Maternal nutritional status predicts adverse birth outcomes among HIV-infected rural Ugandan women receiving combination antiretroviral therapy.
Maternal Nutritional Status Predicts Adverse Birth Outcomes among HIV-Infected Rural Ugandan Women Receiving Combination Antiretroviral Therapy

Sera Young1, Katherine Murray2, Julia Mwesigwa3, Paul Natureeba3, Beth Osterbauer4, Jane Achan5, Emmanuel Arinaitwe3, Tamara Clark4, Veronica Ades6, Albert Plently2, Edwin Charlebois2, Theodore Ruel7, Moses Kamya8, Diane Havlir4, Deborah Cohan6*

1 Division of Nutritional Sciences, Cornell University, Ithaca, New York, United States of America, 2 Center for AIDS Prevention Studies, University of California San Francisco, San Francisco, California, United States of America, 3 Makerere University-University of California San Francisco Research Collaboration, Kampala, Uganda, 4 Department of Medicine, University of California San Francisco, San Francisco, California, United States of America, 5 Department of Paediatrics and Child Health, Makerere University College of Health Sciences, Kampala, Uganda, 6 Department of Obstetrics, Gynecology and Reproductive Sciences, University of California San Francisco, San Francisco, California, United States of America, 7 Department of Pediatrics, University of California San Francisco, San Francisco, California, United States of America, 8 Department of Medicine, Makerere University Medical School, Kampala, Uganda

Abstract

Objective: Maternal nutritional status is an important predictor of birth outcomes, yet little is known about the nutritional status of HIV-infected pregnant women treated with combination antiretroviral therapy (cART). We therefore examined the relationship between maternal BMI at study enrollment, gestational weight gain (GWG), and hemoglobin concentration (Hb) among 166 women initiating cART in rural Uganda.

Design: Prospective cohort.

Methods: HIV-infected, ART-naïve pregnant women were enrolled between 12 and 28 weeks gestation and treated with a protease inhibitor or non-nucleoside reverse transcriptase inhibitor-based combination regimen. Nutritional status was assessed monthly. Neonatal anthropometry was examined at birth. Outcomes were evaluated using multivariate analysis.

Results: Mean GWG was 0.17 kg/week, 14.6% of women experienced weight loss during pregnancy, and 44.9% were anemic. Adverse fetal outcomes included low birth weight (LBW) (19.6%), preterm delivery (17.7%), fetal death (3.9%), stunting (21.1%), small-for-gestational age (15.1%), and head-sparing growth restriction (26%). No infants were HIV-infected. Gaining <0.1 kg/week was associated with LBW, preterm delivery, and a composite adverse obstetric/fetal outcome. Maternal weight at 7 months gestation predicted LBW. For each g/dL higher mean Hb, the odds of small-for-gestational age decreased by 52%.

Conclusions: In our cohort of HIV-infected women initiating cART during pregnancy, grossly inadequate GWG was common. Infants whose mothers gained <0.1 kg/week were at increased risk for LBW, preterm delivery, and composite adverse birth outcomes. cART by itself may not be sufficient for decreasing the burden of adverse birth outcomes among HIV-infected women.

Trial Registration: Clinicaltrials.gov NCT00993031

Citation: Young S, Murray K, Mwesigwa J, Natureeba P, Osterbauer B, et al. (2012) Maternal Nutritional Status Predicts Adverse Birth Outcomes among HIV-Infected Rural Ugandan Women Receiving Combination Antiretroviral Therapy. PLoS ONE 7(8): e41934. doi:10.1371/journal.pone.0041934

Editor: Claire Thorne, UCL Institute of Child Health, University College London, United Kingdom

Received February 2, 2012; Accepted June 29, 2012; Published August 7, 2012

Copyright: © 2012 Young et al. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

Funding: The primary sponsor of the PROMOTE-Pregnant Women and Infants trial is the National Institute for Child Health and Human Development (http://www.nichd.nih.gov/). This nutritional sub-study was supported by PEPFAR (President’s Emergency Plan for AIDS Relief), the Office of the Global AIDS Coordinator, and the Office of AIDS Research. Abbott Laboratories provides Lopinavir/ritonavir for the parent trial. The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Competing Interests: The authors have the following interests to declare: Abbott Laboratories provides Lopinavir/ritonavir for the parent trial. There are no patents, products in development or marketed products to declare. This does not alter the authors’ adherence to all the PLoS ONE policies on sharing data and materials, as detailed online in the guide for authors.

* E-mail: cohand@obgyn.ucsf.edu

Introduction

As the availability of combination antiretroviral therapy (cART) for HIV-infected pregnant women broadens and perinatal HIV transmission is reduced, there are increasing numbers of HIV-exposed, uninfected children worldwide [1]. Studies to date suggest that these children have worse outcomes compared to their HIV-unexposed counterparts [2–6]. Preterm delivery (PTD), low birth weight (LBW), stunting, and other markers of fetal growth...
restriction are important predictors of neonatal mortality, post-neonatal infant mortality, and infant and child morbidity [7–16]. Among the many factors that predict poor gestational outcomes, maternal nutritional status before and during pregnancy has emerged as a major modifiable determinant [15,17–21]. Specifically, pre-pregnancy body mass index (BMI) and gestational weight gain (GWG) have repeatedly been associated with LBW [22–28]. Little is known, however, about the relationship between maternal nutritional status and birth outcomes among HIV-infected women on cART, particularly in the resource-constrained settings of rural sub-Saharan Africa.

