 infants remained in the data set after dropping those with no

follow-up after recruitment (n=17) or with missing (n=3) or indeterminate (n=15)

maternal HIV status. Demographic and infant characteristics were available for those

dropped with missing or indeterminate maternal HIV status. Comparisons between these

and maternal-infant pairs kept in the analysis were made. (TABLE 2) The only

significant difference noted was a greater percentage of infants dropped from the analysis

had electricity in the household. Those children who were dropped from the study tended

to have lower birthweights, but the means between the two groups were not signficant.

TABLE 2: Maternal and infant characteristics of dropped and kept mother-infant pairs

Characteristic	Kept in the Analysis (n=379)	Dropped from Analysis (n=18)	p-value
Maternal			
• Age* (years)	22.2 ± 4.7 (14-40)	21.9 ± 5.1 (15-36)	p=0.72
• CD4+* (µg/L)	720 ± 376 (71-2539)	887 ± 367 (286-1516)	p=0.09
• Electricity	19%	39%	p=0.04
• Gravida*	2.2 ± 1.4 (1-8)	2.2 ± 1.3 (1-5)	p=0.97
• Married	78%	75%	p=0.60
• Maternal education* (years)	8.6 ± 2.6 (0-17)	8.6 ± 2.5 (5-14)	p=0.59
• Husband education* (years)	10.5 ± 2.6 (0-16)	11.2 ± 1.9 (7-14)	p=0.76
• Positive syphilis result at recruitment	15.4%	9.7%	p=0.50
Infant			
• Birthweight* (grams)	3366 ± 109 (1800 – 4462)	3167 ± 451 (2569-4286)	p=0.09
• Gestation* (weeks)	38.5 ± 1.3 (34-42)	38.1 ± 1.6 (35-40)	p=0.30
• Male infant	44%	51%	p=0.60

*means expressed as mean ± standard deviation (range)

Questionable weight measurements in the data set had the original form re-

checked in Kenya. Thirteen weight entries were identified as physiologically improbable

from visual examination of weight versus age graphs for each infant. Six were identified as computer entry error and were corrected. Seven were identified as correctly entered into the data set from the original. These were changed to missing because of the likelihood of entry error onto the original form or measurement error. Additionally, five infants were missing weight measurements, but had been present at the visit. Two of these did have weight measurements taken, but not entered into the data set. Three did not have a weight measurement taken although the infant was present. Lastly, one infant had a weight measurement changed to missing because weight was entered for the visit, but the child was not present. Overall, nineteen entries were re-checked with eight corrections made and eleven changed to missing.

Demographics and Comparison of Covariates by HIV Status

There were 379 mother-infant pairs in the base data set for the analyses. Summary statistics for all women are presented in TABLE 3. There were 191 male infants and 185 female infants (3 infants were missing sex characteristic). The women ranged in age from 14 to 40 years old, with a mean age of 22.2 years. Gestation length ranged from 34 to 42 weeks with a mean of 38.5 weeks. Mean gravida for all women was 2.2, ranging from 1 to 8 pregnancies. Mean number of years of education completed was 8.7 years (range: 0-17). The majority of women (75%) were married, and husbands had completed a mean of 10.5 years of education (range: 0-16). A minority (19%) of women had electricity in the household. Introduction of mixed feeding ranged from 12 to 187 days after birth with a mean of 64 days.

TABLE 3: Maternal and Infant characteristics of HIV-positive and HIV-negative women

Characteristics	All Women (n=379)	HIV- Women (n=110)	HIV+ Women (n=269)
Maternal			
• Age* (years)	22.2 ± 4.7 (14-40)	21.6 ± 4.7 (15-35)	22.5 ± 4.7 (14-40)
• CD4+ (µg/L)	720 ± 376 (71-2539)	949 ± 381 (161-2041)	625 ± 331 (71-2539)✦
• Electricity	19%	18%	19%
• Gravida*	2.2 ± 1.4 (1-8)	2.1 ± 1.5 (1-8)	2.6 ± 1.3 (1-7)
• Married	75%	69%	77%
• Maternal education* (years)	8.7 ± 2.6 (0-17)	9.1 ± 2.4 (0-14)	8.5 ± 2.6 (0-17)✦
• Husband education* (years)	10.5 ± 2.6 (0-16)	10.4 ± 2.6 (2-16)	10.6 ± 2.6 (0-16)
• Positive syphilis result at recruitment	8.2%	2.8%	10.4%✦
Infant			
• Age mixed feeding introduction* (days)	64 ± 42 (12-187)	64 ± 41 (13-187)	64 ± 43 (12-183)
• Gestation* (weeks)	38.5 ± 1.3 (34-42)	38.7 ± 1.2 (36-41)	38.5 ± 1.4 (34-42)
• Male Infant	51%	40%	55%✦

*means expressed as mean ± standard deviation (range)
✦p<0.05 compared with HIV-negative women

Significant differences were noted between HIV-positive women and HIV-negative women for maternal years of education, sex of their infants, syphilis result at recruitment, and CD4+ counts (µg/L) at birth. (TABLE 4) HIV-positive women were more likely to have less education than were HIV-negative women by approximately five months. HIV-positive women were more likely to give birth to a male child. A greater percentage (10.4% vs. 2.8%) of HIV-positive women had a positive syphilis result at recruitment. As expected, mean CD4+ count was also lower in HIV-positive women at birth compared with HIV-negative women. However, HIV-positive women had mean CD4+ counts greater than 500 µg/L and only 12 HIV-positive women had CD4+ counts

less than 200 µg/L. The women by HIV status were similar for age, presence of

electricity, gravida, marital status, and husband educational level. Infants, by maternal

HIV status, had similar lengths of gestation and timing of mixed feeding. All women

were still breastfeeding at the end of the infant's first year of life.

TABLE 4: Maternal and infant characteristics by infant's HIV status at the end of the first year of life

Characteristics	Controls (n=110)	PCR- (n=196)	PCR+ (n=73)
Maternal			
• Age* (years)	21.6 ± 4.7 (15-35)	22.4 ± 4.5 (14-37)	22.8 ± 5.1 (16-40)
• CD4+ (µg/L)	949 ± 381 (161-2041)	662 ± 339* (71-2539)	525 ± 287*✦ (98-1548)
• Electricity	18%	18%	21%
• Gravida*	2.1 ± 1.5 (1-8)	2.3 ± 1.3 (1-7)	2.2 ± 1.4 (1-7)
• Married	69%	78%	74%
• Maternal education* (years)	9.1 ± 2.4 (0-14)	8.5 ± 2.4 (1-17)*	8.3 ± 3.1 (0-14)*
• Husband education* (years)	10.4 ± 2.6 (2-16)	10.5 ± 2.7 (0-16)	10.8 ± 2.2 (6-16)
• Positive syphilis result at recruitment	2.8%	10.6%*	9.9%*
Infant			
• Age mixed feeding introduction* (days)	64 ± 41 (13-187)	67 ± 43 (13-183)	57 ± 42 (12-157)
• Gestation* (weeks)	38.7 ± 1.2 (36-41)	38.5 ± 1.3 (35-42)	38.3 ± 1.5 (34-41)✦
• Male Infant	40%	50%	68%*✦

*means expressed as mean ± standard deviation (range)
*p<0.05 compared with HIV-negative, ✦p<0.05 compared with PCR-

To further characterize the sample, maternal and infant characteristics were also

compared by infant HIV status. (TABLE 4) The infants were divided into three groups

based in HIV status at the end of the first year of life: 110 infants born to HIV-negative

women (Controls); 196 uninfected infants born to HIV-positive women (PCR-); and, 73

HIV-positive infants (PCR+). Lower maternal CD4+ counts at birth for PCR+ infants were observed compared with Controls and PCR- infants. Both PCR- and PCR+ infants had lower maternal CD4+ counts than Controls. All groups had mean CD4+ counts greater than 500 µg/L. A greater percent of PCR+ infants were male than were Controls and PCR- infants. PCR+ infants had significantly shorter gestation than did Controls, but PCR- infants did not differ from Controls. Maternal mean educational level was lower for PCR- and PCR+ infants than Controls. The difference in mean maternal level of education was less than one year. No difference was noted for positive syphilis status at recruitment for mothers of PCR- and PCR+ infants, but HIV-positive mothers in both groups had a greater percentage of positive syphilis results than did mothers of Controls. No differences were observed for gravida, maternal age, marital status, husband education level, presence of electricity or timing of mixed feeding introduction by infant HIV status.

Rates of HIV Transmission

Twenty-seven percent of the infants born to HIV-positive women became positive in the first year of life. Twenty-seven infants (37%) were likely to have been infected at birth and twenty-nine infants (40%) were infected later. Seventeen infants (23%) were infected at the first visit they were tested, but it was not the first scheduled, so it was not possible to reasonably predict whether they were infected at birth or later. The transmission rate for infants presumed infected at birth or during labor and delivery was 10.0% (27/269). Of the infants classified as infected postpartum, the transmission rate

for those infected postpartum was 10.8% (29/269). Age of infection was estimated as the

midpoint between the last visit PCR that tested HIV-negative and the first visit PCR that

tested HIV-positive. Estimated age of infection was skewed toward earlier infection.

(FIGURE 3) The mean estimated age for postpartum infection was 145 days. The

median estimated age of infection was 113 days. Twenty-five percent of the HIV-

positive infants were estimated to have been infected by 73 days. Seventy-five percent of

these infants were infected by 191 days.

FIGURE 3: Estimated age of postpartum infection in days
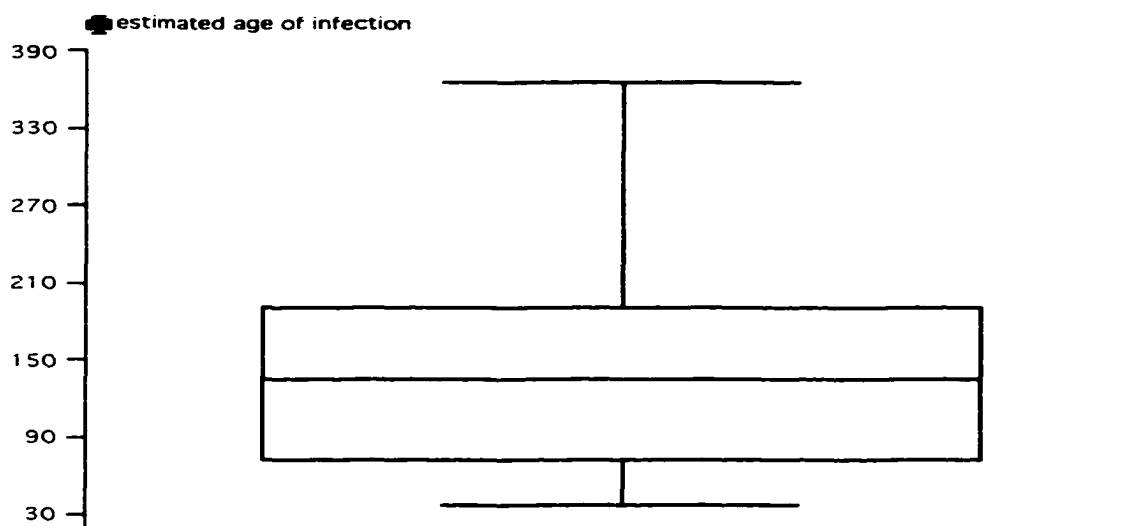
Birthweight

An additional three infants were excluded from the analysis of birthweight,

because birthweight was missing. Infants (Controls) born to HIV-negative women

(n=110) comprised 30% of the sample. Infants born to HIV-positive women were

divided into four groups: 195 uninfected infants born to HIV-positive women (PCR-); 29

infants with a positive PCR results later than their first visit (lPCR+); 27 infants

presumed infected at birth or during labor and delivery because the PCR was positive at

the first visit (fPCR+); and, 17 infants whose PCR was positive at the first test, but this

was not the first scheduled test (ePCR+).

