

## Short- and Long-term Immunological and Virological Outcome in HIV-Infected Infants According to the Age at Antiretroviral Treatment Initiation

Tessa Goetghebuer,<sup>1</sup> Jerome Le Chenadec,<sup>2</sup> Edwige Haelterman,<sup>1</sup> Luisa Galli,<sup>3</sup> Catherine Dollfus,<sup>4</sup> Claire Thorne,<sup>5</sup> Ali Judd,<sup>6</sup> Olivia Keiser,<sup>7,8</sup> Jose Tomas Ramos,<sup>9</sup> Jack Levy,<sup>1</sup> Josiane Warszawski,<sup>2,10</sup> and the European Infant Collaboration Group<sup>a</sup>

<sup>1</sup>Pediatric Department, CHU St Pierre, Brussels, Belgium; <sup>2</sup>Inserm, U822, Le Kremlin-Bicêtre, France; <sup>3</sup>Italian Register, Department of Paediatrics, University of Florence, Italy; <sup>4</sup>P-HP, Hôpital Trousseau, Service d'Hématologie et d'oncologie pédiatrique, Paris, France; <sup>5</sup>MRC Centre of Epidemiology for Child Health, Institute of Child Health, University College London; <sup>6</sup>Collaborative HIV Paediatric Study, MRC Clinical Trials Unit, London, United Kingdom; <sup>7</sup>Data Center of the Swiss HIV Cohort Study, Lausanne, and <sup>8</sup>Institute of Social and Preventive Medicine, University of Bern, Switzerland; <sup>9</sup>Pediatric Department, Hospital Universitario de Getafe, Madrid, Spain; and <sup>10</sup>Université Paris-Sud, Faculté de Médecine Paris-Sud, U822, Le Kremlin-Bicêtre, France

**The clinical benefit of antiretroviral therapy in infants is established. In this cohort collaboration, we compare immunological and virological response to treatment started before or after 3 months of age. Early initiation provides a better short-term response, although evolution after 12 months of age is similar in both groups.**

In the absence of combination antiretroviral therapy (cART), approximately 20% of vertically infected children born in developed countries progress to AIDS in the first year of life [1]. Since cART became available, early initiation of antiretroviral therapy (ART) in infants has been questioned, and policies have varied across Europe. The prevention of disease progression when treatment is started before the age of 3 months was clearly demonstrated in the South African Children with HIV Early Antiretroviral Therapy randomized trial [2] and in the European Infant Collaboration (EIC) [3]. All guidelines have

been updated to recommend early ART in infants infected with human immunodeficiency virus (HIV).

Vertical infection is associated with rapid viral multiplication during the first weeks of life and high viral load (VL) over the first 2 years of life. At birth, the CD4 percentage is high and decreases to reach a plateau around the age of 1 year [4]. CD4 T-cell depletion associated with HIV infection is correlated with clinical progression [1]. The level of plasma HIV RNA has a poorer prognostic significance than CD4 percentage.

Although the clinical benefits of early treatment are recognized, early initiation of cART does not prevent establishment of a reservoir of latently infected cells. It was suggested that early intervention during primary infection leads to better long-term virological suppression (VS) and preserved immune system function [5]. Indeed, when cART is started early in infancy with persistent control of HIV replication, children may become seronegative after the loss of maternal antibodies [5, 6]. Our aim, within the EIC group, was to determine if virological and immunological responses to treatment differ according to the age at which cART is started during first 12 months of life.

### METHODS

#### Study Population

EIC study methods have been described previously [3]. For the present analysis, infants were excluded if they received monotherapy or dual therapy (n = 38), developed AIDS before cART was initiated (n = 7), or did not begin treatment before the age of 12 months (n = 26). The final sample size was 139 infants. A subsequent data merger collected virological and immunological data to the end of 2008.

#### Variables

Demographics, pregnancy data, details of prophylaxis and treatment, and CD4 and VL measurements available since birth were collected. We defined cART as any regimen that included at least 3 antiretroviral drugs. Infants were classified into 3 groups: those starting cART at  $\leq 3$  months of age (0–91 days) (the early-treatment group), those starting at ages 3–6 months, and those starting at ages 6–12 months; the latter 2 groups were considered the deferred-treatment group.