Given that nutritional status is a strong modifiable predictor of birth outcomes, we sought to characterize the baseline nutritional status of pregnant women initiating cART in rural Uganda and examine the associations between their nutritional status and adverse birth outcomes.

Methods

Study Design and Population

We analyzed nutritional data from an ongoing prospective clinical trial in Tororo, Uganda, evaluating malaria outcomes among women randomized to receive an HIV protease inhibitor or non-nucleoside reverse transcriptase inhibitor based cART regimen (NCT009993031, http://clinicaltrials.gov). The protocol for this trial and supporting CONSORT checklist are available as supporting information; see Checklist S1 and Protocol S1. Women with HIV-1 infection and a documented pregnancy between 12 and 28 weeks of gestation were enrolled (Figure S1). Women were excluded if they had ever used cART, had received single-dose nevirapine within 2 years, had prior dose-limited toxicity to trimethoprim-sulfamethoxazole (TS) within 2 weeks, received any contraindicated medications, had any WHO stage 4 diseases, had cardiac abnormalities, or if they had abnormal laboratory values at screening, including hemoglobin (Hb) <7.5 g/dL. All women gave written informed consent. The study protocol was approved by the Faculty of Medicine’s Research and Ethics Committee at Makerere University, the Uganda National Council of Science and Technology, and the Committee on Human Research at the University of California San Francisco.

Study Procedures

Trained study staff collected baseline demographic data and general medical, HIV, and obstetric history. Close birth spacing was defined as ≤2 years between births (either live born or stillbirth) based on self-reported obstetric history. Socioeconomic status (SES) was assessed by performing principle component analysis of a series of questions about possession of a radio, telephone, television, motorcycle, or bicycle. We used the first two components of the principle component analysis which accounted for over 55% of the information contained in the 6 asset holding variables.

Maternal height was measured to the nearest 0.1 cm using a Seca 206 wall-mounted measuring tape and maternal weight was measured to the nearest 500 g using a Seca 876 mechanical scale. Gestational age was estimated based on last menstrual period (LMP) and fetal ultrasound at the screening visit. All women received a fetal ultrasound. Final pregnancy dating was based on ultrasound if the discrepancy between LMP and ultrasound was greater than 1 week in the 1st trimester, 2 weeks in the 2nd trimester or 3 weeks in the 3rd trimester [29]. Ultrasound was used to date 50.5% of second trimester pregnancies (n = 105) and 43.4% of third trimester pregnancies (n = 53), for a total of 48.1% of the 158 pregnancies in these analyses. HIV status was documented with a positive rapid HIV antibody test (Determine, Inverness Medical Japan Co., Japan) plus a confirmatory test (Stat-Pak, Chembio Diagnostic Systems, Inc., NY, USA). All women received multivitamins containing iron and folic acid, iron supplements, prophylactic mebendazole, an insecticide-treated bed net, and were started on either zidovudine/lamivudine/efavirenz or zidovudine/lamivudine/lopinavir/ritonavir. Women were also started on daily TS if they were not already receiving TS prophylaxis prior to study enrollment.

Women returned to the study clinic every four weeks until delivery for scheduled study visits as well as when they experienced adverse events or any health conditions requiring evaluation. At scheduled monthly visits, maternal weight was measured as described above. Laboratory evaluations were regularly conducted throughout pregnancy including Hb, HIV RNA PCR, and CD4/CD8 lymphocyte subsets. Clinical progression of HIV disease was categorized according to 2007 WHO criteria [30]. Adverse events were classified according to the standardized Toxicity Table for Grading Severity of Adult and Pediatric Adverse Events [31]. Clinical malaria was defined using standardized WHO criteria as the presence of fever within the past 24 hours and a positive thick blood smear. At each scheduled visit, women were given a 5-week supply of multivitamins, antiretroviral therapy, and TS. Data were collected on socioeconomic status (SES) during a scheduled visit after enrollment.

If delivery took place in the hospital, trained study staff assessed infant anthropometry immediately after birth. Infant weight was measured to the nearest 10 g using a calibrated digital Seca 354 scale. Birth length was obtained using a locally made infant length board. Head circumference was measured to the nearest 0.1 cm using non-stretchable tape (Seca 212). If delivery occurred outside the study-affiliated hospital and infants were brought to the clinic, the anthropometric assessments were performed by study staff as described above. Only those infants who were measured within 12 hours of birth were included in this analysis. Infant HIV status at birth was determined by HIV-1 DNA PCR (Cobas Amplicor, Roche Diagnostics).

