図 1:HIV 陽性結核患者での治療前体重毎の Kaplan-Meier 生存曲線

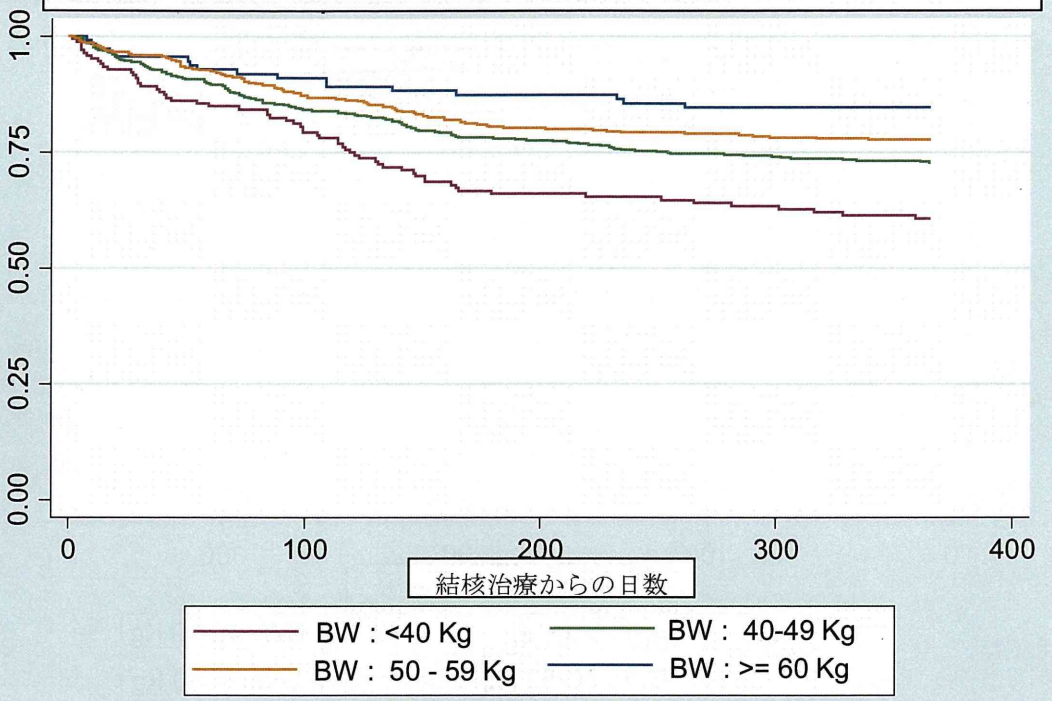


図 2:HIV 陽性結核患者での治療前 BMI 毎の Kaplan-Meier 生存曲線

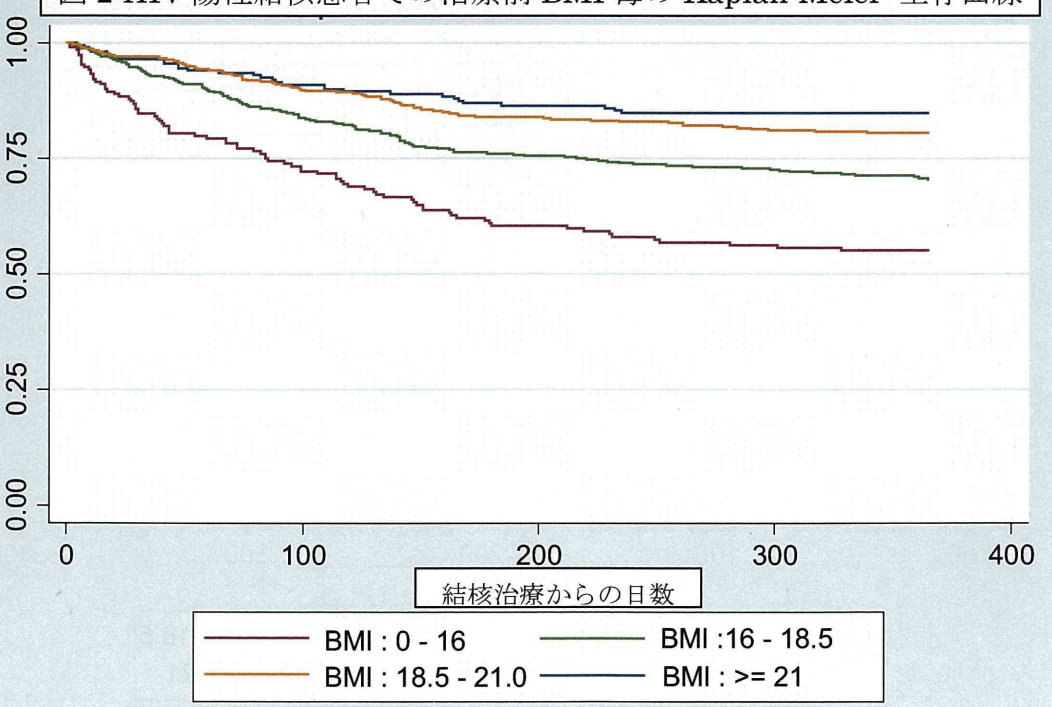


図 3:HIV 陰性結核患者での治療前体重毎の Kaplan-Meier 生存曲線

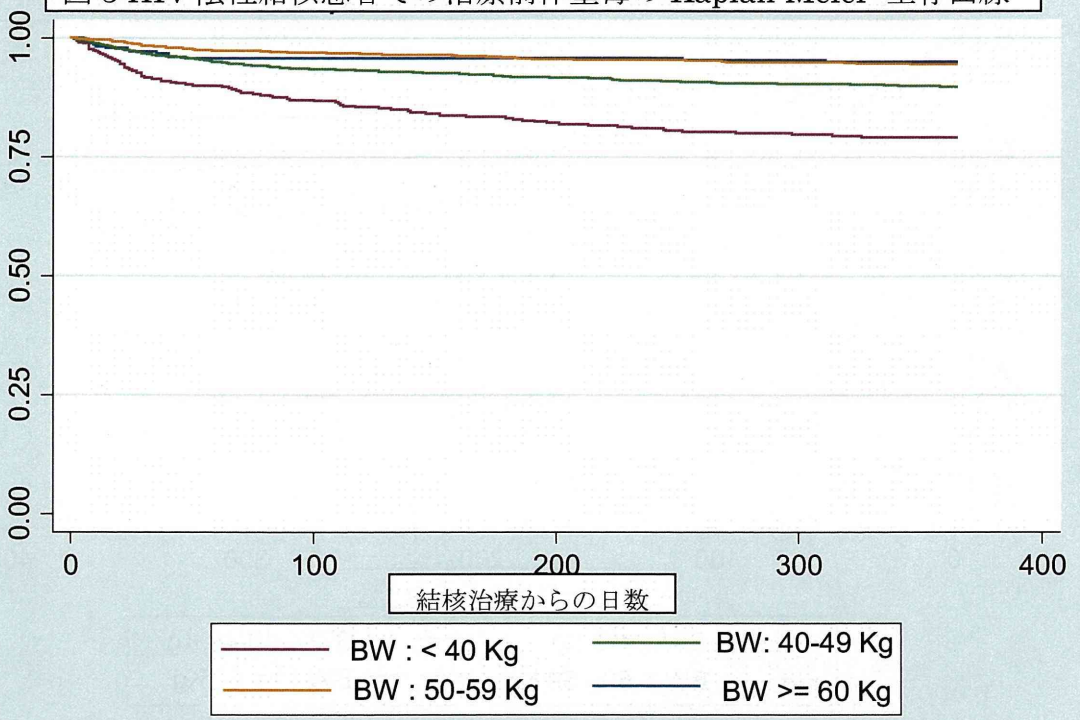
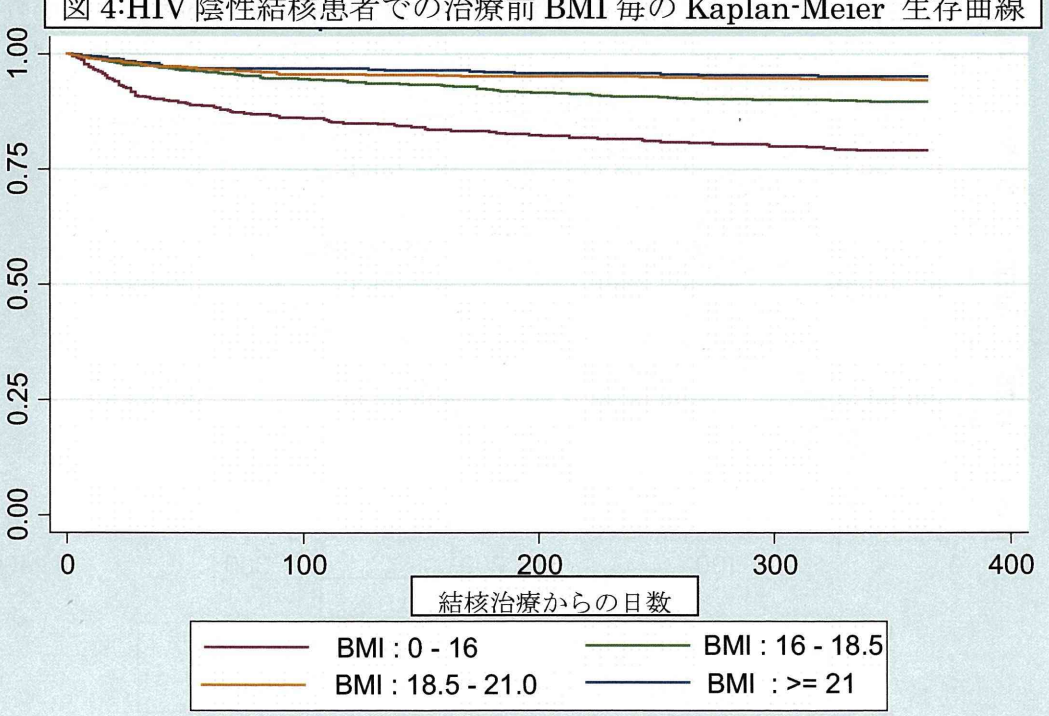


図 4:HIV 陰性結核患者での治療前 BMI 毎の Kaplan-Meier 生存曲線



Ⅳ章
研究成果の刊行に関する一覧表

研究成果の刊行に関する一覧表

(2009年4月1日～2012年3月31日迄)

