Short Communication: Hematological Safety of Perinatal Exposure to Zidovudine in Uninfected Infants Born to HIV Type 1-Infected Women in Thailand

Nelly Briand,^{1,2} Sophie Le Coeur,^{2,3,4} Gonzague Jourdain,^{2,3,5} Somboon Hotrawarikarn,^{2,6}
Surat Sirinontakan,^{2,7} Temsiri Hinjiranandana,^{2,8} Suparat Kanjanavanit,^{2,9}
Patrinee Traisathit,^{2,10} Kenneth McIntosh,^{5,11} and Marc Lallemand,^{2,3,5}
for the Program for HIV Prevention and Treatment Study Group

Abstract

The evolution of hematological parameters in HIV-1-exposed uninfected infants according to various durations of perinatal zidovudine exposure was studied. We used data prospectively collected among 1122 HIV-uninfected formula-fed infants born to HIV-infected mothers who participated in a clinical trial to prevent perinatal transmission in Thailand (PHPT-1). Infants were exposed to different durations of zidovudine both *in utero* and after birth. Hemoglobin level and leukocyte, absolute neutrophil, and lymphocyte counts were measured at birth and at 6 weeks of age. The association between hematological parameters at birth and the duration of zidovudine exposure *in utero* was studied using a linear regression model, and changes between birth and 6 weeks of age and the duration of postnatal zidovudine exposure using mixed effects models. At birth, the hemoglobin level was lower in newborns exposed to zidovudine for more than 7.5 weeks *in utero* (adjusted regression coefficient: -0.6 g/dl; 95% confidence interval: -1.1 to -0.1). Six weeks after birth, the hemoglobin level had decreased faster in infants administered zidovudine for more than 4 weeks (adjusted regression coefficient: -0.1 g/dl; 95% confidence interval: -0.2 to -0.1). The duration of perinatal zidovudine exposure was not associated with the evolution of leukocyte, neutrophil, and lymphocyte counts. Despite the differences in hemoglobin levels, grade 3 or 4 anemia did not significantly differ by maternal or infant zidovudine duration. The clinical impact appeared modest, but longer exposure may warrant close monitoring.

Introduction

ZIDOVUDINE (ZDV) is the most widely used nucleoside reverse transcriptase inhibitor for prevention of mother-to-child transmission (PMTCT) of HIV-1, both because it was shown to be effective in the original placebo-controlled trial¹ and because the World Health Organization (WHO) includes it in all recommended PMTCT regimens.² The recent WHO

recommendation is to begin ZDV, alone or in combination with other antiretrovirals, at 14 weeks or as soon as possible thereafter, and at birth for 6 weeks in the infant.^{2,3}

The use of perinatal ZDV is, however, accompanied by a risk of infant anemia and neutropenia.^{1,4,5} Although these have been widely described, their relation to the length of ZDV administration and the separate effects of antenatal and postnatal exposure have not been investigated in detail. We

¹Institut National d'Études Démographiques (INED), Paris, France.

²Institut de Recherche pour le Développement (IRD), UMI 174, Chiang Mai, Thailand.

³Faculty of Associated Medical Sciences, Chiang Mai University, Chiang Mai, Thailand.

⁴Centre Population et Développement (CEPED), UMR Paris Descartes-IRD-INED, Paris, France.

⁵Harvard School of Public Health, Department of Immunology and Infectious Diseases, Boston, Massachusetts.

⁶Klaeng Hospital, Rayong, Thailand.

⁷Health Promotion Hospital Regional Center I, Bangkok, Thailand.

⁸Somdej Pranangchao Sirikit Hospital, Chonburi, Thailand.

⁹Nakornping Hospital, Chiang Mai, Thailand.

¹⁰Department of Statistics, Faculty of Science, Chiang Mai University, Chiang Mai, Thailand.

¹¹Harvard Medical School, Department of Pediatrics, Boston, Massachusetts.

TABLE 1. ASSOCIATION BETWEEN DURATION OF ZDV EXPOSURE AND CHANGES IN HEMATOLOGICAL MEASUREMENTS IN HIV-UNINFECTED INFANTS