Nutritional Measures

The following markers of maternal nutritional status were assessed: body mass index (BMI) at enrollment, baseline Hb, mean Hb during pregnancy, maternal weight at 24–28 weeks gestation [18], and maternal weight change during pregnancy. Because there are no BMI standards for pregnant women, only non-pregnant adults [32], we categorized BMI at enrollment based on tertiles (<20.43, 20.43–22.59, and >22.59). Hb concentrations were categorized into 3 groups: severe anemia (≤8.5 g/dL), mild-to-moderate anemia (8.5–10.9 g/dL), and no anemia (≥11 g/dL). Because weekly maternal weight gain should be linear in the second and third trimesters [33], weekly maternal weight change was calculated by dividing the weight change between enrollment and last weight before delivery by the number of weeks elapsed between the two. For the categorical GWG variable, weight loss was defined as a rate of weight change ≤0 kg/swk. Those who had a positive average weekly weight gain were categorized as ≤<25th percentile of gainers (<0.1 kg per week) or ≥25th percentile (≥0.1 kg per week). Maternal weight at 7 months gestation was defined as the woman’s weight in kg between 24 and 28 weeks of gestation.

Outcomes

Infant outcomes included LBW, small for gestational age (SGA), stunting, wasting, underweight, preterm delivery, head-sparing growth restriction, and fetal death. LBW was defined as <2500
grams. SGA newborns were those with birth weight <10th percentile for their gestational age [34,35]. Stunting, wasting, and underweight were defined as standardized, sex-specific Z-scores of ≤−2 using length-for-age, weight-for-length and weight-for-age respectively. Z-scores were created using the WHO 2007 SAS Macro package [36]. Head-sparing growth restriction was defined as the presence of LBW, SGA, stunting, wasting, or underweight with a head circumference standardized Z-score of −1 or better [34]. Preterm delivery was defined as birth at less than 37 weeks gestation. Fetal death was defined as either miscarriage (12-20 weeks of gestation) or stillbirth (intrauterine fetal demise >20 weeks of gestation). Lastly, we created a composite dichotomous adverse birth outcome variable including LBW, SGA, stunting, wasting, underweight, preterm delivery, and fetal death.

Statistical Analyses
We restricted the analysis to singleton pregnancies because of the well-established relationship between multiple births and LBW and preterm delivery [37]. In addition, analyses of weight change during pregnancy were restricted to those women whose weights had been measured at least twice before delivery, with their last weight evaluated within two weeks prior to date of delivery.

We calculated descriptive statistics and created scatterplots to assess distribution of the data and to inform regression modeling. Chi-square and Fisher’s exact tests were performed, as appropriate, to test for associations between categorical variables. Wilcoxon signed-rank tests were performed to compare the means of continuous characteristics by categorical variables. For all logistic and linear regression models, univariate analyses were first performed to assess relationships. Subsequently, multivariate logistic regression models were fit with dichotomous outcomes and clinically important predictors: birth spacing (<2 years) and CD4+ count at screening. Multivariate linear regression models were also built for continuous outcome variables. Birth spacing and CD4+ count were included in all models because of their clinical significance, as were any other predictors meeting the p≤0.2 threshold in univariate analysis. Multivariate model inputs were evaluated for significant multi-collinearity and where highly correlated, the variable with strongest association was retained. Model fit was assessed between model versions using differences in model fit was assessed between model versions using differences in the −2 log likelihood and the difference in degrees of freedom between models relative to the Chi-square distribution. All analyses were conducted using SAS version 9.2 (Cary, North Carolina).

Results

Study Population
There were 232 women enrolled in the randomized clinical trial between December 15, 2009 and May 24, 2011. Of these, 166 delivered by May 24, 2011. Of these, 158 (95.2%) were singleton birth and were included in the analysis. Of the 158 births, 13.9% occurred outside the hospital. Birthweight was obtained within 12 hours of birth for 153 (96.8%) of the 158 women with singleton births.

The median age of participants at baseline was 29 years, and during pregnancy were restricted to those women whose weights had been measured at least twice before delivery, with their last weight evaluated within two weeks prior to date of delivery.

We calculated descriptive statistics and created scatterplots to assess distribution of the data and to inform regression modeling. Chi-square and Fisher’s exact tests were performed, as appropriate, to test for associations between categorical variables. Wilcoxon signed-rank tests were performed to compare the means of continuous characteristics by categorical variables. For all logistic and linear regression models, univariate analyses were first performed to assess relationships. Subsequently, multivariate logistic regression models were fit with dichotomous outcomes and clinically important predictors: birth spacing (<2 years) and CD4+ count at screening. Multivariate linear regression models were also built for continuous outcome variables. Birth spacing and CD4+ count were included in all models because of their clinical significance, as were any other predictors meeting the p≤0.2 threshold in univariate analysis. Multivariate model inputs were evaluated for significant multi-collinearity and where highly correlated, the variable with strongest association was retained. Model fit was assessed between model versions using differences in the −2 log likelihood and the difference in degrees of freedom between models relative to the Chi-square distribution. All analyses were conducted using SAS version 9.2 (Cary, North Carolina).

