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## Correlation of Selenium and Zinc Levels to Antiretroviral Treatment Outcomes in Thai HIV-infected Children without Severe HIV Symptoms

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### Abstract

**Background**—Deficiencies in antioxidants contribute to immune dysregulation and viral replication.

**Objective**—To evaluate the correlation of selenium (Se) and zinc (Zn) levels on the treatment outcomes in HIV-infected children.

**Design**—HIV-infected Thai children 1–12 years old, CD4 15–24%, without severe HIV symptoms were included. Se and Zn levels were measured by graphite furnace atomic absorption spectrometry at baseline and 48 weeks. Deficiency cut-offs were Se<0.1  $\mu\text{mol/L}$  and Zn<9.9  $\mu\text{mol/L}$ . Serum ferritin and C-reactive protein (CRP) were performed every 24 weeks. No micronutrient supplement was prescribed.

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#### Financial disclosure and Conflict of interest

All authors declare no conflict of interest and that member of their immediate families do not have a financial interest in or arrangement with any commercial organization that may have a direct interest in the subject matter of this article.

The preliminary analysis of this study was presented as a poster at the 3<sup>rd</sup> International Workshop on HIV Pediatrics 15 – 16 July 2011, Rome, Italy, Poster number P 29.

**Results**—141 children (38.3% male) with a median (IQR) age of 7.3 (4.2–9.0) years, were enrolled. Median baseline CD4% was 20%, HIV-RNA was 4.6 log<sub>10</sub>copies/mL. At baseline, median (IQR) Se and Zn levels were 0.9 (0.7–1.0) μmol/L and 5.9 (4.8–6.9) μmol/L, respectively. None had Se deficiency while all had Zn deficiency.

Over 48 weeks, 97 initiated antiretroviral therapy (ART) and 81% achieved HIV-RNA <50 copies/mL with 11% median CD4 gain. The mean change of Se was 0.06 μmol/L (p = 0.003) and Zn was 0.42 μmol/L (p=0.003), respectively. By multivariate analysis in children who received ART, predictors for greater increase of CD4% from baseline were lower baseline CD4% (p<0.01) and higher baseline Zn level (p=0.02). The predictors for greater decrease of HIV-RNA from baseline were higher baseline HIV-RNA and higher ferritin (both p<0.01). No association of CRP to the changes from baseline of CD4% or HIV-RNA was found.

**Conclusion**—In HIV-infected Thai children without severe immune deficiency who commenced ART, no correlation between selenium and ART treatment outcomes were found. Higher pre-ART Zn levels were associated with significant increases in CD4 percent at 48 weeks.

### Keywords

HIV-infected children; selenium; zinc; CD4%; HIV-RNA; disease progression

## Introduction

Nutritional factors are particularly important in HIV-infected children (1). HIV infection could impair nutritional status by causing a reduced intake and absorption and/or increasing utilization of nutrients. Deficiencies in antioxidants during HIV infection facilitate the development of oxidative stress and may contribute to immune dysregulation and HIV replication (2). Malnourishment could lead to immune dysfunction and higher susceptibility to various infectious diseases (2, 3).

Selenium (Se) and zinc (Zn) are the key trace elements which serve as antioxidants, and play a role in HIV-1 disease progression (2, 4). Se deficiency had been associated with increased mortality among HIV-infected patients (5–10). A randomized trial by Hurwitz et al. reported improvement in CD4 count and HIV-1 RNA suppression after Se supplementation in HIV-infected adults receiving highly active antiretroviral therapy (HAART) (11). Zn deficiency has been associated with decreases in CD4 counts, progression to AIDS, and mortality in HIV-infected adults (12, 13).

Limited data describes changes in Se and Zn levels before and after ART in HIV-infected children (1, 4). Here, we report the correlation of Se and Zn levels in ART-naïve Thai HIV infected children and treatment outcomes after commencement of HAART over 48 weeks.