At birth according to in utero exposure	Univariate analysis			Multivariate linear regression		
	Short (N ^a = 512)	Long (N ^a = 610)	p ^b	Adjusted mean difference ^f	95% Confidence intervals	p ^b
Hemoglobin (g/dl)						
n ^c	202	228		401		
Median (IQR)	15.8 (14.5; 17.2)	15.2 (13.8; 16.8)	<0.01	−0.6	(−1.1 to −0.1)	0.02
Anemia ^d n (%)	24 (11.9)	35 (15.4)	0.30			
Grade 3–4 ^d n (%)	2 (1.0)	5 (2.2)	0.45			
Leukocytes (/mm ³)						
n ^c	202	227		408		
Median (IQR)	16,555 (12,450; 20,800)	15,600 (12,400; 19,770)	0.18	−677	(−1671 to 317)	0.18
Absolute neutrophil counts (/mm ³)						
n ^c	205	232		413		
Median (IQR)	10,106 (7,200; 13,320)	9,551 (6,922; 12,693)	0.50	−169	(−1075 to 738)	0.71
Neutropenia ^e n (%)	22 (10.7)	25 (10.8)	0.99			
Grade 3–4 ^e n (%)	9 (4.4)	9 (3.9)	0.81			
Lymphocytes (/mm ³)						
n ^c	69	90		154		
Median (IQR)	4,620 (3,264; 57,750)	4,152 (3,124; 5,771)	0.34	−268	(−926 to 390)	0.42
From birth to 6 weeks according to postnatal exposure	Short (N ^a = 479)	Long (N ^a = 643)		Adjusted mean difference ^g	95% Confidence intervals	p ^b
Hemoglobin (g/dl)						
n ^c	459	609		813		
Median (IQR)	9.7 (9.0; 10.3)	9.3 (8.6; 10.0)	<0.01	−0.1	(−0.2 to −0.1)	<0.01
Anemia ^d n (%)	190 (41.4)	334 (54.8)	<0.01			
Grade 3–4 ^d n (%)	4 (0.9)	7 (1.2)	0.77			
Leukocytes (/mm ³)						
n ^c	459	613		827		
Median (IQR)	10,300 (8,700; 12,600)	9,900 (8,100; 11,810)	<0.01	−102	(−261 to 56)	0.21
Absolute neutrophil counts (/mm ³)						
n ^c	458	612		838		
Median (IQR)	2,477 (1,760; 3,468)	2,208 (1,521; 3,085)	<0.01	−29	(−162 to 104)	0.67
Neutropenia ^e n (%)	47 (10.3)	99 (16.2)	<0.01			
Grade 3–4 ^e n (%)	10 (2.2)	22 (3.6)	0.21			
Lymphocytes (/mm ³)						
n ^c	157	228		305		
Median (IQR)	6,460 (5,428; 8,160)	6,467 (5,137; 7,947)	0.40	−45	(−194 to 103)	0.55

^aN refers to the total number of mother–infants pairs in the study.

^bp calculated in univariate analysis by Chi-square test or Fisher's test for categorical variables and Student's *t*-test for continuous variables; in multivariate analysis by Wald's test.

^cn includes available samples in each exposure category. For peripheral blood at birth, numbers are smaller than numbers of available mother–infant pairs because many sites used cord blood for hematological values, and these were not included.

^dAnemia here is Grade 1 or greater (<13 g/dl at birth and <9.5 g/dl at 6 weeks). Grade 3–4 anemia: <10 g/dl at birth and <7 g/dl at 6 weeks.

^eNeutropenia here is Grade 1 or greater (<5000/mm³ at birth and <1300/mm³ at 6 weeks). Grade 3–4 neutropenia: <3000/mm³ at birth and <750/mm³ at 6 weeks.

^fCoefficient reflecting *in utero* exposure, short vs. long, adjusted for maternal age, region of residence, maternal hematological parameters at 26 weeks of gestation (before ZDV exposure initiation), CD4 count, HIV-1 RNA viral load, maternal clinical classification, mode of delivery, preterm birth (<37 weeks of gestation), and infant's sex and birth weight.

^gCoefficient reflecting postnatal exposure, short vs. long, from birth to 6 weeks of age, adjusted for maternal age, region of residence, maternal hematological parameters at 26 weeks of gestation (before ZDV exposure initiation), CD4 count, HIV-1 RNA viral load, maternal clinical classification, mode of delivery, preterm birth (<37 weeks of gestation), infant's sex, weight, and hematological parameters at birth.

analyzed the association between the duration of ZDV exposure and the hematological changes in HIV-uninfected infants.

Materials and Methods

We performed a subanalysis of the Perinatal HIV Prevention Trial 1 in Thailand (PHPT-1) (ClinicalTrials.gov Identifier: NCT00386230), a randomized, controlled, double blind clinical trial comparing the efficacy of various durations of zidovudine prophylaxis for the reduction of mother-to-child HIV transmission. Design and results have been published elsewhere.⁶

Four ZDV regimens were compared in a 2×2 factorial design: women received ZDV either from 28 or 35 weeks gestation until delivery and infants either for 3 days or 6 weeks. This subanalysis was restricted to HIV-uninfected singleton infants born to mothers who participated in this trial. We excluded cases with <75% adherence assessed by pill count or bottle weight. Maternal ZDV treatment was categorized as “long” if ≥7.5 weeks or “short” if <7.5 weeks, and infant treatment as “long” if >4 weeks or “short” if 3 days. The 7.5 weeks cut-off was the lowest point observed between the two peaks of the distribution of the maternal treatment durations.

