

Maternal HIV infection is an important health determinant in non-HIV-infected infants

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Objective: To assess morbidity and mortality in HIV-exposed uninfected (HEU) children to help guiding appropriate clinical care and effective preventive interventions.

Design: This is a longitudinal study comparing two cohorts of children; one born to HIV-infected women and the other born to HIV-uninfected women.

Methods: We have analyzed prospectively obtained information on nutritional status, morbidity and mortality from 966 HEU and 909 HIV-unexposed infants followed up until their first 18 months of life at a referral health facility in southern Mozambique. Determinants for adverse health outcomes in HEU children were also assessed using multivariate logistic regression.

Results: Increased incidence of hospital admissions ($P=0.0015$), shorter survival in the first 18 months of life ($P=0.0510$) and moderate and severe malnutrition ($P=0.0006$ and 0.0014 , respectively) were observed among HEU children compared with HIV-unexposed children. Incidence of outpatient attendance in HEU children was associated with being men, older age and the mother being on antiretroviral treatment. Among HEU children, those who were never breastfed, or who were weaned or were partially breastfed, had an increased incidence of hospital admissions compared with children who were exclusively breastfed.

Conclusion: Maternal HIV infection has important health consequences in non-HIV-infected children. As the prevalence of HIV-infected pregnant women is maintained and the proportion of HIV-infected children declines because of the scale-up of antiretroviral treatment during pregnancy and breastfeeding, more focus should be given to the health needs of HEU children to ensure that the post-2015 sustainable development goals are met.

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Introduction

The scale-up of programs for the prevention of mother-to-child transmission of HIV (PMTCT) has increased

remarkably in the last decade, which resulted in a 50% decline of new HIV infections among children since 2010 globally [1]. The currently recommended approach for PMTCT (the so-called Option B+), consisting in the

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initiation of lifelong antiretroviral therapy (ART) for all HIV-infected pregnant and breastfeeding women, is expected to further reduce the incidence of new HIV infections and potentially eliminate mother-to-child transmission (MTCT) of HIV [2]. Although current programmatic efforts are focused in reducing new HIV pediatric infections, the number of children who are HIV-exposed but uninfected is increasing proportionately. Every year, over 1 million infants are born to HIV-infected women worldwide, the majority of them in low-income countries, and most of them will not become infected [3].

HIV-exposed uninfected (HEU) children are increasingly recognized as a group with specific health needs. Direct or indirect HIV exposure *in utero*, *intra partum* or during breastfeeding may confer risks even in the absence of infection. Several studies have described increased mortality and enhanced risk of poor health and growth outcomes in HEU compared with HIV-unexposed children [4–11]. However, the nature of the vulnerability of the HEU children is not entirely clear. In addition to some immunological abnormalities that have been observed in HEU children, exposure to antiretroviral drugs both *in utero* and the neonatal period, feeding practices as well as a particular family environment, might all contribute to poor health outcomes [12].

Mozambique is one of the countries with the highest HIV prevalence and incidence in the world [13]. In some areas, up to 40% of pregnant women attending the antenatal care (ANC) clinics are HIV-infected [14,15]. After more than 10 years since the rollout of antiretrovirals in the country, a 57% reduction in new HIV infections among children has been reported between 2009 and 2013 [16]. However, there are very limited data on health outcomes and survival of HEU children. A better understanding of the impact of maternal HIV infection on the health outcomes of exposed children is needed in low-income settings, where malnutrition and comorbidities are frequent and health monitoring resources are limited. This study compares the survival and health outcomes between two cohorts of HIV-exposed and HIV-unexposed children and identifies determinants for morbidity among the HEU children.

Methods

Study design

The current study compares prospectively obtained data on nutritional status, morbidity and mortality during the first 18 months of life, between two cohorts of children: one cohort born to HIV-infected women and the other born to HIV-uninfected women.