TABLE 5: Maternal and infant characteristics for HIV-negative infants, PCR-negative infants, infants infected after birth, and infants presumed infected at birth

Characteristic	Controls	PCR-	fPCR+	lPCR+	ePCR+
Maternal	(n=110)	(n=195)	(n=27)	(n=29)	(n=17)
• Age* (years)	21.6 ± 4.7	22.4 ± 4.5	22.3 ± 4.8	23.4 ± 5.0	21.6 ± 5.3
• CD4+ (µg/L)*	955 ± 378	660 ± 339*	544 ± 324*✦	536 ± 283*✦	440 ± 260*✦
• CD4+ ≥ 665	63.0%	34.5%*	25.9%*	27.6%*	25.0%*
• CD4+ <665	20.4%	46.2%*	44.4%*	69.0%*	58.3%*
• Electricity	18.5%	18.6%	22.2%	17.2%	25.0%
• Gravida*	2.1 ± 1.5	2.3 ± 1.3	2.1 ± 1.6	2.1 ± 1.1	2.5 ± 1.4
• Gravida ≤ 2	71.3%	59.5%*	74.1%	69.0%	50.0%
• Married	68.5%	77.8%	74.1%	69.0%	75.0%
• Maternal education* (years)	9.2 ± 2.4	8.5 ± 2.4*	8.1 ± 2.9	8.8 ± 3.3	6.9 ± 3.4
• Maternal education less than or equal to 8 years	48.2%	65.0%*	66.7%	51.7%	75.0%
• Husband education*	10.4 ± 2.6	10.4 ± 2.7	11.3 ± 2.4	10.8 ± 2.0	10.1 ± 2.0
• Husband education less than or equal to 12 years	38.5%	41.8%	37.5%	42.9%	48.0%
• Positive syphilis result at recruitment	2.8%	10.3%*	3.7%	17.2%*	8.3%
Infant					
• Age mixed feeding introduction* (days)	64.2 ± 41.5	67.0 ± 42.9	61.0 ± 43.6	57.1 ± 41.5	50.3 ± 50.4
• Gestation* (weeks)	38.7 ± 0.1	38.5 ± 1.3	38.5 ± 1.8	37.9 ± 1.3	38.4 ± 1.4
• Male infant	39.8%	50.3%	74.1%*✦	62.1%*	83.3%*✦

*means expressed as mean ± standard deviation; *p<0.05 compared with HIV-negative, ✦p<0.05 compared with PCR-negative; groups: controls (infants born to HIV- women): PCR- (infants born to HIV+ women who did not have a positive PCR) fpcr+ (PCR positive at first scheduled visit), lPCR+ (first PCR negative with a later positive PCR), ePCR+ (first PCR positive but not at the first scheduled visit)

No significant differences were observed between HIV-infected infants for the maternal and infant characteristics by further characteriztion of infants by HIV status (TABLE 5). HIV-positive women had lower mean CD4+ counts than HIV-negative women, and mothers of infants who became HIV-positive had lower mean CD4+ counts than mothers of PCR- infants. Infants who became HIV-positive were more likely to be male than control and PCR- infants. Mothers of PCR- and IPCR+ infants were more likely to have a positive syphilis result at recruitment than control mothers.

Mean birthweights by maternal HIV-status and infant HIV-status are summarized in TABLE 6. Infants born to HIV-positive women were 0.08 kg lighter than infants born to HIV-negative women, and this difference was marginally significant (p=0.09). Infants who were presumed to be infected at birth were 0.17 kg lighter than infants born to HIV-negative women, and this difference was also marginally significant (p=0.07). No significant differences in birthweight were observed between the other groups of infants by HIV status.

TABLE 6: Unadjusted mean birthweight by maternal and infant HIV status

HIV Status	Birthweight in kilograms (mean ± standard deviation)	Range
Maternal		
• HIV-Negative (n=108)	3.23 ± 0.43	2.14-4.32
• HIV-Positive (n=268)	3.15 ± 0.45*	1.96-4.46
Infant		
• Born to HIV-Negative Women (n=108)	3.23 ± 0.43	2.14-4.32
• PCR-Negative (n=195)	3.16 ± 0.44	2.10-4.46
• Presumed Infected at Birth (n=27)	3.06 ± 0.39✦	2.05-3.51
• Infected Postpartum (n=29)	3.09 ± 0.50	1.96-4.20
• Indeterminate Birth or Postpartum (n=17)	3.19 ± 0.59	2.16-4.00

*p=0.09 compared with HIV-negative, ✦p=0.07 compared with infants born to HIV-negative women

Mean birthweights by maternal HIV-status and infant HIV-status are summarized in TABLE 6. Infants born to HIV-positive women were 0.08 kg lighter than infants born to HIV-negative women, and this difference was marginally significant (p=0.09). Infants who were presumed to be infected at birth were 0.17 kg lighter than infants born to HIV-negative women, and this difference was also marginally significant (p=0.07). No significant differences in birthweight were observed between the other groups of infants by HIV status.

Since both maternal and infant HIV status had marginally significant associations with birthweight in univariate analysis, both were considered for multivariate analysis. Two models were required because maternal HIV status and infant HIV status were collinear. It was desirable to do two models because the significance of maternal status is controversial in the literature and it is of interest to determine if infant HIV status is associated with birthweight. In univariate analysis, the only significant two-way interactions observed were infant HIV status with maternal CD4+ counts and maternal HIV status with maternal CD4+ counts. These interactions were then included in two separate ANCOVA models for birthweight with the other maternal and infant characteristics. The p-value for inclusion was 0.10, so that marginal associations could be identified. Significant predictors of birthweight in the ANCOVA with maternal HIV status were gestation, gravida, sex, syphilis result at recruitment, and interaction between maternal HIV status and maternal CD4+ counts. (TABLE 7) Marginally significant associations were observed for marital status and presence of electricity. Longer

gestation and being male were associated with higher birthweight. Lower gravida,

positive or indeterminate syphilis result, and lowest category of maternal CD4+

interaction with maternal HIV were associated with lower birthweight. The adjusted R-

square value for the ANCOVA was 0.2129.

TABLE 7: Results of stepwise ANCOVA of birthweight with maternal HIV status and
other infant and maternal characteristics

Birthweight (kilograms)	Coefficient	95% Confidence Interval	p-value
Gestation (weeks)	0.115	0.083, 0.146	<0.001
Gravida less than or equal to 2	-0.137	-0.228, -0.045	0.004
Male infant	0.126	0.044, 0.209	0.003
Positive syphilis	-0.261	-0.413, -0.110	0.001
Indeterminate syphilis	-0.109	-0.215, -0.002	0.045
Presence of electricity	0.098	-0.007, 0.203	0.068
CD4+ <473 interaction with HIV-positive woman	-0.130	-0.238, -0.022	0.018
Being married	0.090	-0.010, 0.189	0.076
Constant	-1.24	-.247, -0.023	0.046

Birthweights were adjusted by HIV status for significant covariates signficant in

ANCOVA. (TABLE 8) Within HIV-positive women, only the lowest CD4+ category

had a predicted birthweight significantly lower than the highest CD4+ category.

However, a downward trend toward with decreasing CD4+ category is noted for

birthweights of HIV-positive women. Infants born to HIV-positive women with CD4+

counts less than 900 were predicted to weigh approximately 0.2 kg less than infants born

HIV-positive women with CD4+ counts greater than 900. No trends are evident for

birthweight with HIV-negative women.

TABLE 8: Birthweights (kilograms) and 95% confidence intervals by maternal HIV status and CD4+ category adjusted for gestation, gravida, syphilis result and infant sex

CD4+ Category	HIV-Negative	HIV-Positive
Greater than 900	3.21 (3.09, 3.32)	3.35 (3.21, 3.49)
665-899.9	3.22 (3.05, 3.39)	3.16 (3.05, 3.26)
473-664.9	3.18 (2.97, 3.40)	3.14 (3.04, 3.24)
Less than 473	3.29 (3.02, 3.54)	3.11 (2.99, 3.18)

Similar results were observed for birthweight with the inclusion of infant HIV status. (TABLE 9) Significant predictors for birthweight were gestation, gravida, husband's education, infant sex, marital status, syphilis result, and interaction of CD4+ with infant HIV status. As with the ANCOVA performed with maternal HIV status, longer gestation and being male were associated with higher birthweight. Positive syphilis result, lower gravida, mother not married, husband's education less than or equal to 12 years and presumed birth infection interaction with CD4+ count category less 665 μg/L were associated with lower birthweight.

TABLE 9: Results of stepwise ANCOVA of birthweight with infant HIV status and other infant and maternal characteristics

Birthweight (kilograms)	Coefficient	95% Confidence Interval	p-value
Gestation (weeks)	0.121	(0.090, 0.153)	<0.001
Gravida less than or equal to 2	-0.152	(-0.246, -0.058)	0.002
Male infant	0.125	(0.041, 0.209)	0.004
Positive syphilis	-0.290	(-0.446, -0.133)	<0.001
Indeterminate syphilis	-0.096	(-0.205, 0.013)	0.083
CD4+ <665 with infant presumed infected at birth	-0.348	(-0.589,-0.107)	0.005
Husband's education ≤ 12	-0.127	(-0.225, -0.029)	0.011
Not married	-0.123	(-0.230, -0.015)	0.025
Constant	-1.357	(-2.594, -0.120)	0.032

Adjusted birthweights by infants HIV status and maternal CD4+ category are presented in TABLE 10. fPCR+ infants with maternal CD4+ count less than 665 µg/L had significantly lower adjusted birthweights than control infants. These infants had adjusted birthweights approximately 0.4 kg less than control infants, and fPCR+ infants with higher maternal CD4+ category. Although not significant, a trend for lower adjusted birthweight with lower CD4+ category was observed for the other groups of infants born to HIV-positive women.

TABLE 10: Birthweight in kilograms (95% confidence interval) by infant HIV status and maternal CD4+ category adjusted for gestation, gravida, syphilis result, and sex

CD4+ Category	Controls	PCR-	fPCR+	lPCR+	ePCR+
≥ 665	3.22	3.21	3.28	3.21	3.35
	(3.12, 3.31)	(3.12, 3.31)	(2.95, 3.60)	(2.91, 3.51)	(2.91, 3.51)
< 665	3.25	3.14	2.84	3.16	3.09
	(3.07, 3.43)	(3.05, 3.22)	(2.60, 3.08)	(2.98, 3.34)	(2.79, 3.39)

groups: controls (infants born to HIV- women): PCR- (infants born to HIV+ women who did not have a positive PCR) fpcr+ (PCR positive at first scheduled visit), lPCR+ (first PCR negative with a later positive PCR), ePCR+ (first PCR positive but not at the first scheduled visit)

Growth

Measurements were available for stagewise linear regression for 268 HIV-negative infants and 47 HIV-positive infants. The HIV-negative infants included 101 infants born to HIV-negative women and 167 PCR-negative infants. A total of 64 infants were excluded from the analysis because they died prior to their eleventh visit (n=58), or were still active but missing birthweight (n=3) or final weight (n=3). The excluded infants had a lower mean birthweight (2.98 kg vs. 3.20 kg) and included a greater

proportion of HIV-positive infants (35.6% vs. 12.4%) and infants born to HIV-positive

women (20.4% vs. 8.2%) than infants included in the analysis.