Maternal Weight Gain During Pregnancy
The median weekly GWG during study participation was 0.17 kg (interquartile range [IQR]: 0.06, 0.3). Data on median weekly GWG were unavailable for two participants who were not weighed within two weeks of delivery. Twenty-three women (14.6%) lost weight during pregnancy. The median total GWG during study participation was 3 kg (IQR: 1, 5). In addition, the median maternal weight at 7 months gestation was 58 kg (IQR: 52, 63) and the median BMI at 7 months gestation was 22 (IQR: 20.5, 24.1). Adjusting for birth spacing, baseline CD4 count, baseline weight, mean Hb during pregnancy, and Grade 3 or 4 adverse events, there were no significant predictors of gestational weight gain. (Univariate relationships and the full linear regression model can be found in Table S1).
Obstetric and Neonatal Outcomes

There were no infants infected with HIV at birth. The prevalence of LBW was 19.6%, preterm delivery 17.7%, and fetal death 3.9% (Table 2). Stunting was the most common anthropometric marker of growth restriction seen among live-born newborns (21.1%), followed by SGA (15.1%) and underweight (15.1%). Twenty six percent of live-born infants were diagnosed with head-sparing growth restriction. Seventy three percent of participants experienced a full-term delivery of a live-born infant with normal birth weight.

Risk Factors for Adverse Outcomes

**Low birth weight.** In multivariate analysis adjusting for birth spacing (<2 years) and baseline CD4 count, each kg increase in maternal weight at enrollment was associated with a 30% decreased odds of LBW (aOR 0.70, 95% CI 0.52–0.95, p = 0.022, cf. Table S2). Each cm increase in maternal height was associated with an 8% lower odds of LBW (aOR 0.92, 95% CI 0.85–1.00, p = 0.046). Moreover, women who gained <0.1 kg per week had greater than 6-fold increase in odds of LBW compared to women who gained ≥0.1 kg per week (aOR 6.18, 95% CI 1.80–21.1, p = 0.004). Lastly, each kg decrease in total maternal weight at 7 months gestation was associated with a 38% increased odds of LBW (aOR 1.38, 95% CI 1.03–1.90, p = 0.034).

**Small-for-gestational age.** Adjusting for birth spacing (<2 years), baseline CD4 count, and any GWG, each g/dL increase in mean maternal Hb between enrollment and final measurement was associated with a 52% decreased odds of SGA (aOR 0.48, 95% CI 0.29–0.80, p = 0.004, cf. Table S3).

**Stunting.** After adjusting for birth spacing (<2 years) and baseline CD4 count, risk factors for neonatal stunting included male infant sex (aOR 6.02, 95% CI 1.64–22.06, p = 0.007), gestational age at delivery (aOR 0.51, 95% CI 0.37–0.71, p<0.001) and clinical malaria diagnosed during pregnancy (aOR 0.18, 95% CI 0.03–0.97, p = 0.047, cf. Table S4).

**Head-sparing fetal growth restriction.** There were no statistically significant predictors of head-sparing growth restriction in multivariate analysis adjusting for birth spacing, baseline CD4 count and mean maternal Hb during the study, cf. Table S5.

**Preterm delivery.** Adjusting for birth spacing; baseline CD4 count and history of preterm delivery, GWG <0.1 kg per week was associated with a 4-fold increased odds of preterm delivery (aOR 3.46, 95% CI 1.18–10.15, p = 0.024, cf. Table S6).

**Composite adverse birth outcome.** Lastly, predictors of the composite dichotomous adverse birth outcome included GWG <0.1 kg per week, higher SES status, and birth from June to October (the rainy season), adjusting for CD4 count and birth spacing (Table 3). In particular, gaining <0.1 kg per week was associated with a nearly 3 fold increased odds of an adverse birth outcome (aOR 2.85, 95% CI 1.32–6.15, p<0.01).

**Discussion**

Combination antiretroviral therapy is being delivered to increasing numbers of rural HIV-infected pregnant women in developing countries [38–40]. Goals of treatment include protecting the health of these women and promoting the birth of HIV-uninfected, healthy infants. However, cART alone may not be sufficient to achieve these goals, particularly when these women also often face other challenges besides HIV, including food insecurity and malnutrition [41].

In our cohort of HIV-infected pregnant women in rural Uganda initiating cART, TS and prenatal care, we found evidence of significant nutritional deficiencies. These women had low BMIs upon study entry and well into their pregnancy, despite having relatively preserved CD4 cell counts. Their mean weight gain of 0.17 kg/week was far below the 0.5 kg/week recommended by the Institute of Medicine for underweight women in industrialized countries in the second and third trimester [42]. Although all infants were HIV-uninfected at delivery, adverse birth outcomes were highly prevalent and likely attributable at least in part to poor maternal nutritional status.

Interestingly, HIV severity, measured as baseline CD4 count, viral load, or WHO stage, was not predictive of adverse birth outcomes. This could have been due to our sample size and the relatively small proportion of women with severe immune suppression. Indeed, 50% of our cohort had baseline CD4 counts above 350 and the majority of the women (91.8%) were WHO stage 1.