Study area and population

The study was conducted at the Manhiça District Hospital (MDH), located in a semirural area of the

Maputo Province in southern Mozambique. Since 1996, the Centro de Investigação em Saúde de Manhiça has conducted continuous demographic surveillance covering a population of approximately 95 000 inhabitants in an area of 500 km² [17]. Passive morbidity surveillance was established in parallel at the MDH and peripheral health posts in the area, whereby all pediatric hospital admissions and outpatient attendances are registered onto standardized questionnaires following standard procedures.

The average HIV prevalence figure in 2015 in Mozambique was 11% [18]. However, a community-based survey undertaken in the Manhiça area among adults in 2012 reported nearly 40% of HIV prevalence. The contemporaneous HIV seroprevalence among pregnant women attending the ANC clinics was 30% [15]. A prevalence of 9% of MTCT at first month of age and of 25% at the end of the first year of life has also been reported in the area [19,20]. Before 2013, PMTCT relied on antiretroviral prophylaxis, which consisted in antepartum monotherapy with zidovudine (ZDV), a single-dose nevirapine (NVP) at the onset of labor, and ZDV + lamivudine (3TC) at delivery and for 7 days *post partum*. Daily ZDV or NVP was recommended for infant prophylaxis during breastfeeding. National guidelines recommended ART to HIV-infected pregnant women for their own health if the CD4⁺ T-cell count was less than 350 cells/ μ l and/or they were in 3–4 HIV/AIDS WHO clinical stage. The recommended first-line regimen consisted in the dual nucleoside analog reverse-transcriptase Inhibitors backbone [3TC and either ZDV or stavudine (d4T)] and a nonnucleoside analog reverse-transcriptase Inhibitors [either NVP or efavirenz (EFV)] [21]. In 2013, Mozambique adopted ‘Option B+’, consisting in the initiation of lifelong ART with tenofovir (TDF) + 3TC + EFV in all HIV-positive pregnant and lactating women regardless of their immune status and clinical stage [22]. According to this recommendation, both breastfed and formula-fed infants should receive once-daily NVP from birth to 6 weeks of age. Cotrimoxazol prophylaxis (CTXp) is also recommended from 4 weeks until 2 months after weaning, whereas exclusively breastfeeding is recommended until 6 months of age.

Study procedures

HIV-unexposed children

A total of 909 children born between February 2010 and October 2012 to HIV-uninfected women participating in a trial comparing mefloquine with sulphadoxine-pyrimethamine as intermittent preventive treatment of malaria in pregnancy were included in this study. Results from the trial showed no differences in mortality, morbidity and nutritional status between infants born to women allocated to either study group [23,24]. As part of clinical trial procedures, at delivery, newborns were given a study number to be uniquely identified. Mothers

were asked to bring their child to study health facilities at 1 month of age, or coinciding with the first expanded program on immunization visit, and then at 9 and 12 months of age for study scheduled visits. Both at delivery and at study visits, weight and length were recorded onto standardized questionnaires.

HIV-exposed uninfected children

There were 966 children included in this study who were born to HIV-infected women, followed up at the High-Risk Pediatric Outpatient Clinic (HRPOC) of the MDH and born between January 2010 and December 2013. This study cohort included the children until they became HIV infected, died or ended the follow-up at 18 months of age.

As per national guidelines, children born to HIV-infected women were recommended to attend monthly visits at the HRPOC from the first month of life. In these visits, feeding habits and anthropometric and clinical characteristics were assessed. Informed consent was obtained, and data were collected prospectively onto standardized questionnaires in each of the subsequent visits to the HRPOC. Data from delivery, newborn baseline characteristics and uptake of PMTCT interventions during pregnancy were extracted retrospectively from the infant's Health Card. An HIV-DNA PCR test between 4 and 6 weeks of age was performed, with a confirmatory test on a new sample in those infants who test positive in the PCR. If the PCR test was negative, an HIV-antibody rapid test was performed at 9 months of age, and viral testing was carried out in those children with a positive serological test. The definitive diagnosis of HIV infection through HIV-antibody testing was determined at 18 months of age or when the risk of HIV exposure through breastfeeding ended. Infected infants were referred to the Pediatric HIV Outpatient Clinic for follow-up and ART initiation.