TABLE 11: Characteristics of infants included in stagewise regression by HIV status

Characteristics	HIV-Negative Infants (n=268)	HIV-Positive Infants (n=47)
Gravida		
• Greater than 2 (%)	35.5	34.0
• Equal to 2 (%)	23.1	25.5
• Less than 2 (%)	41.4	40.4
Gestation (weeks)	38.6 ± 1.3 (35-42)	38.5 ± 1.5 (35-41)
Infant sex (% Male)	45.5	63.8✦
Maternal age (years)	22.0 ± 4.6 (14-35)	23.3 ± 5.0✦
Marital status (% married)	76.3	74.5
Maternal education (years)	8.8 ± 2.4 (0-17)	8.6 ± 3.4 (0-14)
• Less than 9 years (%)	58.6	51.1
Husband's education (years)	10.5 ± 2.5 (0-16)	11.0 ± 2.3 (6-16)
• Less than 12 years (%)	32.7	25.0
Presence of electricity (%)	18.7	25.5
Estimated age of first mixed feeding (days)		
• 0-23.5 (%)	24.3	29.8
• 24-64.5 (%)	24.7	27.7
• 65-101 (%)	27.4	19.1
• 101.5-170 (%)	23.6	23.4

*means expressed as mean ± standard deviation (range)
✦p<0.05 and ✦p<0.10 compared with HIV-negative infants

Since a larger number of infants were excluded, comparisons of maternal and

infant characteristics were performed by infant HIV status to characterize the data set

used for analysis. (TABLE 11) The only significant differences observed were a greater

percentage of HIV-positive infants were male and mothers of HIV-positive infants were

slightly older than mothers of HIV-negative infants. Characteristics which were likely to

influence birthweight only, such as maternal CD4+ counts at birth and syphilis result at

recruitment, were not compared because they were not included in the analysis.

FIGURE 4: Graph of final weight versus birthweight and fitted regression line

+ HIV-positive infants (n=47)   · HIV-negative infants (n=268)

final weight = 0.808*birthweight + 6.013
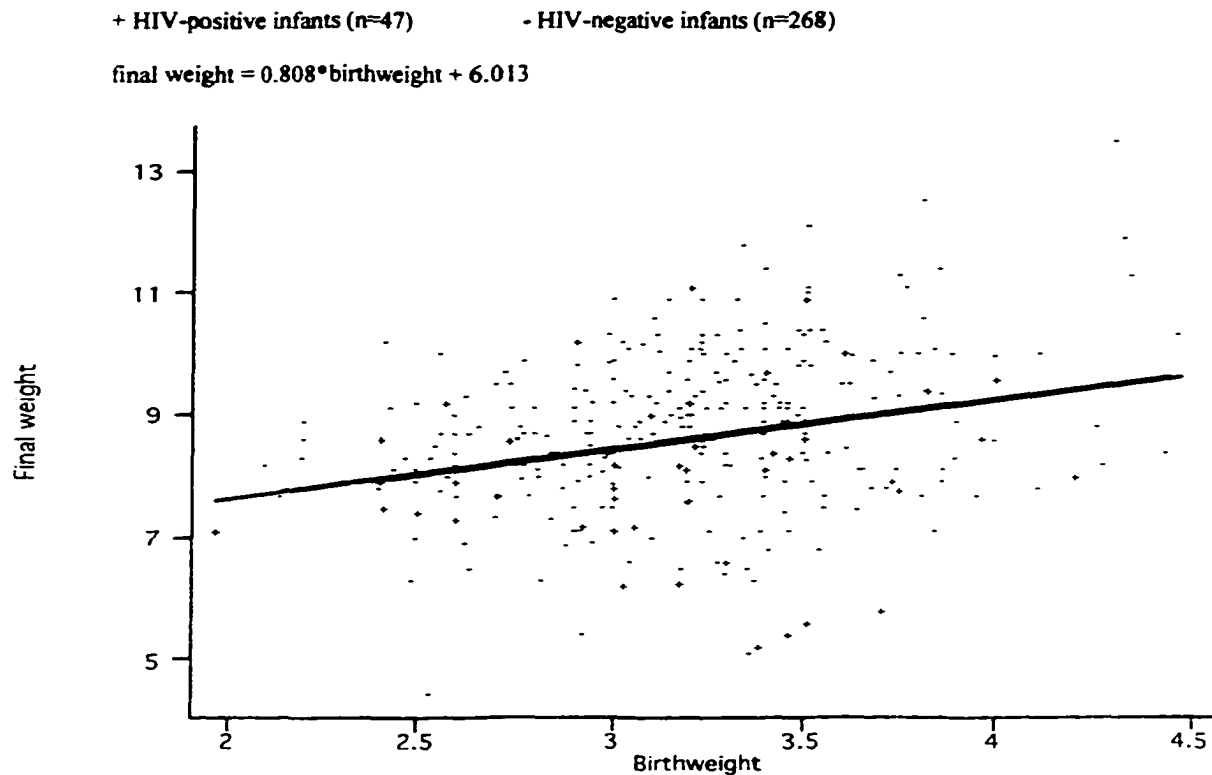
Birthweight did not differ between HIV-negative and HIV-positive infants

included in the analysis. Mean birthweight was 3.21 (range: 2.10-4.46) for HIV-

negative infants and 3.18 (range: 1.96-4.20) for HIV-positive infants. Final weight was

defined as the latest weight measurement taken at or after the infant's eleventh visit.

Final age corresponding with final weight for the infants ranged from 39.2 to 52.7 weeks.

Mean final age was 47.4 weeks for HIV-positive infants and 48.3 weeks for HIV-

negative. Final weight had a significant correlation of 0.2753 with birthweight

(p<0.001). The adjusted R-squared from the regression of final weight on birthweight

was 0.0729. The graph of the fitted regression line and residuals is presented in FIGURE

4. Although a tight linear fit was not achieved, comparisons with fractional polynomial

regression indicated that a linear model was appropriate and no gain would be made with

power terms in the model.

TABLE 12: Significance of univariate regression of residuals upon potential covariates

Potential Covariates	p-value
HIV-positive infant	<0.001
Estimated age HIV-positive	
• 11.5-21.5 days	
• 22-59.5 days	0.485
• 60-140.5 days	0.155
• 141-366 days	0.670
HIV-positive woman	0.290
Gravida	
• Greater than 2	
• Equal to 2	0.104
• Less than 2	0.012
Male infant	0.008
Gravida equal to 2 with male infant	0.952
Gravida less than 2 with male infant	0.056
Final age (Boxcox lamba=12.672)	0.006
Maternal age (Boxcox lamba=-0.6745)	0.144
Married	0.462
Maternal education less than equal to 8 years	0.195
Husband's education less than 12 years	0.075
Presence of electricity	0.031
Estimated age of first mixed feeding	
• 0-23.5 days	
• 24-64.5 days	0.934
• 65-101 days	0.452
• 101.5-170 days	0.770

The results of univariate regression of the residuals upon potential covariates is summarized in TABLE 12. Covariates significant in univariate analyses were infant HIV status, gravida, sex of the infant, final age of the infant, and presence of electricity. An interaction between gravida and sex was marginally significant (p=0.056). Timing of HIV transmission, maternal HIV status, maternal or husband's education, marital status, and age of first mixed feeding were not significant predictors for the residuals.

Covariates with a p-value less than 0.1 in univariate analysis, plus the interaction between sex and gravida, were included in multivariate analysis. Results of multivariate analysis are summarized in TABLE 13. Adjustments were made to aid in interpretation of the effects of the various covariates. Adjustments for final age and presence of electricity by sex, gravida, and infant HIV status are summarized in TABLE 14. The residuals are the deviation, positive or negative, that an infant's final weight is from the weight that is predicted for the infant based on birthweight. A negative residual would indicate lower growth and a positive residual would indicate higher growth than expected from the infant's birthweight. Only being HIV-negative and mother with gravida equal to one was predictive for a positive residual for female infants. There was trend toward a larger negative residual with increased gravida for HIV-negative and HIV-positive female infants. Within gravida groups, a larger negative residual was predicted for HIV-positive female infants. For male infants, being HIV-negative was predictive for a positive residual across all gravida groups, suggesting that being male is protective against the gravida trends observed with female infants. Negative residuals were

predicted for HIV-positive male infants, but not for their HIV-negative counterparts. No

obvious trends were observed for the effects of gravida upon male infants.

TABLE13: Results of stagewise linear regression of residuals from regression of final weight upon birthweight with potential covariates

Residuals	Coefficient	95% Confidence Interval	p-value
HIV-positive infant	-0.772	-1.140, -0.404	<0.001
Gravida=2	0.394	-0.081, 0.870	0.103
Gravida=1	0.842	0.412, 1.27	<0.001
Male infant	0.763	0.326, 1.200	0.001
Gravida=2 with male infant	0.083	0.615, 0.781	0.815
Gravida=1 with male infant	-0.692	-1.290, -0.094	0.024
Presence of electricity	0.430	0.107, 0.753	0.009
Final age	0.012	0.005, 0.019	<0.001
Constant	-5.130	-7.536, -2.723	<0.001

TABLE 14: Adjusted residuals by infant HIV status, infant sex, and gravida

Sex and HIV Status	Gravida >2	Gravida = 2	Gravida = 1
Female			
• HIV-negative	-0.500	-0.168	0.223
• HIV-positive	-1.305	-0.857	-0.691
Male			
• HIV-negative	0.181	0.720	0.361
• HIV-positive	-0.735	-0.095	-0.435

TABLE 15: Adjusted residuals for female infants by gravida and HIV status

HIV Status	Gravida >2	Gravida = 2	Gravida = 1
HIV-negative	-0.386	0.010	0.411
HIV-positive	-1.078	-0.564	-0.246

Separate regressions for male and female infants would have reduced the power

for each regression, so male and female infants were included in the same regression to

create a model. However, a separate regression was performed with female infants to

assess whether the trend in lower residuals with higher gravida persisted or if it was a reflection of being included in an analysis with male infants who normally have higher weight gains. The trend in negative residuals for HIV-negative female infants with higher gravida did persist for those with gravida greater than two, but not equal to two, after the exclusion of males infants. (TABLE 15)

Infants were scheduled to have growth assessed at four week intervals. Analysis of growth at these intervals for the first year of life was also performed by regressing potential covariates upon weight for each four-week age. Infants were divided into five groups: infants born to HIV-negative women (Controls): infants born to HIV-positive women who did not become HIV-positive in the first year of life (PCR-); infants presumed to be infected at birth (fPCR+); infants infected postpartum (lPCR+); and, infants indeterminate birth or postpartum (ePCR+). The number of infants in each group for each age is summarized in TABLE16.

TABLE 16: Frequency of infants by HIV status and age

Infant Age (weeks)	Controls	PCR-	fPCR+	lPCR+	ePCR+
4	110	196	27	29	17
8	109	196	24	29	17
12	108	192	23	29	16
16	107	184	21	29	15
20	107	182	19	29	13
24	105	179	19	28	12
28	104	179	19	28	12
32	103	175	19	27	10
36	103	173	15	27	10
40	103	172	15	27	9
44	103	169	14	26	9
48	98	161	11	23	7
52	90	153	9	18	6

Unadjusted mean predicted weights by infant HIV for each age are displayed in FIGURE 5. Control and PCR- infants have similar weights throughout the first year of life. Their weight growth is higher than that of infants who became HIV-positive. Weight growth is similar between the groups of infants who became HIV-positive.

