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## Sex differences in the effects of maternal vitamin supplements on mortality and morbidity among children born to HIV-infected women in Tanzania

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### Abstract

We examined whether there are sex differences in the effect of vitamin supplements on birth outcomes, mortality, and morbidity by two years of age among children born to HIV-infected women in Tanzania. A randomized placebo-controlled trial was conducted among 959 mother-infant pairs. HIV-infected pregnant women were randomly assigned to receive a daily oral dose of one of four regimens: multivitamins (vitamins B-complex, C, and E), vitamin A plus  $\beta$ -carotene, multivitamins including vitamin A plus  $\beta$ -carotene, or placebo. Supplements were administered during pregnancy and continued after delivery. The beneficial effect of multivitamins on decreasing the risk of low birth weight was stronger among girls (RR = 0.39, 95% CI 0.22 – 0.67) compared to boys (RR = 0.81, 95% CI 0.44 – 1.49; *p* for interaction = 0.08). Maternal multivitamin supplements resulted in 32% reduction in mortality among girls (RR = 0.68, 95% CI 0.47 – 0.97), whereas no effect was found among boys (RR = 1.20, 95% CI 0.80 – 1.78; *p* for interaction = 0.04). Multivitamins had beneficial effects on the overall risks of diarrhea that did not differ by sex. Vitamin A plus  $\beta$ -carotene alone increased the risk of HIV transmission, but had no effect on mortality, and we found no sex differences in these effects. Sex differential effects of multivitamins on mortality may be due to sex related differences in the immunological or genetic

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KK conducted the data analyses, interpreted the results, and wrote the initial draft of the manuscript. GM and KM participated in the study implementation and supervision in the field. EV, RJB, and EH participated in the data analyses and the data interpretation. WWF is the principal investigator and contributed to the study design, implemented the study, analyzed and interpreted the data. None of the authors had any conflicts of interest.

factors. More research is warranted to examine the effect of vitamins by sex and better understand biological mechanisms mediating such effects.

## Keywords

Vitamin A; multivitamins; sex; child mortality; HIV

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## Introduction

Vitamin A supplementation in children aged 6 months to 5 years has been shown to reduce mortality by 24 to 30%<sup>(1-3)</sup>. However, benefits for supplementing young infants less than 6 months of age have been inconclusive<sup>(4)</sup>; Benn and colleagues speculated that the lack of beneficial effects could be due to differences in the effect of supplements by sex<sup>(5)</sup>. Three previous trials found that neonatal vitamin A supplementation may have a beneficial effect on mortality in boys but no effect in girls<sup>(6-8)</sup>. A recent study found that sex differences in the effects of vitamin A on mortality depends on the different dosages of vitamin A, and a lower dosage may be beneficial among girls<sup>(9)</sup>. Studies that examine sex differential effects of other vitamins are still scarce. The possible mechanisms are not understood, but could be due to sex-related differences in the developing immune system or the degree of micronutrient deficiencies by sex<sup>(10)</sup>. It has also been hypothesized that vitamins may enhance the effect of the nonspecific immune modulation induced by live vaccines, which may have sex differential survival effects<sup>(11)</sup>.

Children born to HIV-infected women are at high risk of mortality; however, no studies have examined sex differential effects of vitamin supplement among children born to HIV-infected mothers. Nearly 2 million children were infected with HIV and 270,000 died of AIDS worldwide in 2007<sup>(12)</sup>. Almost 90% of all HIV-infected children live in sub-Saharan Africa. We have previously reported that maternal multivitamin supplements showed no effect on overall mortality among children born to HIV-infected mothers in Tanzania<sup>(13)</sup>. Vitamin A plus  $\beta$ -carotene alone increased the risk of vertical HIV transmission. In this paper, we examined whether there are sex differences in the effect of maternal supplementation of multivitamins or vitamin A plus  $\beta$ -carotene on birth outcomes, mortality, and morbidity among children born to HIV-infected mothers.

## Methods

### Study design and population

From April 1995 to July 1997, 1078 HIV-infected pregnant women were enrolled in a randomized, double-blind, placebo-controlled trial at four prenatal clinics in Dar es Salaam, Tanzania. Details of the study design have been published<sup>(13-15)</sup>. In brief, women were eligible if they were HIV-infected, pregnant between 12 and 27 weeks' gestation age at enrollment, resided in Dar es Salaam, and had consented to participate in the trial. We tested HIV-1 serostatus by enzyme-linked immunosorbent assay (ELISA; Wellcozyme, Murex Biotech Ltd, Dartford, UK) and confirmed positive results by Western blot (Bio-Rad Laboratories Ltd, Hertfordshire, UK). Eligible women were randomly assigned in a two-by-two factorial design to receive a daily oral dose of one of four regimens: (1) multivitamins (20 mg B-1, 20 mg B-2, 25 mg B-6, 100 mg niacin, 50  $\mu$ g B-12, 500 mg vitamin C, 30 mg vitamin E, and 0.8 mg folic acid); (2) vitamin A (5000 IU preformed) plus  $\beta$ -carotene (30 mg); (3) multivitamins including vitamin A plus  $\beta$ -carotene in the same doses as above; or (4) placebo. The supplements were administered from enrollment throughout the pregnancy and continued after delivery. At delivery, women in groups 1 and 3 received an additional oral dose of vitamin A (200,000 IU), whereas women in groups 2 and 4 were given a