発表者氏名	論文タイトル名	発表誌名	巻号	ページ	出版年
Koyanagi A, Humphrey JH, Moulton LH, et al.	Effect of early exclusive breastfeeding on morbidity among infants born to HIV-negative mothers in Zimbabwe	The American Journal of Clinical Nutrition	89(5)	1375-82	2009
Koyanagi A, Ruff AJ, Moulton LH, Ntozini R, Mutasa K, Iliff P, Humphrey JH; ZVITAMBO Study Group.	Postpartum plasma CD4 change in HIV-positive women: implications for timing of HAART initiation	AIDS Research and Human Retroviruses	26(5)	547-52	2010
Koyanagi A,Shibuya K.	What do we really know about adult mortality worldwide?	Lancet	375(9727)	1668-70	2010
Koyanagi A, Humphrey JH, Ntozini R, Nathoo K, Moulton LH, Iliff P, Mutasa K, Ruff A, Ward B and the ZVITAMBO Study Group.	Morbidity among human immunodeficiency virus-exposed but uninfected, human immunodeficiency virus-infected, and human immunodeficiency virus-unexposed infants in Zimbabwe before availability of highly active antiretroviral therapy	The Pediatric Infectious Disease Journal	30(1)	45-51	2011
Koyanagi A, Humphrey JH, Moulton LH, Ntozini R, Mutasa K, Iliff P, Ruff AJ and the Zvitambo Study Group.	Predictive value of weight loss on mortality of HIV-positive mothers in a prolonged breastfeeding setting	AIDS Research and Human Retroviruses	27(11)	1141-8	2011
Ota E, Wariki WMV, Mori R, Hori N, Shibuya K.	Behavioral interventions to reduce the transmission of HIV infection among sex workers and their clients in high-income countries	Cochrane Database of Systematic Reviews	Issue 12. Art. No.: CD006045.	DOI: 10.1002/14651858.CD006045.pub3.	2011

Nababan H, Ota E, Wariki WMV, Koyanagi A, Ezoe S, Shibuya K, Tobe-Gai R.	Structural and community-level interventions for increasing condom use to prevent HIV and other sexually transmitted infections (Protocol)	Cochrane Database of Systematic Reviews	Issue 11. Art. No.: CD003363.	DOI: 10.1002/14651858.CD003363.pub2.	2011
Shibuya K, Hashimoto H, Ikegami N, Nishi A, Tanimoto T, Miyata H, Takemi K, Reich MR.	Future of Japan's system of good health at low cost with equity: beyond universal coverage	Lancet	378(9798)	1265-73	2011
Llano R, Kanamori S, Kunii O, Mori R, Takei T, Sasaki H, Nakamura Y, Kurokawa K, Hai Y, Chen L, Takemi K, Shibuya K.	Re-invigorating Japan's commitment to global health: challenges and opportunities	Lancet	378(9798)	1255-64	2011
Kario K, Nishizawa M, Hoshide S, Shimpo M, Ishibashi Y, Kunii O, Shibuya K.	Development of a disaster cardiovascular prevention network	Lancet	378(9797)	125-7	2011
Ikegami N, Yoo BK, Hashimoto H, Matsumoto M, Ogata H, Babazono A, Watanabe R, Shibuya K, Yang BM, Reich MR, Kobayashi Y.	Japanese universal health coverage: evolution, achievements, and challenges	Lancet	378(9796)	1106-15	2011
Ikeda N, Saito E, Kondo N, Inoue M, Ikeda S, Satoh T, Wada K, Stickley A, Katanoda K, Mizoue T, Noda M, Iso H, Fujino Y, Sobue T, Tsugane S, Naghavi M, Ezzati M, Shibuya K.	What has made the population of Japan healthy?	Lancet	378(9796)	1094-105	2011

Kantipong P, Yamada N, Nampaisan O, Moolphate S, Yanai H, Harnsuthivatchakul C.	Predictors of antiretroviral therapy regimen changes in Northern Thailand	Bulltein of Department of Medical Services			2011 (in press)
Pitabut N, Mahasirimongkol S, Yanai H, Ridruechai C, Sakurada S,Dhepakson P, Kantipong P, Piyaworawong S, Moolphate S, Hansudewechakul C,Yamada N, Keicho N, Okada M, Khusmith S.	Decreased granulysin and increasedinterferon-gamma levels in plasma of patients with newly diagnosed and relapse tuberculosis	Microbiology and Immunology	55(8)	565-73	2011
Ridruechai C, Sakurada S, Yanai H, Yamada N, Kantipong P, Piyaworawong S, Dhepakson P, Khusmith S, Keicho N.	Association between circulating full-length osteopontin and IFY-g with disease status of tuberculosis and response to successful treatment	The Southeast Asian Journal of Tropical Medicine and Public Health	42(4)	876-889	2011
Wariki WMV, Ota E, Mori R, Koyanagi A, Hori N, Shibuya K.	Behavioral interventions to reduce the transmission of HIV infection among sex workers and their clients in low- and middle-income countries	Cochrane Database of Systematic Reviews	Issue 2. Art. No.: CD005272.	DOI: 10.1002/14651858.CD005272.pub3.	2012

V章
代表的関連刊行物・別刷

See corresponding editorial on page 1281.

Effect of early exclusive breastfeeding on morbidity among infants born to HIV-negative mothers in Zimbabwe[1–3]

Ai Koyanagi, Jean H Humphrey, Lawrence H Moulton, Robert Ntozini, Kuda Mutasa, Peter Iliff, Robert E Black, and the ZVITAMBO Study Group

ABSTRACT

Background: Early exclusive breastfeeding (EBF) is recommended by the World Health Organization, but EBF rates remain low throughout the world. For infants born to breastfeeding HIV-positive mothers, early EBF is associated with a lower risk of postnatal transmission than is feeding breast milk together with other liquids or foods. No studies conducted in Africa have reported any benefits of EBF for infants born to HIV-negative women.

Objective: The objective was to compare the rate of sick clinic visits by infants aged 43–182 d according to breastfeeding exclusivity [EBF, predominant breastfeeding (PBF), and mixed breastfeeding (MBF)].

Design: We compared rates of all-cause clinic visits and clinic visits related to diarrhea and lower respiratory tract infection (LRTI) among a cohort of 9207 infants of HIV-negative mothers during 2 age intervals: 43–91 and 92–182 d according to exclusivity of breastfeeding. Breastfeeding exclusivity was defined in 2 ways ("ever since birth" and "previous 7 d") and was assessed at 43 and 91 d.

Results: EBF between birth and 3 mo was significantly protective against diarrhea between 3 and 6 mo of age with the "ever since birth" definition [incidence rate ratios (IRRs) of 8.83 (95% CI: 1.07, 65.53) and 8.76 (95% CI: 1.13, 68.09) for PBF and MBF, respectively] and with the "previous 7 d" definition [2.04 (95% CI: 1.11, 3.77) and 2.05 (95% CI: 1.13, 3.72) for PBF and MBF, respectively]. The adverse effect of MBF on LRTI visits was weaker, reaching borderline significance only by the "ever since birth" definition during the 43–91-d interval (IRR: 1.91; 95% CI: 0.99, 3.67).

Conclusion: Early EBF is associated with a significant reduction in sick clinic visits, especially those due to diarrhea. *Am J Clin Nutr* 2009;89:1375–82.

INTRODUCTION

The 2003 *Lancet* series on child survival identified the promotion of exclusive breastfeeding (EBF) during the first 6 mo of life and continued breastfeeding to 12 mo as the single most effective preventive public health intervention for reducing mortality among children aged <5 y (1). More recently, the 2008 *Lancet* series on maternal and child undernutrition estimated that 1.06 million child deaths (10% of all mortality in children aged <5 y) are attributable to nonexclusive breastfeeding in the first 6 mo of life (2). Most of the data supporting these analyses came from studies conducted in Asia and Latin America (3–6). In Africa, where >40% of the world's deaths of children aged <5 y occur each year, breastfeeding is nearly universal and prolonged, but EBF is rare (7). Over the past 8 y, accumulating evidence that early EBF substantially lowers the risk of breastfeeding-associated HIV transmission more than does predominant breastfeeding (PBF) or mixed breastfeeding (MBF) (8–10) has led to renewed enthusiasm for EBF promotion in sub-Saharan Africa, at least for HIV-infected mothers. Two studies in Ghana have examined EBF-associated health benefits for infants born to HIV-negative mothers. The first study observed higher neonatal mortality among MBF and PBF infants than among EBF infants, although the risk associated with PBF was not statistically significant. The second study (a multisite study in which 31% of the infants were Ghanaian) found no difference in mortality risk between EBF and PBF infants and no difference in hospitalization rates between EBF and either PBF or MBF infants (11). The authors of that study concluded that efforts should focus on sustaining high rates of PBF rather than encouraging mothers to practice strict EBF.

The ZVITAMBO (Zimbabwe Vitamin A for Mothers and Babies) trial enrolled 14,110 mother-infant pairs and assessed early infant feeding practices. Among the 4495 mothers who were HIV-positive at delivery, 2060 of their infants were HIV-negative (by polymerase chain reaction) at 6 wk and continued to be breastfed. Compared with early EBF, PBF and MBF infants before 3 mo of age had, respectively, 2.6 and 4.0 times greater risks of breastfeeding-associated HIV transmission by 6 mo and 1.6 and 2.6 times greater risks of transmission by 18 mo (9).

[1] From the Johns Hopkins Bloomberg School of Public Health, Department of International Health, Baltimore MD (AK, JHH, LHM, and REB), and the ZVITAMBO Study Team, Harare, Zimbabwe (JHH, RN, KM, and PI).

[2] The ZVITAMBO project was supported by the Canadian International Development Agency (R/C Project 690/M3688), US Agency for International Development (cooperative agreement number HRN-A-00-97-00015-00 between Johns Hopkins University and the Office of Health and Nutrition–USAID), and a grant from the Bill and Melinda Gates Foundation, Seattle, WA. Additional funding was received from the Rockefeller Foundation (New York, NY) and BASF (Ludwigshafen, Germany).

[3] Reprints not available. Address correspondence to JH Humphrey, #1 Borrowdale Road, Borrowdale, Harare, Zimbabwe. E-mail: jhumphrey@zvitambo.co.zw.

Received August 8, 2008. Accepted for publication January 8, 2009.
First published online April 1, 2009; doi: 10.3945/ajcn.2008.26810.

These findings indicated that early introduction of solid foods and nonhuman milk (MBF) conveys an especially high risk of HIV transmission, but that feeding even nonmilk fluids (PBF) increases transmission risk. In the present study, we investigated whether EBF is also associated with lower morbidity among infants in the same study population whose mothers tested HIV-negative at delivery and remained HIV-negative throughout the breastfeeding period. The results of this study would help clarify whether universal promotion of early EBF would benefit all infants, whether HIV-exposed or not.

SUBJECTS AND METHODS

The design of the ZVITAMBO trial was described elsewhere (9, 12). In brief, 14,110 mother-infant pairs were recruited within 96 h of delivery at 14 hospitals and clinics in greater Harare between November 1997 and January 2000. The vast majority of Harare residents had tap water and sanitation facilities at the time of the study. Mother-infant pairs were eligible if neither had an acutely life-threatening condition, the infant was a singleton with a birth weight $\geq$1500 g, and the mother planned to stay in Harare after delivery. Written informed consent was obtained. Information on baseline characteristics was obtained by questionnaire or transcription from medical records. Gestational age was estimated (13). Infant birth weight (model 727; Seca Hanover, MD) and maternal midupper arm circumference (MUAC) were measured (14). Household income was adjusted for inflation and converted to US dollars.

