

Time to Viral Suppression in Perinatally HIV-Infected Infants Depends on the Viral Load and CD4 T-Cell Percentage at the Start of Treatment

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Background: Interventions aiming for an HIV cure would benefit from rapid elimination of virus after the onset of antiretroviral therapy (ART), by keeping the latent HIV reservoir small.

Setting: We investigated HIV suppression in 312 perinatally infected infants starting ART within 6 months after birth from the EPPICC (European Pregnancy and Paediatric HIV Cohort Collaboration).

Methods: To better understand kinetic differences in HIV suppression among infants, we investigated their individual viral load (VL) decay dynamics. We identified VL decay patterns and determined times to viral suppression (TTS). For infants with strictly declining VLs (n = 188), we used parameter fitting methods to estimate baseline VLs, decay rates, and TTS. We subsequently identified the parameters determining TTS by linear modeling.

Results: The majority of infants suppress HIV VL after the onset of ART. Some children experienced a long TTS due to an “erratic” VL decay pattern. We cannot exclude that this is partly due to treatment complications and subsequent treatment changes, but these children were characterized by significantly lower CD4 percentages (CD4%) at start of treatment compared with those with a “clean” VL decline. Focusing on this “clean” subset, the TTS could be predicted by mathematical modeling, and we identified baseline VL and CD4% as the major factors determining the TTS.

Conclusions: As VL steeply increases and CD4% constantly decreases in untreated HIV-infected infants, the progression of an HIV infection is largely determined by these 2 factors. To prevent a further disease progression, treatment should be initiated early after contracting HIV, which consequently shortens TTS.

Key Words: virological suppression, decay dynamics, perinatal HIV, early ART, mathematical model, mechanisms

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INTRODUCTION

Since the discovery of HIV in the 1980s, there is still no cure for HIV.¹ HIV research has resulted in effective antiretroviral therapy (ART) in which a combination of drugs successfully controls the virus. ART reduces HIV replication^{2,3} and allows for the recovery of target cells (largely CD4⁺ T cells).⁴ Once HIV infects a CD4⁺ T cell, it either uses the cell for active replication or inserts viral DNA into the host’s DNA and forms a latent reservoir. Since the virus is quiescent in the latent reservoir, ART fails to affect these established reservoirs, and once treatment stops, latent HIV reservoirs can get reactivated leading to the production of new HIV particles.⁵ Latent HIV reservoirs are the main hurdle in achieving HIV cure.

Current HIV research aims to prevent the establishment of viral reservoir and to reduce its size and diversity. An HIV reservoir is established during the early stages of an HIV infection.⁶ To keep the reservoir small and homogenous, it seems essential to keep the viral exposure time to a minimum by an early starting of ART.⁷ An ART initiation soon after contracting the infection can be accomplished in perinatally HIV-infected infants who are tested and diagnosed soon after birth if the HIV status of the mother is known.⁸ Thus, perinatally HIV-infected infants are the ideal target group for an early-ART initiation,⁹ resulting in the reduction of the viral reservoir size in these infants.^{8,10}

Although early ART typically reduces the viral load (VL), the time to viral suppression (TTS) varies in infants. In 23 reviewed infant studies, the percentage of infants achieving viral suppression ranged from 19% to 81% with a median TTS of 6 months.¹¹ Significant predictors of a fast TTS in infants have been found to be age, VL, and CD4 percentage (CD4%) at start of ART (baseline).¹² The mechanisms underlying these differences in viral suppression in infants are unknown. As a follow-up to previous epidemiological analyses,^{11,12} we aimed to understand how the baseline factors of age, VL, and CD4% contribute to the observed differences in TTS among infants on ART. Therefore, we investigated individual VL decay dynamics from the beginning of treatment until viral suppression was established in perinatally HIV-infected infants receiving ART within the first 6 months of life. These longitudinal measurements were collected by the European Pregnancy and Paediatric HIV Cohort Collaboration (EPPICC).^{12,13}