In both cohorts, information on hospital admissions and outpatient attendances during the first 18 months of life was obtained from the MDH passive morbidity surveillance system. Data on mortality were extracted from the demographic surveillance system in place.

Laboratory methods

HIV status was assessed from dried blood spot on filter paper using the Amplicor HIV-1 DNA-PCR kit (Roche Diagnostics, Branchburg, New Jersey, USA). Determine HIV-1/2 Rapid Test (Abbott Laboratories, Chicago, Illinois, USA) and Uni-Gold Rapid Test (Trinity Biotech, Dublin, County Wicklow, Ireland) were used for HIV serological testing on whole blood collected by finger prick. Hemoglobin was determined using mobile devices in capillary blood samples [HemoCue (www.eurotrol.com) and Hemocontrol (www.ekfdiagnostics.com)]. Thick and thin blood films were stained and read for

Plasmodium species detection according to standard, quality-controlled procedures [25].

Data management, statistical methods and definitions

Data were double-entered using the OpenClinica Enterprise software for clinical data management (www.openclinica.com).

Low birth weight (LBW) was defined as weight less than 2500 g at birth and preterm as a birth less than 37 weeks of gestation. Gestational age was assessed by the Ballard's score at delivery. Transformation of child anthropometric data to *z*-scores was performed using the least mean squares method and the reference data available from the 2000 Centers for Disease Control and Prevention growth reference in the United States [26,27]. Moderate malnutrition was defined as less than -2 SD for either weight-for-age, height-for-age or weight-for-height *z*-scores, and severe malnutrition as any of the previous *z*-scores under -3 SD [28]. Causes of hospital admission were classified according to the international classification of disease (ICD-10) coding system.

Minimum community-based incidence rates for the morbidity outcomes (outpatient attendances, hospital admissions and moderate/severe malnutrition) were calculated as the number of cases in children resident in the study area divided by the total children-years at risk (CYARs). CYARs were estimated from the demographic surveillance system databases. Children did not contribute to the numerator or denominator in the following situations: for a period of 15 days after each hospital admission, when they were outside the study area, after seroconversion (only in the HEU group) or after death. Cox proportional hazard models were used to assess the differences in mortality between the different age groups. Negative binomial regression models with random intercept were estimated to assess the association of HIV-exposure and other potential risk factors with the incidence of the studied outcomes. All the variables (sex, age, birth weight, type of delivery, location of delivery, gestational age, mother on CTXp, breastfeeding, maternal ART, child PMTCT and child CTXp) were included in the multivariate models to assess their adjusted associations with the incidence of outpatient attendances and hospital admissions among HEU children as they have all have been described in literature as potential confounders. Cox regression with left truncation (or delayed entry) was used for the evaluation of the associations with time to death.

The statistical software for analysis was STATA 14.1 (Stata Corp., College Station, Texas, USA).

Ethics statement and participants' safety

Protocols from both HEU and HIV-unexposed cohorts were reviewed and approved by the Ethics Committees

Table 1. Baseline characteristics of HIV-exposed and HIV-unexposed newborns at birth.