placebo. The active treatment and placebo tablets were indistinguishable. Compliance with the study regimens was assessed by pill count. On average, 83% of participants complied over 2 years from randomization<sup>(13,14)</sup>. Women and infants received the standard prenatal and child care services in Tanzania. Daily folate and iron and weekly malaria prophylaxis were provided during pregnancy. All infants received 100,000 IU of vitamin A at 6 months of age and twice that amount every 6 months thereafter. Antiretroviral therapy was not available in this setting at the time of the study. Breastfeeding was almost universally adopted.

Information on the women's sociodemographic characteristics and obstetric history was obtained at baseline. A study physician performed a complete medical examination, and a study nurse measured the women's weight, height, and mid-upper arm circumference. The stage of HIV disease was determined based on the World Health Organization Staging System<sup>(16)</sup>. Maternal complete blood counts and absolute T cell counts were measured at baseline. CD4+ and CD8+ cell counts were measured using the FACScout system (Becton Dickinson, San Jose, CA, USA). Hemoglobin was measured using either a CBC5 Coulter counter (Coulter Corp., Miami, FL, USA) or the cyanmethemoglobin method with a colorimeter (Corning, Inc., Corning, NY, USA). Plasma levels of vitamins A and E were measured using reversed-phase high-performance liquid chromatography (HPLC)<sup>(17)</sup>. At delivery, a research midwife weighed the infants to the nearest 10 g on a standard beam balance. Gestational age was based on the date of the last menstrual period.

Mortality and morbidity surveillance of children was performed at monthly clinic visits. For women who missed their clinic visits, a home visit was made. We obtained the vital status of women and children from neighbors or relatives. At each clinic visit, mothers were asked about the number of days, if any, that the child had signs of diarrheal or respiratory disease during the previous month. Diarrhea was defined as  $\geq 3$  watery stools in the prior 24 hours. Acute diarrhea included all episodes with at least 1 day but less than 14 days of diarrhea. Acute diarrhea was classified as dysentery, which included all episodes of diarrhea with mucus or blood, or as watery diarrhea, which included all other episodes. Episodes of respiratory illness were classified in four ways: the occurrence of cough alone; cough and fever; "cough plus" (defined as cough with  $\geq 1$  of the following signs: difficulty breathing, chest retractions, and refusal to eat, drink, or breastfeed.); or cough with rapid respiratory rate on the day of visit.

For diagnosis of HIV infection in infants, blood samples were collected at birth, at 6 weeks, and every 3 months thereafter. A child was determined to be HIV-1 infected if either a peripheral blood mononuclear cell specimen tested positive using a polymerase chain reaction at any point or a plasma specimen obtained at 18 months of age or older tested positive using ELISA and was confirmed by a Western blot test. The serum concentrations of vitamin B-12 were measured in infants by a competitive magnetic separation assay on the Technico Immuno-1 analyzer<sup>(18)</sup>.

Primary outcomes of interest for this study were all-cause child mortality and HIV infection. Secondary outcomes were birth outcomes and child morbidity. Birth outcomes included stillbirths (delivery of a dead baby at or after 28 weeks' gestation), birth weight, low birth weight (birth weight less than 2500g), preterm birth (delivery before 37 weeks), and small for gestational age (birth weight below 10th percentile of weight for gestational age<sup>(19)</sup>). Morbidity included diarrhea and respiratory infections. We also examined the serum concentration of vitamin B-12 in children as an outcome.

This study was conducted according to the guidelines laid down in the Declaration of Helsinki, and the study protocol was approved by the Research and Publications Committee

of Muhimbili University of Health and Allied Sciences, the Ethical Committee of the National AIDS Control Program of the Tanzanian Ministry of Health, and the Institutional Review Board of the Harvard School of Public Health. Written informed consent was obtained from all participants.

## Data analyses

Of the 1078 HIV-infected women who were enrolled, 3 were not pregnant, 6 died before delivery, and 27 were lost to follow-up before delivery. Of the remaining 1042 women, 78 pregnancies resulted in fetal deaths (29 miscarriage and 49 stillbirths), 25 gave birth to twins, and 939 gave birth to singletons. For the analyses, we included 40 singleton stillbirths and 919 live singletons with known sex of the infant. Among those singletons, 855 had at least one specimen for HIV testing.