MATERIAL AND METHODS

Study Inclusion Criteria

We received virological, immunological, sociodemographic, and treatment-related data of 499 infants born between 1997 and 2013 from the EPPICC.^{12,13} These infants were perinatally infected with HIV-1, and 469 started standard ART within 6 months of age. The standard ART regime was composed of either a boosted protease inhibitor or non-nucleoside reverse transcriptase inhibitor and 2 or 3 nucleoside reverse transcriptase inhibitors.¹² To study VL dynamics shortly after the initiation of ART, we required at least one VL measurement within 90 days after the first ART initiation. This requirement was fulfilled by a subset of 312 infants (see Figure S1, Supplemental Digital Content, <http://links.lww.com/QAI/B428>).

Defining Baseline Values, Viral Suppression, and Decay Dynamics

For each infant, we determined age, VL, and CD4⁺ T-cells as a percentage of total lymphocytes (CD4%) at baseline. As the EPPICC was an observational study, baseline VL and CD4% measurements were not available for most infants. To enrich the baseline information, we extrapolated missing measurements of baseline VL and baseline CD4% from the closest measurement within 10 days before ART initiation (see Table S1, Supplemental Digital Content, <http://links.lww.com/QAI/B428>). We chose a short period of 10 days because the VL changes rapidly during acute infections, making more extended extrapolations unreliable.

We restricted our analysis of VL dynamics to the time points before viral suppression. Viral suppression is defined as 2 consecutive VL measurements below 400 HIV RNA copies/mL.¹² We defined the “time to viral suppression” (TTS) as the time to the first measurement <400 copies/mL and considered the second measurement <400 copies/mL to be the endpoint of our investigation. We further characterized 3 VL decay patterns describing the VL dynamics from ART initiation

to TTS: “immediate” VL suppression reached TTS at the first measurement after ART initiation, “monotonic” VL suppression reached TTS with strictly decreasing VL measurements over all consecutive time points, and “erratic” VL suppression reached TTS in a nonmonotonic manner, including increasing VL measurements that increased between consecutive time points. Infants showing an immediate or monotonic VL decay pattern are referred to as the “clean” subset.

Mathematical Model and Statistics

For infants with a “clean” VL decay pattern, we applied parametric fitting methods to estimate baseline VL and to mathematically determine the TTS for each infant. We used Monolix (2018R1, www.lixoft.com) to perform a nonlinear mixed-effects analysis and fitted a phenomenological model of exponential decay to the logarithmically transformed (\log_{10}) VL measurements:

$$V = V_0 e^{-\gamma t} \quad (1)$$

where V_0 is the baseline \log_{10} (VL), and γ is the decay rate of \log_{10} (VL). This model, adapted from Ásbjörnsdóttir et al,¹⁴ can capture the typical “biphasic” decline of the HIV VL during treatment.¹⁵ We applied random effects on both parameters and obtained for each infant a mean estimate and SD for each parameter. These individual parameter sets were used to calculate an expected TTS by solving Equation 1 for the time at which the threshold of \log_{10} (400 copies/mL) is reached. We opted for this phenomenological model because the data for some infants were too sparse to fit the classical mechanistic biexponential model.¹⁵

Statistical analyses were performed with R (Version 3.4.4, R Core Team 2018, <https://www.r-project.org>). We used linear regression and the Spearman correlation test to examine the associations between the different parameters. We quote Spearman correlation coefficients ρ with a significance level of 5%. We assessed cross-sectional time courses with locally estimated scatterplot smoothing (LOESS regressions). Differences in decay patterns were compared using two-sided Wilcoxon-tests. Pearson correlation tests were used to compare estimates derived from fits to experimental measurements. In these cases, we quote the coefficients of determination, R^2 , with a significance level of 5%. Finally, we performed multivariable linear models with the inclusion of intercepts, by first performing univariate analyses, and subsequently considering all univariably significant variables in a multivariable model. We then performed backward stepwise regression, selecting on the basis of the Akaike information criterion. For all cases, we quote β -coefficients and P values with a significance level of 5%.