Maternal nutritional predictors of preterm delivery and growth restriction have primarily been evaluated among HIV-infected women not receiving cART. The Pregnancy and HIV Study Group of 177 ARV-naive women in Rwanda found that each kg increment in final weight before delivery was associated with a 6% decreased odds of LBW [43]. Villamor et al. found that low maternal weight at first prenatal visit was associated with lower mean birth weight and SGA but not preterm delivery among 1002 ARV-naive women in Tanzania [44]. Similar to our study, the prevalence of gestational weight loss was 10%, and weight loss was associated with LBW, preterm delivery and fetal death. In Zambia, Bandu et al. found infant birth weight increased by 28.3 g for every unit increase in BMI at 36 weeks of gestation; they did not assess the risk of preterm delivery or other markers of growth restriction [41]. Finally, Mehta et al. analyzed outcomes of 2294 ARV-naive pregnant women enrolled in HIVNET 024 [27]. They found enrollment maternal BMI in the lowest tertile to be associated with LBW and preterm delivery. As observed in our.

---

**Table 2. Obstetric and Fetal Outcomes (n = 158)**

| Male sex (n = 155) | 83 (53.6%) |
|-------------------|-----------|
| Overall low birth weight (n = 153) | 30 (19.6%) |
| Overall preterm delivery | 28 (17.7%) |
| Full term, normal birth weight (n = 152) | 111 (73.0%) |
| Full term, low birth weight (n = 152) | 13 (8.6%) |
| Preterm, normal birth weight (n = 152) | 11 (6.6%) |
| Preterm, low birth weight (n = 152) | 12 (7.9%) |
| Miscarriage (12–20 weeks) (n = 152) | 1 (0.6%) |
| Stillbirth (>20 weeks) (n = 152) | 5 (3.3%) |
| Composite adverse obstetric/fetal outcome | 64 (40.5%) |
| Live born infants only (n = 152) | 132 (86.5%) |
| Stunting (LAZ*≤ −2) at birth (n = 142) | 30 (21.1%) |
| Small for gestational age (<10th percentile) | 23 (15.1%) |
| Underweight (WAZ*≤ −2) at birth | 23 (15.1%) |
| Wasting (WLZ*≤ −2) at birth (n = 128) | 11 (8.6%) |
| Small head circumference (HCZ*≤ −2) at birth (n = 150) | 10 (6.7%) |
| Head-sparing growth restriction (n = 150) | 39 (26.0%) |

*All data are represented as n (%). LAZ: length-for-age Z-score. WAZ: weight-for-age Z-score. WLZ: weight-for-length Z-score. HCZ: head circumference Z-score. doi:10.1371/journal.pone.0041934.t002
study, weight gain <0.1 kg per week was associated with increased risk of LBW.

Very few studies have evaluated nutritional predictors of pregnancy outcomes among HIV-infected women on cART. Ekouevi and colleagues studied 151 pregnant women receiving cART as part of the ANRS Ditrame Plus and the MTCT-Plus Projects in Cote d’Ivoire [45]. Similar to the Pregnancy and HIV Study Group in Rwanda, these researchers found maternal BMI at delivery to be predictive of LBW. In particular, the odds of LBW was 2.43 fold higher among women with a delivery BMI <25. Conversely, Powis et al recently reported that change in BMI one month following the initiation of cART in pregnancy was not significantly associated with preterm delivery among 530 HIV-infected pregnant women in Botswana [46].

It is reasonable to postulate that pregnant women treated with cART would have improved nutritional status compared to those without access to cART. Women receiving effective cART should experience less HIV morbidity, including diarrhea and wasting, which should outweigh the toxicity of the antiretroviral agents. However, our study and the two others examining nutritional markers among pregnant women receiving cART can neither support nor refute this assumption because all women received cART, and it would be unethical to randomize to non-cART treatment regimens. Furthermore, even if cART does improve nutritional status, we demonstrate in this cohort that there remain significant nutritional deficiencies and that these are associated with poor birth outcomes.

Mechanisms to explain these poor outcomes are likely numerous and not yet fully understood. For example, head-sparing, or asymmetric, growth restriction is thought to be due to preferential blood flow to the brain in the setting of placental insufficiency [47]. Indeed, the inverse association between head-sparing growth restriction and weekly GWG suggests a nutritional basis for this placental insufficiency.

Much work is needed to determine factors that contribute to low GWG and weight loss among HIV-infected pregnant women, including the impact of initiating HAART during pregnancy versus use of HAART prior to conception. It is also necessary to develop strategies to identify those women at greatest risk for poor birth outcomes. Pre-pregnancy BMI has consistently been associated with adverse birth outcomes. However, because most women do not know their pre-pregnancy weight and do not have regular access to preconception care, this indicator is not clinically useful. In order to identify another relevant maternal anthropometric predictor of adverse fetal outcomes, Kelly and colleagues conducted a meta-analysis of 25 studies including over 111,000 births worldwide [18]. They found that low maternal weight attained at 7 months gestation was a significant risk factor for fetal growth restriction, particularly among women with below average pre-pregnancy weight. Indeed in our study, those women with low weight at 7 months gestation were at particularly high risk of LBW. Because increased GWG in the 3rd trimester was associated with a diminished odds of LBW, preterm delivery, and overall adverse birth outcome, these women with low weight at 7 months may benefit from a nutritional intervention.