Intent-to-treat analyses were performed. In order to examine the effects of vitamin supplements on birth outcomes by sex, linear regression models were used for birth weight, and binomial regression models with a log link function were used for stillbirths, low birth weight, preterm birth, and small for gestational age. We used Cox proportional hazard models to investigate the effects of supplements on all-cause mortality, HIV infection, and the combined endpoints of all-cause mortality or HIV infection. We examined the risk of mortality within the strata of HIV infection as a time-varying covariate by using a counting process data structure. Likelihood ratio tests were used to assess the statistical significance of the interactions by sex and treatment effects. The Kaplan-Meier method was used to construct survival curves. The effects of supplements on diarrhea and respiratory infections were examined using generalized estimating equations with an exchangeable working covariance structure. A log link function with a binomial working covariance was used in these models. All statistical analyses were conducted using SAS version 9.1 (SAS Institute, Cary, NC).

## Results

Within strata of sex, treatment groups were comparable with respect to maternal characteristics at baseline (Table 1). The mean age of women was  $24.7 \pm 4.7$  years at baseline. The majority (80%) of pregnant women were in stage I of HIV disease and 12% had a CD4 cell count  $< 200$  cells/mm<sup>3</sup>.

### Effect of vitamin supplements on birth outcomes by sex

We have previously reported that multivitamin supplements significantly reduced the risks of fetal death and low birth weight<sup>(15)</sup>. Multivitamins decreased the risk of stillbirths non-differentially by sex ( $p$  for interaction between sex and regimen = 0.99; Table 2). The beneficial effect of multivitamins on birth weight was stronger among girls (3051g vs. 2893g; mean difference = 158 g) compared to boys (3074g vs. 3033g; mean difference = 41g;  $p$  for interaction = 0.10). Multivitamin supplementation reduced the incidence of low birth weight by 61% among girls (RR = 0.39, 95% CI 0.22 – 0.67) and 19% among boys (RR = 0.81, 95% CI 0.44 – 1.49;  $p$  for interaction = 0.08). Multivitamins significantly reduced the risk of being small for gestational age among both sexes ( $p$  for interaction = 0.66). We have previously found no effects of vitamin A plus  $\beta$ -carotene on fetal death, low birth weight, or small for gestational age<sup>(15)</sup>; and we found no differential effect on these birth outcomes by sex.

### Effect of vitamin supplements on mortality and HIV infection by sex

The effect of multivitamin supplements on child mortality among live births by 24 months of age differed by sex ( $p$  for interaction = 0.04, Table 3). Multivitamin supplements resulted

in 32% reduction in mortality among girls (RR = 0.68, 95% CI 0.47 – 0.97), whereas no evidence of the effect was found among boys (RR = 1.20, 95% CI 0.80 – 1.78; Figure 1). We observed similar effects when stillbirths were included in the analysis. The effects of multivitamin on the risk of HIV transmission or HIV free survival did not differ by sex (p for interaction = 0.73 and 0.68, respectively). We have previously found that maternal vitamin A plus  $\beta$ -carotene supplements had no effect on child mortality by 24 months, but increased the overall risk of HIV transmission<sup>(13)</sup>. We did not observe any differences between boys and girls in the effects of vitamin A plus  $\beta$ -carotene on mortality or the risk of mother-to-child transmission of HIV (p for interaction = 0.82 and 0.22, respectively; Table 4).

We also examined the effect of supplements by four treatment arms, and we did not find any interactions between the multivitamins and the vitamin A plus  $\beta$ -carotene regimens. Compared with placebo, multivitamins alone reduced mortality among girls (RR = 0.58, 95% CI 0.34 – 0.97), but not among boys (RR = 1.45, 95% CI 0.81 – 2.58; p for interaction = 0.02). Multivitamins together with vitamin A plus  $\beta$ -carotene also showed sex-differential effects (girls RR = 0.69, 95% CI 0.42 – 1.14 and boys RR = 1.33, 95% CI 0.74 – 2.39; p for interaction = 0.09). However, vitamin A plus  $\beta$ -carotene supplements showed no effect among girls and boys when compared with placebo (girls RR = 0.88, 95% CI 0.54 – 1.43 and boys RR = 1.33, 95% CI 0.74 – 2.40; p for interaction = 0.29).