RESULTS

Early-Treated Infants Differ in Their TTS

From 312 infants of the EPPICC cohorts fulfilling our inclusion criteria (see Methods), 276 infants showed viral suppression (see Figure S1, Supplemental Digital Content, <http://links.lww.com/QAI/B428>). These infants started

standard ART at a median age of 82 days [interquartile range, IQR = (34–121)] with median baseline values of 5.3 \log_{10} (VL) [IQR = (4.2–5.9), $n = 128$] and 33 CD4% [IQR = (22.5–42.5), $n = 99$], and were virally suppressed within a median 132 days [IQR = (64–283), Fig. 1A]. Their distribution of baseline values and sociodemographic characteristics was indistinguishable from the 36 infants who showed no viral suppression in the data available (age: $P = 0.105$, VL: $P = 0.047$, CD4%: $P = 0.671$, open circles in Fig. 1B–D and see Table S1, Supplemental Digital Content, <http://links.lww.com/QAI/B428>). We excluded these 36 infants from the further analysis, as our objective was to study the VL dynamics from first treatment initiation until viral suppression. Thus, the majority of infants (88%) in the EPPICC cohorts starting early ART suppressed their VLs, but they did not suppress VLs equally rapidly. We aimed to understand these differences.

Baseline Conditions and VL Decay Pattern Lead to Differences in TTS

To understand the wide range of observed TTS, we assessed differences among the 276 viral suppressors. First, we examined correlations between observed TTS and baseline measurements. Infants with a shorter TTS started treatment at a younger age ($P < 9 \times 10^{-9}$, Fig. 1B), a lower baseline VL ($P < 2 \times 10^{-5}$, Fig. 1C), and a higher baseline CD4% ($P < 7 \times 10^{-4}$, Fig. 1D). All these associations are in agreement with a previous analysis by the EPIICAL consortium, using a somewhat larger selection of infants from the same cohorts.¹² Thus, the age, VL, and CD4% at which infants start treatment are clearly correlated with differences in TTS.

We then studied how individual VL decay dynamics lead to viral suppression. We classified the data into 3 decay patterns (Fig. 2A): 47 infants showed immediate VL suppression after ART initiation (immediate), 141 infants had a VL declining in a strictly monotonic manner (monotonic),