At enrollment, women were tested for HIV by an algorithm incorporating 2 enzyme-linked immunosorbent assays and Western blot as previously described (12). Women who tested negative at baseline were retested at every subsequent blood draw to identify those who seroconverted postpartum. Hemoglobin was measured in women enrolled from October 1998 to the end of the study ($\approx$60% of the total sample) by using a hemoglobinometer (HemoCue, Mission Viejo, CA).

Follow-up visits were conducted at 6 wk, 3 mo, and every 3 mo thereafter up to 24 mo. At each scheduled visit, mothers were asked if, since the previous visit, their infant had been taken to a health care provider for treatment of an illness. The date and reason for each visit were determined from records completed by the health care provider who attended the infant, or, when these records were not available, by maternal report. If a child was sick when making a scheduled study visit, or if a sick child presented to the research clinic between scheduled visits, free treatment was provided by the study. Public sector health care was free of charge for children aged <5 y when recruitment for the trial began. However, as economic conditions declined during the period of the study, fees were sometimes charged and medicines were occasionally out of stock. The Medical Research Council of Zimbabwe, Medicines Control Authority of Zimbabwe, Johns Hopkins Bloomberg School of Public Health Committee on Human Research and Montreal General Hospital Ethics Committee approved the ZVITAMBO trial protocol.

Definition of breastfeeding

Detailed infant feeding information was collected at baseline, 6 wk (28–56 d), and 3 mo (77–105 d). At each of these 3 visits, the mothers were asked whether any of 22 items had ever been given to the infant since birth. These included nonmilk liquids (eg, plain water, sugar water, and juice), nonhuman milks (eg, infant formula and cow milk), solid foods (eg, porridge, fruit, vegetables, and eggs), and medicines (eg, gripe water, traditional, and prescribed Western oral medicines). Because enrollment occurred anytime between birth and 96 h, to avoid misclassification due to age at baseline, at 6 wk, mothers were also asked how the infant was fed during the entire first 96 h; this information was considered together with the data collected at baseline in classifying baseline feeding practice. At 6 wk and 3 mo, the mother was also asked whether any of the 22 foods had been given to the infant within the previous 7 d.

The World Health Organization definitions for EBF, PBF, and MBF were used (15). EBF was defined as breast milk and Western oral medications only, PBF was defined as breast milk and nonmilk liquids including oral rehydration solution (ORS), and MBF was defined as breast milk and nonhuman milk or solid food. For each infant, the breastfeeding pattern for 2 intervals (birth to 6 wk and birth to 3 mo) was defined in 2 ways:

1) "Ever since birth" definition: data collected at 2 time points (baseline and 6 wk) were used for the 6-wk definition, and data collected at 3 time points (baseline, 6 wk, and 3 mo) were used for the 3-mo definition. This definition was applied only among infants who were breastfeeding and provided complete dietary data. (EBF infants: only received breast milk and Western medicine at all time points; PBF infants: received nonmilk liquid at at least one time point but never received nonhuman milk or solid food; MBF infants: received nonhuman milk or solid food at at least one time point.)
2) "Previous 7 d" definition: infants were classified into EBF, PBF, or MBF categories based on one 7-d diet history at 6 wk and 3 mo.

Statistical analysis

Statistical analysis was conducted by using Stata version 9.2 (StataCorp LP, College Station, TX). Baseline characteristics were compared between EBF, PBF, and MBF infants defined by the 7-d history at 3 mo. Between-group differences were tested by using chi-square and Kruskal-Wallis tests for categorical and continuous variables, respectively.

Preliminary analyses showed that a significantly greater proportion of MBF infants than EBF infants at baseline had an APGAR score <8, had a birth weight <2500 g, had a gestational age <37 wk, and had been delivered by a mode other than normal vaginal delivery. For nearly all (642/645) of these MBF infants at baseline, the food that resulted in their being classified as MBF was commercial infant formula. Because this formula feeding was most likely part of clinical care required by their vulnerable medical condition, we reasoned that subsequent morbidity more likely reflected poor health at birth rather than MBF at birth. To avoid this reverse causality, we excluded these 645 infants from the "ever since birth" definition in order to not overestimate the deleterious effects of MBF.

When the trial began, little information was available on breastfeeding-associated HIV transmission. When the United Nations Program on HIV/AIDS produced new guidelines in June 1998, we conducted formative research to inform an intervention to educate and counsel mothers about infant feeding and HIV. This intervention (16) promoted EBF to 6 mo for all HIV-negative mothers and HIV-positive mothers who chose to

breastfeed and was, respectively, partially and fully implemented by September and November 1999. Thus, EBF rates were higher among infants enrolled later; therefore, all regression analyses were adjusted for enrollment date.

Child-years of observation were calculated for 2 time periods: 43 through 91 d and 92 through 182 d of age; infants were censored on the date of loss to follow-up or death. We used these periods for the primary analyses to avoid reverse causality resulting from overlapping exposure and outcome periods. The number of total sick clinic visits and cause-specific visits for lower respiratory tract infection (LRTI) and diarrhea were summed for each child during each time interval. The incidence of total and cause-specific clinic visits was calculated for the 43–91 d-interval stratified by breastfeeding practice (EBF, PBF, and MBF) at 6 wk, as defined by each definition ("ever since birth" and "previous 7 d"). These calculations were repeated for the 92–182-d interval stratified by breastfeeding practice at 3 mo. Incidence rate ratios (IRRs) were calculated with EBF as the reference group by using negative binomial regression to adjust for overdispersion.

Important covariates were identified by stepwise selection of variables (with entry and retention levels of $P = 0.20$ and 0.10, respectively) in a negative binomial regression in which feeding group was forced into the model. In addition, baseline characteristics that were unevenly distributed among the breastfeeding groups were assessed for their confounding effect. Because of the small number of events in the cause-specific models, only the strongest covariates were retained. To address the possibility of breastfeeding status changing as a result of illness, a sensitivity analysis that excludes all sick clinic visits occurring within 7 d of breastfeeding status assessment was conducted.

We also hypothesized that if the breastfeeding mode changed because of infant disease or vulnerability and because infants who were sick in the past also tend to be sick subsequently, PBF and MBF in the past may be a marker of vulnerability. For example, provision of liquid or solid food because of loss of appetite due to disease or mixing of bitter medicine into sweet liquids or provision of ORS all shift children into PBF or MBF because of disease. To address this issue, we conducted a sensitivity analysis where we added a binary variable that was "ever having had sick clinic visit due to diarrhea or LRTI in last 42 or 91 d" as adjustment covariate to cause-specific regression analyses. A P value < 0.05 was considered to be significant.

RESULTS

Of the 14,110 mothers enrolled in the ZVITAMBO trial, 9207 tested HIV-negative at baseline and did not seroconvert during follow-up. Infant feeding data were provided by 6665 and 6878 HIV-negative mothers at 6 wk and 3 mo, respectively. After the exclusion of baseline MBF infants and mother-infant pairs who came for their visit more than ± 14 d from the scheduled visit date, 4965 and 4425 mothers were classified by the "ever since birth" definition at 6 wk and 3 mo, respectively (**Table 1**). A total of 5470 and 5398 mothers provided a 7-d feeding history within 14 d of the scheduled visit date at 6 wk and 3 mo, respectively (Table 1). Mothers who were primiparous and younger and those with a male infant were less likely to exclusively breastfeed; other baseline characteristics did not differ across the 3 feeding groups (**Table 2**). Baseline characteristics were similarly distributed across feeding groups when defined by the "ever since birth" definition at 3 mo or when defined by either definition at 6 wk (data not shown). The background mortality rate was low; 99.6% of the 5398 infants who provided "previous 7 d" breastfeeding status at 3 mo were alive at 6 mo.

TABLE 1
Breastfeeding exclusivity rates at 6 wk and 3 mo of age according to 2 definitions[1]

	"Ever since birth"			
"Previous 7 d"	EBF[2]	PBF[2]	MBF[2]	Total
6 wk [*n* (%)]				
EBF	403 (61.4/100.0)	237 (36.1/7.4)	16 (2.5/1.2)	656
PBF	0	2949 (93.2/92.6)	214 (6.8/15.6)	3163
MBF	0	0	1146 (100.0/83.3)	1146
Total	403	3186	1376	4965
3 mo [*n* (%)]				
EBF	147 (30.8/100)	296 (62.1/19.5)	34 (7.1/1.2)	477
PBF	0	1225 (78.0/80.5)	346 (22.0/12.5)	1571
MBF	0	0	2377 (100.0/86.2)	2377
Total	147	1521	2757	4425

[1] EBF, exclusive breastfeeding; PBF, predominant breastfeeding; MBF, mixed breastfeeding. "Previous 7 d" refers to breastfeeding practice during the previous 7 d, and "ever since birth" refers to breastfeeding practice since birth.

[2] The first value in parentheses corresponds to the proportion of infants classified as EBF, PBF, or MBF by the "previous 7 d" definition at 6 wk and 3 mo; the second value in parentheses corresponds to the proportion of infants classified as EBF, PBF, or MBF by the "ever since birth" definition at 6 wk and 3 mo.

Breastfeeding practice, household income, and sick clinic visit rates were all strongly associated with date of enrollment into the trial. The EBF rate (during both time intervals defined by either definition) was highest among infants enrolled in the last quartile, reflecting introduction of the EBF promotion intervention (**Table 3**). Median household income fell and the proportion of families living on <$1/d rose over the recruitment period. Rates of sick clinic visits fell, for both total and cause-specific visits and for the first and second 3-mo age intervals. This may reflect the declining economy because the proportion of sick clinic visits conducted at the study clinic (where treatment was free and transport costs were reimbursed) rose over the recruitment period.

Food items consumed by >1% of the PBF and MBF infants with the "previous 7 d" definition at 3 mo are illustrated in **Figure 1**. Water, cooking oil, juice, traditional medicine, and gripe water were the most common liquids that shifted infants from EBF to PBF. Porridge was the most common food item that shifted infants from PBF to MBF; however, most MBF infants also consumed water, cooking oil, and juice.