Characteristics at birth	HIV-exposed		HIV-nonexposed		P value
	N	n (%)	N	n (%)	
Sex (males)	966	508 (53)	909	460 (51)	0.3905 ^a
Birth weight (median, IQR ^b)	509	3000 (500)	906	3100 (500)	0.2420 ^c
LBW ^d (<2500g)	509	43 (8)	906	68 (8)	0.5269 ^a
Gestational age (median, IQR ^b)	395	39 (3)	738	38 (3)	0.8713 ^c
Preterm (<37 weeks)	395	74 (19)	738	103 (14)	0.0348 ^a
Delivery location					
Maternity	513	468 (91)	756	682 (90)	0.5420 ^a
Outside maternity	513	45 (9)	756	74 (10)	
Type of delivery					
Vaginal	510	479 (94)	754	707 (94)	0.9105 ^a
Caesarean section	510	31 (6)	754	47 (6)	

^aChi-squared test.^bInterquartile range.^cWilcoxon rank-sum test.^dLow birth weight.

from the Hospital Clinic of Barcelona (Spain) and local regulatory authorities and National Ethics Review Committee from Mozambique. The HIV-unexposed children trial was conducted under the provisions of the Declaration of Helsinki and in accordance with Good Clinical Practices guidelines set up by the WHO and by the International Conference on Harmonization.

Results

Baseline characteristics of study children at birth

A total of 966 HEU and 909 HIV-unexposed children were followed up for their first 18 months of life. There were 53 (508/966) and 51% (460/909) of men among HEU and HIV-unexposed children, respectively. There was no statistical significant difference in mean birth weight between HEU children [3000 g (interquartile range (IQR): 500)] and HIV-unexposed children [3100 g (IQR: 500); $P=0.2420$]. The prevalence of LBW was 8% in both groups. The proportion of preterm newborns was significantly higher in HEU children (19%, 74/395) compared with HIV-unexposed children (14%, 103/738; $P=0.0348$). The proportion of institutional deliveries was similar in both groups (91%, 468/513 in HEU children and 90%, 682/756 in HIV-unexposed children, respectively) with no difference either in the frequency of cesarean section by cohort, which was 6% in both groups (Table 1).

Mortality, morbidity and malnutrition among study children

A significantly lower incidence of outpatient attendances during the first 18 months of life was observed among HEU children compared with HIV-unexposed children [incidence rate ratio (IRR)=0.77, 95% confidence interval (CI): (0.71, 0.83); $P<0.0001$]. In contrast, hospital admissions [IRR=1.45, 95% CI: (1.15, 1.83); $P=0.0015$] and episodes of moderate or severe

malnutrition [IRR=2.14, 95% CI: (1.39, 3.30); $P=0.0006$ and IRR=2.62, 95% CI: (1.45, 4.75); $P=0.0014$, respectively] were more frequent among HEU children. A trend toward increased mortality was observed in the HEU cohort with 2.20 deaths per 100 children-year at risk compared with 1.16 deaths per 100 children-year at risk in the HIV-unexposed cohort [hazard ratio=1.87, 95% CI: (1.00, 3.52); $P=0.0510$] (Table 2). In both HEU and HIV-unexposed cohorts, mortality was higher in infants less than 6 months of age than in older children. Among HEU children, mortality rate was 2.67 per 100 CYARs [95% CI: (0.67, 10.66)] in neonates and 4.94 [95% CI: (3.07, 7.94)] in the 1–6-month age group, whereas among HIV-unexposed children, mortality rate in neonates was 5.28 [95% CI: (1.98, 14.07)] and 1.67 [95% CI: (0.75, 3.72)] in children between 1 and 6 months of age (Table 3).

Cause-specific hospital admissions are presented in Supplementary Table 1, <http://links.lww.com/QAD/B86>. Higher incidences of diarrhea and moderate or severe malnutrition were observed among the HEU cohort compared with the HIV-unexposed one [IRR=2.10, 95% CI: (1.17, 3.63); $P=0.0128$; IRR=2.14, 95% CI: (1.39, 3.30); $P=0.0006$ and IRR=2.62, 95% CI: (1.45, 4.75); $P=0.0014$, respectively].