## Material and Methods

### Study Design and Population

This is a sub-study in The Pediatric Randomized Early versus Deferred Initiation in Cambodia and Thailand study (The PREDICT study, clinicaltrials.gov identification number NCT00234091). Briefly, the inclusion criteria for PREDICT study were HIV infected

children, aged 1–12 years, Center for Disease Control and Prevention clinical category N (no HIV symptoms) A (mild HIV symptoms) or B (moderate HIV symptoms) (14, 15), CD4 15%–24%, hemoglobin  $\geq 7.5$  g/dL, no active infections and naïve to ART at enrolment.

After enrolment, children were randomized to either the immediate arm in which they started HAART at week 0 or the deferred arm in which they started HAART when CD4% dropped to  $< 15\%$  or if the child developed CDC category C events. In this sub-study analysis in Thai HIV-infected children were categorized into 2 groups: 1) those who initiated HAART during the 48 weeks (HAART group) and 2) those who did not initiate HAART over the duration of the study (No HAART group). All children were followed up every 12 weeks for weight, height and clinical evaluation. A complete blood count, CD4% and cell count were collected every 12 weeks. Plasma HIV-RNA, serum ferritin and C-reactive protein (CRP) were performed every 24 weeks. CRP  $\geq 10$  mg/L is defined as having inflammation (16).

## Ethics

This study was approved by local and the Thai Ministry of Public Health institutional review boards. All caregivers gave consent prior to enrollment.

## Methods for Se and Zn level measurement

Se and Zn level were measured by graphite furnace atomic absorption spectrometry; Hitachi Z8200 at Faculty of Tropical Medicine laboratory, Mahidol University, Bangkok. The detection limit of Se was  $0.02 \mu\text{mol/L}$  and the measurable concentration range was  $0.06$  to  $1.91 \mu\text{mol/L}$ . The detection limit of Zn was  $0.765 \mu\text{mol/L}$  with the measurable concentration range between  $2.295$  to  $45.9 \mu\text{mol/L}$ . Se deficiency was defined as level  $< 0.1 \mu\text{mol/L}$  (17). Zn deficiency was defined by Zn  $< 9.9 \mu\text{mol/L}$  in children  $< 10$  years, Zn  $< 10.7 \mu\text{mol/L}$  in males  $\geq 10$  years, and Zn  $< 10.15 \mu\text{mol/L}$  in females  $\geq 10$  years (18). In this study, acid citrate dextrose (ACD) plasma samples collected before HAART commencement and at week 48 were used for Se and Zn level measurement.

## Statistical analysis

Baseline characteristics of ART-naïve children are summarized using median and interquartile range (IQR) for quantitative variables and percentages for categorical variables. Associations between Se and Zn level and baseline characteristics of the patients were analyzed using linear regression models. Changes in Se and Zn levels from baseline to week 48 were assessed by a paired-t test.

Analyses for the effects of Se and Zn level and other clinical factors on the treatment outcomes were conducted for children who commenced HAART. Se and Zn level at baseline were categorized in quartiles before fitting linear regression models. The two primary endpoints were the absolute change in CD4 percent and change in HIV-RNA  $\log_{10}$  copies/mL from baseline. Predictors with a p-value less than 0.10 in univariate analysis were included in multivariate models. Multicollinearity of predictors was assessed using the variance inflation factor (VIF), and predictors with VIF  $> 5$  were removed from

the final model. All analyses were done using STATA/IC version 11.2 (Statacorp LP, TX, USA).

## Results

### Baseline characteristics

A total of 141 children were enrolled and their baseline characteristics are shown in Table 1. All children were infected with HIV via mother-to-child transmission. Six (4%) children had weight for height z-score less than  $-2$  z-score. At baseline, none had Se deficiency with a median (IQR) level of 0.9 (0.7–1.0)  $\mu\text{mol/L}$ . All children had Zn deficiency with median (IQR) level 5.9 (4.7–6.9)  $\mu\text{mol/L}$ . No correlation between Se and Zn levels at baseline was found (coefficient = 0.01, 95% CI  $-0.01$  to 0.04;  $p = 0.2$ ; Pearson's correlation coefficient (Rho) = 0.1).