Between 43 and 182 d, the infants included in the "ever since birth" and "previous 7 d" definitions made a total of 4951 and 5690 sick clinic visits, respectively; the all-cause sick clinic rate was similar by the 2 definitions (282.3 visits/100 child-years and 276.1 visits/100 child-years, respectively). In addition, with the use of either definition, the all-cause visit rate was higher during the 43–91-d interval than during the 92–182-d interval (**Table 4**). Upper respiratory infection and skin disease were the most common causes of sick clinic visits, accounting for 45.5% and 12.6% respectively, of the total visits among children included in the "previous 7 d" definition at 3 mo. Because breastfeeding practice was not significantly associated with sick clinic visits

TABLE 2
Baseline characteristics by breastfeeding practice at 3 mo by the "previous 7 d" definition[1]

	EBF	PBF	MBF	*P* value
Infant characteristics				
Male sex	257 (46.6)	980 (51.6)	1577 (53.6)	0.009[2]
5-min APGAR <8	13 (2.5)	74 (4.0)	107 (3.7)	0.265[2]
Birth weight <2500 g	72 (13.1)	247 (13)	327 (11.2)	0.106[2]
Birth weight (g)	3012.9 ± 480.5[3]	3006.7 ± 464.2	3010.1 ± 443.1	0.978[4]
Gestational age <37 wk[5]	42 (7.7)	132 (7.0)	172 (5.9)	0.136
Mode of delivery not normal vaginal[6]	61 (11.2)	214 (11.4)	319 (11.0)	0.882[2]
Maternal characteristics				
Hemoglobin				0.216[2]
<70 g/L	6 (1.6)	16 (1.6)	26 (1.7)	
70–120 g/L	156 (41.1)	448 (44.1)	737 (47.2)	
>120 g/L	218 (57.4)	553 (54.4)	800 (51.2)	
Marital status				0.535[2]
Married or stable	524 (95.5)	1786 (94.2)	2783 (94.8)	
Separated or widowed	9 (1.6)	29 (1.5)	36 (1.2)	
Single or never married	16 (2.9)	81 (4.3)	118 (4.0)	
Age				0.003[2]
<20 y	98 (17.8)	438 (23.1)	674 (22.9)	
20–34 y	389 (70.5)	1305 (68.9)	2037 (69.3)	
>34 y	65 (11.8)	151 (8.0)	227 (7.7)	
Education <8 y	95 (17.3)	337 (17.8)	531 (18.1)	0.897[2]
Occupation				0.058[2]
Unemployed	444 (80.6)	1607 (84.7)	2405 (81.8)	
Domestic worker	32 (5.8)	84 (4.4)	149 (5.1)	
Other	75 (13.6)	206 (10.9)	387 (13.2)	
MUAC <23 cm[7]	51 (9.3)	219 (11.6)	328 (11.2)	0.311[2]
Parity				<0.001[2]
1	161 (29.2)	879 (46.3)	1329 (45.1)	
2–4	341 (61.8)	887 (46.7)	1414 (48.0)	
≥5	50 (9.1)	134 (7.1)	203 (6.9)	
Vital status of previous child[8]				0.024[2]
Alive	375 (96.2)	956 (94.6)	1497 (92.8)	
Dead	15 (3.9)	55 (5.4)	116 (7.2)	
Religion				0.136[2]
Apostolic or Zion	169 (30.7)	545 (28.7)	880 (29.9)	
Protestant or Catholic	338 (61.3)	1139 (60.0)	1725 (58.6)	
Other	44 (8.0)	214 (11.3)	340 (11.5)	
Other characteristics				
Household income (US$)[9]	79.1 (51.9–136.4)[10]	82.2 (53.9–133.6)	78.5 (50.7–133.6)	0.522[4]
No. of subjects whose household income was available	440	1453	2246	
Paternal education <8 y	38 (7.0)	126 (6.8)	232 (8.1)	0.217[2]
Paternal occupation				0.468[2]
Unemployed	44 (8.1)	136 (7.3)	205 (7.1)	
Domestic worker	158 (29.2)	515 (27.5)	741 (25.6)	
Skilled manual	205 (37.8)	739 (39.4)	1179 (40.7)	
Other	135 (24.9)	484 (25.8)	775 (26.7)	
Date of enrollment				<0.001[2]
25 Nov 1997–15 Jun 1998	106 (19.2)	563 (29.6)	833 (28.3)	
16 Jun 1998–31 Dec 1998	82 (14.9)	464 (24.4)	832 (28.2)	
1 Jan 1999–15 Jul 1999	89 (16.1)	408 (21.5)	663 (22.5)	
16 Jul 1999–31 Jan 2000	275 (49.8)	465 (24.5)	618 (21.0)	

[1] Data are *n* (%) unless otherwise stated. Less than 2% of data are missing for all characteristics, except for hemoglobin and household income. EBF, exclusive breastfeeding; PBF, predominant breastfeeding; MBF, mixed breastfeeding. "Previous 7 d" refers to breastfeeding practice during the previous 7 d.
[2] *P* value calculated by using a chi-square test across all 3 groups by "previous 7 d" breastfeeding definition.
[3] Mean ± SD (all such values).
[4] *P* value calculated by using a Kruskal-Wallis test across all 3 groups by "previous 7 d" breastfeeding definition.
[5] Calculated by using the method of Capurro et al (13).
[6] Includes breech, forceps, vacuum, and cesarean deliveries.
[7] Midupper arm circumference; method described by Gibson (14).
[8] Primiparous mothers excluded.
[9] Adjusted for inflation.
[10] Median; 25th–75th percentile in parentheses (all such values).

TABLE 3
Exclusive breastfeeding (EBF) rate, incidence of sick clinic visits, and household income according to date of enrollment[1]

	Quartile of enrollment[2]			
	1	2	3	4
Proportion of EBF infants to 6 wk [% (*n*)]				
"Ever since birth" definition[3]	6.9 (1327)	4.6 (1275)	6.3 (1026)	14.1 (1337)
"Previous 7 d" definition[4]	11.4 (1431)	9.4 (1403)	10.1 (1196)	20.8 (1440)
Proportion of EBF infants to 3 mo [% (*n*)]				
"Ever since birth" definition[3]	2.5 (1161)	0.8 (1109)	1.8 (915)	7.8 (1163)
"Previous 7 d" definition[4]	7.3 (1249)	6.1 (1224)	7.4 (1073)	20.6 (1257)
Household income (US$/mo)[5,6]				
Median	82.0	86.9	74.2	74.2
<$1.00/d [% (*n*)]	6.7 (1086)	6.9 (985)	10.4 (937)	12.5 (1131)
Sick clinic visits [no. per 100 child-years (child-years of observation)][6]				
All-cause, 0–6 mo	412.4 (744.6)	328.2 (682.5)	301.0 (576.0)	250.9 (674.5)
Diarrhea-specific, 0–6 mo	26.2 (744.6)	19.9 (682.5)	19.4 (576.0)	10.5 (674.5)
LRTI-specific, 0–6 mo	79.5 (744.6)	59.8 (682.5)	49.3 (576.0)	32.8 (674.5)
All-cause, 0–3 mo	525.9 (374.2)	402.6 (343.2)	400.3 (289.0)	324.9 (338.3)
All-cause, 3–6 mo	297.8 (370.4)	252.9 (339.3)	201.1 (287.0)	176.4 (336.2)
Proportion of all sick clinic visits in children aged 0–6 mo				
At research clinic [% (*n*)]	8.6 (2497)	16.3 (1712)	29.2 (1393)	37.5 (1426)

[1] Tests of general association across the 4 enrollment quartiles: proportions with EBF, household income <$1.00/d, and sick clinic visits at research clinic were tested by using chi-square tests (3 df); median income by using a Kruskal-Wallis test; and incidence of sick clinic visits by using negative binomial regression (3 df Wald test); the result for each test was $P < 0.001$. LRTI, lower respiratory tract infection.
[2] Quartile 1 (25 November 1997–15 June 1998), 2 (16 June 1998–31 December 1998), 3 (1 January 1999–15 July 1999), and 4 (16 July 1999–31 Jan 2000).
[3] Definition refers to all foods consumed since birth.
[4] Definition refers to all foods consumed during the previous 7 d.
[5] Adjusted for inflation.
[6] Based on infants who provided "previous 7 d" breastfeeding information at 3 mo.

for either of these causes (data not shown) and because both are rarely life threatening, our analysis focused on visits for LRTI and diarrhea. Based on the "previous 7 d" data, the visit rate for LRTI was higher in the 43–91-d interval than in the 92–182-d interval (63.5 and 46.4 visits/100 child-year, respectively), whereas the diarrhea-specific visit rate increased with infant age (14.6 and 26.3 visits/100 child-years during the 43–91-d and 92–182-d intervals, respectively). Values for all of these rates were similar when calculated based on infants included in the "ever since birth" feeding definition (Table 4).

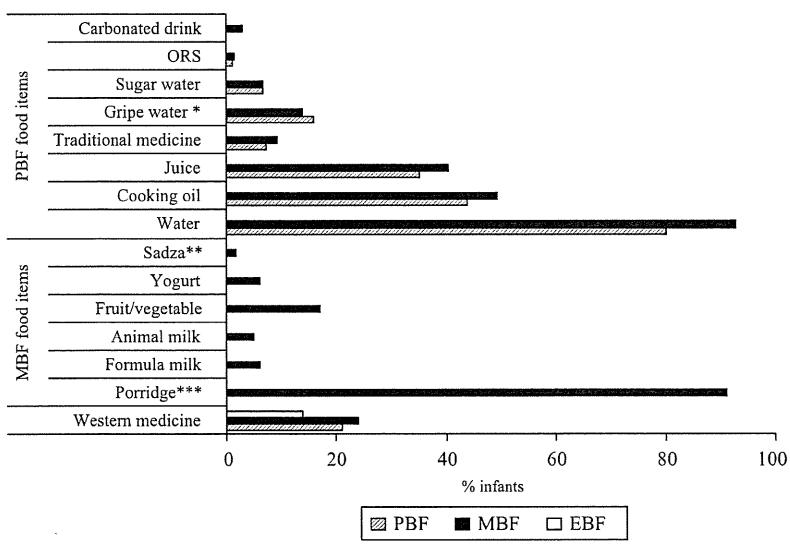


FIGURE 1. Food items consumed by >1% of the infants by breastfeeding practice during the previous 7 d at 3 mo of age. Percentage of infants who had consumed that item. EBF, exclusive breastfeeding ($n = 552$); MBF, mixed breastfeeding ($n = 2946$); PBF, predominantly breastfeeding ($n = 1900$); ORS, oral rehydration solution. *Home remedy for colic and other stomach symptoms. Consists of dill, alcohol, and sodium bicarbonate. **Thickened porridge or dumpling made of maize meal. ***Porridge usually refers to diluted sadza.