FIGURE 5: Unadjusted weights for each age by infant HIV status

Key
O mother negative   + PCR negative   △ fPCR+   ◊ lPCR+   □ ePCR+
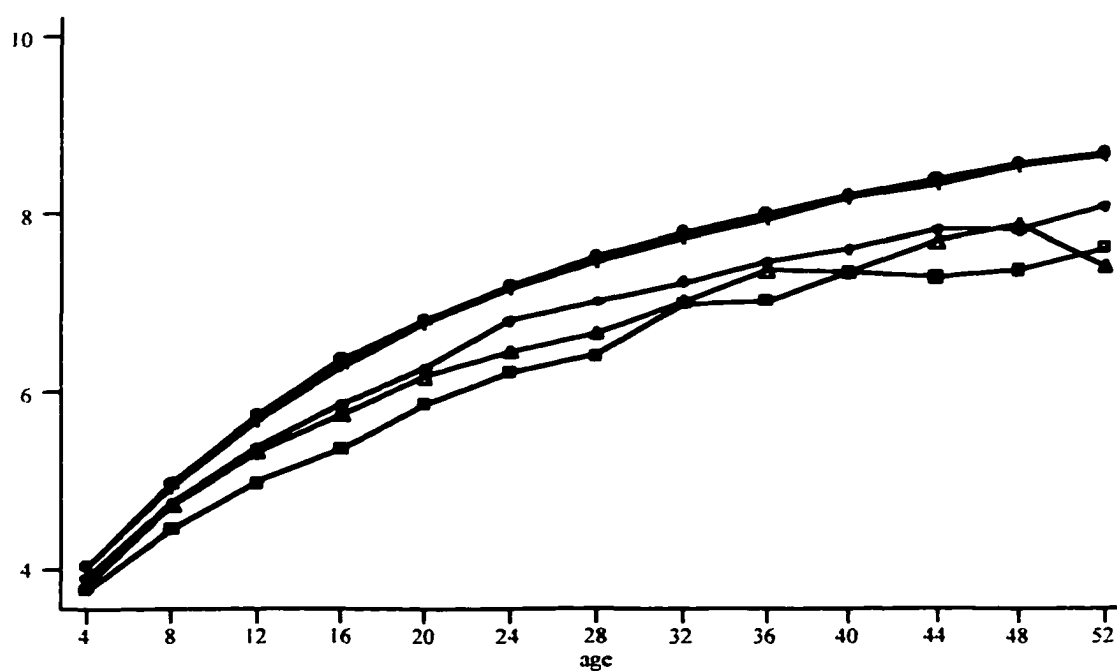
HIV status was then included in ANCOVA for each age with other potential covariates for weight. Coefficients for covariates that were significant are presented in TABLE 17. Detailed results for each ANCOVA, including R-squared values, p-values, and confidence intervals are presented in APPENDIX 1. Higher coefficients for weight were associated with male infants, gravida two or less, presence of the electricity, and higher birthweight. The adjusted weights for these covariates by HIV status are presented in FIGURE 6. No differences were observed in adjusted weights for control and PCR- infants. All groups of infants who became HIV-positive had lower weights at 12 weeks of age throughout the end of the first year of life. Although not significant, lPCR+ infants exhibited higher weights than fPCR+ and ePCR+ infants, with the weight growth of these two groups appearing to be similar.

TABLE 17: Coefficients of significant covariates for ANCOVA of weight for each age in weeks

Age	PCR-	fPCR+	lPCR+	ePCR+	Bweight*	Gravida ≤ 2	Electricity	Male
4	.052	-.052	-.036	-.191	**.939**	.016	.075	.055
8	.026	-.0187	-.159	**-.482**	**.890**	.076	**.152**	**.172**
12	-.013	**-.402**	**-.294**	**-.774**	**.945**	.147	.159	**.256**
16	-.032	**-.662**	**-.474**	**-1.090**	**.925**	**.204**	**.305**	**.280**
20	-.005	**-.772**	**-.470**	**-1.015**	**.869**	**.261**	**.358**	**.279**
24	-.003	**-.860**	-.348	**-1.062**	**.915**	**.327**	**.404**	**.405**
28	-.012	**-.980**	**-.442**	**-1.191**	**.917**	**.348**	**.414**	**.418**
32	-.004	**-.911**	**-.509**	**-1.058**	**.865**	**.428**	**.389**	**.441**
36	.022	**-.854**	-.450	**-1.220**	**.862**	**.390**	**..378**	**.438**
40	.033	**-1.051**	**-.505**	**-1.078**	**.876**	**.398**	**.369**	**.3448**
44	.011	**-.902**	-.466	**-1.342**	**.908**	**.434**	**.326**	**.457**
48	.051	**-.795**	**-.572**	**-1.380**	**.889**	**.480**	**.329**	**.435**
52	.028	**-1.412**	-.429	**-1.313**	**.881**	**.438**	**.438**	**.525**

Bweight = Birthweight in kilograms; significant (p<0.05) in bold
groups: controls (infants born to HIV- women): PCR- (infants born to HIV+ women who did not have a positive PCR) fpcr+ (PCR positive at first scheduled visit), lPCR+ (first PCR negative with a later positive PCR), ePCR+ (first PCR positive but not at the first scheduled visit)

FIGURE 6: Adjusted weights for each age by infant HIV status

Key
O mother negative   + PCR negative   △ lPCR+   ◊ lPCR+   □ ePCR+
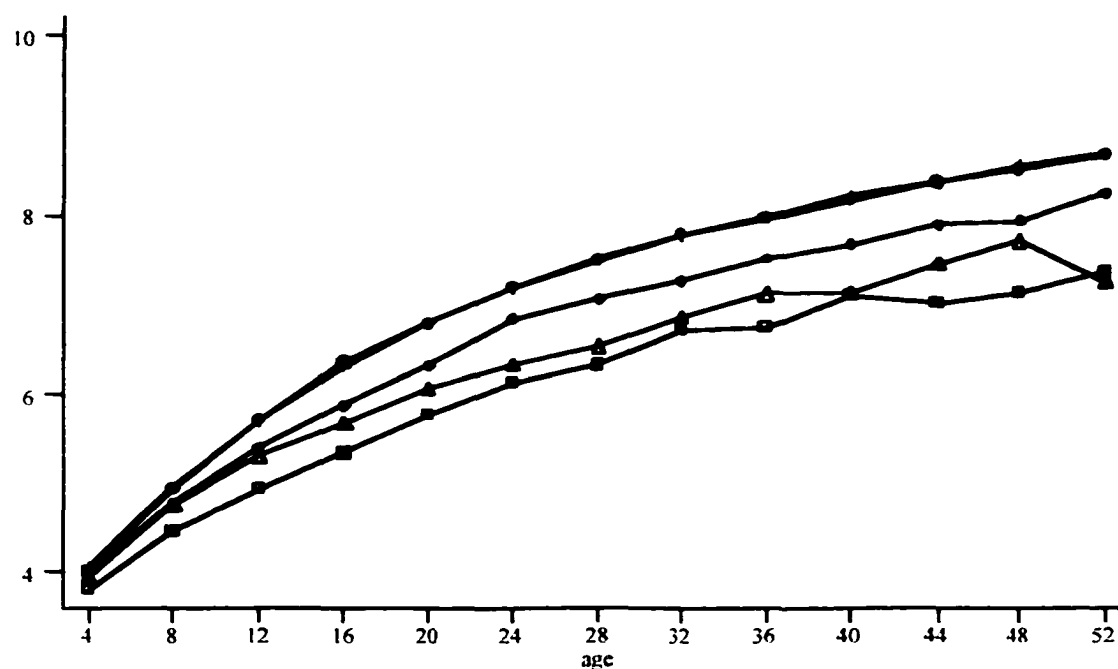
Infants with postpartum infections (lPCR+) were further divided into groups by

the estimated time of infection. Those with estimated age of infection less than five

months were classified as early postpartum, and those later than five months classified as

late postpartum. These two groups did not have adjusted weights significantly different

throughout the first year of life. Although a non-significant trend toward higher weight

was observed for early postpartum infants after 20 weeks of age.

FIGURE 7: Adjusted weights for each age for infants infected postpartum, divided by early and late postpartum

Key
O Early Postpartum (n=17)   + Late Postparum (n=12)   Δ All Postpartum (n=29)
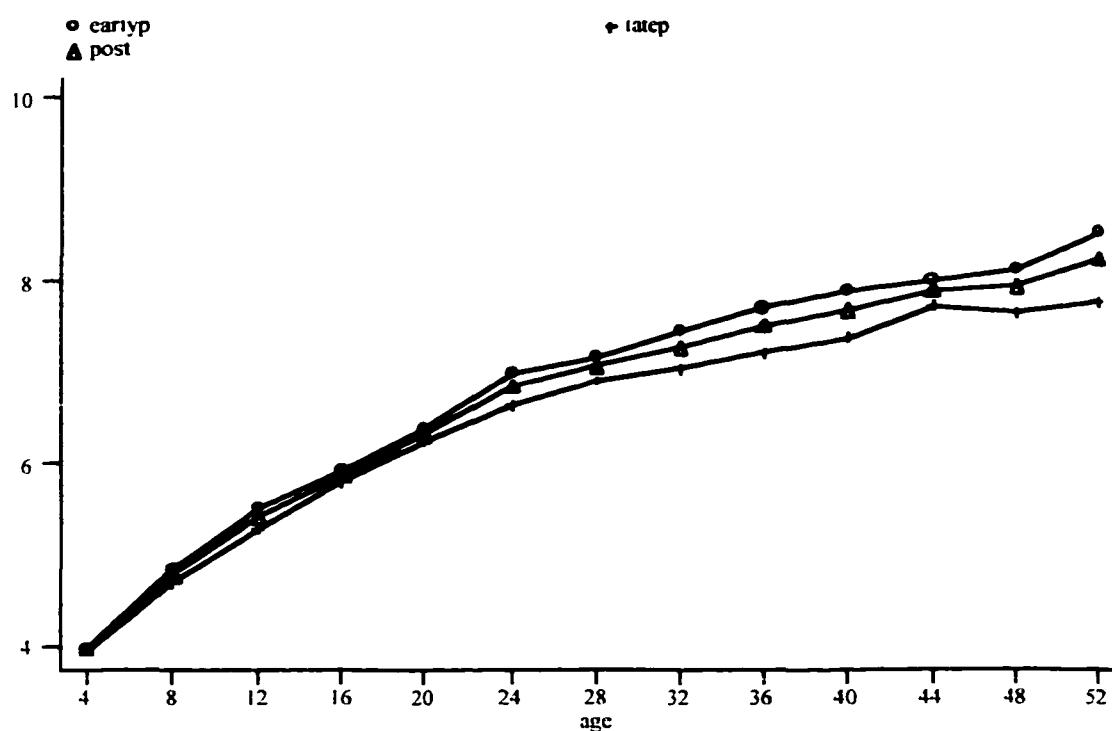
Mortality

There were 70 infants who died within the first year of life. Twenty-nine (36.7%

of total infants that died) were infants who were HIV-positive, thirty-two (16.3%) were

PCR-negative infants, and nine (8.2%) were infants born to HIV-negative women. The

actual age of death is not known for these infants. The number of infants who were

classified as having died is summarized in TABLE 18.

TABLE 18: Infant classification by HIV status

Classification	Controls	PCR-	HIV+
Active	88	135	30
Died	9	32	29
Out of Study	13	29	14

groups: controls (infants born to HIV- women); PCR- (infants born to HIV+ women who did not have a positive PCR), HIV+ (infants born to HIV+ women who had a positive PCR in the first year of life)

Infant HIV status at eight weeks of age was analyzed in logistic regression with other maternal and infant characteristics to asses the significance with later mortality. Predicted weight and being an infant born to an HIV-positive woman were the significant predictors for later mortality (TABLE 19). Each kilogram increased in weight corresponded to a sixty percent lower risk of later mortality. Infants born to HIV-positive women, but still PCR-negative had an almost three-fold greater risk for later mortality compared with infants born to HIV-negative women, and infants who were PCR-positive by eight weeks of age had an almost 18-fold greater risk of later mortality compared with control infants. No confounding or effect modification was observed between HIV status and predicted weight.