Preterm delivery, LBW, neonatal stunting, SGA, and wasting are strong predictors of infants’ future health trajectories [48]. With the increased availability of cART during pregnancy and breastfeeding, there is an expanding generation of HIV-exposed, uninfected children. Nutritional interventions that increase maternal weight gain during pregnancy have the potential to decrease the burden of a range of adverse birth outcomes among women infected with HIV. As such, the improvement of maternal nutritional status may be a golden opportunity to not only protect the health of the mother, but to improve birth outcomes and create a thriving generation of HIV-exposed, uninfected offspring.

There are several limitations to our study. Small sample size may have impaired our ability to find statistically significant predictors of head-sparing fetal growth restriction and other poor birth outcomes. Our findings may not be generalizable to other cohorts of HIV-infected pregnant women receiving cART (e.g. [38,39,40]) because our participants were older, nearly all were multigravidae.

Table 3. Predictors of Composite Adverse Obstetric/Fetal Outcome1, Univariate and Multivariate Analysis.

| Outcome (%) | OR  | P   | aOR2 | 95% CI | P   |
|-------------|-----|-----|------|--------|-----|
| Birth spacing <2 years | 77 (44.4%) | 1.19 | 0.80 | 0.89 | 0.20–3.92 | 0.88 |
| Baseline CD4 count (continuous) | 64 (40.8%) | 1 | 0.95 | 1 | 0.99–1.00 | 0.84 |
| Maternal weight at 7 months gestation | 62 (40%) | 0.97 | 0.15 | 0.95 | 0.91–1.00 | 0.07 |
| Weekly weight gain | | | | | | |
| <0.1 kg/week | 28 (52%) | 2.12 | 0.03 | 2.85 | 1.32–6.15 | <0.01 |
| 0.1 kg/week or greater | 34 (33.7%) | – | 1 |
| Higher SES* | 30 (47%) | 1.67 | 0.13 | 2.61 | 1.20–5.67 | 0.02 |
| Season of birth | | | | | | |
| June to October | 16 (28.6%) | 0.45 | 0.03 | 0.33 | 0.15–0.74 | <0.01 |
| November to May | 48 (47.1%) | 1 | – | 1 |

1 Composite adverse obstetric/fetal outcome includes any of the following: low birth weight, SGA, stunting, wasting, underweight, preterm delivery, and fetal death.
2 Adjusted odds ratio using multivariate logistic regression, adjusting for all variables listed in table.
3 Per kg increment in maternal weight at 7 months gestation.
4 Indicator variable for household being in the upper quartile of either the first or second component of the SES principal component analysis based on possession of a radio, telephone, television, motorcycle, bicycle or none of the above.

95% CI: Confidence Interval.
P: p-value.
aOR: Adjusted Odds Ratio.

doi:10.1371/journal.pone.0041934.t003
and mean BMI was lower [38–40]. Further, our results may not be generalizable to those women on cART prior to conceiving. We excluded multiple births because of its known effect on adverse birth outcomes, which may further limit the generalizability of our findings. Our finding that higher SES was associated with an increased odds of an adverse birth outcome may be spurious because the SES measure was generated using principal component analysis of specific asset holding questions and not a validated poverty scale. Such a measurement could have been vulnerable to unmeasured confounding. Finally, we analyzed data from an ongoing randomized trial and differences by study treatment arm cannot be addressed until study completion and data unblinding.

In conclusion, initiating cART during pregnancy among HIV-infected women in rural Uganda successfully prevented HIV transmission to their infants but did not prevent poor nutritional status during pregnancy that independently predicted poor birth outcomes. More attention is needed to characterize the scope and causes of nutritional deficiencies among this population and to design interventions that improve both the health of HIV-infected mothers and optimize the health and development of their offspring.

Supporting Information
Figure S1 CONSORT 2010 flow diagram for NCT00993031.
(DOC)
Table S1 Univariate and multivariate logistic regression models of weekly gestational weight gain.
(DOC)
Table S2 Univariate and multivariate logistic regression models of low birthweight.
(DOC)