TABLE 4
Illness-associated infant clinic visits for all causes, lower respiratory tract infection (LRTI), and diarrhea according to feeding mode between birth and 6 wk or 3 mo according to the "ever since birth" and "previous 7 d" definitions[1]

		Total				LRTI-specific				Diarrhea-specific			
Feeding mode	Days of observation	No. of visits	IRR[2]	95% CI	*P*	No. of visits	IRR[3]	95% CI	*P*	No. of visits	IRR[4]	95% CI	*P*
"Ever since birth"													
43–91-d[5]													
EBF	19,616	155	1.00	—	—	19	1.00	—	—	4	1.00	—	—
PBF	154,856	1531	1.14	(1.93, 1.41)	0.197	267	1.64	(0.88, 3.05)	0.121	72	2.17	(0.70, 6.76)	0.182
MBF	66,686	642	1.11	(0.89, 1.38)	0.366	137	1.91	(0.99, 3.67)	0.052	24	1.73	(0.52, 5.75)	0.371
Total[6]	241,158	2328 (352.9/100 cy)				423 (64.1/100 cy)				100 (15.2/100 cy)			
92–182 d[7]													
EBF	13,369	67	1.00	—	—	9	1.00			1	1.00	—	—
PBF	137,641	900	1.17	(0.85, 1.63)	0.333	169	1.50	(0.58, 3.91)	0.406	101	8.38	(1.07, 65.53)	0.043
MBF	248,607	1656	1.19	(0.86, 1.65)	0.285	333	1.55	(0.60, 3.99)	0.364	192	8.76	(1.13, 68.09)	0.038
Total[6]	399,617	2623 (239.7/100 cy)				511 (46.7/100 cy)				294 (26.9/100 cy)			
"Previous 7 d"													
43–91-d[8]													
EBF	34,872	298	1.00	—	—	46	1.00	—	—	13	1.00	—	—
PBF	169,261	1663	1.08	(0.92, 1.26)	0.347	295	1.24	(0.80, 1.94)	0.336	68	1.03	(0.50, 2.12)	0.942
MBF	61,543	598	1.05	(0.88, –1.25)	0.560	121	1.33	(0.81, 2.19)	0.254	25	1.05	(0.46, 2.39)	0.901
Total[6]	265,676	2559 (351.8/100 cy)				462 (63.5/100 cy)				106 (14.6/100 cy)			
92–182 d[9]													
EBF	49,623	244	1.00	—	—	53	1.00	—	—	16	1.00	—	—
PBF	171,873	1130	1.20	(1.00, 1.44)	0.045	197	0.88	(0.55, 1.43)	0.620	133	2.04	(1.11, 3.77)	0.022
MBF	265,341	1757	1.23	(1.03, 1.46)	0.021	368	1.04	(0.65, 1.65)	0.884	201	2.05	(1.13, 3.72)	0.019
Total[6]	486,837	3131 (234.9/100 cy)				618 (46.4/100 cy)				350 (26.3/100 cy)			

[1] EBF, exclusive breastfeeding; PBF, predominant breastfeeding; MBF, mixed breastfeeding; IRR, incidence rate ratio; cy, child-years. 95% CIs and *P* values calculated by negative binomial regression. "Previous 7 d" refers to breastfeeding practice during the previous 7 d, and "ever since birth" refers to breastfeeding practice since birth.
[2] Adjusted for enrollment date, parity, 5-min APGAR score, and religion.
[3] Adjusted for enrollment, parity, and sex.
[4] Adjusted for enrollment date.
[5] Morbidity by breastfeeding practice at 6 wk ($n = 4965$).
[6] Total incidence rate: sick clinic visits for all causes, LRTI, and diarrhea per 100 cy.
[7] Morbidity by breastfeeding practice at 3 mo ($n = 4425$).
[8] Morbidity by breastfeeding practice at 6 wk ($n = 5470$).
[9] Morbidity by breastfeeding practice at 3 mo ($n = 5398$).

On the basis of the stricter "ever since birth" definition, infants who were predominantly and mixed breastfed before 6 wk of age had, respectively, 1.64 (95% CI: 0.88, 3.05) and 1.91 (95% CI: 0.99, 3.67) times more LRTI-specific clinic visits and 2.17 (95% CI: 0.70, 6.76) and 1.73 (95% CI: 0.52, 5.75) times more diarrhea-specific clinic visits during the subsequent 43–91-d interval than did infants who were exclusively breastfed during the first 42 d of life (Table 4). These differences were not as apparent when the "previous 7 d" definition was used. During the 92–182-d interval, infants who had been predominantly and mixed breastfed during the first 91 d of life made ≈20% more all-cause sick clinic visits based on either feeding group definition, although these differences were statistically significant only on the basis of the "previous 7 d" definition. This higher rate of total visits was primarily driven by diarrhea-specific visits, which were significantly higher based on the "ever since birth" definition (≈8 times) and based on the "previous 7 d" definition (≈2 times) in both the PBF and MBF infants than in the EBF infants. There was no significant difference in LRTI-specific visits between feeding groups based on either feeding definition during the 92–182-d period. A sensitivity analysis, excluding all sick clinic visits that occurred within 7 d from the breastfeeding assessment date, showed that the effect size remained similar (data not shown) and adjustment for vulnerability of the child in the past also did not modify the effect size (data not shown).

DISCUSSION

In this study, early EBF was associated with significantly fewer sick clinic visits than was early PBF or MBF among non-HIV–exposed infants. The magnitude and significance of this association was particularly strong for diarrhea-specific visits.

Several methodologic features of this analysis strengthened our conclusions. First, comparison of subsequent morbidity according to previous breastfeeding practice minimized the effect of reverse causality. Second, we excluded infants who had been fed infant formula soon after birth most likely as a part of neonatal intensive care; inclusion of these infants would likely have overestimated the adverse consequences of MBF. Third, we conducted a sensitivity analysis that excluded sick clinic visits that occurred within 7 d of breastfeeding practice assessment and another analysis that adjusted for infant vulnerability in the past; it was confirmed that the effect size was similar. Fourth, we used 2 breastfeeding definitions that have different strengths and limitations and often correlate poorly (17). Finally, we controlled for the effect of secular trends.

Two mechanisms have been described to explain the protective effects of EBF compared with those of PBF or MBF. First, non–breast milk fluids and foods are often contaminated with pathogens when prepared under unhygienic conditions (18–20), which results in diarrhea and subsequent malnutrition. Second, because breast milk contains a wide array of antiinfective properties, reduced breast milk volume would result in reduced intakes of these factors and leave the infant more vulnerable. Data are quite consistent, demonstrating a displacement effect by formula, animal milk, and solid food (21, 22). However, recent studies indicate that PBF liquids do not result in lower breast milk intake (23, 24). In our study, the deleterious effects of PBF and MBF were of remarkably similar magnitude in all the analyses that we conducted. This finding suggests that the excess diarrhea associated with PBF and MBF was more likely to be due to accompanying pathogens than to a reduced intake of breast milk and its antiinfective factors. Previous studies observed higher morbidity and mortality rates due to diarrhea among MBF infants than among EBF infants but similar rates among PBF infants and EBF infants in Bangladesh (5), in Peru (25), and in a pooled analysis from India, Ghana, and Peru (11). This may be because the food items that shift infants from EBF to PBF and MBF classifications differ depending on the setting. Future studies should examine the particular foods that contribute to PBF and MBF in their communities, as we did in this analysis.

The association between breastfeeding exclusivity and sick clinic visits was more modest for LRTI-specific visits than for visits for diarrhea, reaching marginal significance only among MBF infants during the 43–91-d interval when the "ever since birth" definition was used (IRR: 1.91; 95% CI: 0.99, 3.67; $P = 0.052$). No adverse effect on LRTI associated with either PBF or MBF was observed in the pooled analysis (11), and promotion of EBF was not associated with a reduction in morbidity due to respiratory infections (26). Two previous studies reported a protective effect of EBF compared with MBF on respiratory infections; however, in one of these studies, nonbreastfed infants were included in the MBF group (5), and, in the other study, the periods of morbidity observation and breastfeeding status overlapped completely, so reverse causality may have occurred (25). A possible explanation for the weaker association between EBF and respiratory infections may be that it is mediated by a chain of events where diarrhea is caused by PBF or MBF, which, in turn, gives rise to subsequent malnutrition and susceptibility to respiratory infections.

Several limitations of this study deserve mention. First, because breastfeeding practice was not randomized, the possibility of residual confounding exists because of unmeasured factors. Second, we did not collect data on the frequency or quantity of non–breast milk foods consumed by infants, nor did we collect data on the age at introduction of non–breast milk foods. Therefore, we were unable to estimate whether there is a threshold of nonexclusivity or infant age associated with increased morbidity. Third, our analysis used the breastfeeding practice that preceded the period of observation of outcome as the exposure variable; however, this may have resulted in misclassification of exposure, because the breastfeeding practice preceding the observation period may not necessarily reflect the breastfeeding practice during the period of observation. Finally, most of the clinic visits were conducted in public sector clinics, so we were reliant on nonresearch staff and the mothers themselves for morbidity reports.

The recommendation of exclusive breastfeeding from birth to 6 mo has emerged from the integration of the benefits of exclusive breastfeeding on a wide range of health outcomes, including infant growth, late return of maternal menstruation, and postpartum weight loss (27). Although our study was not intended to assess the optimal duration of exclusive breastfeeding, our study supports and strengthens current recommendations to promote early EBF among all infants. Cluster randomized controlled studies of promotion of exclusive breastfeeding in Mexico (28), India (29), and Belarus (26) found reductions in diarrhea in the intervention group.

Exclusive breastfeeding among HIV-negative Zimbabwean mothers is associated with significant health benefits to their

infants. These findings, together with our earlier observation of reduced postnatal HIV transmission among HIV-exposed infants whose mothers practiced early EBF (9), indicate that universal promotion of EBF is very likely to improve infant health. Our findings are particularly notable because our study was conducted among relatively well-educated urban women, the vast majority of whom had access to tap water, toilet facilities, and electricity.

Members of the ZVITAMBO Study Group, in addition to the named authors are as follows: Henry Chidawanyika, John Hargrove, Agnes I Mahomva, Florence Majo, Lucie C Malaba, Michael T Mbizvo, Faith Mzengeza, Kusum J Nathoo, Mary Ndhlovu, Ellen Piwoz, Lidia Propper, Phillipa Rambanepasi, Andrea J Ruff, Naume Tavengwa, Brian J Ward, Lynn S Zijenah, Clare D Zunguza, and Partson Zvandasara.

The authors' responsibilities were as follows—AK: analyzed the data and wrote the manuscript; JHH: designed the study and provided advice; RN and LHM: provided statistical advice; KM: conducted laboratory work; and PI and REB: provided advice and consultation. None of the authors had a commercial interest or any other association that might have posed a conflict of interest with the results presented in this article.