TABLE 19: Adjusted odds ratios (95% confidence interval) for mortality by weight and HIV status at eight weeks of age

Age (weeks)	Predicted Weight (n=109)	PCR+ (n=223)	PCR- (n=43)
8 (n=307)	0.39 (0.24, 0.62)	17.87 (6.05, 52.75)	2.83 (1.17, 6.84)

Pearson chi-square goodness of fit: p=0.5900; area under the ROC curve = 0.7755
groups: PCR+ (infants who had a positive PCR by eight weeks of age); PCR- (infants born to HIV+ women but not having a positive PCR- by eight weeks of age)

SUMMARY AND CONCLUSIONS

Women did not differ by HIV status for most demographic characteristics in this analysis. HIV-positive women were more likely to test positive for syphilis than HIV-negative women. The presence of at least one sexually transmitted disease among HIV-positive women has been previously observed (17). There is no prevailing demographic profile of HIV-positive women as compared with HIV-negative women in developing countries. In this sample, HIV-positive women had a lower mean educational level than HIV-negative women. A greater proportion of male infants were born to HIV-positive women than HIV-negative women. There is no known biological reason for this observation. HIV-positive women had lower mean CD4+ counts than did HIV-negative women. This is expected because CD4+ levels decrease with HIV infection progression. No other demographic or health differences were observed between the women by HIV status.

Twenty-seven percent of the infants born to HIV-positive women became HIV-positive. At least ten percent were infected postpartum, with the majority of these infections estimated to have occurred in the first four months of life. These were infants who did not test positive for HIV at their first visit, but tested HIV-positive later. It is likely that more than ten percent were infected postpartum, because without an early infant HIV test it is not possible to distinguish between infants infected from breastfeeding early or those infected in utero or intrapartum. These rates are similar to those seen in other studies. Based upon a critical review of the literature, De Cock et. al.

estimated overall absolute rate of transmission in breastfeeding populations through six months of age to be 25% to 35% (52). Intrauterine transmission rate was estimated to be 5% to 10%. Intrapartum transmission rate was estimated to be 10% to 20%. Postpartum transmission rate within two months after birth was estimated to be 5% to 10%. After two months, the estimated transmission rate was 1% to 5% (52). Although similar to these rates, the postpartum transmission rate is at the higher end of the expected range. Since some of the infants considered indeterminate for presumed birth or postpartum infection were postpartum, this may reflect the healthier status of the HIV-positive women in this study, as compared with cohorts including all HIV-positive women. It could be speculated that since the health of the mother is important for intrauterine and intrapartum transmission, that this type of tranmission may be reduced in a group of healthier mothers. However, the health of the mother and infant may be important in postpartum transmission, so a higher relative transmission rate may follow this reduced birth transmission with healthier mothers. This would have important implications for interventions focused on reducing intrauterine and intrapartum transmission in breastfeeding populations.

Studies conducted in developing countries comparing mean birthweight by maternal HIV status have observed significant differences (13, 14). In a cohort in Rwanda which included 627 pregnant women with 318 of the women being HIV-positive, unadjusted mean birthweights were 0.119 kg lower for infants born to HIV-positive women, compared to infants born to HIV-negative women (13) In a cohort of women in the Democratic Republic of Congo, infants were divided into groups by HIV

status (14). Both groups of infants born to HIV-positive women had significantly lower

mean birthweights than infants born to HIV-negative women, after adjustment for

maternal stature. Contrary to these studies and the original hypothesis, birthweight by

maternal HIV status did not differ in this sample after adjustment for other factors. The

difference in results may partially reflect the difference in adjustment for other factors

and that this study included healthier women.

Results of the analysis of birthweight with maternal HIV status suggest the health

of the mother is an important component of the relationship of HIV status with

birthweight. This is illustrated by the significance of the interaction of the lowest CD4+

category with positive HIV status. Birthweight of infants with mothers in higher CD4+

categories, possibly indicating better overall health, were not affected by positive

maternal HIV status. This suggests that maternal HIV status alone does not seem to

adversely affect birthweight. Further indication that disease progression may be an

important factor associated with birthweight is that AZT administration during pregnancy

decreases the incidence of LBW (54).

The ability to assess birthweight by infant HIV status in developing countries has

been limited because of the use of antibody testing. Although an early PCR test was not

conducted, the best possible distinction was made between infants who were likely to

have been infected at birth or during labor and delivery and infants infected postpartum.

Similar results to the regression with maternal HIV status were observed when

birthweight was compared by infant HIV status. Infant HIV status alone was not a

significant predictor for birthweight. Interaction between presumed birth infection and

maternal CD4+ counts less than 665 µg/L was a significant predictor for lower birthweight. This could suggest a relationship between the health of the mother and early transmission with birthweight. Also, low numbers of the infants in the lPCR+ and ePCR+ groups may have limited the power to detect other differences in birthweight with regard to HIV status and maternal CD4+ levels between them and the other groups.

Gestation, gravida, infant sex and maternal syphilis result were also significant predictors of birthweight. Gestation and infant sex are expected predictors of birthweight. Longer gestation is normally associated with higher birthweight. Male infants are likely to weigh more at birth than female infants. Gravida less than or equal to two was negatively associated with birthweight in both regression analyses. Gravida and birthweight have not been known to be evaluated in other studies. Lower gravida may indicate a younger mother. In this study, gravida and maternal age were significantly and positively correlated. A younger mother suggests that the mother herself may not be physically mature or a lower level of experience. These factors could result in lower infant birthweight. A positive maternal syphilis result at recruitment was also significantly and negatively associated with birthweight. This is difficult to interpret because women who tested positive for syphilis received treatment. In other studies of syphilis and birthweight, no association was observed between mean birthweight of infants born to women with and without syphilis (55, 56). The results of both birthweight analyses provide support for the idea that maintaining health of HIV-positive women, including treatment of concurrent infections and antiretroviral therapy when possible, is important for better birthweight outcomes.

Stagewise regression was utilized to assess the impact of infant HIV status on growth at one year of age. The benefit of stagewise regression is that it eliminates confounding if a variable effects birthweight in a different way than it effects postnatal growth (57). The results of the stagewise regression analysis suggest that being HIV-positive was independently associated with weight at one year of age. HIV-positive infants had negative adjusted mean residuals, indicating that weight gain over the first year of life was less than what would have been predicted from birthweight for these infants. HIV-negative male infants had positive adjusted mean residuals. An interesting relationship for HIV-negative female infants and gravida was observed. HIV-negative female infants with a gravida greater than one had negative adjusted mean residuals. The negative residuals for HIV-negative females with gravida greater than two persisted when females were considered separately from males. The overall trend toward lower residuals for females infants with increasing gravida was still evident after separate analysis of female infants. The mechanism by which gravida influences weight of infants is not clear, but could be speculated to relate to the number of children and food allocation. Gravida may be an indirect indicator of the number of children in the household. More children in the household may influence the amount of food available for each child. Although all infants are breastfed, this is an important consideration because mean introduction of complementary foods for this population is at approximately two months after birth. In addition to HIV status, gravida and sex, the presence of electricity was associated with a positive residual. This suggests the SES of the household may influence infant weight at one year of age. This has been observed with LBW infants in

Brazil. Socioeconomic variables, such as housing variables, income, father's literacy and presence and crowding, explained 21.4% of the variation in maximal weight-for-age z-score gain at one year of age (58). However, the authors speculated that SES was acting prenatally. The results of this analysis are contrary to that speculation. By utilizing stagewise regression, the results suggest that the presence of electricity is acting upon infant weight gain postnatally.

Further information regarding present HIV status and weight was available from this study because timing of infection after birth could be reasonably predicted. After adjustment for other factors known to affect growth, infants who became HIV-positive had lower weight than controls at 12 weeks of age throughout the end of the first year of life. This suggests that HIV status, independent of other factors, is a predictor of lower weight growth compared with uninfected infants. This corresponded to a 1.42 kg and 1.31 kg lower predicted weight at one year of age compared to controls for infants presumed infected at birth and infants indeterminate for presumed birth or postpartum infection. This is a larger difference between HIV-positive infants and uninfected infants than two other studies conducted in developing countries. In the Democratic Republic of Congo, HIV-positive infants were estimated to weigh 0.86 kg less at one year of age than infants born to HIV-negative women (14). Weight was considered as the outcome variable with infant HIV status and other factors in an analysis of infants in Malawi (34). The coefficient for HIV-positive status was –0.63 kg in a model with infant sex, infant age, maternal vitamin A status, and maternal body mass index. These results indicate

HIV-positive status of the infant has a significant negative impact on weight gain of infants in the first year of life, which could influence infant morbidity and mortality.

However, these other studies did not separate infants based on timing of infection. In this study, infants known to be infected postpartum exhibited growth lower than Controls and PCR- infants, but higher after 12 weeks than the other groups of infants infected with HIV earlier. Although this was not significant, it suggests a role in the timing of infection with growth. However, within these known to be infected postpartum, similar growth was observed regardless of whether infection occurred before or after five months of age. A small number of infants in each group may have limited the power to detect differences. It could also be speculated that the length of time infected is important for growth.

Similar to other studies conducted, PCR-negative infants were observed to have predicted weight similar to infants born to HIV-negative women. PCR-negative infants followed for twenty months in the Democratic Republic of Congo and for forty months in Rwanda had weight growth patterns similar to the infants born to HIV-negative women. (14, 33) This suggests that while infant HIV status is an important predictor for weight, maternal HIV status is not significantly associated with infant growth.

HIV-status of the mother and infant, and predicted weight at eight weeks of age were significantly associated with later mortality of infants. The mechanism by which maternal HIV status influences later mortality is not known. It could be speculated tht this is related to the health of the mother. If the mother's health were to decline throughout the first year of infant life, this could impact the mortality of the infant. Being

HIV-positive at eight weeks, was associated with an almost 18-fold higher risk of later death for infants. This suggests that early infection is an important component of mortality in HIV-infected infants. HIV status and growth failure have been observed to be significant predictors of mortality in other studies. Infants born to HIV-positive and HIV-negative mothers were followed from birth until 25 months of age in Uganda to assess growth and mortality (29). A significantly higher proportion of HIV-positive infants died before 25 months of age compared with uninfected infants born to HIV-positive women and infants born to HIV-negative women. Of the HIV-positive infants, an average weight-for-age z-score over the study period of less than −1.5 translated into almost five-fold greater risk of mortality compared with infants with an average z-score greater than −1.5. The question is whether HIV-status alone is predictive for mortality or is mortality for HIV-positive infants a result of impaired growth. No effect modification or confounding was observed between these two variables. This suggests that HIV status and weight are independent predictors of later mortality.

The results of these analyses provide insight into important considerations for future research. Among those, associations between birthweight and intrauterine and intrapartum HIV transmission remain to be fully elucidated. It is also of interest to determine whether lower birthweight or growth impairment after birth are risk factors for postpartum transmission. Additionally, whether infant sex influences growth of HIV-infected children is not known. It would be important to determine mechanisms by which maternal HIV status influences infant mortality, independent of infant HIV status.

In summary, there were no differences in birthweight of the infants solely by either maternal or infant HIV status. An interaction of HIV status and maternal CD4+ counts was predictive for infant birthweight, suggesting a role for the overall health of the mother with birthweight. After birth, uninfected infants born to HIV-positive women had growth similar to infants born to HIV-negative women. However, HIV-positive status was predictive for lower growth of infants. HIV-positive infants gained less weight than other infants. At the end of the first year of life, this translated to approximately one kilogram of difference between HIV-positive infants and uninfected infants. Lower growth and being HIV-positive were both independently predictive of later mortality. These results underscor the importance of continued growth monitoring of HIV-infected children, and the establishment of growth monitoring programs in areas with high HIV prevalence if such programs are not yet in existence.