Risk factors for outpatient attendances and hospital admissions in HIV-exposed uninfected children

Results from the univariate and multivariate analyses for outpatient attendances are presented in Table 4. In the multivariate analysis, female children had lower incidence of outpatient attendances [IRR=0.82, 95% CI: (0.67, 0.99); $P=0.0386$] compared with men, whereas children whose mothers were on ART during pregnancy had higher incidence of this outcome [IRR=1.33, 95% CI: (1.01, 1.75); $P=0.0428$] compared with children whose mothers were not on ART. Incidence of outpatient

Table 2. Incidence of outpatient visits, hospital admissions, mortality and malnutrition in HIV-exposed uninfected and unexposed children in first 18 months of life.

Incidences	HIV-exposed uninfected, N = 966				HIV-unexposed, N = 909				Incidence rate ratio	95% CI	P value
	Cases	CYAR ^a	Incidence rate (per 100 CYARs)	95% CI	Cases	PYAR	Incidence rate (per 100 CYARs)	95% CI			
Outpatient visits	3063	1233.28	248.36	(239.72, 257.32)	4157	1287.46	322.88	(313.22, 332.85)	0.77	(0.71, 0.83)	<0.0001
Hospital admissions	207	1233.68	16.78	(14.64, 19.23)	152	1292.70	11.76	(10.03, 13.78)	1.45	(1.15, 1.83)	0.0015
Mortality	27	1226.11	2.20	(1.51, 3.21)	15	1288.41	1.16	(0.70, 1.93)	1.87 ^b	(1.00, 3.52)	0.0510
Malnutrition											
Moderate ^c	77	1238.88	6.22	(4.97, 7.77)	39	1297.26	3.01	(2.20, 4.11)	2.14	(1.39, 3.30)	0.0006
Severe ^d	44	1240.04	3.55	(2.64, 4.77)	18	1298.12	1.39	(0.87, 2.20)	2.62	(1.45, 4.75)	0.0014

95% CI, 95% confidence interval.

^aChildren-years at risk.

^bHazard ratio.

^cWeight-for-age (WAZ), length-for-age (LAZ) or weight-for-length (WHZ) z-scores under -2 SD.

^dWAZ, LAZ or WHZ under -3 SD.

attendances increased with age from 6 to 18 months ($P=0.0001$).

Results from the univariate and multivariate analyses for hospital admissions are shown in Table 5. In the multivariate analysis, the incidence of hospital admissions decreased in older age groups, with children in the 12 to less than 18-month age group having the lowest IRR compared with those in the 0 to less than 1-month age group [IRR = 0.13, 95% CI: (0.03, 0.64); $P=0.0517$]. Children who were not exclusively breastfed, those who had been weaned and children who were never breastfed showed an increased incidence of hospital admissions compared with children who were exclusively breastfed [IRR = 2.21, 95% CI: (0.70, 6.99); IRR = 6.74, 95% CI: (1.87, 24.31) and IRR = 2.63, 95% CI: (0.92, 7.52); $P=0.0177$].

Discussion

There are very limited data on health outcomes and survival in HEU children in Mozambique, one of the countries with the highest HIV prevalence of the world.

A previously conducted study in the same area of smaller sample size also compared morbidity and mortality between HEU children and HIV-unexposed children. However, this study was performed only 4 years after initiation of ART rollout in the country, and it did not evaluate health determinants among HEU children [19]. The findings of the current study show that HEU children are more likely to be undernourished, be admitted to hospital and die within the first 18 months of life compared with HIV-unexposed children. All of this shows that maternal HIV infection continues to have important adverse consequences on children's health even if the infection is prevented in the children.

In our study, mortality rate in HEU children almost doubled that of HIV-unexposed children. Cohort studies from other sub-Saharan countries have shown that, before the availability of ART, HEU children had a higher mortality risk than HIV-unexposed children [4,6,29]. However, reports on mortality in HEU and HIV-unexposed children in the ART era are scarce, and results are heterogeneous [19,30–33]. Mortality rate in HEU children in this study (2.2 deaths per 100 live births per year at risk) was lower than that reported from other countries in sub-Saharan Africa in the ART era, though

Table 3. Incidence of mortality by age group in HIV-exposed uninfected and HIV-unexposed children in first 18 months of life.