### Effect of vitamin supplements on morbidity by sex

We have previously found that multivitamin supplements significantly reduced the overall risks of diarrhea by 17%, and also had beneficial effects on the risks of acute diarrhea, watery diarrhea, and dysentery<sup>(14)</sup>. These effects on diarrhea-related endpoints did not significantly differ by sex (Table 5). Multivitamin supplementation had no effect on the risks of respiratory infections and there were no interactions between the multivitamins regimen and sex. We have previously reported no effect of vitamin A plus  $\beta$ -carotene on the risks of diarrhea-associated outcomes<sup>(14)</sup>. However, boys may have benefited from vitamin A plus  $\beta$ -carotene with regards to acute diarrhea and watery diarrhea (p for interaction = 0.07 and 0.04, respectively; Table 5). We have previously found that supplementation with vitamin A plus  $\beta$ -carotene reduced the risk of cough with a rapid respiratory rate, a proxy for pneumonia, but no effects were observed for other respiratory end points<sup>(14)</sup>. We found no sex differences in the effects on respiratory infections.

### Effect of vitamin supplements on micronutrient status by sex

We have previously reported that maternal multivitamin supplements increased serum concentrations of vitamin B-12 in their infants at 6 weeks and 6 months<sup>(18)</sup>. The beneficial effects were similarly observed among girls and boys. At 6 weeks, serum vitamin B-12 was 185 pmol/l higher among girls with maternal multivitamins (441±190 pmol/l) compared to girls without maternal multivitamins (256±107 pmol/l). Similarly, serum vitamin B-12 was 166 pmol/l higher among boys with multivitamins (406±183 pmol/l) compared to boys without multivitamins (240±98 pmol/l). These beneficial effects of multivitamin supplements on the concentration of vitamin B-12 was sustained through 6 months (mean difference = 150 pmol/l among girls and 105 pmol/l among boys).

## Discussion

Supplementation of HIV-infected women during pregnancy and lactation with vitamins B-complex, C, and E at multiples of the recommended dietary allowances reduced the risk of mortality among girls by 32% but no beneficial effect was observed among boys. Multivitamins had beneficial effects on the risk of diarrhea, and the beneficial effect did not



differ by sex. Vitamin A plus  $\beta$ -carotene alone increased the risk of HIV transmission but had no effect on mortality, and we found no sex differences in these effects.

Our study did not find sex differences in the effect of vitamin A plus  $\beta$ -carotene supplements on mortality. However, we found that vitamin A plus  $\beta$ -carotene may have lowered the risks of diarrhea among boys. Previous trials demonstrated that vitamin A supplements reduced the severity of diarrheal disease, which may be due to the mediation of vitamin A on strengthening the integrity of mucosal epithelia in the gut and improve immune function<sup>(2)</sup>. Our findings conflict with previous evidence suggesting that neonatal vitamin A supplements may be associated with a reduced mortality only in boys, but it is consistent in regards to diarrhea morbidity<sup>(5)</sup>. Benn hypothesized that vitamin A may interact with routine childhood vaccinations; the beneficial effect of vitamin A may be weakened by diphtheria-pertussis-tetanus (DPT) vaccination especially for girls<sup>(11)</sup>. Most infants were vaccinated with DPT in our study, and we did not observe any differences in the effect of vitamin A plus  $\beta$ -carotene on mortality. In contrast to previous trials, our study consisted of children of HIV-infected mothers. Furthermore, mothers received daily vitamin A plus  $\beta$ -carotene supplements in our study, whereas in other studies young infants received a large dose of vitamin A at birth or at the time of routine vaccinations. Vitamin A supplementation to HIV-infected pregnant women should be avoided, because of the increased risk of HIV transmission observed in our study and Zimbabwe<sup>(13,20)</sup>.

The beneficial effect of multivitamin supplements on reducing the risk of low birth weight was stronger among girls compared to boys, and this suggests that sex differential effects of multivitamins may have started during pregnancy. Previous trials also found evidence consistent with our results. The effect of micronutrient supplements on birth weight gain was stronger among girls born to presumably HIV-negative women in Nepal (girls, 108 g vs. boys, 44 g) and birth size gain was stronger among girls in Zimbabwe (girls, 0.5 cm vs. boys, 0.02 cm)<sup>(21,22)</sup>. A recent large trial showed that multivitamins reduced the incidence of low birth weight among HIV-negative women in Tanzania<sup>(23)</sup>. In this trial, girls had gained slightly more weight at birth than boys (girls, 72 g vs. boys, 54 g; unpublished results).