Hemoglobin (g/dl) and absolute total leukocyte, neutrophil, and lymphocyte counts (cells/mm³) were measured in infants on peripheral blood at birth and at the 6-week visit. Anemia and neutropenia were defined using established grades and thresholds.⁷

We used linear regression models to study the association between the duration of ZDV exposure *in utero* and hematological parameters at birth, and mixed effects models to study the changes between birth and 6 weeks of age according to the duration of postnatal ZDV exposure.

Analyses were conducted using SAS statistical software (version 9.1; SAS, Institute Inc., Cary, NC). $p < 0.05$ was considered statistically significant.

Results

The analysis was performed on 1122 singleton HIV-uninfected infants after exclusion of 123 infected or indeterminate infants, 20 twins, and 144 infants whose mothers had inadequate adherence. During pregnancy, the median (interquartile range) duration of ZDV administration was 4.4 (3.4–5.4) weeks in the *short* group ($n = 512$) and 11.3 (10.1–12.3) weeks in the *long* group ($n = 610$). After birth, the median duration of ZDV administration was 3 days in the *short* group ($n = 479$) and, using the actual date of the 6-week visit, 6.6 (6.1–6.9) weeks in the *long* group ($n = 643$).

Neonates exposed *in utero* for ≥7.5 weeks had a significantly lower peripheral blood hemoglobin level (median time from birth to blood draw, 5 h) than those exposed for a shorter time (Table 1). Total leukocyte, absolute neutrophil, and lymphocyte counts were also lower in longer-exposed infants, but the differences were not statistically significant. The associations between *in utero* exposure and hematological parameters remained the same after adjustment for maternal age, region of residence, maternal hematological parameters at 26 weeks of gestation, CD4 count, HIV-1 RNA viral load, maternal clinical classification, mode of delivery, preterm birth, and infant's sex and birthweight (Table 1).

Between birth and 6 weeks, the hemoglobin level decreased faster in infants exposed to a long postnatal ZDV regimen

compared to infants exposed for 3 days (Table 1). This effect was significant whatever the duration of *in utero* exposure. Total leukocyte and absolute neutrophil counts also decreased significantly more rapidly in infants exposed to longer postnatal ZDV (Table 1). After adjustment for the same factors as above and for hematological parameters at birth, only the association with hemoglobin level remained significant. The evolution of lymphocyte count did not differ according to postnatal exposure to ZDV.

The percentage of neonates with anemia or neutropenia did not differ at birth in neonates exposed to the long versus short ZDV regimen. However, at 6 weeks, anemia and neutropenia were significantly more frequent in infants exposed to a long postnatal regimen. The rates of grade 3 or 4 anemia (hemoglobin <7 g/dl) and neutropenia (neutrophils <750/mm³) were, however, not different between the two groups, 1.2% versus 0.9% ($p = 0.77$) and 3.6% versus 2.2% ($p = 0.18$), respectively.

Discussion

In this study, infants exposed to ZDV *in utero* for over 7.5 weeks had a lower hemoglobin level at birth, and infants exposed longer after birth had a faster decrease in hemoglobin level from birth to 6 weeks of age, with these effects being independent of each other. Similar negative trends were observed for leukocyte, absolute neutrophil, and lymphocyte counts. Grade 3 or 4 anemia and neutropenia were uncommon at birth and at 6 weeks, with no difference according to the duration of ZDV exposure.

In a secondary intent-to-treat analysis, using the randomized instead of actual duration of zidovudine exposure *in utero* and after birth (data not shown), we found similar results.

No previous study has analyzed separately the effect of various lengths of maternal and infant ZDV exposure in HIV-uninfected infants, but our results are consistent with those of previous clinical trials in which ZDV or placebo was randomly assigned.^{1,4,5}

In conclusion, our results confirm that perinatal ZDV, a major antiretroviral component for PMTCT of HIV, induces a small but noticeable decrease in hematological parameters in infants, that a longer maternal ZDV treatment is associated with a lower hemoglobin level at birth, and that the effects of antenatal and postnatal exposure are independent. However, although independent, these effects may be additive. As the 2009 WHO guidelines are now recommending initiation of antiretrovirals as early as 14 weeks of gestation in addition to 6 weeks of infant exposure, it will be important to pay attention to factors that might aggravate drug-induced hematological toxicity, such as iron deficiency, maternal blood loss, or preexisting anemia.