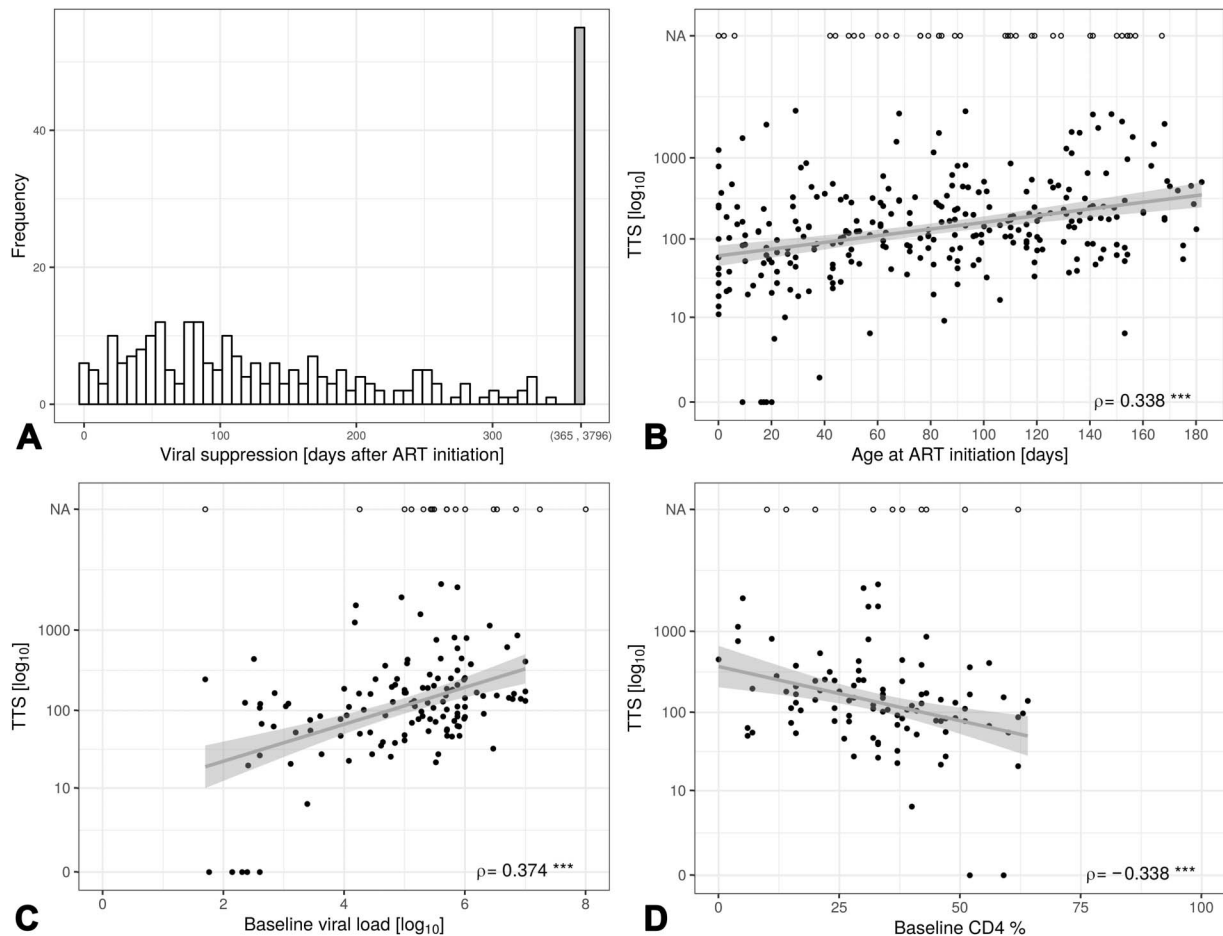


FIGURE 1. TTS is associated with baseline measurements. A, Histogram of TTS with a bin-width of 7 days. TTS longer than one year are summarized in one bin. B–D, TTS correlations with (B) age, (C) observed VL, and (D) observed CD4% at ART initiation. To cover the wide range TTS is shown on a logarithmic scale. Filled circles represent infants experiencing viral suppression; open circles represent infants not achieving viral suppression, their TTS is noted with NA. Linear regressions with 95% confidence intervals are shown in grey. The Spearman correlation test was performed, and significant correlation coefficients ρ are given (P -value < 0.001 ***).

and 88 infants showed an “erratic” VL with irregular and intermittent increases in the VL (erratic). As expected, infants with a “clean” (immediate or monotonic) decay pattern suppressed the virus in a significantly shorter time than infants with an erratic decay pattern ($P < 2.2 \times 10^{-16}$, Fig. 2B). Infants with erratic decay patterns had, according to their medical records, more changes in treatment (mean = 2.89, SD = 2.26, $P < 2.2 \times 10^{-16}$) and accumulated treatment interruptions (mean = 0.28, SD = 0.55), suggesting treatment-related challenges such as poor adherence, drug resistance, toxicity, or irregular drug administration. Thus, long TTS in infants with erratic VL declines are at least partly due to treatment complications. Consequently, the VL dynamics of infants with erratic VL decay patterns provide no reliable information regarding the factors associated with VL suppression after first ART initiation because we cannot exclude the possibility that the observed TTS might be a response to later initiated ART. Therefore, we removed these infants from the analysis.