Several possible mechanisms may explain sex differential effects of multivitamins on pregnancy outcomes and survival. The thymus plays a central role in the development of T lymphocytes. HIV-infected infants often have their thymus disrupted by HIV<sup>(24)</sup>. A small thymus size at birth is associated with impaired cellular immunity and malnutrition, and it is a strong risk factor for infant mortality<sup>(25,26)</sup>. Girls have smaller thymus indices than boys<sup>(25)</sup>. Girls may be more susceptible to maternal malnutrition and would have benefited more from multivitamins than boys during fetal development. In our study, low birth weight girls were more prevalent than boys in the groups that did not receive multivitamins, and multivitamins decreased the risk of low birth weight by 61% among girls and 19% among boys. Thymus size and thymic hormone activity are reduced among low birth weight infants<sup>(25,27)</sup>. Particularly among children born to HIV-infected mothers, low birth weight is a strong risk factor for infant mortality<sup>(28)</sup>. The differences in the effect of multivitamin supplements on infant girls compared to boys may be due to their varying immune systems. Infants primarily have Th-2 immune response at birth and later shift into Th1 response<sup>(29,30)</sup>. The balance between Th-1 and Th-2 responses is important, and the response biased towards Th-2 may increase risk of infection and mortality during infancy. Supplementations with vitamins B-6, C, and E alters the Th-2 immune response to a proinflammatory Th-1 cytokine-regulated response with enhanced innate immunity<sup>(31,32)</sup>. The beneficial effect of multivitamins may have been stronger among girls than boys because girls have predominantly Th-2 mediated immunity<sup>(33-35)</sup>. Evidence shows that girls tend to have excess mortality from Th-2 response-exacerbating infectious diseases, where as

boys from Th-1 response-exacerbating diseases<sup>(33)</sup>. Female sex hormones may suppress immune responses and affect susceptibility to infections<sup>(34)</sup>. Other possible explanations are hormone mediated differences in metabolism or genetic differences by sex.

Little is known about sex-related differences in the effects of vitamins on nutritional status of infants. Because our study found that multivitamin supplements had similar effects on improving vitamin B-12 status in girls and boys, sex differences in the immunological or genetic factors may have led to differential effects of multivitamins on survival.

In conclusion, maternal multivitamin (vitamins B-complex, C, and E) supplements reduced the risk of mortality among girls but not among boys born to HIV-infected women. Even though we observed sex differential effects on child mortality, multivitamins are recommended to all HIV-infected pregnant and lactating women, because of the beneficial effects among mothers on delaying HIV disease progression<sup>(36)</sup>. Antiretroviral therapy was not available when the trial was conducted. Therefore, we may not be able to generalize our results to mothers who received antiretroviral therapy. Furthermore, our findings may not be generalizable among children of HIV-negative women. More research is warranted to examine the effects of vitamin supplements by sex and elucidate biological mechanisms of sex differential effects on morbidity and mortality.