REFERENCES

1. Jones G, Steketee RW, Black RE, Bhutta ZA, Morris SS; Bellagio Child Survival Study Group. How many child deaths can we prevent this year? Lancet 2003;362:65–71.
2. Black RE, Allen LH, Bhutta ZA, et al. Maternal and child under nutrition: global and regional exposures and health consequences. Lancet 2008;371:243–60.
3. World Health Organization. The optimal duration of exclusive breastfeeding: a systematic review. Geneva, Switzerland: World Health Organization, 2001.
4. Ashraf RN, Jalil F, Zaman S, et al. Breast feeding and protection against neonatal sepsis in a high risk population. Arch Dis Child 1991;66:488–90.
5. Arifeen S, Black RE, Antelman G, Baqui A, Caulfield L, Becker S. Exclusive breastfeeding reduces acute respiratory infection and diarrhea deaths among infants in Dhaka slums. Pediatrics 2001;108:E67.
6. Bhutta ZA, Yusuf K. Early-onset neonatal sepsis in Pakistan: a case control study of risk factors in a birth cohort. Am J Perinatol 1997;14:577–81.
7. UNICEF. Progress for children: report card on nutrition. Available from: http://www.unicef.org/progressforchildren/2006n4/index_breastfeeding.html#note13 (cited 1 October 2007).
8. Coovadia HM, Rollins NC, Bland RM, et al. Mother-to-child transmission of HIV-1 infection during exclusive breastfeeding in the first 6 months of life: an intervention cohort study. Lancet 2007;369:1107–16.
9. Iliff PJ, Piwoz EG, Tavengwa NV, et al. Early exclusive breastfeeding reduces the risk of postnatal HIV-1 transmission and increases HIV-free survival. AIDS 2005;19:699–708.
10. Coutsoudis A, Kubendran P, Spooner E, Kuhn L, Coovadia HM, for the South African Vitamin A Study Group. Influence of infant feeding patterns on early mother-to-child transmission of HIV-1 in Durban, South Africa: a prospective cohort study. Lancet 1999;354:471–6.
11. Bahl R, Frost C, Kirkwood BR, et al. Infant feeding patterns and risks of death and hospitalization in the first half of infancy: multicentre cohort study. Bull World Health Organ 2005;83:418–26.
12. Humphrey JH, Iliff PJ, Marinda ET, et al. Effects of a single large dose of vitamin A, given during the postpartum period to HIV-positive women and their infants, on child HIV infection, HIV-free survival, and mortality. J Infect Dis 2006;193:860–71.
13. Capurro H, Konichezky S, Fonseca D, Caldeyro-Barcia R. A simplified method for diagnosis of gestational age in the newborn infant. J Pediatr 1978;93:120–2.
14. Gibson RS. Principles of nutritional assessment. New York, NY: Oxford, 1990.
15. World Health Organization. Indicators for assessing breastfeeding practices. Report of an informal meeting, 11–21 June 1991. Geneva, Switzerland: WHO, Division of Diarrhoeal and Acute Respiratory Disease Control, 1991. (WHO/CDD/SER/91.14.)
16. Piwoz EG, Iliff PJ, Tavengwa N, et al. An education and counseling program for preventing breast-feeding-associated HIV transmission in Zimbabwe: design and impact on maternal knowledge and behavior. J Nutr 2005;135:950–5.
17. Aarts C, Kylberg E, Hornell A, Hofvander Y, Gebre-Medhin M, Greiner T. How exclusive is exclusive breastfeeding? A comparison of data since birth with current status data. Int J Epidemiol 2000;29:1041–6.
18. Afifi ZE, Nasser SS, Shalaby S, Atlam SA. Contamination of weaning foods: organisms, channels, and sequelae. J Trop Pediatr 1998;44:335–7.
19. Black RE, Brown K, Becker S, Alim A, Merson M. Contamination of weaning foods and transmission of enterotoxigenic *Escherichia coli* diarrhea in children in rural Bangladesh. Trans R Soc Trop Med Hyg 1982;76:259–64.
20. Motarjemi Y, Kaferstein F, Moy G, Quevedo F. Contaminated weaning food: a major risk factor for diarrhoea and associated malnutrition. Bull World Health Organ 1993;71:79–92.
21. Drewett R, Amatayakul K, Wongsawasdii L, et al. Nursing frequency and the energy intake from breast milk and supplementary food in a rural Thai population: a longitudinal study. Eur J Clin Nutr 1993;47:880–91.
22. Cohen RJ, Brown KH, Canahuati J, Rivera LL, Dewey KG. Effects of age of introduction of complementary foods on infant breast milk intake, total energy intake, and growth: a randomised intervention study in Honduras. Lancet 1994;344:288–93.
23. Haisma H, Coward WA, Albernaz E, et al. Breast milk and energy intake in exclusively, predominantly, and partially breast-fed infants. Eur J Clin Nutr 2003;57:1633–42.
24. Thiombiano-Coulibaly N, Rocquelin G, Eymard-Duvernay S, Zougmore ON, Traore SA. Effects of early extra fluid and food intake on breast milk consumption and infant nutritional status at 5 months of age in an urban and a rural area of Burkina Faso. Eur J Clin Nutr 2004;58:80–9.
25. Brown KH, Black R, Lopez de Romana G, Creed de Kanashiro H. Infant-feeding practices and their relationship with diarrheal and other diseases in Huascar (Lima), Peru. Pediatrics 1989;83:31–40.
26. Kramer MS, Chalmers B, Hodnett ED, et al. Promotion of Breastfeeding Intervention Trial (PROBIT): a randomized trial in the Republic of Belarus. JAMA 2001;285:413–20.
27. Kramer MS, Kakuma R. The optimal duration of exclusive breastfeeding: a systematic review. Adv Exp Med Biol 2004;554:63–77.
28. Morrow AL, Guerrero ML, Shults J, et al. Efficacy of home-based peer counselling to promote exclusive breastfeeding: a randomised controlled trial. Lancet 1999;353:1226–31.
29. Bhandari N, Bahl R, Mazumdar S, Martines J, Black RE, Bhan MK. Effect of community-based promotion of exclusive breastfeeding on diarrhoeal illness and growth: a cluster randomised controlled trial. Lancet 2003;361:1418–23.

AIDS RESEARCH AND HUMAN RETROVIRUSES
Volume 26, Number 5, 2010

DOI: 10.1089/aid.2009.0138

Postpartum Plasma CD4 Change in HIV-Positive Women: Implications for Timing of HAART Initiation

Ai Koyanagi,[1] Andrea J. Ruff,[1] Lawrence H. Moulton,[1] Robert Ntozini,[2] Kuda Mutasa,[2] Peter Iliff,[2] Jean H. Humphrey,[1,2] and the ZVITAMBO Study Group

Abstract

CD4 counts increase during the postpartum period and may not correctly identify HAART-eligible HIV-positive women. HAART eligibility when defined by two CD4 cutoffs (<200 and <350 cells/μl) measured at two time points (within 96 h of delivery and 6 weeks) in postpartum HIV-positive women was compared. Among HIV-positive women who had CD4 at delivery and 6 weeks ($n = 423$), time to Stage 3 or 4 opportunistic infection or death was compared using Cox regression between three groups of women: (1) CD4 <200 cells/μl at delivery and 6 weeks, (2) CD4 <200 cells/μl at delivery but ≥200 cells/μl at 6 weeks, and (3) CD4 ≥200 cells/μl at delivery and at 6 weeks. The analysis was repeated using the CD4 <350 cells/μl cut-off. CD4 counts increased by a median (IQR) of 70 (1–178) cells/μl between delivery and 6 weeks and decreased thereafter to approximately delivery levels at 12 months. Only 60% and 61% who had CD4 <200 cells/μl and CD4 <350 cells/μl, respectively, at delivery also had those levels at 6 weeks. Among those with CD4 <350 cells/μl at both delivery and 6 weeks, the risk of death or Stage 3 or 4 disease was 5.27 (95% CI 1.85–14.96) times higher than those with CD4 <350 at delivery but ≥350 cells/μl at 6 weeks. The use of CD4 counts immediately postpartum to define HAART eligibility may lead to substantial misclassification.

Introduction

In the course of HIV infection, plasma CD4 cell counts decrease on average 75 cells/μl per year.[1] Conversely, CD4 cell counts rise in women following pregnancy, due to resolution of physiologic hemodilution in both HIV-uninfected and HIV-infected women.[2–4] The WHO guidelines currently recommend CD4 <200 cells/μl as an absolute indication of highly active antiretroviral therapy (HAART) initiation and CD4 <350 cells/μl as a benchmark to consider treatment.[5,6]

A recent publication reported that using CD4 counts at 32 weeks of gestation as HAART eligibility criteria leads to substantial misclassification of HAART eligibility when compared to CD4 values at 1 month postpartum.[7] Using WHO clinical staging and CD4 counts, 28.3% of women in this study were HAART eligible according to their baseline CD4 values whereas only 17.2% were eligible according to their postpartum CD4 values. The authors pointed out that CD4 percentage may be a more accurate indicator of immune status in pregnant and postpartum women than absolute CD4 since the former is less affected by hemodynamic changes associated with pregnancy and postpartum. Misclassification of HAART eligibility or premature initiation of HAART may lead to increased viral resistance, waste of resources, non-compliance, and unnecessary adverse events.[8–12] In this article, we describe the change in absolute CD4 cell count postpartum up to 12 months and the potential misclassification of HAART eligibility if using CD4 cell counts measured immediately postpartum.

Materials and Methods

ZVITAMBO trial

Details of the ZVITAMBO trial have been previously published.[13–15] Briefly, 14,110 mother–infant pairs were recruited within 96 h of delivery in greater Harare, Zimbabwe between November 1997 and January 2000. Mothers were eligible if they did not have a life threatening condition and had planned to stay in Harare after enrollment. Written informed consent was obtained. Baseline characteristics were collected from hospital records and a questionnaire. Follow-up was conducted at 6 weeks and 3 months and 3-monthly intervals through 12–24 months. At delivery, women were tested for HIV by an algorithm incorporating two parallel

[1]Johns Hopkins Bloomberg School of Public Health, Department of International Health, Baltimore, Maryland.
[2]ZVITAMBO Study Team, Harare, Zimbabwe.

ELISAs and Western blot. CD4 cells were counted by FACScount (Becton Dickinson). Among a randomly selected subgroup of approximately 10% of HIV-positive women, CD4 cells were also counted at 6 weeks and at 3, 6, 9, and 12 months. Weight was measured at each follow-up visit but not at delivery. At each visit, a 7-day morbidity history was elicited that included oral thrush and chronic diarrhea, and mothers were asked if they had been hospitalized or visited a clinic for treatment of an illness since their previous visit.

The causes and dates of these health care visits were determined from medical records, if available, or by maternal history. Data were available to identify the following Stage 3 opportunistic infections: chronic diarrhea, recurrent or persistent oral candidiasis (presence of at least two oral thrush episodes during follow-up with a ≥14 days interval between the two episodes), pulmonary tuberculosis (TB), and severe bacterial infections (e.g., pneumonia, meningitis, and sepsis). All severe bacterial infections were diagnosed at a clinic or hospital. Data were available to identify the following Stage 4 opportunistic infections: HIV wasting syndrome (simultaneous presence of ≥10% weight loss relative to any previous weight measurement during study and chronic diarrhea as defined in Stage 3), recurrent severe pneumonia (≥2 episodes of pneumonia during follow-up with ≥14 days interval between the two episodes), esophageal candidiasis, extrapulmonary TB, and Kaposi's sarcoma (all diagnosed during a clinic visit or hospitalization). Antiretroviral therapy (ART) was not available during the trial either as prophylaxis or treatment.