Baseline VL and CD4 percentages were measured before 3 months of age in both groups. This corresponded to the pretreatment values in the early treatment group, which were measured at a median age 37 days (interquartile range [IQR], 12–54 days) for VL and 39 days (IQR, 17–56 days) for CD4

Received 19 July 2011; accepted 15 November 2011; electronically published 23 December 2011.

<sup>a</sup>The European Infant Collaboration Group is described in the Acknowledgments.

Correspondence: Tessa Goetghebuer, MD, Pediatric Department, CHU St Pierre, 322 rue Haute, 1000 Brussels, Belgium (tessa\_goetghebuer@stpierre-bru.be).

**Clinical Infectious Diseases** 2012;54(6):878–81

© The Author 2011. Published by Oxford University Press on behalf of the Infectious Diseases Society of America. All rights reserved. For Permissions, please e-mail: journals.permissions@oup.com.

DOI: 10.1093/cid/cir950

percentage. In the deferred treatment group, baseline values were selected at the age closest to the median ages before age 3 months. Outcomes were VL and CD4 percentage at 6, 12, 18, 24, and 48 months of age. As the threshold for viral detection varied along the study period, VS was defined as VL <500 copies/mL.

### Statistical Analysis

Evolution of CD4 percentage and log<sub>10</sub> VL from baseline to 48 months was described separately for the 3 groups, using smoother techniques [7]. We added the reference curve of CD4 percentage by age established among HIV-exposed, uninfected (HEU) children included in the national French Perinatal Cohort (N = 11 851) and the European Collaborative Study (ECS) (N = 1028) [8]. CD4 z scores generated from these populations as reference were compared with zero at 6, 12, and 24 months in early- and deferred-treatment groups.

The time from treatment initiation to first VS was estimated in each group by the Kaplan-Meier method, censored at last follow-up using the log-rank test for significance. A hazard ratio for the associations with early versus deferred treatment was estimated using the Cox model.

Levels of quantitative CD4 percentage and log<sub>10</sub> VL, and proportion of VL <500 copies/mL at baseline and measured at 6 months ( $\pm 1.5$  month) and 12, 18, 24, and 48 months ( $\pm 3$  months) were compared in the 2 groups. The  $\chi^2$  test, Fisher exact test, or Monte Carlo method was used for categorical data, and the Student *t* test, analysis of variance, Mann-Whitney *U* test, and Kruskal-Wallis test were used for continuous data.

The analysis was intent-to-treat, using SAS (version 9.1; SAS Institute, Cary, North Carolina) and R software (Foundation for Statistical Computing).

## RESULTS

Baseline characteristics of the 139 infants were evenly distributed between the 3 treatment groups including cohort of origin, year of birth, geographical origin of the mother, mode of delivery, antenatal and postnatal ART prophylaxis, maternal VL nearest delivery, gestational age, sex, and baseline infant CD4 cell count and VL (Supplementary Table 1).

### Immunological Response

The CD4 percentage diminished less markedly over the first year of life in the early-treatment group compared with the deferred-treatment groups (Figure 1a). However, CD4 percentages were consistently lower in all groups of HIV-infected infants compared with HEU infants, and median CD4 z score values remained significantly lower in both early- and deferred-treatment groups at the ages of 6, 12, and 24 months (Supplementary Table 2). Median nadir CD4 cell count and CD4 percentages were significantly higher in the early-treatment group until the age of 6 months, but not thereafter.

### Virological Response

A lower peak VL was observed in the early-treatment group infants compared with the 2 other groups (Figure 1b). Kaplan-Meier survival analysis showed that time from treatment initiation to first VS was significantly shorter in the early-treatment group (crude hazard ratio [HR], 1.8 [95% confidence interval {CI}, 1.2–2.7]; *P* = .003; HR adjusted for baseline VL and cohort = 1.7 [95% CI; 1.0–2.9]; *P* = .05) (Figure 1c).