Age	HIV-exposed uninfected			HIV-unexposed		
	Mortality rate ^a (95% CI)	HR ^b (95% CI)	P value ^c	Mortality rate ^a (95% CI)	HR ^b (95% CI)	P value
0 to <1 month	2.67 (0.67, 10.66)	1	0.0155	5.28 (1.98, 14.07)	1	0.0302
1 to <6 months	4.94 (3.07, 7.94)	1.92 (0.43, 8.51)		1.67 (0.75, 3.72)	0.25 (0.07, 0.94)	
6 to <12 months	1.22 (0.51, 2.93)	0.55 (0.11, 2.82)		0.92 (0.35, 2.46)	0.12 (0.03, 0.57)	
12 to <18 months	0.76 (0.24, 2.34)	0.47 (0.08, 2.62)		0.24 (0.03, 1.69)	0.04 (0.00, 0.51)	
Total	2.20 (1.51, 3.21)	–	–	1.16 (0.70, 1.93)	–	–

95% CI, 95% confidence interval.

^aPer 100 children-years at risk.

^bHazard ratio.

^cCox proportional hazard models.

Table 4. Risk factors for outpatient visits in HIV-exposed uninfected children.

Variable	n	Categories	Univariate analysis			Multivariate analysis ^a		
			Incidence rate ratio	(95% confidence interval)	P value	Incidence rate ratio	(95% confidence interval)	P value
Sex	684	Male	1		0.0122	1		0.0386
		Female	0.82	(0.71, 0.96)		0.82	(0.67, 0.99)	
Age	909	0 to <1 month	1		<0.0001	1		0.0001
		1 to <6 months	1.07	(0.76, 1.51)		0.96	(0.54, 1.70)	
		6 to <12 months	1.61	(1.16, 2.23)		1.60	(0.89, 2.86)	
		12 to <18 months	1.54	(1.11, 2.14)		1.40	(0.77, 2.52)	
Birth weight	494	≥2500 g	1		0.2404	1		0.4885
		<2500 g	1.18	(0.89, 1.57)		1.13	(0.79, 1.72)	
Type of delivery	495	Vaginal	1		0.2861	1		0.1263
		Cesarean	1.20	(0.86, 1.67)		1.35	(0.92, 1.98)	
Location of delivery	497	Maternity	1		0.0106	1		0.6494
		Home	0.57	(0.39, 0.82)		0.80	(0.50, 1.28)	
		Other	0.85	(0.37, 1.92)		0.00	–	
Gestational age (weeks)	384	≥37 weeks	1		0.7255	1		0.5085
		<37 weeks	0.96	(0.76, 1.21)		0.91	(0.70, 1.19)	
Mother on CTXp ^b	474	No	1		0.4436	1		0.2340
		Yes	0.94	(0.79, 1.11)		0.89	(0.73, 1.08)	
Breastfeeding	684	Exclusive	1		0.0038	1		0.2446
		Partial	1.17	(0.95, 1.43)		0.82	(0.61, 1.09)	
		Weaned	1.38	(1.09, 1.74)		0.88	(0.62, 1.24)	
		Never did	1.46	(1.15, 1.84)		1.01	(0.74, 1.38)	
Maternal ART ^c	684	No	1		0.0803	1		0.0428
		Yes	1.23	(0.98, 1.54)		1.33	(1.01, 1.75)	
Child PMTCT ^d	684	No	1		0.0081	1		0.9348
		Yes	1.24	(1.06, 1.44)		1.01	(0.74, 1.38)	
Child CTXp ^b	684	No	1		0.0042	1		0.1423
		Yes	1.39	(1.11, 1.75)		1.35	(0.90, 2.03)	