Statistical analysis

Statistical analysis was conducted using Stata Version 9.2 (StataCorp LP, Texas). Overall, 14,110 women were recruited and 4495 women were HIV positive at delivery. CD4 was counted at follow-up in a random subsample of approximately 10% of the HIV-positive women. To describe accurately the postpartum trajectory of CD4 counts in the first year, we restricted analyses that involved delivery, 6 week, and 12 month data to those with CD4 data at all those time points. Wilcoxon signed rank tests were used to test change in CD4 distribution between delivery and 6 weeks or 12 months and between 6 weeks and 12 months. McNemar's test was used to test pairwise changes in the proportion of those with CD4 <200 or <350 cells/μl at delivery, 6 weeks, and 12 months.

We also calculated the sensitivity, specificity, positive and negative predictive value, and 95% confidence intervals using information on 423 women who had CD4 measured at both delivery and 6 weeks. We considered CD4 counts at 6 weeks to be the gold standard indicator of immune status at delivery and used CD4 cut-off points of 150, 175, and 200 cells/μl and 300, 325, and 350 cells/μl at delivery to assess its correlation with, and sensitivity, specificity, and predictive values in identifying CD4 counts with cut-off points of 200 and 350 cells/μl at 6 weeks, respectively, among 423 women who had CD4 data available at both time points. Finally, Kaplan–Meier methods were also used to estimate cumulative risk of Stage 3 or 4 disease or death by CD4 count at delivery and 6 weeks.

The 423 women who had CD4 counts at delivery and 6 weeks were divided into three groups: (1) CD4 <200 cells/μl at delivery and 6 weeks, (2) CD4 <200 cells/μl at delivery and CD4 ≥200 cells/μl at 6 weeks, and (3) CD4 ≥200 cells/μl at both delivery and 6 weeks. The censoring date was the last date of follow-up, the first date in which Stage 3 or 4 disease was identified, or the date of death, whichever occurred earlier. We assessed the time to Stage 3 or 4 disease or death between 6 weeks and 24 months using the Cox proportional hazards model and present the 95% confidence intervals. The analysis was repeated by using the CD4 <350 cells/μl cut-off point.

Ethical approval

Ethical approval was granted from the Medical Research Council of Zimbabwe, Medicines Control Authority of Zimbabwe, the Committee on Human Research of the Johns Hopkins University Bloomberg School of Public Health, and the Ethics Committee of the Research Institute of the McGill University Health Center.

Results

A total of 226 women had complete CD4 count information at delivery, 6 weeks, and 12 months. The median (IQR) number of days between delivery and blood sampling at 6 weeks and 12 months was 43 (42–46) days and at 12 months was 365 (365–368). As illustrated in Fig. 1, the median CD4 cell count increased between delivery and 6 weeks but gradually decreased by about the same magnitude between 6 weeks and 12 months such that the counts at delivery and 12 months did not significantly differ. Among the 27 women who had CD4 <200 cells/μl at delivery, 12 (44.4%) had CD4 ≥200 cells/μl at 6 weeks and among these 12 women, 7 (58.3%) still had CD4 ≥200 cells/μl at 12 months. Among the 78 women who had CD4 <350 cells/μl at delivery, 34 (43.6%) had CD4 ≥350 cells/μl at 6 weeks and among these 34 women, 21 (61.8%) still had CD4 ≥350 cells/μl at 12 months (Table 1).

Between delivery and 6 weeks postpartum, CD4 cell count increased in 75.5% (170/226) of the women (Fig. 2). Between delivery and 12 months partum, CD4 cell count increased in 46% (104/226) and decreased in 54% (122/226) of the women. In contrast, 179 (79.2%) women had lower CD4 counts at 12 months than at 6 weeks. At 6 weeks, a smaller proportion of women had CD4 values of <200 cells/μl (6.6% vs. 12.0%; $p = 0.0005$) or <350 cells/μl (22.1% vs. 34.5%; $p = 0.0000$) compared to delivery. However, at 12 months, the proportions of women with CD4 <200 cells/μl and CD4 <350 cells/μl were not different compared to delivery (12.0% vs. 12.0%; $p = 1.0000$ and 35.0 vs. 34.5%; $p = 1.0000$), respectively. Based on the finding that CD4 counts on average increase between delivery and 6 weeks, we investigated the utility of using lower CD4 cut-off points at delivery than the conventional cut-off points of 200 and 350 cells/μl to identify HAART eligible women. Among those who had CD4 counts <150 cells/μl at delivery, 83% of the women also had CD4 counts <200 cells/μl at 6 weeks but only 60% of the women who had CD4 <200 cells/μl at delivery also had CD4 <200 cells/μl at 6 weeks (Table 2, see PPV values). However, CD4 <150 cells/μl at delivery identified only 79% of women who had CD4 <200 cells/μl at 6 weeks whereas CD4 <200 cells/μl at delivery correctly identified 91% (Table 2, see Sensitivity values). The positive predictive values of all delivery cut-off points in identifying women with CD4 <350 cells/μl at 6 weeks were low (61–68%).

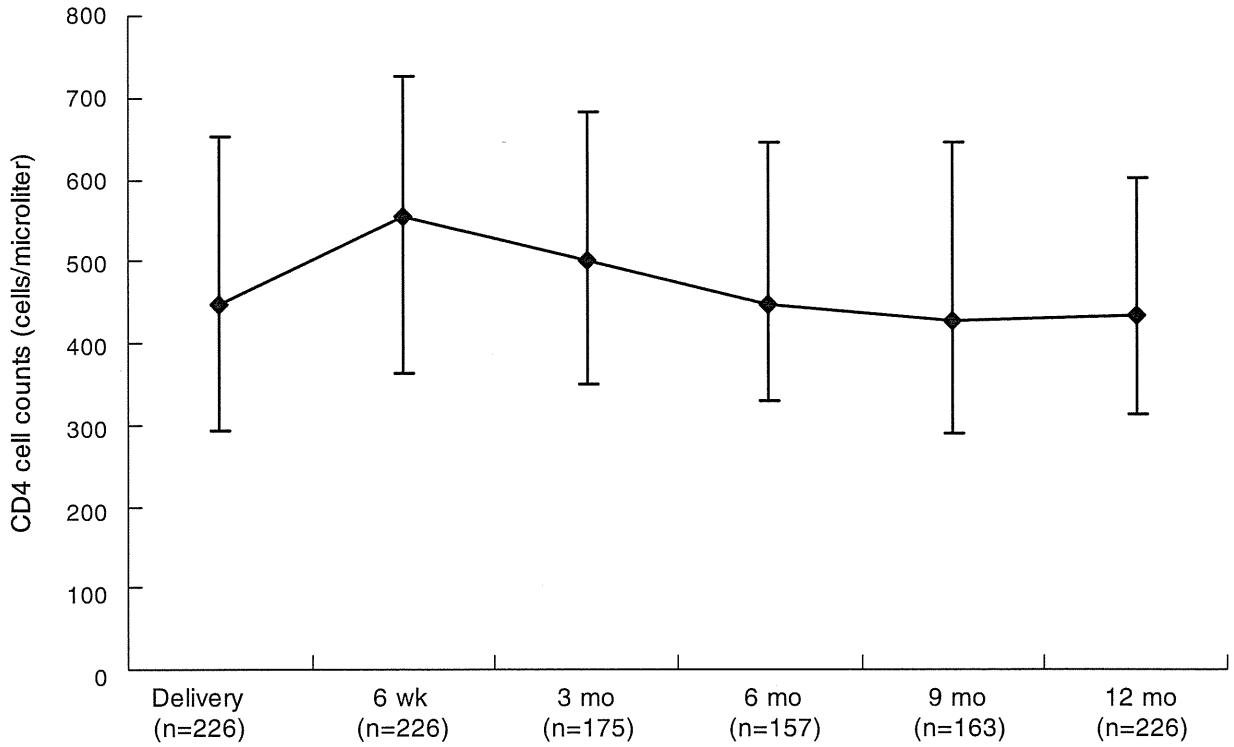


FIG. 1. Median and interquartile ranges of CD4 cell counts at delivery, 6 weeks, and 3, 6, 9, and 12 months postpartum. Only those who had information on CD4 counts at delivery, 6 weeks, and 12 months are included.

As shown in Fig. 3, women with CD4 counts that were <200 cells/μl at delivery and remained so at 6 weeks had the highest risk of death or Stage 3 or 4 opportunistic infection (HR 5.57; 95% CI 3.11–9.98; $p = 0.000$) compared to those who had CD4 ≥200 cells/μl at both time points. Compared to women who started with CD4 counts <200 at delivery and then had CD4 counts ≥200 cells/μl at 6 weeks, the risk of death or Stage 3 or 4 disease in women with persistently low CD4 counts at both delivery and 6 weeks was higher but not statistically significant (HR 2.84; 95% CI 0.96–8.45; $p = 0.060$). Having CD4 counts <350 cells/μl at both time points was also associated with a significantly higher risk (HR 5.27; 95% CI 1.85–14.96; $p = 0.002$) compared to those with CD4 <350 cells/μl at delivery and CD4 ≥350 cells/μl at 6 weeks.

Discussion

We have demonstrated that the majority of HIV-infected women in this cohort had higher CD4 counts at 6 weeks postpartum than they did within 4 days of delivery. Forty percent (26/65) of women with CD4 counts <200 cells/μl shortly after delivery had counts of >200 cells/μl at 6 weeks postpartum indicating that using CD4 count immediately postpartum as HAART eligibility criteria could lead to substantial misclassification of HAART eligibility. The magnitude of CD4 increase observed between delivery and postpartum was similar to other studies conducted in Africa.[2–4,7] In our study, the proportion with CD4 counts <200 and <350 cells/μl at delivery was 15.4% and 39.2%, respectively, and this decreased to 10.2% and 26.7%, respectively, by 6 weeks.

TABLE 1. POSTPARTUM CD4 CHANGE IN HIV-POSITIVE WOMEN[a]

N = 226 (total)	*Delivery*	*6 weeks*	*12 months*
Median (IQR[b]) (cells/μl)	448 (293–651)	553 (362–727)	432 (312–602)
Median difference from delivery (IQR) (cells/μl)		70 (1–178)[c]	−14 (−125–77)
Median difference from 6 weeks (IQR) (cells/μl)			−87 [−187–(−14)][c]

[a]The 226 women who had CD4 count available at delivery, 6 weeks, and 12 months are included.
[b]IQR, interquartile range.
[c]Significantly different from 0 by sign rank test ($p < 0.05$).