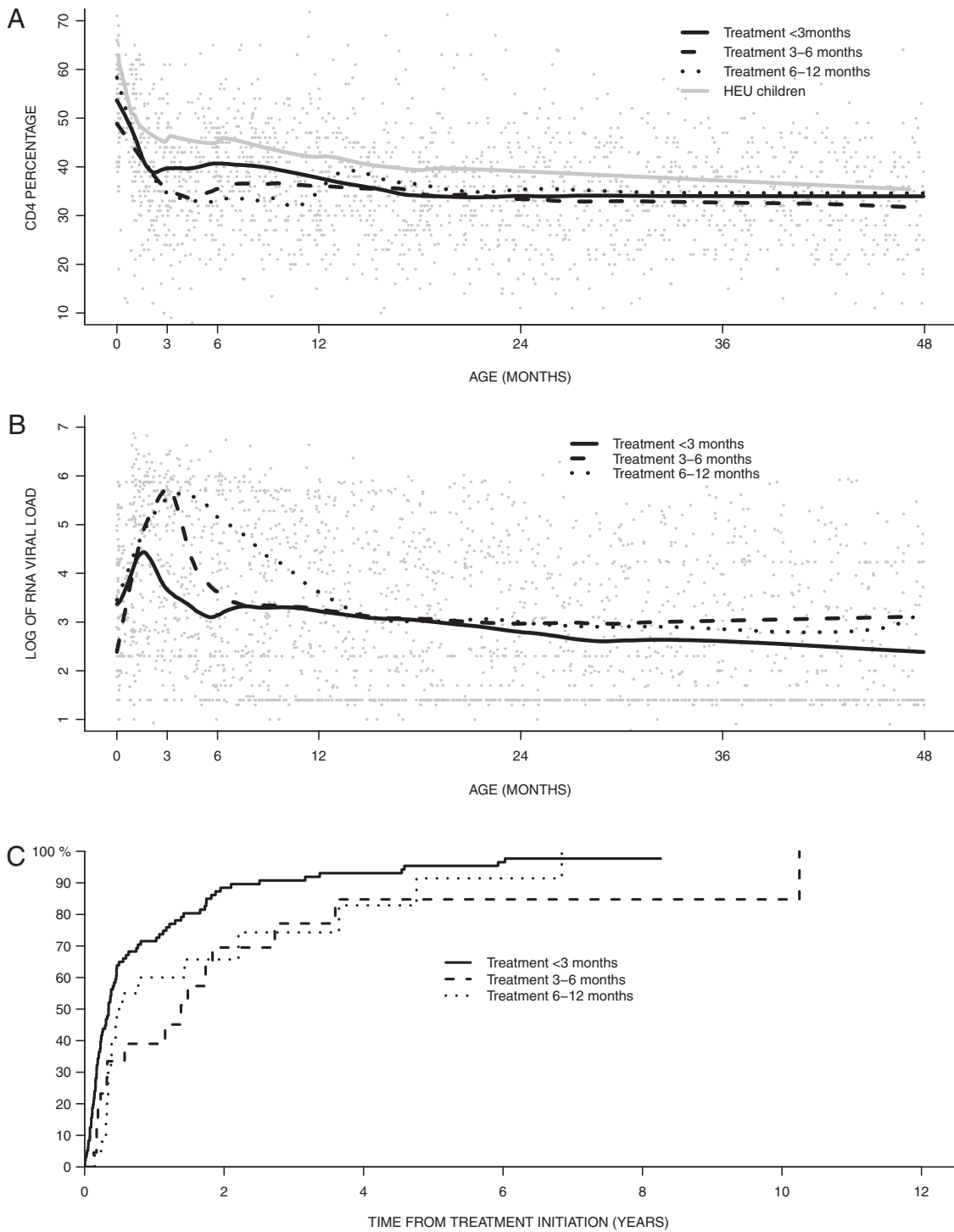
Although similar at baseline, the zenith VL and median VL at 6 months were significantly lower in the early-treatment group, compared with the deferred-treatment group. Moreover, in the early-treatment group, the zenith VL was significantly correlated with the starting age for cART (*r* = 0.48, *P* < .001). There was a trend to lower VL between 12 and 48 months in the early-treatment group compared with the deferred-treatment group.