Erratic VL Patterns are Associated With Significantly Lower Baseline CD4%

Next, we asked whether the exclusion of infants with erratic VL decay dynamics would introduce a bias in our analysis regarding the baseline conditions. To address this issue, we

compared the different decay patterns by the age and the conditions at which infants started treatment. Although the age at start of treatment ($P = 0.213$, Fig. 2C) and baseline VL ($P = 0.465$, Fig. 2D) was comparable between infants with erratic and clean VL declines, infants with an erratic decay pattern started treatment with a significantly lower baseline CD4% ($P = 0.032$, Fig. 2E). We found no significant differences in baseline CD4% between immediate suppressors and infants with a monotonic decay pattern ($P = 0.065$, Fig. 2E). Infants with an immediate suppression started treatment significantly earlier ($P = 0.015$, Fig. 2C) and with a significantly lower baseline VL ($P < 7 \times 10^{-5}$, Fig. 2D) than monotonic decliners. Thus, we acknowledge that infants with an erratic decay pattern are likely biased towards significantly lower baseline CD4% than infants with an immediate or monotonic VL suppression.

Mathematical Modeling Estimates Missing Baseline VL and Determines TTS More Precisely

To further determine with more precision which factors determine TTS during a successful initial ART, we focused on the “clean” subsets of infants with an immediate or monotonic decay pattern. To estimate additional parameters describing these VL decay dynamics, and to more precisely determine their TTS rather than defining it as the time of the first measurement of VL