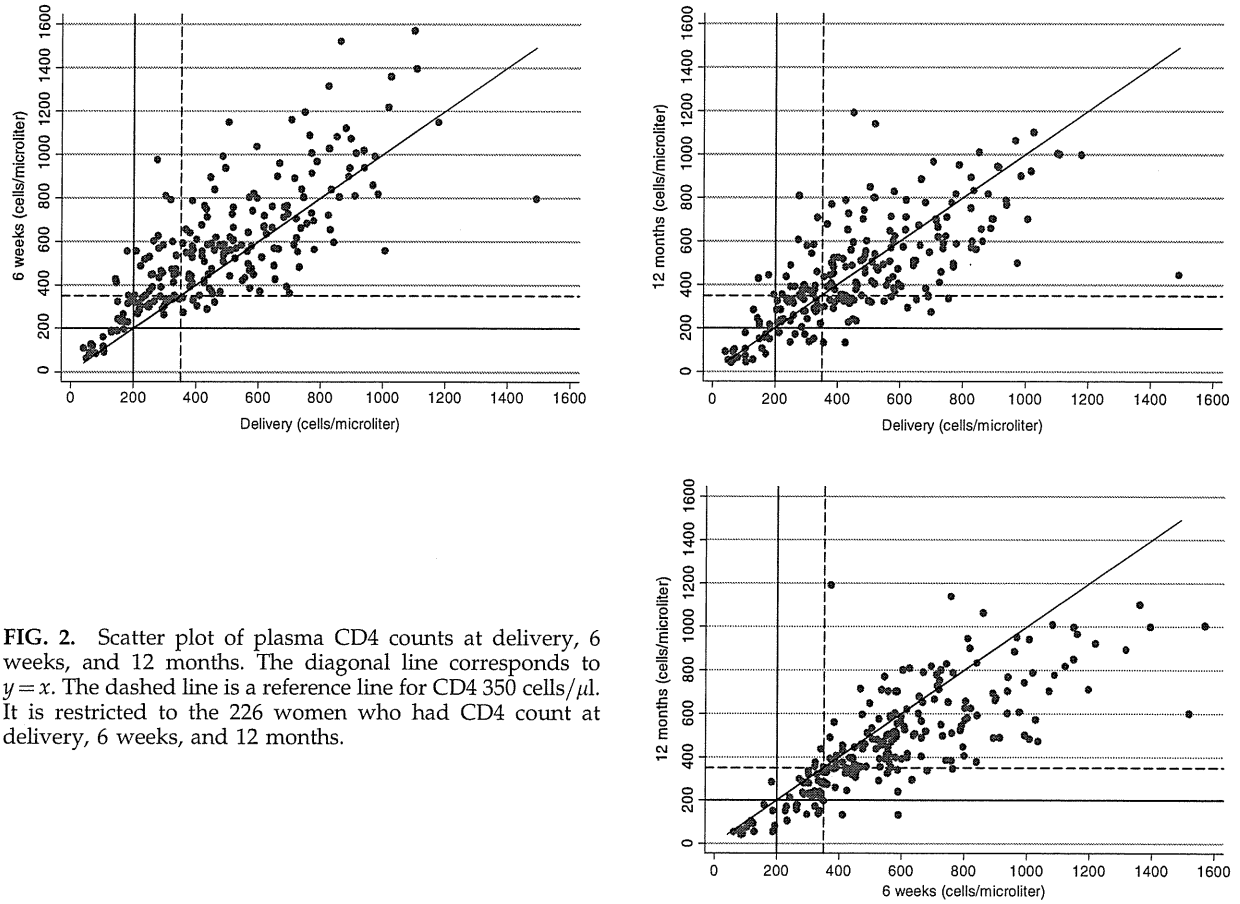


FIG. 2. Scatter plot of plasma CD4 counts at delivery, 6 weeks, and 12 months. The diagonal line corresponds to $y = x$. The dashed line is a reference line for CD4 350 cells/μl. It is restricted to the 226 women who had CD4 count at delivery, 6 weeks, and 12 months.

A study conducted in Ivory Coast with similar sample size also found similar results where 17.8% and 48.3% of the women had CD4 <200 and <350 cells/μl at 32 weeks gestation but at 1 month postpartum the proportion decreased to 9.5% and 28.9%, respectively.[7] Our study showed that CD4 200 and 350 cells/μl cut-off points at delivery correctly identified only 60% (95% CI 47–72) and 61% (95% CI 54–69) of women who had CD4 counts less than the same cut-off at 6 weeks. Of note, our study provides additional information regarding the association between CD4 counts obtained early during the postpartum period and risk of progression of HIV. We demonstrated that those who were HAART eligible based on CD4 counts at delivery but no longer so based on values at 6 weeks had a lower risk of Stage 3 or 4 opportunistic infections or death compared to those who were persistently HAART eligible at both time points.

The initiation of HAART is based on clinical, immunologic, and virologic indications, which in turn are associated with risk of progression of HIV. The consequences of initiation of HAART in those who are not in need of it for their health include potential unnecessary adverse events and depletion of scarce resources in settings with limited treatment options.[10,11,16] In our study, the proportion of women who were HAART eligible by the CD4 200 and 350 cells/μl cut-off points was almost identical at delivery and 12 months, meaning that values at delivery are a reflection of CD4 counts after 1 year. This finding was not surprising since it is known that CD4 counts decrease on average 75 cells/μl per year[1] and the median increase in CD4 counts between delivery and 6 weeks that we observed was 70 cells/μl. Also, more than half of the women who were HAART eligible at delivery but no longer so at 6 weeks were not HAART eligible even by 12 months.

The following may be proposed as possible solutions to correctly identify women during the postpartum period who would be in need of long-term HAART. First, although predictive values are known to be influenced by prevalence, we investigated the possibility that use of lower CD4 values than the conventional CD4 cut-off points for HAART eligibility at delivery might lead to less misclassification or higher positive predictive values. Compared to the conventional cut-off point of CD4 200 cells/μl, use of 175 cells/μl increased the positive predictive value from 60% to 72% while maintaining the same sensitivity of 91%. However, compared to the 350 CD4 cell/μl cut-off point, the positive predictive value increased only from 61% to 68% by using a 300 CD4 cell/μl cut-off point at the expense of lowering the sensitivity from 90% to 78%. Because this low level of sensitivity is not acceptable, we could not identify any cut-off point that might be of any benefit for the CD4 350 cells/μl cut-off point. Since women generally return to the clinic at 6 weeks for a postnatal health care visit for themselves and a vaccination for their infant, this would be a convenient time to check their CD4 count and then make

TABLE 2. NUMBER AND PERCENTAGE OF WOMEN BY CD4 CUTOFFS 200 AND 350 CELLS/μL AND TIME POSTPARTUM[a]

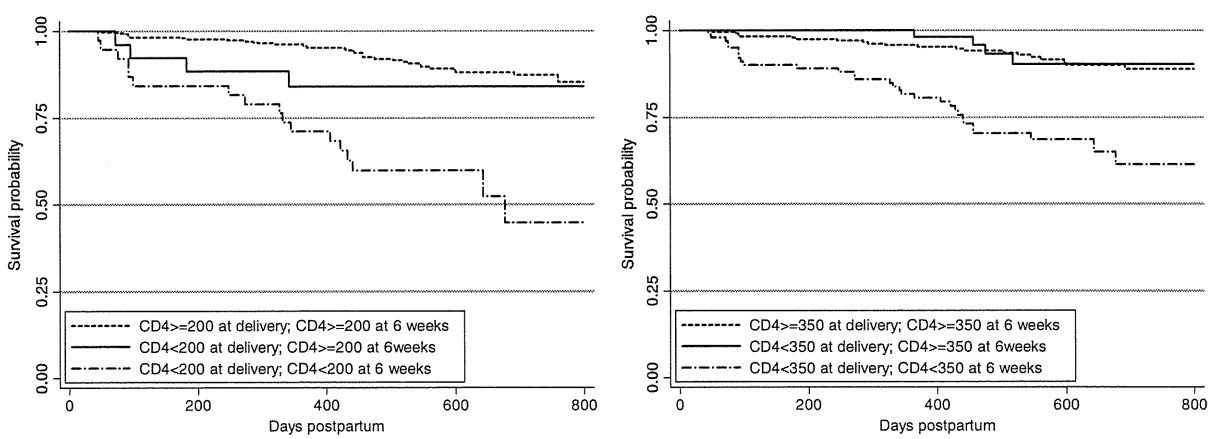
CD4 at delivery (cells/μl)	*CD4 at 6 weeks (n) (cells/μl)*		*Sensitivity (95% CI)*	*Specificity (95% CI)*	*PPV[b] (95% CI)*	*NPV[b] (95% CI)*
	<200	≥200				
<200	39	26	91 (78–97)	93 (90–95)	60 (47–72)	99 (97–100)
≥200	4	354				
<175	39	15	91 (78–97)	96 (94–98)	72 (58–84)	99 (97–100)
≥175	4	365				
<150	34	7	79 (64–90)	98 (96–99)	83 (68–93)	98 (96–99)
≥150	9	373				
	<350	≥350				
<350	102	64	90 (83–95)	79 (74–84)	61 (54–69)	96 (92–98)
≥350	11	246				
<325	97	53	86 (78–92)	83 (78–87)	65 (56–72)	94 (91–97)
≥325	16	257				
<300	88	41	78 (69–85)	87 (82–90)	68 (59–76)	91 (88–94)
≥300	25	269				

[a]423 women who had CD4 cell counts available at delivery and 6 weeks are included.
[b]PPV, positive predictive value; NPV, negative predictive value.

a treatment decision. Finally, it has been reported that CD4 percentage rather than absolute CD4 value is a better indicator of immune status as it remains stable even in the presence of hemodilution.[2,7] CD4 percentage is used to decide HAART eligibility in children[17] and further studies on the utility of this indicator in pregnant and postpartum women in deciding HAART eligibility are warranted.

Acknowledgments

The ZVITAMBO project was supported by the Canadian International Development Agency (CIDA) (R/C Project 690/M3688), United States Agency for International Development (USAID) (cooperative agreement number HRN-A-00-97-00015-00 between Johns Hopkins University and the Office



CD4 at delivery (cells/μ)	CD4 at 6 weeks (cells/μl)	Median (IQR) CD4 at delivery (cells/μl)	Median (IQR) CD4 at 6 weeks (cells/μl)	n	No. of OI[§]	No. of death	HR	95%CI	p value
≥200	≥200	477 (331-644)	558 (422-731)	354	30	4	1.00		
<200	≥200	168 (149-185)	275 (236-352)	26	4	0	1.96	(0.69-5.53)	0.203
<200	<200	104 (70-130)	118 (104-160)	39	15	2	5.57	(3.11-9.98)	<0.001
≥350	≥350	567 (459-721)	615 (524-812)	246	18	2	1.00		
<350	≥350	281 (243-317)	453 (400-568)	64	3	1	0.84	(0.29-2.45)	0.744
<350	<350	177 (110-251)	235 (129-299)	102	27	3	4.40	(2.50-7.79)	<0.001

FIG. 3. Kaplan–Meier survival curves of time to first Stage 3 or 4 opportunistic infection or death after 42 days postpartum by CD4 count at delivery and at 6 weeks. The 423 women who had CD4 count available at delivery and 6 weeks are included. [§]Opportunistic infections. Refer to the text for definitions.