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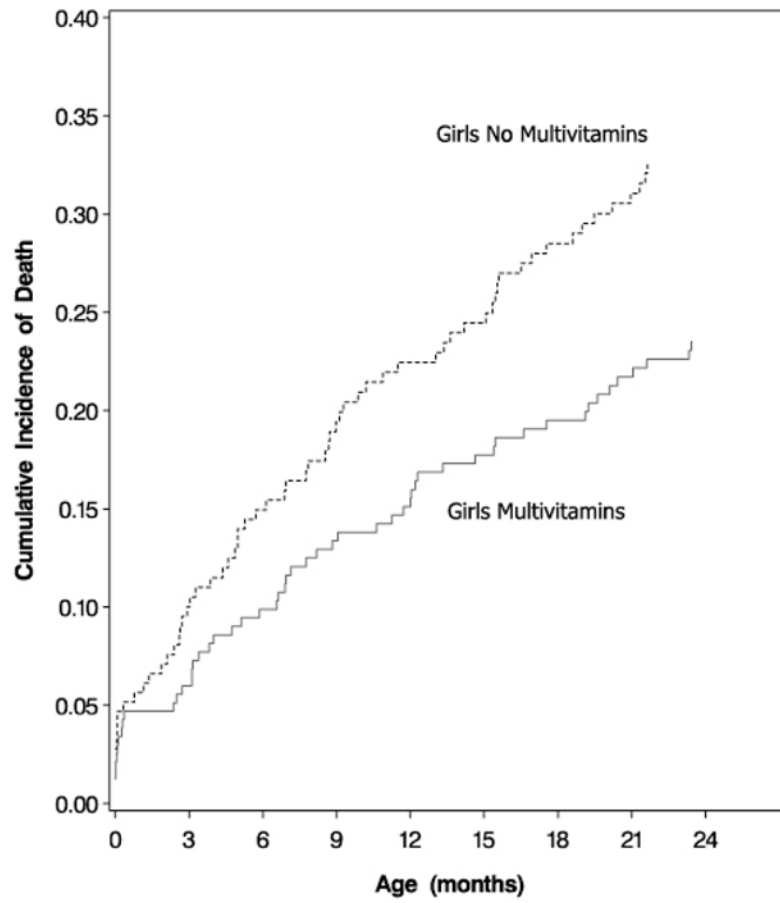
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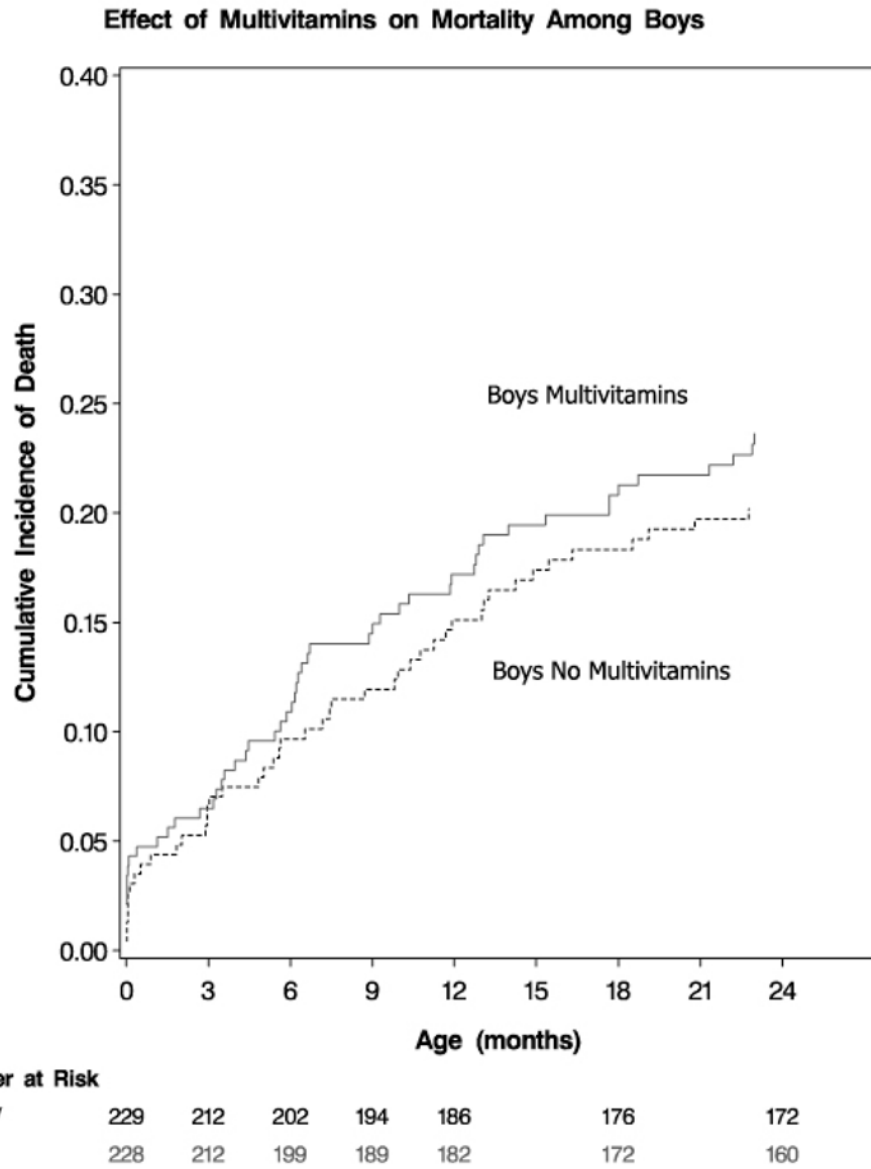
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**Effect of Multivitamins on Mortality Among Girls**



Number at Risk							
No MV	210	183	172	161	154	141	131
MV	233	217	207	199	194	182	172