APPENDIX 1: DETAILED INFORMATION REGARDING PREDICTED WEIGHT ANCOVAS AT FOUR WEEK INTERVALS AS STATA OUTPUT

Key to covariates

agekid	age of the infant in days
pweight	predicted weight from linear splines
i.tgrp	dummy variables for infant by HIV status
Itgrp_1	infants presumed infected at birth
Itgrp_2	infants infected postpartum
Itgrp_3	infants indeterminate for presumed birth infection or postpartum infection
Itgrp_4	infants born to HIV-positive women who did not become HIV-positive
electr	presence of electricity
gravc	gravida less than or equal to 2
sex	male infant

```
STATA  Output
4 weeks
. xi: regress pweight i.tgrp electr bweight gravc sex if agekid==28
i.tgrp                  Itgrp_0-4    (naturally coded; Itgrp_0 omitted)
```

Source	SS	df	MS	
Model	67.0889061	8	8.38611326	
Residual	51.5444805	357	.144382298	
Total	118.633387	365	.325022977	

Number of obs =	366
F(8, 357) =	58.08
Prob > F =	0.0000
R-squared =	0.5655
Adj R-squared =	0.5558
Root MSE =	.37998

pweight	Coef.	Std. Err.	t	P>|t|	[95% Conf.	Interval]
Itgrp_1	-.0522428	.0848116	-0.616	0.538	-.219036	.1145503
Itgrp_2	-.0358925	.0826033	-0.435	0.664	-.1983428	.1265577
Itgrp_3	-.1913966	.1032516	-1.854	0.065	-.3944545	.0116612
Itgrp_4	.0517637	.0466121	1.111	0.268	-.0399051	.1434326
electr	.0750425	.0506802	1.481	0.140	-.0246268	.1747118
bweight	.0009373	.0000461	20.336	0.000	.0008467	.001028
gravc	.0162913	.0429516	0.379	0.705	-.0681786	.1007612
sex	.0546085	.0410852	1.329	0.185	-.026191	.1354081
_cons	.9572268	.160731	5.955	0.000	.6411283	1.273325

8 weeks

. xi: regress pweight i.tgrp electr bweight gravc sex if agekid==56
i.tgrp   Itgrp_0-4   (naturally coded; Itgrp_0 omitted)

Source	SS	df	MS
Model	67.8688699	8	8.48360873
Residual	101.025479	349	.289471287
Total	168.894349	357	.473093415

```
Number of obs =    358
F( 8,  349) =  29.31
Prob > F     = 0.0000
R-squared    = 0.4018
Adj R-squared = 0.3881
Root MSE     = .53803
```

pweight	Coef.	Std. Err.	t	P>|t|	[95% Conf. Interval]	
Itgrp_1	-.1816645	.1282678	-1.416	0.158	-.4339397	.0706107
Itgrp_2	-.1587828	.1172488	-1.354	0.177	-.3893859	.0718202
Itgrp_3	-.4817929	.1464185	-3.291	0.001	-.7697666	-.1938192
Itgrp_4	.025609	.066599	0.385	0.701	-.105377	.1565949
electr	.1523045	.0724049	2.104	0.036	.0098996	.2947094
bweight	.00089	.000066	13.488	0.000	.0007602	.0010198
gravc	.0762289	.0617495	1.234	0.218	-.0452189	.1976768
sex	.171748	.0587295	2.924	0.004	.0562397	.2872564
_cons	1.928767	.2300603	8.384	0.000	1.476288	2.381246

12 weeks

. xi: regress pweight i.tgrp electr bweight gravc sex if agekid==84
i.tgrp   Itgrp_0-4   (naturally coded; Itgrp_0 omitted)

Source	SS	df	MS
Model	82.1511912	8	10.2688989
Residual	158.864814	340	.467249452
Total	241.016005	348	.692574727

```
Number of obs =    349
F( 8,  340) =  21.98
Prob > F     = 0.0000
R-squared    = 0.3409
Adj R-squared = 0.3253
Root MSE     = .68356
```

pweight	Coef.	Std. Err.	t	P>|t|	[95% Conf. Interval]	
Itgrp_1	-.4017399	.1666316	-2.411	0.016	-.7294987	-.0739812
Itgrp_2	-.2941358	.1491674	-1.972	0.049	-.5875429	-.0007287
Itgrp_3	-.7735763	.1908754	-4.053	0.000	-1.149022	-.398131
Itgrp_4	-.0133484	.0853065	-0.156	0.876	-.1811434	.1544465
electr	.1588337	.0928199	1.711	0.088	-.0237398	.3414073
bweight	.0009453	.0000856	11.047	0.000	.000777	.0011136
gravc	.1470054	.0798814	1.840	0.067	-.0101187	.3041295
sex	.2557245	.075643	3.381	0.001	.1069373	.4045117
_cons	2.436585	.3002203	8.116	0.000	1.846062	3.027108

16 weeks
. xi: regress pweight i.tgrp electr bweight gravc sex if agekid==112
i.tgrp   Itgrp_0-4   (naturally coded; Itgrp_0 omitted)

Source	SS	df	MS
Model	90.6933032	8	11.3366629
Residual	178.248545	327	.545102585
Total	268.941849	335	.802811488

Number of obs = 336
F(8, 327) = 20.80
Prob > F = 0.0000
R-squared = 0.3372
Adj R-squared = 0.3210
Root MSE = .73831

pweight	Coef.	Std. Err.	t	P>ItI	[95% Conf.	Interval]
Itgrp_1	-.6617225	.1881081	-3.518	0.000	-1.031777	-.2916678
Itgrp_2	-.473735	.1614916	-2.933	0.004	-.7914285	-.1560414
Itgrp_3	-1.009736	.2125127	-4.751	0.000	-1.4278	-.591671
Itgrp_4	-.0316138	.0933476	-0.339	0.735	-.2152514	.1520237
electr	.3050176	.102349	2.980	0.003	.103672	.5063631
bweight	.0009246	.0000948	9.755	0.000	.0007382	.0011111
gravc	.2035784	.087845	2.317	0.021	.0307657	.3763911
sex	.2799292	.0831883	3.365	0.001	.1162774	.443581
_cons	3.063346	.3339226	9.174	0.000	2.406439	3.720254

20 weeks
. xi: regress pweight i.tgrp electr bweight gravc sex if agekid==140
i.tgrp   Itgrp_0-4   (naturally coded; Itgrp_0 omitted)

Source	SS	df	MS
Model	85.6093731	8	10.7011716
Residual	228.643389	325	.70351812
Total	314.252762	333	.943701989

Number of obs = 334
F(8, 325) = 15.21
Prob > F = 0.0000
R-squared = 0.2724
Adj R-squared = 0.2545
Root MSE = .83876

pweight	Coef.	Std. Err.	t	P>ItI	[95% Conf.	Interval]
Itgrp_1	-.7217361	.2229854	-3.237	0.001	-1.160413	-.2830591
Itgrp_2	-.4699353	.1785375	-2.632	0.009	-.8211703	-.1187003
Itgrp_3	-1.014688	.2497813	-4.062	0.000	-1.50608	-.5232958
Itgrp_4	-.0051649	.1058638	-0.049	0.961	-.2134298	.2031
electr	.3580484	.116403	3.076	0.002	.1290499	.587047
bweight	.0008686	.0001073	8.094	0.000	.0006575	.0010797
gravc	.2609927	.099883	2.613	0.009	.064494	.4574915
sex	.2793149	.0945816	2.953	0.003	.0932454	.4653844
_cons	3.642153	.3793897	9.600	0.000	2.895783	4.388522

24 weeks

. xi: regress pweight i.tgrp electr bweight gravc sex if agekid==168
i.tgrp   Itgrp_0-4   (naturally coded; Itgrp_0 omitted)

Source	SS	df	MS
Model	99.4904384	8	12.4363048
Residual	247.609093	319	.776204054
Total	347.099532	327	1.06146646

Number of obs = 328
F(8, 319) = 16.02
Prob > F = 0.0000
R-squared = 0.2866
Adj R-squared = 0.2687
Root MSE = .88102

pweight	Coef.	Std. Err.	t	P>|t|	[95% Conf. Interval]	
Itgrp_1	-.8601618	.2343766	-3.670	0.000	-1.321281	-.3990426
Itgrp_2	-.3476851	.1902476	-1.828	0.069	-.7219837	.0266135
Itgrp_3	-1.062194	.2716586	-3.910	0.000	-1.596663	-.5277254
Itgrp_4	-.0025776	.1119663	-0.023	0.982	-.2228632	.2177081
electr	.4039538	.1226305	3.294	0.001	.162687	.6452206
bweight	.0009149	.0001143	8.002	0.000	.00069	.0011399
gravc	.3270299	.1050918	3.112	0.002	.1202693	.5337905
sex	.4049024	.1002486	4.039	0.000	.2076705	.6021343
_cons	3.783654	.402061	9.411	0.000	2.992627	4.57468

28 weeks

. xi: regress pweight i.tgrp electr bweight gravc sex if agekid==196
i.tgrp   Itgrp_0-4   (naturally coded; Itgrp_0 omitted)

Source	SS	df	MS
Model	108.665766	8	13.5832207
Residual	285.31122	321	.88882
Total	393.976986	329	1.19749844

Number of obs = 330
F(8, 321) = 15.28
Prob > F = 0.0000
R-squared = 0.2758
Adj R-squared = 0.2578
Root MSE = .94277

pweight	Coef.	Std. Err.	t	P>|t|	[95% Conf. Interval]	
Itgrp_1	-.9799246	.2509572	-3.905	0.000	-1.473653	-.4861961
Itgrp_2	-.4418868	.2037282	-2.169	0.031	-.8426979	-.0410757
Itgrp_3	-1.191391	.2908844	-4.096	0.000	-1.763672	-.6191107
Itgrp_4	-.012216	.1198056	-0.102	0.919	-.2479194	.2234874
electr	.4135459	.1302824	3.174	0.002	.1572307	.669861
bweight	.0009167	.0001218	7.525	0.000	.000677	.0011564
gravc	.3479381	.1121434	3.103	0.002	.1273092	.5685671
sex	.4179834	.1069884	3.907	0.000	.2074965	.6284704
_cons	4.067545	.4282407	9.498	0.000	3.225032	4.910058

32 weeks
```
. xi: regress pweight i.tgrp electr bweight gravc sex if agekid==224
i.tgrp                   Itgrp_0-4    (naturally coded; Itgrp_0 omitted)
```

Source	SS	df	MS			
Model	94.7126115	8	11.8390764			
Residual	300.087043	314	.955691219			
Total	394.799654	322	1.22608588			

Number of obs = 323
F(8, 314) = 12.39
Prob > F = 0.0000
R-squared = 0.2399
Adj R-squared = 0.2205
Root MSE = .97759

pweight	Coef.	Std. Err.	t	P>ltl	[95% Conf.	Interval]
Itgrp_1	-.9111327	.2545808	-3.579	0.000	-1.412032	-.4102329
Itgrp_2	-.5089578	.2149871	-2.367	0.019	-.9319552	-.0859605
Itgrp_3	-1.058337	.3263152	-3.243	0.001	-1.700378	-.4162968
Itgrp_4	-.0044858	.1252466	-0.036	0.971	-.2509144	.2419429
electr	.3894295	.1378414	2.825	0.005	.11822	.6606391
bweight	.0008654	.0001308	6.614	0.000	.000608	.0011228
gravc	.4281785	.1179132	3.631	0.000	.1961787	.6601783
sex	.4405349	.1123659	3.921	0.000	.2194495	.6616202
_cons	4.449473	.4592811	9.688	0.000	3.545815	5.35313

36 weeks
```
. xi: regress pweight i.tgrp electr bweight gravc sex if agekid==252
i.tgrp                   Itgrp_0-4    (naturally coded; Itgrp_0 omitted)
```

Source	SS	df	MS			
Model	86.5433941	8	10.8179243			
Residual	331.765013	305	1.08775414			
Total	418.308407	313	1.33644859			