The correlations between baseline Se and Zn and other baseline data are shown in Table 2. Baseline Se and Zn levels were not significantly different between HARRT and No HAART groups,  $p$ -value 0.38 and 0.79 respectively. Baseline Se was positively correlated with age (coefficient 0.02, 95% CI = 0.01–0.04,  $p < 0.001$ , Rho = 0.31) and negatively associated with plasma HIV-RNA (coefficient  $-0.09$ , 95% CI =  $-0.15$  to  $-0.04$ ,  $p = 0.001$ , Rho =  $-0.27$ ). Baseline Zn was positively correlated with hemoglobin level (coefficient 0.26, 95% CI = 0.02 to 0.50,  $p = 0.034$ , Rho = 0.18).

### Treatment outcomes over week 48

No death or loss to follow up was reported during the sub-study. A total of 44 children were still ART-naïve at the end of this study and 97 children had started HAART (70 started at baseline, and 27 started due to confirmed  $\text{CD4} < 15\%$ ). Median (IQR) duration of HAART in these 27 children in deferred arm who started HAART was 45 (44–47) weeks.

Se and Zn levels in children categorized by HAART commencement status are shown in Table 3. The difference in Se level changes (95% CI) between HAART and No HAART groups was  $-0.01$  ( $-0.08$  to 0.06),  $p = 0.78$ . Difference in Zn level changes (95% CI) between HAART and No HAART groups was  $-0.5$  ( $-0.06$  to 1.0),  $p = 0.03$ .

### Children in HAART group (N=97)

The HAART regimens were nevirapine-based 92%, efavirenz-based 3%, and lopinavir/ritonavir-based 5%. The NRTIs were zidovudine 98%, lamivudine 100%, and abacavir 3%. Over the study period, 5 children had CDC clinical progression from A to B, and 1 from B to C. At 48 weeks, 81% had HIV-RNA  $< 50$  copies/mL and median (IQR)  $\text{CD4}$  gain was 11 (8–14)%. No child had Se deficiency at week 48. The mean change in Se was 0.06  $\mu\text{mol/L}$  ( $p = 0.003$ ). Zn level increased by 0.42  $\mu\text{mol/L}$  ( $p = 0.003$ ) but 95 (98%) were still deficient.

By multivariate analysis, predictors for greater increase of  $\text{CD4}\%$  from baseline to week 48 were lower baseline  $\text{CD4}\%$  ( $p < 0.01$ ) and higher baseline Zn level ( $p = 0.02$ ) (Table 4). The predictors for greater decrease of HIV-RNA from baseline to 48 were higher baseline HIV-RNA and higher baseline ferritin (both  $p < 0.01$ ) (Table 4).

### Children in No HAART group (N=44)

One child in this group had CDC clinical progression from A to B. At week 48, the median (IQR) CD4%, CD4 count and HIV-RNA changes were 0.4 (−2.7 to 2.6)%, −49.5 (−126.5 to −61.5) cells/mm<sup>3</sup> and −0.005 (−0.3 to 0.3) log<sub>10</sub>copies/mL, respectively.

None had Se deficiency and all still had Zn deficiency at week 48. Se level was significantly increased (p=0.02) while Zn level was unchanged (Table 3). In multivariate analysis, we found no association of baseline characteristics, including Zn and Se levels with the change in CD4% or HIV-RNA in children in the No HAART group (all p>0.05; data not shown)

## Discussion

In ART-naïve, Thai HIV-infected children with mild to moderate immune deficiency and no AIDS symptoms, none had Se deficiency but all had Zn deficiency. Higher baseline Se was associated with lower plasma HIV-RNA before HAART initiation but not significantly associated with disease progression and treatment outcomes. Higher baseline Zn was associated with better improvement in CD4% at 48 weeks after commencement of HAART after adjusting for baseline CD4%.

The prevalence of Se deficiency is influenced by geography, nutritional status, and severity of HIV/AIDS. The prevalence of Se deficiency among HIV-infected patients differs substantially with all patients with AIDS being deficient in one study (19) but no patients successfully treated being deficient in another report (20). Se deficiency has been associated with increased mortality among HIV-infected patients (5–10). There are limited data of the prevalence of Se deficiency in ART-naïve HIV-infected children from previous publication to compare with. In our study, no children had Se deficiency which is similar to a report in non HIV-infected children from the Northeast of Thailand (17). However, the Se level in those children was higher than in our study. In our study, higher baseline Se was associated with lower plasma HIV-RNA before ART initiation but did not influence disease progression and treatment outcomes over the 48 weeks of the study.