^aN=364.^bCotrimoxazol prophylaxis.^cAntiretroviral therapy.^dPrevention of mother-to-child transmission of HIV.

comparable with the rate previously observed in this setting (2.5 deaths per 100 live births per year at risk) [19,34]. Importantly, the mortality rate observed in HEU children may be underestimated as only children who were passively followed up at the HRPOC were included in the analysis, and therefore those who may have died before follow-up initiation or not engaged in healthcare were not part of it. This is supported by the mortality rate distribution observed by age group, whereby in the first month of life, death rate in HIV-unexposed children was twice as high as that in HEU children, whereas this was the opposite in the other age group. This may be explained by neonatal deaths occurring before initiation of follow-up at the HRPOC among HEU children not being captured. Contrarily, estimates in HIV-unexposed children could be regarded as minimum estimates, as they refer to children who were enrolled in a clinical trial in which healthcare and follow-up might have been better than those in the general population.

The risk of all-cause hospital admissions in the first 18 months of life was significantly higher in HEU children compared with HIV-unexposed children, which is consistent with the findings of previous studies conducted in the context of available antiretrovirals for PMTCT [7,9,32]. Diarrhea (loose or watery stools more

than three times in 1 day according to ICD-10 coding system) was more frequent among HEU compared with HIV-unexposed children. There is no conclusive evidence as to whether the risk of diarrhea is different in HEU compared with HIV-unexposed children in sub-Saharan Africa [5,19,30,35,36]. Increased risk of diarrhea-related morbidity in HEU might be a consequence of suboptimal breastfeeding rather than of HIV exposure itself. In contrast, as in the previous study performed in the same setting, the incidence of outpatient attendances was higher in HIV-unexposed children compared with HEU. This might be explained by the antibacterial effect of the routinely administered CTXp to HEU children from 6 weeks of age [19]. The special clinical follow-up of HEU children at the HRPOC might have also reduced outpatient healthcare utilization by HEU children.

HEU children were more likely to have higher incidence of both malnutrition-related hospital admissions and overall moderate and severe malnutrition during the first 18 months of life compared with HIV-unexposed children. HIV exposure during pregnancy has been associated with poor pregnancy outcomes such as preterm birth or LBW compared with nonexposed children [19,37–40]. In this study, prevalence of preterm birth was higher in HEU children compared with HIV-unexposed

Table 5. Risk factors for hospital admissions in HIV-exposed uninfected infants.

Variable	<i>n</i>	Categories	Univariate analysis			Multivariate analysis ^a		
			Incidence rate ratio	(95% confidence interval)	<i>P</i> value	Incidence rate ratio	(95% confidence interval)	<i>P</i> value
Sex	684	Male	1		0.6541	1		0.8055
		Female	0.90	(0.58, 1.41)		0.93	(0.51, 1.70)	
Age	909	0 to <1 month	1		0.0254	1		0.0517
		1 to <6 months	0.39	(0.17, 0.85)		0.37	(0.09, 1.52)	
		6 to <12 months	0.34	(0.16, 0.72)		0.24	(0.01, 1.05)	
		12 to <18 months	0.33	(0.15, 0.70)		0.13	(0.03, 0.64)	
Birth weight	494	≥2500	1		0.9033	1		0.5567
		<2500	0.95	(0.40, 2.26)		0.68	(0.19, 2.42)	
Type of delivery	495	Vaginal	1		0.9695	1		0.5521
		Cesarean	0.98	(0.35, 2.77)		1.39	(0.47, 4.16)	
Location of delivery	497	Maternity	1		0.6799	1		0.8717
		Home	0.60	(0.18, 1.95)		0.66	(0.14, 3.11)	
		Other	1.22	(0.15, 9.54)		0.00	–	
Gestational age (weeks)	384	≥37 weeks	1		0.9279	1		0.5864
		<37 weeks	1.03	(0.53, 2.01)		1.24	(0.57, 2.69)	
Mother on CTXp ^b	474	No	1		0.5228	1		0.6979
		Yes	0.85	(0.52, 1.40)		0.89	(0.48, 1.64)	
Breastfeeding	684	Exclusive	1		0.3682	1		0.0177
		Partial	0.95	(0.45, 2.04)		2.21	(0.70, 6.99)	
		Weaned	1.48	(0.63, 3.46)		6.74	(1.87, 24.31)	
		Never did	1.48	(0.68, 3.20)		2.63	(0.92, 7.52)	
Maternal ART ^c	684	No	1		0.6262	1		0.5378
		Yes	0.81	(0.35, 1.87)		1.37	(0.51, 3.70)	
Child PMTCT ^d	684	No	1		0.9461	1		0.5552
		Yes	0.98	(0.62, 1.57)		0.77	(0.31, 1.86)	
Child CTXp ^b	684	No	1		0.0242	1		0.9356
		Yes	0.51	(0.29, 0.92)		0.95	(0.28, 3.22)	