References
1. Filteau S (2009) The HIV-exposed, uninfected African child. Trop Med Int Health 14: 278–287.
2. Marinda E, Humphrey JH, Iliff PF, Mutasa K, Nathoo KJ, et al. (2007) Child mortality according to maternal and infant HIV status in Zimbabwe. Pediatr Infect Dis J 26: 519–526.
3. Brahmbhatt H, Kajogi Z, Walwire-Mangen F, Serwadda D, Lutalo T, et al. (2006) Mortality in HIV-infected and uninfected children of HIV-infected and uninfected mothers in rural Uganda. J Acquir Immune Defic Syndr 41: 504–508.
4. Makasa M, Kasonka L, Chisenga M, Sinkala M, Chintu C, et al. (2007) Early growth of infants of HIV-infected and uninfected Zambian women. Trop Med Int Health 12: 594–602.
5. Shapiro RL, Lockman S, Kim S, Smeaton L, Rahkola JT, et al. (2007) Infant morbidity, mortality, and breast milk immunologic profiles among breastfeeding HIV-infected and HIV-uninfected women in Botswana. J Infect Dis 196: 562–569.
6. Zaha B, Whitworth J, Marston M, Nakayiingi J, Ruberantwari A, et al. (2005) HIV and mortality of mothers and children: evidence from cohort studies in Uganda, Tanzania, and Malawi. Epidemiology 16: 275–280.
7. Wei R, Msamanga GI, Spiegelman D, Hertzmark E, Baylin A, et al. (2004) Association between low birth weight and infant mortality in children born to human immunodeficiency virus 1-infected mothers in Tanzania. Pediatr Infect Dis J 23: 530–535.
8. Iscovoldou N, Varsami M, Syggelous A (2010) Neonatal outcome of preterm delivery. Ann N Y Acad Sci 1205: 130–134.
9. Nannan N, Norman R, Hendricks M, Dhanasy MA, Bradhaw D (2007) Estimating the burden of disease attributable to childhood and maternal undernutrition in South Africa in 2000. S Afr Med J 97: 733–739.
10. Kramer MS, Barros FC, Demissie K, Liu S, Kucz K, et al. (2005) Does reducing infant mortality depend on preventing low birthweight? An analysis of temporal trends in the Americas. Paediatr Perinat Epidemiol 19: 445–451.
11. Pattinson RC (2003) Why babies die—a perinatal care survey of South Africa, 2000–2002. S Afr Med J 93: 445–450.
12. van der Mei J, Volmer M, Boersma ER (2000) Growth and survival of low birthweight infants from 0 to 9 years in a rural area of Ghana. Comparison of moderately low (1,501–2,000 g) and very low birthweight (1,000–1,500 g) infants and a local reference population. Trop Med Int Health 5: 371–377.
13. Manji KP, Massawe AW, Mgone JM (1998) Birthweight and neonatal outcome at the Muhimbili Medical Centre, Dar es Salaam, Tanzania. East Afr Med J 75: 382–387.
14. McCormick MC (1985) The contribution of low birth weight to infant mortality and childhood morbidity. N Engl J Med 312: 82–90.
15. Kramer MS (1987) Determinants of low birth weight: methodological assessment and meta-analysis. Bull World Health Organ 65: 663–737.
16. Ruel MT (2003) The natural history of growth failure: Importance of intrauterine and postnatal periods. In: Martorell R, Haschke F, eds. Nutrition and growth. Nestlé Nutrition Workshop Series, Pediatric Program. Philadelphia: Lippincott Williams & Wilkins. 123–158.
17. Eldabby EM, Shamalsich G (2008) The effect of maternal anthropometric characteristics and social factors on gestational age and birth weight in Sudanese newborn infants. BMC Public Health 8: 244.
18. Kelly A, Kevany J, de Onis M, Shah PM (1996) A WHO Collaborative Study of Maternal Anthropometry and Pregnancy Outcomes. Int J Gynaecol Obstet 53: 219–233.
19. Kramer MS (1987) Intrauterine growth and gestational duration determinants. Pediatrics 80: 502–511.
20. Viswanathan M, Siegel-Sez AM, Moos MK, Deierlein A, Muzamrud S, et al. (2008) Outcomes of maternal weight gain. Evit Rep Technol Assess (Full Rep): 1–223.
21. Cesay SM, Prentice AM, Coyle TJ, Ford F, Weaver LT, et al. (1997) Effects on birth weight and perinatal mortality of maternal dietary supplements in rural Gambia: 5 year randomised control trial. BMJ 313: 786–790.
22. Kruger HS (2005) Maternal anthropometry and pregnancy outcomes: a proposal for the monitoring of pregnancy weight gain in outpatient clinics in South Africa. Curationis 28: 40–49.
23. Villamor E, Msamanga G, Spiegelman D, Peterson KE, Atteman G, et al. (2003) Pattern and predictors of weight gain during pregnancy among HIV-1-infected women from Tanzania. J Acquir Immune Defic Syndr 32: 560–569.
24. Abrams B, Newman V (1991) Small-for-gestational-age birth: maternal predictors and comparison with risk factors of spontaneous preterm delivery in the same cohort. Am J Obstet Gynecol 161: 785–790.

Table S3 Univariate and multivariate logistic regression models of small for gestational age.
(DOC)
Table S4 Univariate and multivariate logistic regression models of stunting.
(DOC)
Table S5 Univariate and multivariate logistic regression models of head-sparing growth restriction.
(DOC)
Table S6 Univariate and multivariate logistic regression models of preterm delivery.
(DOC)
Protocol S1 Trial protocol.
(DOC)

Checklist S1 CONSORT checklist.

Acknowledgments
The authors would like to sincerely thank the women who have participated in the PROMOTE-Pregnant Women and Infants trial. We would also like to acknowledge the dedicated PROMOTE study team, members of the Makerere University-University of California San Francisco Research Collaboration, and the midwives at Tororo District Hospital.

Author Contributions
Conceived and designed the experiments: SY EC TR DH DC. Wrote the manuscript: SY VA JA EC TR MK DH DC.