Number of obs = 314
F(8, 305) = 9.95
Prob > F = 0.0000
R-squared = 0.2069
Adj R-squared = 0.1861
Root MSE = 1.043

pweight	Coef.	Std. Err.	t	P>ltl	[95% Conf.	Interval]
Itgrp_1	-.8542213	.302632	-2.823	0.005	-1.449732	-.2587105
Itgrp_2	-.4500399	.2335819	-1.927	0.055	-.9096759	.0095962
Itgrp_3	-1.219995	.3483164	-3.503	0.001	-1.905403	-.5345881
Itgrp_4	.0222232	.1349212	0.165	0.869	-.2432711	.2877174
electr	.3772555	.1488552	2.534	0.012	.0843424	.6701686
bweight	.0008624	.0001428	6.038	0.000	.0005813	.0011434
gravc	.3903487	.1285946	3.035	0.003	.1373038	.6433937
sex	.4376068	.1210606	3.615	0.000	.199387	.6758265
_cons	4.678861	.5042375	9.279	0.000	3.686636	5.671085

40 weeks
. xi: regress pweight i.tgrp electr bweight gravc sex if agekid==280
i.tgrp   Itgrp_0-4   (naturally coded; Itgrp_0 omitted)

```
    Source |      SS       df       MS              Number of obs =     315
-----------+------------------------------         F( 8,    306) =    9.98
     Model |  92.8328725      8  11.6041091         Prob > F      = 0.0000
  Residual |  355.786797    306  1.16270195         R-squared     = 0.2069
-----------+------------------------------         Adj R-squared = 0.1862
     Total |   448.61967    314  1.42872506         Root MSE      = 1.0783
```

```
   pweight |     Coef.   Std. Err.       t     P>|t|     [95% Conf. Interval]
-----------+----------------------------------------------------------------
    Itgrp_1 | -1.051029   .3030294    -3.468   0.001    -1.647314   -.454744
    Itgrp_2 | -.5053551   .2418821    -2.089   0.038    -.9813178   -.0293925
    Itgrp_3 | -1.078232   .3776619    -2.855   0.005    -1.821375   -.3350892
    Itgrp_4 |  .0326407   .1396045     0.234   0.815    -.2420656    .3073471
     electr |  .3693659   .1536959     2.403   0.017     .0669313    .6718005
    bweight |  .0008763    .000146     6.001   0.000     .000589     .0011636
      gravc |  .3981469   .1330101     2.993   0.003     .1364167    .6598771
        sex |  .4478254   .1246991     3.591   0.000     .2024492    .6932016
      _cons |  4.834618   .5181547     9.330   0.000     3.815021    5.854215
```

44 weeks
. xi: regress pweight i.tgrp electr bweight gravc sex if agekid==308
i.tgrp   Itgrp_0-4   (naturally coded; Itgrp_0 omitted)

```
    Source |      SS       df       MS              Number of obs =     312
-----------+------------------------------         F( 8,    303) =    9.50
     Model |  94.2197994      8  11.7774749         Prob > F      = 0.0000
  Residual |  375.805295    303   1.2402815         R-squared     = 0.2005
-----------+------------------------------         Adj R-squared = 0.1793
     Total |  470.025094    311  1.51133471         Root MSE      = 1.1137
```

```
   pweight |     Coef.   Std. Err.       t     P>|t|     [95% Conf. Interval]
-----------+----------------------------------------------------------------
    Itgrp_1 | -.9018913   .3220649    -2.800   0.005    -1.535658   -.2681243
    Itgrp_2 |  -.465686   .2535766    -1.836   0.067    -.9646802    .0333082
    Itgrp_3 | -1.341935   .3900371    -3.441   0.001    -2.109459   -.5744108
    Itgrp_4 |  .0113834   .1441391     0.079   0.937    -.272257     .2950238
     electr |  .3257501    .159985     2.036   0.043     .0109278    .6405725
    bweight |  .0009079   .0001507     6.026   0.000     .0006114    .0012044
      gravc |  .4344245   .1385798     3.135   0.002     .1617239    .7071251
        sex |  .4566491   .1293737     3.530   0.000     .2020643    .7112338
      _cons |  4.896902   .5368593     9.121   0.000     3.840457    5.953346
```

48 weeks
. xi: regress pweight i.tgrp electr bweight gravc sex if agekid==336
i.tgrp   Itgrp_0-4   (naturally coded; Itgrp_0 omitted)

Source	SS	df	MS			Number of obs = 292
						F(8, 283) = 9.22
Model	90.9805274	8	11.3725659			Prob > F = 0.0000
Residual	348.985414	283	1.23316401			R-squared = 0.2068
						Adj R-squared = 0.1844
Total	439.965941	291	1.51191045			Root MSE = 1.1105

pweight	Coef.	Std. Err.	t	P>ItI	[95% Conf. Interval]	
Itgrp_1	-.795365	.356606	-2.230	0.027	-1.497302	-.0934281
Itgrp_2	-.5718896	.2645159	-2.162	0.031	-1.092558	-.0512214
Itgrp_3	-1.380309	.436234	-3.164	0.002	-2.238984	-.5216336
Itgrp_4	.0510239	.1474038	0.346	0.729	-.2391231	.3411709
electr	.3289375	.1646735	1.998	0.047	.0047971	.6530779
bweight	.0008884	.0001561	5.691	0.000	.0005812	.0011957
gravc	.480424	.1418487	3.387	0.001	.2012117	.7596363
sex	.4350669	.1326146	3.281	0.001	.1740308	.6961031
_cons	5.090059	.5581058	9.120	0.000	3.991494	6.188625

52 weeks
. xi: regress pweight i.tgrp electr bweight gravc sex if agekid==364
i.tgrp   Itgrp_0-4   (naturally coded; Itgrp_0 omitted)

Source	SS	df	MS			Number of obs = 271
						F(8, 262) = 10.05
Model	98.1185842	8	12.264823			Prob > F = 0.0000
Residual	319.888067	262	1.22094682			R-squared = 0.2347
						Adj R-squared = 0.2114
Total	418.006651	270	1.54817278			Root MSE = 1.105

pweight	Coef.	Std. Err.	t	P>ItI	[95% Conf. Interval]	
Itgrp_1	-1.419973	.3920607	-3.622	0.000	-2.191964	-.6479818
Itgrp_2	-.4285081	.2954178	-1.451	0.148	-1.010203	.1531871
Itgrp_3	-1.31309	.4676859	-2.808	0.005	-2.233991	-.3921883
Itgrp_4	.0281098	.1522114	0.185	0.854	-.2716035	.3278231
electr	.413743	.1720845	2.404	0.017	.0748984	.7525875
bweight	.0008812	.0001583	5.568	0.000	.0005696	.0011929
gravc	.4381843	.145771	3.006	0.003	.1511526	.725216
sex	.5249491	.1381844	3.799	0.000	.2528557	.7970425
_cons	5.237668	.5614531	9.329	0.000	4.132134	6.343203

REFERENCES

1. UNAIDS. World AIDS Report December 1999. www.unaids.org/

2. McCORMICK M.C. (1985) The contribution of low birth weight to infant mortality and childhood morbidity. *New Engl. J. Med.* 312: 82-90.

3. BLOLAND, P., SLUTSKER, L., SKETEKEE, R.W., WIRIMA, J.J., HEYMENN, D.L., & BREMAN, J.G. (1996) Rates and risk factors for mortality during the first two years of life in rural Malawi. *Am. J. Trop. Med. Hyg.* 55: 82-86.

4. KUHN, L., ABRAMS, E.J., MATHESON, P.B., THOMAS, P.A., LAMBERT, G., BAMJI, M., GREENBURG, B., STEKETEE, R.W., THEA, D.M., & THE NEW YORK CITY PERINATAL HIV TRANSMISSION COLLABERATIVE STUDY GROUP (1997) Timing of maternal-infant HIV transmission: association between intrapartum factors and early polymerase chain reaction results. *AIDS* 11: 429-435.

5. BRYSON, Y.J., LUZURIAGA, K., SULLIVAN, J.L., & WARA, D.W. (1992) Proposed definitions for in utero versus intrapartum transmission of HIV-1. *New Engl. J. Med.* 327: 1246-1247.

6. KUHN, L., ABRAMS, E.J., CHINCHILLA, M., TSAI, W.Y., & THEA, D.M. (1996) Sensitivity of HIV-1 DNA polymerase chain reaction in the neonatal period. *AIDS* 10: 1181-1182.

7. ALGER, L.S., FARLEY, J.J., ROBINSON, B.A., HINES, S.E., BERCHIN, J.M., & JOHNSON, J.P. (1993) Interactions of human immunodeficiency virus infection and pregnancy. *Obstet. Gynecol.* 82: 787-796.

8. JOHNSTONE, F.D., RAAB, G.M., & HAMILTON, B.A. (1996) The effect of human immunodeficiency virus interaction and drug use on birth characteristics. *Obstet. Gynecol.* 88: 321-326.

9. BUTZ, A., HUTTON, N., & LARSON, E. (1991) Immunoglobulins and growth parameters at birth of infants born to HIV seropositive and seronegative women. *Am. J. Public Health* 81: 1323-1326.

10. MINKOFF, H.L., HENDERSON, C., MENDEZ, H., GAIL, M.H., HOLMAN, S., WILLOUGHBY, A., GOEDETT, J.J., RUBINSTEIN, A., STRATTON, P., WALSH, J.H., & LANDESMAN, S.H. (1990) Pregnancy outcomes among mother infected with human immunodeficiency virus and uninfected control subjects. *Am. J. Obstet. Gynoceol.* 162: 1598-1604.

11. SELWYN, P.A., SCHOENBAUM, E.E., DAVENRY, K., ROBERTSON, V.J., FEINGOLD, A.R., SHULMAN, J.F., MAYERS, M.M., KLEIN, R.S., FRIEDLAND, G.H., & ROGERS, M.F. (1989) Prospective study of human immunodeficiency virus infection and pregnancy outcomes in intravenous drug users. *JAMA* 261: 1289-1294.

12. MARKSON, L.E., TURNER, B.J., HOUCHENS, R., SILVERMAN, N.S., COSLER, L., & TAKYI, B.K. (1996) Association of maternal HIV infection with low birth weight. *J. Acquir. Immune Defic. Syndr. Human Retrovirol.* 13: 227-234.

13. JOHNSTONE, F.D., MACCALLUM, L., BRETTLE, R., INGLIS, J.M., & PEUTHERER, J.F., Does infection with HIV affect the outcome of pregnancy? *Br. Med. J.* 296: 467

14. BULTERYS, M., CHAO, A., MUNYEMANA, S., KURAWIGE, J-B., NAWROCKI, P., HABIMANA, P., KAGERUKA, M., MUKANTABANA, S., MBARUTSO, E., DUSHIMIMANA, A., & SAAH, A. (1994) Maternal human immunodeficiency virus 1 infection and intrauterine growth: a prospective cohort study in Butare, Rwanda. *Pediatr. Infect. Dis. J.* 13: 94-100.

15. BAILEY, R.C., KAMENGA, M.C., NSUAMI, M.J., NIEBURG, P., & ST. LOUIS, M.E. (1999) Growth of children according to maternal and child HIV, immunological and disease characteristics: a prospective cohort study in Kinshasa, Democratic Republic of Congo. *Int. J. Epidemiol.* 28: 532-540.

16. HALSEY, N.A., BOULOS, R., HOLT, E., RUFF, A., BRUTUS, J-R., KISSINGER, P., QUINN, T.C., COBERLY, J.S., ADRIEN, M., BUOLOS, C., AND THE CDS/JHU AIDS PROJECT TEAM (1990) Transmission of HIV-1 infections from mothers to infants in Haiti. *JAMA* 264: 2088-2092.

17. LEROY, V., LADNER, J., NYIRAZIRAJE, M., DE CLERCQ, A., BAZUBAGIRA, A., VAN DE PERRE, P., KARITA, E., AND DABIS, F. (1998) Effect of HIV-1 infection on pregnancy outcome in women in Kigali, Rwanda, 1992-1994. *AIDS* 12: 643-650.