Data from randomized controlled trials describing the effect of Se supplementation on CD4 count and HIV RNA in HIV-infected adults are conflicting and may be confounded by ART use. For example in one study, CD4 count and HIV-RNA suppression was significantly improved in 74% of 262 adults receiving HAART after Se supplement (11). Another study in pregnant HIV-infected women reported no significant effect, but only 3.4% were receiving HAART (21). This suggests that HAART plus selenium supplementation may have more effect on immunologic and virologic outcomes than selenium supplementation alone. No previous publication of Se supplement in HIV-infected children was found (4). In our study where no micronutrient supplementation was prescribed, we found that Se levels in both groups increased by approximately the same amount over the duration of the study.

Zn deficiency has been reported in between 30–51% of HIV infected adults (22–24). Sixty percent of Ugandan HIV-infected children aged 1–5 years who were not on HAART had Zn deficiency (25). Udomkesmalee et al. reported that 70% of non HIV-infected children from the Northeast part of Thailand had Zn deficiency (26). Different sampling collection

techniques may have led to this difference. Another potential confounder is that Zn levels are influenced by inflammation status. Plasma zinc was lower in HIV-infected adults with inflammation compared to adults without inflammation (27). However, the proportion of children in our study with high CRP was less than 9% and no association between baseline Zn and CRP was found.

There are limited data from randomized trials describing the effect of Zn supplementation and treatment outcomes in HIV-infected children. Bobat et al. reported no effect of Zn supplementation on CD4 counts and HIV-RNA levels in 96 HIV-infected children in South Africa (28). However, they did not measure the plasma Zn levels in their study subjects. Zn supplement may benefit HIV-infected children who are Zn deficient and this association warrants further investigated.

Our study has some limitations, and some findings need to be interpreted with caution. The results of this study cannot be extrapolated to HIV-infected children with underlying severe malnutrition or clinical of AIDS, or to other continents which may have a different prevalence of micronutrient deficiency in general population. Our study was performed only in Thai due to the availability of samples. Therefore, we do not have information of Se and Zn in Cambodian HIV-infected children. In addition, HIV-infected children in the No HAART group who did not need to start treatment during the study may represent a group of slower progressors with a better prognosis compared to those in the HAART group, and therefore the findings on Se and Zn may be confounded. The strengths of our study are the multicentre randomized control trial design in ART-naïve HIV-infected children without AIDS, the availability of Se and Zn levels before and after HAART commencement as well as levels from children who did not receive HAART as a comparator group. In addition, no micronutrient supplementation was used in our study.

In conclusion, in Thai HIV-infected children, who are ART-naïve without AIDS symptoms, CD4 15–24%, none had Se deficiency but all had Zn deficiency. Higher Se levels were associated with lower plasma HIV-RNA at baseline. However, there was no correlation of Se levels to disease progression or treatment outcomes over 48 weeks. Higher baseline Zn was correlated with better improvement of CD4% over 48 weeks in those children who started HAART after adjusting for baseline CD4%.

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## Appendix

The following investigators, clinical centers, and committees participated in the Pediatric Randomized of Early versus Deferred Initiation in Cambodia and Thailand (PREDICT trial).

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**Table 1**  
Baseline characteristics of antiretroviral therapy naïve 141 HIV-infected children