Acknowledgments

All authors contributed significantly to this work. N. Briand performed the statistical analyses for this study and contributed to its design and interpretation. She also drafted the manuscript. S. Le Coeur, G. Jourdain, K. McIntosh, P. Traisathit, and M. Lallemand contributed to the design and contributed to the acquisition of data, analysis, and interpretation. S. Hotrawarikarn, S. Sirinontakan, T. Hinjiranandana, and S. Kanjanavanit participated in the follow-up of the

mothers and the children as part as coinvestigators of the PHPT-1 clinical trial. M. Lallemand was the principal investigator of the PHPT-1 and was responsible for the entire scientific process of the study. All authors revised the paper and approved the final version.

This study was funded by the Eunice Kennedy Shriver National Institute of Child & Human Development, National Institutes of Health (R01 HD 33326), USA; Fogarty International, USA; Ministry of Public Health, Thailand; Institut de Recherche pour le Développement (IRD), France; Institut National d'Études Démographiques, France; and the Thailand International Development Cooperation Agency funded the clinical trial. The French National Agency for AIDS Research (ANRS) provided the first author (N.B.) with a Ph.D. scholarship. None of the sponsors was directly involved in the collection, analysis, and interpretation of data, writing of the paper, and decision to submit it for publication.

Presented at the 4th conférence francophone VIH/Sida, March, 29–31, 2007, Paris.

We would like to thank the investigators who participated in the trial (the number of women enrolled at each hospital is given in parentheses): Rayong (161): S.L. Weerawatgoompa, V. Karnchanamayul, C. Tantiyaworawongse, S. Ariyadej; Chiang Rai Provincial (140): R. Hansudewechakul, J. Achalapong, R. Srismith, P. Wattanaporn (deceased); Prapokklao (117): S. Phongpanich, P. Yuthavisuthi, C. Ngampiyasakul, S. Sooksengchai; Phayao Provincial (100): S. Bhakeechep, J. Hemvuttiphan, L. Thambhitak, V. Lattiwongsakorn; Banglamung (87): J. Ithisukanan, S. Siritathandon, K. Boonrod, S. Piyaman; Chonburi (72): P. Pinchan, N. Chotivanich, S. Hongsiworn, P. Kittikoon; Bhumibhol Adulyadej (66): S. Prommat, S. Nimkarn, S. Charnivises, V. Suraseranivong, P. Layangool, B. Gulakirt; Chiang Kham (57): C. Putiyanun, S. Limsuwan, S. Charkrit; Mae Sai (55): H. Sirichai, P. Jindaapilukkul, S. Kunkongkapan; Hat Yai (47): S. Lamlerkittikul, B. Warachit, K. Veerapradist; Chachoengsao (43): A. Kanjanasing, R. Keawsonthi, C. Jirawison; Klaeng (42): S. Hotrawarikarn, S. Techapalokul, A. Palasudhi; Bamrasnaradura (42): P. Tunthanathip, S. Sirikawin, A. Chaovavanich; Mae Chan (41): S. Tantanarat, S. Piyaworawong; Phan (40): P. Lersuangpanya, S. Jeungphichanwanit, T. Jhangjit; Health Promotion Center Region I (38): I. Tangtitawong, S. Sirinontakan, V. Japikanond, B. Ngamsiriudom; Somdej Prapinklao (38): S. Suphanich, S. Maitrisathit, N. Tawornpanit, N. Kalawantavanich; Nopparat Rajathanee (35): T. Chanpoo, S. Ruangsirinorn, N. Thamanavat, P. Hotrarapavanond; Phranangkla (33): S. Mookmakkul, S. Wanwaisart, S. Hongyok; Banchang (32): S. Tragarnool, S. Chutimanukul, G. Kunawudhi; Somdej Prangchao Sirikit (31): W. Pornkijprasarn, T. Hinjiranandana, S.N. Nakorn; Nakornping (30): V. Gomutbutra, P. Leelanitkul, K. Kunsuikmengrai, S. Kanjanavanit, S. Kahintapongs; Lamphun (28): K. Pagdi, W. Matanasaravoot, R. Somsamai, W. Chavengchaiyong;

Buddhachinaraj (24): P. Thanomrat, W. Wannapira, W. Ar-tong, C. Chaipat; Chiang Khong Royal Crown Prince (15): C. Taiyaitiang, S. Parinya; Phayamengrai (12): P. Tanti-wattanakul, T. Onchomjum (deceased); McCormick (11): C. Tangchaitrong, S. Suwansarakul, C. Phimphilai.

We would also like to acknowledge the Perinatal HIV Prevention trial staff: study monitoring, safety monitoring, data management, logistics, and support staff.

Author Disclosure Statement

No competing financial interests exist.

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Address correspondence to:

Nelly Briand

CESP

Inserm U1018

82 rue du Général Leclerc

94276 Le Kremlin-Bicêtre Cedex, France

E-mail: nelly.briand@inserm.fr