## DISCUSSION

The prognosis of HIV-infected children has improved considerably since the introduction of cART in 1996 [9]. The EIC has demonstrated that, in routine clinical practice in industrialized countries, the risk of AIDS/death was 5 times lower in infants starting ART before 3 months of age compared with those whose treatment was deferred, and the difference was significant until the age of 60 months [3]. Several reports previously demonstrated that early initiation of treatment and low-baseline VL were significant predictors of long-term VS [10–12]. In this article, we report that early initiation of cART was associated with a lower VL peak despite similar baseline VL. Moreover, we showed that the virological response was more rapid when cART was started before 3 months of age. There was a significant correlation between the age at treatment initiation and the zenith VL in this latter group, suggesting that the sooner the treatment is started, the better the virological control. This observation is important, because VL peak may be correlated with the establishment of a reservoir of latently infected cells.

Regarding the immunological response to treatment, we observed that the decrease of CD4 cell count was less pronounced and the CD4 percentages were higher in the early-treatment group until 12 months of age than in the deferred-treatment group. However, the CD4 percentages remained significantly lower in all groups of HIV-infected infants than in HEU controls up to the age of 2 years.

In this report, the better immunological and virological evolution observed in the early-treatment group compared with the deferred-treatment group was no longer detectable by age 12 months, possibly because of lack of power due to sample size. However, it is possible that the functional competences of CD4 cells are influenced by early initiation of therapy and may maintain an impact on the clinical outcome after the age of 12 months.



**Figure 1.** Comparison of immunological and virological evolution according to age at treatment initiation. *A*, Evolution of CD4 percentage in human immunodeficiency virus (HIV)-infected infants according to age at treatment initiation and in HIV-exposed uninfected infants (HEU). *B*, Evolution of viral load (VL) from birth in different groups according to age at treatment initiation. *C*, Kaplan-Meier survival analysis: time to attaining undetectable VL (VL <500 copies/mL) from the time of treatment initiation according to age at treatment initiation.

To limit potential selection and information biases, the study included HIV-infected infants prospectively followed from birth to exclude the few long-term nonprogressor

children diagnosed later. We analyzed only infants who received cART as first treatment before AIDS developed to evaluate response to cART in asymptomatic children. The

2 groups did not differ significantly for the type of first cART regimen used.

In conclusion, this analysis of the EIC study indicates that cART initiated before the age of 3 months, in addition to the strong and durable impact on the clinical outcome previously described, results in more rapid control of early virological replication and preservation of CD4 lymphocytes up to age 12 months compared with deferred treatment. After the age of 12 months, immunovirological outcomes did not differ significantly between the groups. Because early initiation of treatment in HIV vertically infected infants is now recommended in all guidelines, efforts should be made to establish the diagnosis of infection as soon as possible after birth.

## Supplementary Data

Supplementary materials are available at *Clinical Infectious Diseases* online ([http://www.oxfordjournals.org/our\\_journals/cid/](http://www.oxfordjournals.org/our_journals/cid/)). Supplementary materials consist of data provided by the author that are published to benefit the reader. The posted materials are not copyedited. The contents of all supplementary data are the sole responsibility of the authors. Questions or messages regarding errors should be addressed to the author.

## Notes

**Acknowledgments.** We are indebted to the infants included in this study and to their parents, and to all physicians who participated in collecting clinical data. We thank Dr A. Marchant for his critical review of the manuscript.