Characteristics <sup>†</sup> (n=141)	Total (n=141)	HAART group (n=97)	No HAART group (n=44)	p-value
Age (years)	7.3 (4.2, 9.0)	7.3 (4.0–8.8)	7.3 (4.5–9.1)	0.47
Male	54 (38.3%)	41 (42.3%)	13 (29.6%)	0.15
CDC clinical classification N:A:B	3:88:50 (2:62:36%)	2:58:37 (2:38:60%)	1:30:13 (2:30:68%)	0.61
Weight (kg)	19 (15, 24)	18.8 (14.3, 23)	19.8 (15.1, 24.3)	0.49
Height (cm)	114 (98, 125)	114 (95, 125)	115 (101, 125)	0.38
Weight for age z-score	-1.0 (-1.6, -0.4)	-1.0 (-1.6, -0.4)	-1.0 (-1.4, -0.5)	0.84
Height for age z-score	-1.3 (-2.0, -0.6)	-1.4 (-2.2, -0.5)	-1.1 (-1.8, -0.6)	0.28
Weight for height z-score	-0.1 (-0.8, 0.4)	-0.1 (-0.7, 0.4)	-0.2 - (-0.9, 0.3)	0.35
CD4%	20 (16.0, 22.6)	18 (16, 22)	22 (19, 25)	<b>&lt;0.01</b>
HIV-RNA (log <sub>10</sub> copies/mL)	4.6 (4.1, 5.0)	4.8 (4.3, 5)	4.5 (3.9, 4.8)	<b>0.01</b>
Number of children with HIV-RNA 5 log <sub>10</sub> copies/mL	43 (30.5%)	36 (37%)	7 (16%)	<b>0.01</b>
Hemoglobin (g/dL)	11.6 (10.9, 12.1)	11.5 (10.5, 12.1)	11.7 (11.2, 12.2)	0.08
Number of children with hemoglobin 7.5 –10.0 g/dL	11 (7.8%)	9 (9.3%)	2 (4.6%)	0.33
Ferritin( $\mu$ g/L); n=140	53.7 (31.7, 86.1)	58.7 (35.1, 98.0)	40.6 (29.4, 72.2)	<b>0.03</b>
CRP (mg/L); n=103	1 (0.3, 2.7)	0.1 (0.4, 3.6)	0.9 (0.3, 1.4)	0.07
CRP>10 mg/L	9 (8.7%)	9 (13%)	0 (0%)	<b>0.03</b>
Selenium level ( $\mu$ mol/l)	0.9 (0.7, 1.0)	0.8 (0.7, 1.0)	0.9 (0.7, 1.0)	0.38
% selenium deficient	0%	0%	0%	-
Zinc level ( $\mu$ mol/l)	5.9 (4.7, 6.9)	5.9 (4.6, 6.9)	5.9 (4.8, 6.6)	0.80
% zinc deficient	100%	100%	100%	-

Note: Selenium deficiency is defined as selenium <0.1  $\mu$ mol/l (17), zinc deficiency is defined by the following criteria (18): zinc <9.9  $\mu$ mol/l in children <10 years, zinc <10.7  $\mu$ mol/l in males 10 years, and zinc <10.15  $\mu$ mol/l in females 10 years

CRP: C-reactive protein

<sup>†</sup>Data are presented as median (IQR)

**Table 2**

Correlation between selenium and zinc levels and characteristics at baseline

Baseline characteristics	Coefficient	95% CI	<i>p</i> -value
<b>Selenium (<math>\mu\text{mol/l}</math>)</b>			
Age (year)	0.02	(0.01, 0.04)	<0.001
Gender			
Female	Ref.		
Male	-0.02	(-0.10, 0.06)	0.57
CDC clinical classification			
N	Ref.		
A	0.13	(-0.14, 0.39)	0.35
B	0.14	(-0.13, 0.41)	0.31
Weight for age z-score	-0.002	(-0.04, 0.04)	0.92
Height for age z-score	0.001	(-0.03, 0.04)	0.97
Weight for height z-score	-0.005	(-0.04, 0.03)	0.81
Hemoglobin (g/dL)	0.03	(-0.01, 0.06)	0.17
Ferritin ( $\mu\text{g/L}$ ) <sup>I</sup>	-0.05	(-0.16, 0.06)	0.35
CRP (mg/L) <sup>I</sup>	-0.02	(-0.09, 0.05)	0.59
Having CRP>10 mg/L	-0.07	(-0.22, 0.09)	0.41
CD4%	0.003	(-0.004, 0.01)	0.45
HIV-RNA log <sub>10</sub> copies/mL	-0.09	(-0.15, -0.04)	<b>0.001</b>
<b>Zinc (<math>\mu\text{mol/l}</math>)</b>			
Age (year)	0.02	(-0.06, 0.11)	0.59
Gender			
Female	Ref.		
Male	0.04	(-0.49, 0.56)	0.89
CDC clinical classification			
N	Ref.		
A	-1.01	(-2.79, 0.77)	0.27
B	-0.96	(-2.77, 0.84)	0.29
Weight for age z-score	0.13	(-0.13, 0.39)	0.32
Height for age z-score	0.20	(-0.03, 0.44)	0.08
Weight for height z-score	-0.03	(-0.29, 0.23)	0.80