**Figure 1.** Effect of maternal multivitamin supplementation on child mortality among live births according to sex

Table 1

Baseline characteristics of mothers according to treatment assignment

Characteristics	Multivitamins						Vitamin A plus $\beta$ -carotene												
	Girls			Boys			Girls			Boys									
	Yes (N = 241)	SD	Mean or %	No (N = 228)	SD	Mean or %	Yes (N = 239)	SD	Mean or %	No (N = 230)	SD	Mean or %	Yes (N = 249)	SD	Mean or %	No (N = 241)	SD	Mean or %	
Woman's age (years)	24.9	4.7	24.5	5.0	24.4	4.5	24.7	4.8	24.5	4.6	25.0	5.0	24.9	4.9	24.2	4.5	24.2	4.5	
Education (%)																			
None or adult	7.1		10.1		8.7		5.6		8.8		8.3		5.2		9.1		5.0		
1 – 4 years	5.8		4.4		4.6		5.6		3.4		7.0		5.2		5.0		5.0		
5 – 8 years	74.3		75.0		75.1		81.5		74.1		75.2		79.9		76.8		76.8		
> 8 years	12.9		10.5		11.6		7.2		13.8		9.6		9.6		9.1		9.1		
Nulliparous (%)	32.5		36.7		35.3		31.8		37.8		31.1		30.0		37.3		37.3		
WHO HIV disease stage (%)																			
I	80.0		82.5		82.2		76.3		79.5		83.0		78.3		80.1		80.1		
II and III	20.0		17.5		17.8		23.7		20.5		17.0		21.7		19.9		19.9		
CD4 cell count (cells/mm <sup>3</sup> )	419	192	410	202	425	203	425	212	398	179	432	212	423	214	428	201	428	201	
CD8 cell count (cells/mm <sup>3</sup> )	730	307	771	330	754	332	723	326	778	322	721	313	715	304	762	353	762	353	
Hemoglobin (g/dL)	9.3	1.7	9.5	1.7	9.4	1.6	9.6	1.8	9.4	1.7	9.4	1.7	9.4	1.7	9.6	1.7	9.6	1.7	
Plasma vitamin A ( $\mu$ g/dL)	24.3	8.8	24.5	10.4	24.5	9.5	25.7	10.6	23.5	8.9	25.2	10.3	25.2	9.1	24.9	10.9	24.9	10.9	
Plasma vitamin E ( $\mu$ mol/dL)	9.8	3.4	9.9	3.1	10.0	2.5	9.5	2.9	10.2	3.2	9.5	3.2	9.7	2.6	9.9	2.8	9.9	2.8	
Midupper arm circumference (cm)	25.9	2.9	25.5	3.1	25.5	2.8	25.8	2.9	25.6	2.9	25.7	3.0	25.6	2.8	25.6	2.9	25.6	2.9	
Weight (kg)	57.9	9.0	56.7	9.6	57.3	8.8	57.0	8.7	57.3	9.5	57.3	9.1	57.0	8.5	57.2	9.0	57.2	9.0	
BMI (kg/m <sup>2</sup> )	23.5	3.1	23.1	3.5	23.5	3.3	23.2	3.2	23.3	3.3	23.3	3.3	23.4	3.2	23.3	3.3	23.3	3.3	

**Table 2**  
Effect of maternal multivitamins supplementation on birth outcomes according to sex

Endpoint	Multivitamins												P <sup>int.†</sup>		
	Girls						Boys								
	Yes		No		RR*	95% CI	P	Yes		No		RR*		95% CI	P
N	%	N	%	N				%	N	%					
Stillbirth	5	2.1%	11	4.8%	0.43	(0.15, 1.22)	0.11	7	2.9%	17	6.8%	0.43	(0.18, 1.01)	0.05	0.99
Low birth weight	16	7.6%	37	19.6%	0.39	(0.22, 0.67)	<0.001	17	8.1%	21	10.0%	0.81	(0.44, 1.49)	0.50	0.08
Preterm delivery	53	22.5%	56	25.8%	0.87	(0.63, 1.21)	0.41	56	23.9%	60	25.9%	0.93	(0.67, 1.27)	0.63	0.79
Small for gestational age	19	9.0%	35	18.5%	0.49	(0.29, 0.82)	0.007	14	6.6%	24	11.4%	0.58	(0.31, 1.10)	0.09	0.66

\* Relative risks (RR) and 95% CI are obtained from binomial regression model, and mean difference and 95% CI are obtained from linear regression model.

† P<sup>int.</sup> test for interaction between multivitamins regimen and sex from the likelihood ratio test.



**Table 3**  
Effect of maternal multivitamins supplementation on child mortality and HIV infection according to sex

Endpoint	Multivitamins												P <sub>int</sub> <sup>‡</sup>
	Girls						Boys						
	Yes		No		P		Yes		No		P		
n	N*	n	N*	RR <sup>†</sup>	95% CI	n	N*	n	N*	RR <sup>†</sup>	95% CI	P	
Mortality including stillbirths	59	241	77	228	0.64	(0.46, 0.90)	60	241	62	249	0.99	(0.69, 1.41)	0.95
Mortality among live births													
All children	54	236	66	217	0.68	(0.47, 0.97)	53	234	45	232	1.20	(0.80, 1.78)	0.38
HIV-negative <sup>§</sup>	12	147	19	135	0.55	(0.26, 1.17)	18	156	20	164	0.80	(0.41, 1.56)	0.50
HIV-positive <sup>§</sup>	36	75	32	57	0.76	(0.47, 1.23)	28	61	22	60	1.27	(0.72, 2.24)	0.41
Total HIV infection <sup>//</sup>	75	222	57	192	1.16	(0.82, 1.64)	61	217	60	224	1.05	(0.73, 1.50)	0.85
Total HIV infection or mortality including stillbirths	98	241	102	228	0.87	(0.66, 1.15)	93	241	100	249	0.95	(0.71, 1.26)	0.66

\* n, number of endpoints; N, number of individuals at risk.

<sup>†</sup> Relative risks (RR), 95% CI, and p-values are obtained from Cox proportional hazards model.

<sup>‡</sup> P<sub>int</sub>, test for interaction between multivitamins regimen and sex from the likelihood ratio test.

<sup>§</sup> Time-varying HIV status from birth among live births.

<sup>//</sup> Total HIV infection includes intrauterine, intrapartum, and breastfeeding transmission.