^a*N* = 364.^bCotrimoxazol prophylaxis.^cAntiretroviral therapy.^dPrevention of mother-to-child transmission of HIV.

children, though data on gestational age at the time of delivery were only available in one-third of the HEU infants. However, the role of HIV exposure during pregnancy, delivery and breastfeeding on postnatal growth remains uncertain. Previous to availability of antiretroviral drugs, several studies showed poor growth outcomes in HEU children compared with HIV-unexposed children [41,42]. However, reports in which antiretroviral drugs for PMTCT were used either found no differences in growth outcomes between HEU and HIV-unexposed children or if they existed were most likely due to different feeding practices rather than to HIV exposure [43,44].

The incidence of outpatient attendances in HEU children increased with age. Although exclusive breastfeeding is considered sufficient to provide adequate nutrition and immunological protection to infants during the first 6 months, the likely inadequacy, both in quantity and nutritional quality of weaning foods, together with exposure to infectious diseases may compromise the infant's health onward [45–48]. Another risk factor for outpatient attendance among HEU children was having a mother on ART during pregnancy. At the time of the study, women had to meet immunological and clinical criteria to start HIV treatment, being those more

immunosuppressed or with advanced clinical stage the ones who were eligible for ART. Offspring born to women with advanced HIV infection and eventually at higher risk of maternal mortality during childhood might be particularly vulnerable. In addition to an adverse social and economic background, compared with healthy mothers, those who are immunodeficient or clinically unwell might be less able to provide appropriate child care, adequate duration of breastfeeding or more likely to carry infectious pathogens harmful to their infants [49,50].

The main limitation of this study is the difference in the follow-up between both cohorts. The HEU cohort was followed up by passive surveillance at the HRPOC, and the HIV-unexposed cohort included children participating in a clinical trial who were actively followed up from birth. Another limitation is that information on feeding practices was not available for HIV-unexposed children limiting the availability to infer their effect on the differences between HEU and HIV-unexposed children. On the other hand, undetected HIV infections occurring in between the recommended HIV testings in HEU children might have influenced the results, as HIV-infected children tend to have worst health outcomes compared with HEU children [51]. Information on HEU

children was obtained from passive surveillance at the HRPOC, and consequently there were missing data for some of the independent variables, which may limit the interpretation of the multivariate analysis.

In conclusion, increased morbidity and mortality was observed in HEU children compared with HIV-unexposed children. New programs and interventions must take into consideration the long-term health needs of this growing population; they should also address transmission from mother to child, not only in terms of infant infections avoided, but also in terms of child survival. Due to the scale-up of option B+, MTCT of HIV is reducing and therefore the number of HIV-infected children. However, the prevalence of HIV-infected mothers is maintained, and thus the number of HEU children who have specific health needs that should be focused on to ensure that post-2015 sustainable development goals are achieved.

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Conflicts of interest

There are no conflicts of interest.

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