Baseline characteristics	Coefficient	95% CI	<i>p</i> -value
Hemoglobin (g/dL)	0.26	(0.02, 0.50)	<b>0.03</b>
Ferritin ( $\mu\text{g/L}$ ) <sup><i>l</i></sup>	0.30	(-0.43, 1.03)	0.42
CRP (mg/L) <sup><i>l</i></sup>	0.16	(-0.29, 0.61)	0.49
Having CRP>10 mg/L	-0.46	(-1.43, 0.51)	0.35
CD4%	-0.04	(-0.09, 0.01)	0.09
HIV-RNA log <sub>10</sub> copies/mL	-0.08	(-0.46, 0.31)	0.69

Linear regression was used with significance level of 0.05

<sup>*l*</sup>Log<sub>10</sub> transformed were calculated

CRP: C-reactive protein

Table 3

Selenium and zinc levels by HAART commencement status

Study group	n	Baseline Mean (SD)	Week48 Mean (SD)	Mean change	95%CI	p-value <sup>1</sup>
<b>Selenium (<math>\mu\text{mol/l}</math>)</b>						
HAART group	97	0.83 (0.23)	0.89 (0.21)	0.06	(0.02–0.10)	<b>0.003</b>
No HAART group	44	0.87 (0.24)	0.94 (0.22)	0.07	(0.01–0.12)	<b>0.02</b>
<b>Zinc (<math>\mu\text{mol/l}</math>)</b>						
HAART group	97	5.85 (1.61)	6.26 (1.45)	0.42	(0.15–0.70)	<b>0.003</b>
No HAART group	44	5.78 (1.35)	5.64 (1.19)	-0.12	(-0.53–0.28)	0.54

<sup>1</sup> Paired t-test were used with significance level of 0.05

HAART: highly active antiretroviral therapy

HAART group was defined as children received HAART within 48 weeks,

No HAART group was defined as no HAART initiation over 48 weeks

Univariate and multivariate regression models showing relationship of baseline characteristics to change CD4% and HIV-RNA changes in 97 HIV-infected children 48 weeks after commencing HAART

**Table 4**

	Univariate			Multivariate		
	Coef	95%CI	p-value	Coef	95%CI	p-value
<b>Change in CD4% from baseline to week 48</b>						
<b>Baseline Se quartile<sup>1</sup></b>			0.26 <sup>†</sup>			
Q1	Ref.					
Q2	-2.52	(-5.77, 0.74)	0.13			
Q3	-1.93	(-5.22, 1.35)	0.25			
Q4	-3.16	(-6.52, 0.20)	0.07			
<b>Baseline Zn quartile<sup>1</sup></b>						
Q1	Ref.		0.05 <sup>†</sup>	Ref.		
Q2	0.03	(-3.26, 3.33)	0.99	-0.04	(-3.09, 3.00)	0.98
Q3	2.20	(-1.18, 5.57)	0.20	2.00	(-1.13, 5.13)	0.21
Q4	3.92	(0.76, 7.09)	<b>0.02</b>	3.69	(0.74, 6.63)	<b>0.02</b>
<b>Age</b>	-0.13	(-0.52, 0.27)	0.53			
<b>Gender</b>						
Female	Ref.					
Male	-0.71	(-3.12, 1.70)	0.56			
<b>CDC clinical classification</b>						
N	Ref.		0.20 <sup>†</sup>			
A	-4.51	(-12.84, 3.82)	0.29			
B	-6.21	(-14.63, 2.20)	0.15			
<b>Weight for age z-score</b>	-0.53	(-1.75, 0.70)	0.39			
<b>Height for age z-score</b>	-0.49	(-1.56, 0.58)	0.36			
<b>Weight for height z-score</b>	-0.14	(-1.46, 1.17)	0.83			
<b>Hemoglobin (g/dL)</b>	-0.26	(-1.31, 0.79)	0.62			