**Table 4**  
Effect of maternal vitamin A plus  $\beta$ -carotene supplementation on child mortality and HIV infection according to sex

Endpoint	Vitamin A plus $\beta$ -carotene												P <sub>int.</sub> <sup>‡</sup>		
	Girls						Boys								
	Yes		No		P	95% CI	Yes		No		RR <sup>†</sup>	95% CI		P	
n	N*	n	N*	n			N*	n	N*						
Mortality including stillbirths	70	239	66	230	1.00	(0.71, 1.40)	0.98	63	249	59	241	1.07	(0.75, 1.53)	0.71	0.76
Mortality among live births															
All children	63	232	57	221	1.03	(0.72, 1.48)	0.86	51	237	47	229	1.10	(0.74, 1.63)	0.65	0.82
HIV-negative <sup>§</sup>	15	138	16	144	0.99	(0.47, 2.08)	0.98	17	151	21	169	1.00	(0.52, 1.94)	0.99	0.99
HIV-positive <sup>§</sup>	35	73	33	59	0.79	(0.49, 1.28)	0.34	31	74	19	47	1.30	(0.72, 2.32)	0.38	0.21
Total HIV infection//	73	211	59	203	1.20	(0.85, 1.69)	0.31	74	225	47	216	1.65	(1.14, 2.38)	0.01	0.22
Total HIV infection or mortality including stillbirths	108	239	92	230	1.15	(0.87, 1.52)	0.34	106	249	87	241	1.27	(0.95, 1.69)	0.11	0.64

\* n, number of endpoints; N, number of individuals at risk.

<sup>†</sup> Relative risks (RR), 95% CI, and p-values are obtained from Cox proportional hazards model.

<sup>‡</sup> P<sub>int.</sub> test for interaction between vitamin A plus  $\beta$ -carotene regimen and sex from the likelihood ratio test.

<sup>§</sup> Time-varying HIV status from birth among live births.

// Total HIV infection includes intrauterine, intrapartum, and breastfeeding transmission.

**Table 5**

Effect of maternal multivitamins or vitamin A plus  $\beta$ -carotene supplements on the incidence of diarrhea and respiratory infection in children according to sex

Endpoint	Multivitamins						Vitamin A plus $\beta$ -carotene					
	Girls			Boys			Girls			Boys		
	RR* (95% CI)	P	P <sup>int.†</sup>	RR* (95% CI)	P	P <sup>int.†</sup>	RR* (95% CI)	P	RR* (95% CI)	P	P <sup>int.†</sup>	
Diarrhea												
All	0.91 (0.74, 1.13)	0.39	0.80 (0.66, 0.97)	0.02	0.37	1.11 (0.90, 1.38)	0.32	0.85 (0.71, 1.03)	0.10	0.07		
Acute	0.89 (0.72, 1.10)	0.30	0.81 (0.67, 0.98)	0.03	0.52	1.08 (0.87, 1.34)	0.48	0.83 (0.69, 1.00)	0.06	0.07		
Watery	0.91 (0.68, 1.21)	0.50	0.77 (0.59, 1.00)	0.05	0.40	1.13 (0.84, 1.50)	0.42	0.75 (0.58, 0.97)	0.03	0.04		
Dysenteric	0.89 (0.67, 1.17)	0.39	0.86 (0.66, 1.13)	0.27	0.87	1.03 (0.78, 1.36)	0.82	0.89 (0.68, 1.17)	0.41	0.48		
Respiratory infection												
Cough	1.04 (0.92, 1.17)	0.57	1.02 (0.92, 1.13)	0.75	0.74	1.12 (0.99, 1.26)	0.06	0.96 (0.86, 1.07)	0.45	0.06		
Cough and fever	1.04 (0.85, 1.28)	0.70	0.99 (0.82, 1.20)	0.95	0.73	1.10 (0.89, 1.35)	0.38	0.94 (0.78, 1.14)	0.56	0.28		
“Cough plus” <sup>‡</sup>	1.10 (0.81, 1.50)	0.55	0.81 (0.60, 1.08)	0.15	0.15	1.13 (0.83, 1.54)	0.43	0.99 (0.74, 1.33)	0.97	0.51		
Cough and rapid respiratory rate	1.36 (0.86, 2.16)	0.19	0.97 (0.60, 1.56)	0.90	0.32	0.82 (0.52, 1.28)	0.38	0.62 (0.38, 1.00)	0.05	0.40		

\* Relative risk (RR), 95% CI, and p-values are obtained from generalized estimating equations using a log link function with a binomial variance assumption.

<sup>†</sup> P<sup>int.</sup> test for interaction between regimen and sex from the robust score test.

<sup>‡</sup> “Cough plus” defined as cough with  $\geq 1$  of the following signs: difficulty breathing, chest retractions, and refusal to eat, drink, or breastfeed.