EIC group: Albert Faye, AP-HP, Hôpital Robert Debré, Service de pédiatrie générale, Paris, France; Stephane Blanche, AP-HP, Hôpital Necker, Service d'immunologie pédiatrique, Paris, France; Clara Gabiano, Italian Register, Department of Paediatrics, University of Turin, Italy; Diana Gibb, Hannah Castro, Trinh Duong, Collaborative HIV Paediatric Study, MRC Clinical Trials Unit, London; Carlo Giaquinto, Martina Penazzato, Department of Pediatrics, University of Padova, Italy; Magdalena Marczyńska, Infectious Disease Hospital, Medical University Warsaw, Poland; Luminita Ene, Hospital for Infectious Diseases "Dr Victor Babes," Bucharest, Romania; Marc Hainaut, Paediatric Department, CHU St Pierre, Brussels, Belgium; Henriette Scherpbier, Emma Children's Hospital, Academic Medical Center, Amsterdam, Netherlands; Uwe Wintergerst, University Children's Hospital, Munich, Germany; Véronique Schmitz, Hôpital La Citadelle, Liège, Belgium; Bénédicte Brichard, Hôpital St Luc, Brussels, Belgium; Gwenda Verweel, Erasmus MC Sophia Children's Hospital, Rotterdam, Netherlands; Pat Tookey, National Study of HIV in Pregnancy and Childhood, MRC Centre of Epidemiology for Child Health, Institute of Child Health, UCL, London.

**Financial support.** This work was supported by the Paediatric European Network for the Treatment of AIDS (PENTA), which has previously received funding from the European Union Sixth Framework Programme (grant agreement PENTA/ECS 018865); the European Union Seventh Framework Programme (FP7/2007–2013) under EuroCoord grant agreement 260694; the French National Agency for AIDS and Hepatitis Research; and the Smiles Foundation, Belgium.

**Potential conflicts of interest.** All authors: No conflicts.

All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

## References

1. Gray L, Newell ML, Thorne C, Peckham C, Levy J. Fluctuations in symptoms in human immunodeficiency virus-infected children: the first 10 years of life. *Pediatrics* **2001**; 108:116–22.
2. Violarì A, Cotton MF, Gibb DM, et al. Early antiretroviral therapy and mortality among HIV-infected infants. *N Engl J Med* **2008**; 359: 2233–44.
3. Goetghebuer T, Haelterman E, Le Chenadec J, et al. Effect of early antiretroviral therapy on the risk of AIDS/death in HIV-infected infants. *AIDS* **2009**; 23:597–604.
4. Bunders M, Cortina-Borja M, Newell ML. Age-related standards for total lymphocyte, CD4+ and CD8+ T cell counts in children born in Europe. *Pediatr Infect Dis J* **2005**; 24:595–600.
5. Luzuriaga K, Bryson Y, Krogstad P, et al. Combination treatment with zidovudine, didanosine, and nevirapine in infants with human immunodeficiency virus type 1 infection. *N Engl J Med* **1997**; 336: 1343–9.
6. Hainaut M, Peltier CA, Gerard M, Marissens D, Zissis G, Levy J. Effectiveness of antiretroviral therapy initiated before the age of 2 months in infants vertically infected with human immunodeficiency virus type 1. *Eur J Pediatr* **2000**; 159:778–82.
7. Friedman J. A variable span scatterplot smoother. Laboratory for Computational Statistics. Stanford University technical report. *JASA* **1984**; 1–16.
8. Bunders M, Thorne C, Newell ML. Maternal and infant factors and lymphocyte, CD4 and CD8 cell counts in uninfected children of HIV-1-infected mothers. *AIDS* **2005**; 19:1071–9.
9. Doerholt K, Duong T, Tookey P, et al. Outcomes for human immunodeficiency virus-1-infected infants in the United Kingdom and Republic of Ireland in the era of effective antiretroviral therapy. *Pediatr Infect Dis J* **2006**; 25:420–6.
10. Luzuriaga K, McManus M, Mofenson L, Britto P, Graham B, Sullivan JL. A trial of three antiretroviral regimens in HIV-1-infected children. *N Engl J Med* **2004**; 350:2471–80.
11. Chiappini E, Galli L, Tovo PA, et al. Virologic, immunologic, and clinical benefits from early combined antiretroviral therapy in infants with perinatal HIV-1 infection. *AIDS* **2006**; 20:207–15.
12. Van der Linden D, Hainaut M, Goetghebuer T, et al. Effectiveness of early initiation of protease inhibitor-sparing antiretroviral regimen in human immunodeficiency virus-1 vertically infected infants. *Pediatr Infect Dis J* **2007**; 26:359–61.