	Univariate			Multivariate		
	Coef	95%CI	p-value	Coef	95%CI	p-value
<b>Change in CD4% from baseline to week 48</b>						
Ferritin <sup>2</sup> (µg/L)	2.2	(-0.86, 5.30)	0.16			
<b>CRP<sup>2</sup>(mg/L)</b>						
CRP <=10	Ref.					
CRP >10	2.26	(-1.90, 6.42)	0.28			
<b>Baseline CD4%</b>	-0.40	(-0.61, -0.19)	<0.01	-0.33	(-0.54, -0.13)	<0.01
<b>Baseline HIV-RNA log<sub>10</sub> copies/mL</b>	2.03	(0.33, 3.74)	0.02	1.63	(0, 3.26)	0.05
<b>Change in HIV-RNA log<sub>10</sub> copies/mL from baseline to week 48</b>						
	Univariate			Multivariate		
	Coef	95%CI	p-value	Coef	95%CI	p-value
<b>Baseline Se quartile<sup>1</sup></b>			0.04 <sup>†</sup>			
Q1	Ref.					
Q2	0.03	(-0.52, 0.58)	0.91	-0.15	(-0.59, 0.30)	0.51
Q3	-0.01	(-0.57, 0.55)	0.97	-0.31	(-0.76, 0.14)	0.17
Q4	0.71	(0.14, 1.28)	0.02	0.17	(-0.31, 0.65)	0.48
<b>Baseline Zn quartile<sup>1</sup></b>			0.84 <sup>†</sup>			
Q1	Ref.					
Q2	-0.06	(-0.65, 0.54)	0.85			
Q3	-0.27	(-0.88, 0.34)	0.38			
Q4	-0.09	(-0.66, 0.49)	0.77			
<b>Age</b>	0.02	(-0.05, 0.09)	0.52			
<b>Gender</b>						
Female	Ref.					
Male	0.15	(-0.26, 0.57)	0.46			
<b>CDC clinical classification</b>			0.64 <sup>†</sup>			
N	Ref.					



	Univariate			Multivariate		
	Coef	95%CI	p-value	Coef	95%CI	p-value
<b>Change in HIV-RNA log<sub>10</sub> copies/mL from baseline to week 48</b>						
A	-0.70	(-2.17, 0.76)	0.34			
B	-0.63	(-2.11, 0.85)	0.40			
<b>Weight for age z-score</b>						
	0.11	(-0.10, 0.32)	0.31			
<b>Height for age z-score</b>						
	0.10	(-0.09, 0.28)	0.31			
<b>Weight for height z-score</b>						
	0.03	(-0.19, 0.26)	0.77			
<b>Hemoglobin (g/dL)</b>						
	-0.09	(-0.27, 0.10)	0.35			
<b>Ferritin<sup>2</sup> (µg/L)</b>						
	-0.65	(-1.17, -1.13)	<b>0.02</b>	-0.87	(-1.28, -0.45)	<b>&lt;0.01</b>
<b>CRP<sup>2</sup> (mg/L)</b>						
CRP<=10	Ref.					
CRP>10	0.02	(-0.70, 0.74)	0.96			
<b>Baseline CD4%</b>						
	0.01	(-0.03, 0.04)	0.78			
<b>Baseline HIV-RNA log<sub>10</sub> copies/mL</b>						
	-0.83	(-1.09, -0.58)	<b>&lt;0.01</b>	-0.87	(-1.12, -0.62)	<b>&lt;0.01</b>

Interpretation:

- Predictors for greater increase of CD4% from baseline to week 48 were lower baseline CD4% and higher baseline Zn level.
- Predictors for greater decrease of HIV-RNA from baseline to 48 were higher baseline HIV-RNA and higher baseline ferritin.

<sup>1</sup> Se and Zn level at baseline were categorized in quartiles before fitting linear regression models.

<sup>2</sup> Log<sub>10</sub> transformed

<sup>†</sup> Overall p-values from F-statistics

Coef: Coefficient, Ref: reference, Se: selenium, Zn: Zinc, CRP: C-reactive protein