Downloaded from www.plosone.org
25. Adam I, Babiker S, Mohamed AA, Salih MM, Prins MH, et al. (2008) Low body mass index, anaemia and poor perinatal outcome in a rural hospital in eastern Sudan. J Trop Pediatr 54: 202–204.
26. Frederick IO, Williams MA, Sales AE, Martin DP, Killien M (2008) Pre-pregnancy body mass index, gestational weight gain, and other maternal characteristics in relation to infant birth weight. Matern Child Health J 12: 557–567.
27. Mehta S, Manji KP, Young AM, Brown ER, Chasela C, et al. (2008) Nutritional indicators of adverse pregnancy outcomes and mother-to-child transmission of HIV among HIV-infected women. Am J Clin Nutr 87: 1639–1649.
28. Ehrenberg HM, Dierker L, Milluzzi C, Mercer BM (2003) Low maternal weight, failure to thrive in pregnancy, and adverse pregnancy outcomes. Am J Obstet Gynecol 189: 1726–1730.
29. (2009) ACOG Practice Bulletin No. 101: Ultrasonography in pregnancy. Obstet Gynecol 113: 451–461.
30. (2007) WHO Case Definitions of HIV for Surveillance and Revised Clinical Staging and Immunological Classification of HIV-Related Disease In Adults and Children. Division of AIDS (2009) Toxicity table for grading the severity of adult and pediatric adverse events version 1.0 - December 2004 (Clarification dated August 2009). http://rsc.tech-res.com/Document/safetyandpharmacovigilance/Table_for_Grading_Severity_of_Adult_Pediatric_Adverse_Events.pdf. Accessed December 21, 2011.
31. World Health Organization (2006, updated 2011) Global Database on Body Mass Index. http://apps.who.int/bmi/index.jsp?introPage=intro_3.html. Accessed December 21, 2011.
32. Rasmussen KM, Abrams B, Bodnar LM, Butte NF, Catalano PM, et al. (2010) Recommendations for weight gain during pregnancy in the context of the obesity epidemic. Obstet Gynecol 116: 1191–1195.
33. de Onis M, Onyango AW, Borghi E, Siyam A, Chou A, et al. (2007) Development of a WHO growth reference for school-aged children and adolescents. Bull World Health Organ 85: 660–667.
34. Martin JA, Hamilton BE, Sutton PD, Ventura SJ, Mathews TJ, et al. (2010) Births: final data for 2007. Natl Vital Stat Rep 58: 1–85.
35. Marazzi CM, Germano P, Liotta G, Guidotti G, Loureiro S, et al. (2007) Implementing anti-retroviral triple therapy to prevent HIV mother-to-child transmission: a public health approach in resource-limited settings. Eur J Pediatr 166: 1305–1307.
36. de Vincenzi I (2011) Triple antiretroviral compared with zidovudine and single-dose nevirapine prophylaxis during pregnancy and breastfeeding for prevention of mother-to-child transmission of HIV-1 (Kesho Bora study): a randomised controlled trial. Lancet Infect Dis 11: 171–180.
37. Toro PL, Katyal M, Carter RJ, Myer L, Es-Sadr WM, et al. (2010) Initiation of antiretroviral therapy among pregnant women in resource-limited countries: CD4+ cell count response and program retention. AIDS 24: 515–524.
38. Banda Y, Chapman V, Goldenberg RL, Chi BH, Vermund SH, et al. (2007) Influence of body mass index on pregnancy outcomes among HIV-infected and HIV-uninfected Zambian women. Trop Med Int Health 12: 856–861.
39. Rasmussen KM, Yakine AL, editors (2009) Weight Gain During Pregnancy: Reexamining the Guidelines. Washington, D.C.: National Academies Press.
40. Castetbon K, Ladner J, Leroy V, Chauvala M, Karita E, et al. (1999) Low birthweight in infants born to African HIV-infected women: relationship with maternal body weight during pregnancy. Pregnancy and HIV Study Group (EGE). J Trop Pediatr 45: 152–157.
41. Villamor E, Dreyfuss ML, Baylin A, Maamanga G, Fawzi WW (2004) Weight loss during pregnancy is associated with adverse pregnancy outcomes among HIV-1 infected women. J Nutr 134: 1424–1431.
42. Ekouevi DK, Coffie PA, Becquet R, Tonwe-Gold B, Horo A, et al. (2008) Antiretroviral therapy in pregnant women with advanced HIV disease and pregnancy outcomes in Abidjan, Cote d’Ivoire. AIDS 22: 1815–1820.
43. Powis K, Kitch D, Ogwu A, Hughes M, Lockman S, et al. (2011) Protease Inhibitor-based ART Was Associated with Pre-term Delivery, but Not Adverse Infant Outcomes, in a Randomized MTCT Prevention Study in Botswana. Abstract #746. 18th Conference on Retroviruses and Opportunistic Infections. Boston, MA.
44. al-Ghazali W, Chita SK, Chapman MG, Allan LD (1989) Evidence of redistribution of cardiac output in asymmetrical growth retardation. Br J Obstet Gynaecol 96: 697–704.
45. Markestad T, Vik T, Ahlsten G, Gehe-Reidhun M, Skjerven R, et al. (1997) Small-for-gestational-age (SGA) infants born at term: growth and development during the first year of life. Acta Obstet Gynecol Scand Suppl 165: 93–101.