18. SUTTON, M.Y., STERNBERG, M, NSUAMI, M., BEHETS, F., & ST. LOUIS, M.E. (1999) Trichomoniasis in pregnant human immunodeficiency virus-infected Congolese women: prevalence, risk factors, and association with low birth weight. *Am. J. Obstet. Gynecol.* 181: 656-662.

19. TAHA, T.E.T., DALLABETTA, G.A., CANNER, J.K., CHIPHANGWI, J.D., LIOMBA, G., HOOVER, D.R., & MIOTTI, P.G. (1995) The effect of human immunodeficiency virus infection on birthweight, and infant and child mortality in urban Malawi. *Int. J. Epidemiol.* 24: 1022-1029.

20. BROCKLEHURST, P. & FRENCH, R. (1998) The association between maternal HIV infection and perinatal outcome: a systematic review of the literature and meta-analysis. *Br. J. Obstet. Gynecol.* 105: 836-848.

21. THE EUROPEAN COLLABERATIVE STUDY (1995) Weight, height and human immunodeficiency virus infection in young children of infected mothers. *Pediatr. Infect Dis. J.* 14: 685-690.

22. MILLER, T.L., EVANS, S.J., ORAV, E.J., MORRIS, V., MCINTOSH, K. & WINTER, H.S. (1993) Growth and body composition in children infected with the human immunodeficiency virus. *Am. J. Clin. Nutr.* 57: 588-592.

23. MOYE, J., RICH, K.C., KALISH, L., SHEON, A.R., DIAZ, C., COPPER, E.R., PITT, J., & HANDELSMAN, E. (1996) Natural history of somatic growth in infants born to women infected by human immunodeficiecy virus. *J. Pediatr.* 128: 58-69.

24. POLLACK, H., GLASBERG, H., LEE, E., NIRENBERG, A., DAVID, R., KRASINSKI, K., BORKOWSKY, W., & OBERFIELD, S. (1997) Impaired early growth of infants perinatally infected with human immunodeficiency virus: correlation with viral load. *J. Pediatr.* 130: 915-922.

25. SAAVEDRA, J.M., HENDERSON, R.A, PERMAN, J.A., HUTTON, N., LIVINGSTON, R.A., YOLKEN, R.H. (1995) Longitudinal assessment of growth in children born to mothers with human immunodeficiency virus infection. *Arch. Pediatr. Adolesc. Med.* 149: 497-502.

26. MOCK, P.A., SHAFFER, N., BHADRAKOM, C., SIRIWASIN, W., CHOTPITAYSUNONDH, T., CHEARSKUL, S., YOUNG, N.L., ROONGPISUTHIPONG, A., CHINAYON, P., KALISH, M.L., PAREKH, B., & MASTRO, T.D. (1999) Maternal viral load and timing of mother-to-child HIV transmission, Bangkok, Thailand. *AIDS* 13: 407-414.

27. THEA, D.M, ST. LOUIS, M.E., ATIDO, U., KANJINGA, K., KEMBO, B., MATONDO, M., TSHIAMALA, T., KAMENGA, C., DAVACHI, F., BROWN, C., RAND, W.M., & KEUSCH, G.T. (1993) A prospective study of diarrhea and HIV-1 infection among 429 Zairian infants. *N Engl. J. Med.* 329: 1696-1702.

28. IKEOGU, M.O., WOLF, B., & MATHE, S. (1997) Pulmonary manifestations in HIV seropositivity and malnutrition in Zimbabwe. *Arch. Dis. Child* 76: 124-128.

29. TAHA, T.E.T., KUMWENDA, N.I., BROADHEAD, R.L., HOOVER, D.R., GRAHAM, S.M., VAN DER HOVEN, L., MRKAKIS, D., LIOMBA, G.N., CHIPHANGWI, J.D., & MIOTTI, P.G. (1999) Mortality after the first year of life among human immunodeficiency virus type-1-infected and uninfected children. *Pediatr. Infect. Dis. J.* 18: 689-694.

30. BERHANE, R., BAGENDA, D., MARUM, L., ACENG, E., NDUGWA, C., BOSCH, R.J., & OLNESS, K. (1997) Growth failure as a prognostic indicator of mortality in pediatric HIV infection. *Pediatrics* 100: e7.

31. FONTANA, M., ZUIN, G., PLEBANI, A., BASTONI, K., VISCONTI, G., & PRINCIPI, N. (1999) Body composition in HIV-infected children: relations with disease progression and survival. *Am. J. Clin. Nutr.* 69: 1282-1286.

32. CAREY, V.J., YONG, F.H, FRENKEL, L.M., & MCKINNEY JR., R.E. (1998) Pediatric AIDS prognosis using somatic growth velocity. *AIDS* 12: 1361-1369.

33. MCKINNEY JR., R.E., WILFERT, C., & THE AIDS CLINICAL TRIALS GROUP PROTOCOL 043 STUDY GROUP (1994) Growth as a prognostic indicator in children with human immunodeficiency virus infection treated with zidovudine. *J. Pediatr.* 125: 728-733.

34. LEPAGE, P., MSELLATI, P., HITIMANA, D-G., BAZUBAGIRA, A., VAN GOETHEM, C., SIMONON, A., KARITA, E., DEQUAE-MERCHADOU, L., VAN DE PERRE, P., & DABIS, F. (1996) Growth of human immunodeficiency type-1 infected and uninfected children: a prospective cohort study in Kigali, Rwanda, 1988 to 1993. *Pediatr. Infect. Dis. J.* 15: 479-485.

35. SEMBA, R.D., MIOTTI, P., CHIPHANGWI, J.D., HENDERSON, R., DALLABETA, G., YANG, L-P., & HOOVER, D. (1997) Maternal vitamin A deficiency and child growth failure during human immunodeficiency virus infection. *J. Acquir. Immune Defic. Syndr. Human Retrovirol.* 14: 219-222.

36. MCKINNEY JR., R.E., ROBERTSON, W.R., & THE DUKE PEDIATRIC AIDS CLINICAL TRIALS UNIT (1993) Effect of human immunodeficiency virus infection on the growth of young children. *J. Pediatr.* 123: 579-582.

37. PETERS, V.B., ROSH, J.R., MUGRDITCHIAN, L., BIRNBAUM, A.H., BENKOV, K.J., HODES, D.S., & LELEIKO, N.S. (1998) Growth failure as the first expression of malnutrition in children with human immunodeficiency virus infection. *Mt. Sinai. J. Med.* 65: 1-4.

38. WAIBALE, P., BROWLIN, S.J., MORTIMER JR., E.A., & WHALEN, C. (1999) The effect of human immunodeficiency virus-1 infection and stunting on measles immunoglobulin-G levels in children vaccinated against measles in Uganda. *Int. J. Epidemiol.* 28: 341-346.

39. AGOSTONI, C., ZUCCOTTI, G.V., GIOVANNINI, M., DECARLIS, S., GIANNI, M.L., PIACENTINI, E., D'AURIA, E., & RIVA, E. (1998) Growth in the first two years of uninfected children born to HIV-1 seropositive mothers. *Arch. Dis. Child.* 79: 175-178.

40. GARG, S. & MILLER, T.L. (1999) Special considerations for the pediatric patient, in *Nutritional Aspects of HIV Infection.* Miller, T.L. & Gorbach, S.L., Ed., Oxford University Press, New York, chap. 12.

41. Miller, T.L., Evans, S.E., Vasquez, I., Orav, E.J. (1997) Dietary intake is an important predictor of nutritional status in HIV-infected children. *Pediatr. Res.*41: 85A (abstract 497).

42. Miller, T.L., Orav, E.J., Martin, S.R., Cooper. E.R., McIntosh, K., Winter, H.S. (1991) Malnutrition and carbohydrate malabsorption in children infected by HIV. *Gastroenterology* 100: 1296-1302.

43. Zuin, G., Fontana, M., Monti, S., Marchisio, P., Beretta, P., Principi, N. (1992) Malabsorption of different lactose load in children with human immunodeficiency virus infection. *J. Gastroenterol. Nutr.* 15: 408-412.

44. Cunningham-Rundles, S., Kim, S.H., Dnistrian, A., Noroski, L., Menendez-Botet, C., Grassey, C.B., Hinds, G., & Cervia, J.S. (1996) Micronutrient and cytokine interaction in congenital pediatric HIV infection. *J. Nutr.* 126: 2674S-2679S.

45. Schwartz, L.J., St. Louis, Y., Wu, R., Wiznia, A., Rubinstein, A., & Saenger, P. (1991) Endocrine function in children with human immunodeficiency virus infection. *AJDC* 145: 330-333.

46. Laue, L., Pizzo, P.A., Butler, K., & Cutler Jr., G.B. (1990) Growth and neuroendocrine dysfunction in children with acquired immune deficiency syndrome. *J. Pediatr.* 117: 541-545.

47. Tamura. S., Udagawa, N., Takahasi, N., Miyaura, C., Tanaka, S., Yamada, Y., Koishihara, Y., Ohsugi, Y., Kumuki, K., Taga, T., Kishimoto, T., & Suda, T. (1993) Soluble interleukin-6 receptor triggers osteoclast formation by interleukin 6. *Proc. Natl. Acad. Sci. USA* 90: 11924-11928.

48. Breen, E.C., Rezai, A.R., Nakajima, K., Beall, G.N., Mittyasi, R.T., Huano, T., Kishimoto, T., & Martinez-Maza, O. (1990) Infection with HIV is associated with elevated IL-6 levels and production. *J. Immunol.* 144: 480-484.

49. Gurram, M., Chimule, N., Wang, X.P., Ponugoti, N., & Pahwa, S. (1994) Increased spontaneous secretion of interleukin 6 and tumor necrosis factor alphaby peripheral blood lymphocytes of human immunodeficiency virus-infected children. *Pediatr. Infect. Dis. J.* 13: 496-501.

50. Mata, L. (1992) Diarrheal disease as a cause of malnutrition. *Am. J. Trop. Med. Hyg.* 47: 16-27.

51. Brown, K.H., Stallings, R.Y., de Kanashiro, H.C., de Romana, G.L., & Black, R.E. (1990) Effects of common illnesses on infants' energy intakes from breast milk and other foods during longitudinal community-based studies in Huascar (Lima), Peru. *Am. J. Clin. Nutr.* 52: 1005-1013.

52. Janini, L.M., Pieniazek, D., Peralta, J.M., Schecter, M., Vicente, A.C.P., dela Torre N., Pieniazek N.J., Kalish, M.L., Schochetman, G., & Rayfield, M.A. (1996) Identification of single and dual infections with distinct subtypes of human immunodeficiency virus type 1 by using restriction fragment length polymorphism analysis. *Virus Genes* 13: 69-81.

53. De Cock, K.M., Fowler, M.G., Mercier, E., de Vincenzi, I., Saba, J., Hoff, E., Alnick, D.J., Rogers, M., & Shaffer, N. (2000) Prevention of mother-to-child HIV transmission in resource-poor countries. *JAMA* 283: 1175-1182.

54. The European Collaberative Study. (1998) Is zidovudine therapy in pregnant HIV-infected women associated with birthweight? *AIDS* 13: 119-124.

55. Donders, G.G., Desmyter, J., De Wet, D.H., & Van Assche, F.A. (1993) The association of gonorrhoea and syphilis with premature birth and low birthweight. *Genitourin. Med.* 69: 98-101.

56. How, J.H. & Bowditch, J.D. (1994) Syphilis in pregnancy: experience from a rural aborginal community. *Aust. N. Z. J. Obstet. Gynaecol.* 34: 383-389.

57. Esrey, S.A., Casella, G., & Habicht, J-P. (1990) The use of residuals for longitudinal data analysis: the example of child growth. *Am. J. Epidemiol.* 131: 365-372.

58. Ashworth, A., Morris, S.S., & Lira, P.I.C. (1997) Postnatal growth patterns of full-term low birth weight infants in northeast Brazil are related to socioeconomic status. *J. Nutr.* 127: 1950-1956.