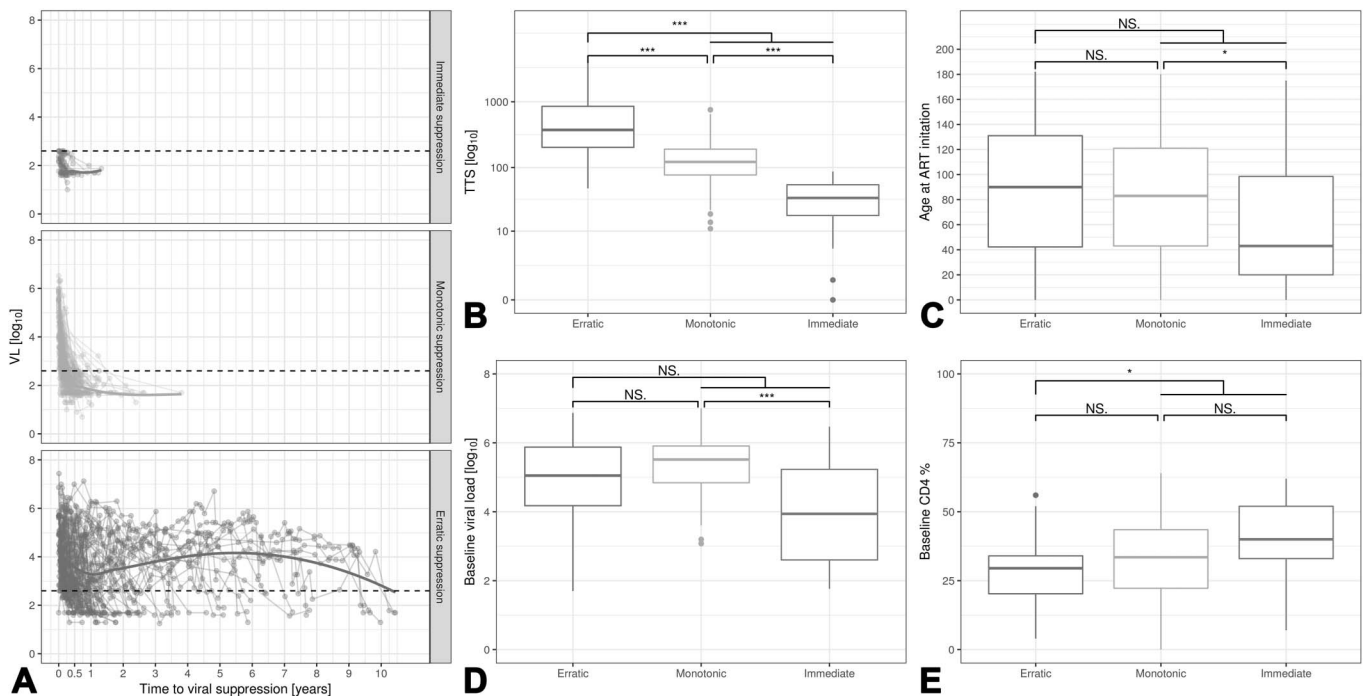


FIGURE 2. Association of VL decay patterns with TTS, age, baseline VL, and baseline CD4%. A, The initial VL decay after initiation of standard ART is shown for 276 infants distinguished by their decay pattern: immediate ($n = 47$), monotonic ($n = 141$), and erratic suppression ($n = 88$). The measurements belonging to the same individual are connected by lines. The dashed line represents the threshold VL of 400 copies/mL and the thick lines represent the LOESS regressions for each category. B–E, Boxplots distinguished by VL decay pattern (erratic, monotone, immediate) show differences in the distribution of (B) time to viral suppression (TTS), (C) age, (D) observed baseline VL, and (E) observed baseline CD4% at ART initiation. Two-sided Wilcoxon tests were performed, and significance levels are shown (P -value ≥ 0.05 : NS, P -value < 0.05 : * P -value < 0.01 : ** P -value < 0.001 : ***).

<400 copies/mL, we performed mathematical modeling (see Figure S2, Supplemental Digital Content, <http://links.lww.com/QAI/B428>). By fitting the phenomenological model (Equation 1), we inferred the baseline VL for all 188 infants with a “clean” VL decay kinetic (population estimate V_0 , fixed \pm SD of the random effect: 4.52 ± 0.196); these estimated VLs were in good correspondence with the available measured values ($R^2 = 0.85$, $P < 2.2 \times 10^{-16}$, $n = 87$, see Figure S3A, Supplemental Digital Content, <http://links.lww.com/QAI/B428>). We also estimated the slope parameter γ , which is the readout for the VL decline, showing huge individual variability (population estimate γ , fixed \pm SD of the random effect: 0.01 ± 0.705). The residuals of both parameter estimates were uncorrelated (see Figure S3B, Supplemental Digital Content, <http://links.lww.com/QAI/B428>). From these parameters, we calculated a TTS; these were (as expected) shorter than those derived from the measurement times ($R^2 = 0.52$, $P < 2.2 \times 10^{-16}$, see Figure S3C, Supplemental Digital Content, <http://links.lww.com/QAI/B428>), as viral suppression occurs before an undetectable measurement. The median estimated TTS for the clean subset was 54 days [IQR = (32–82), see Figure S3D, Supplemental Digital Content, <http://links.lww.com/QAI/B428>]. We confirmed the previously observed correlations of age ($P = 4.793 \times 10^{-12}$, Figure S3A), estimated baseline VL ($P < 2.2 \times 10^{-16}$, see Figure S4B, Supplemental Digital Content, <http://links.lww.com/QAI/B428>), and measured baseline CD4% ($P = 0.005$, $n = 69$, see Figure S4C, Supplemental Digital Content, <http://links.lww.com/QAI/B428>) with the TTS estimated by data fitting.¹² In addition, TTS negatively correlated with the slope parameter ($P < 2.2 \times 10^{-16}$, see Figure S4D, Supplemental Digital Content, <http://links.lww.com/QAI/B428>). All these parameters are highly correlated with each other (see Figure S4–S6, Supplemental Digital Content, <http://links.lww.com/QAI/B428>). Thus, mathematical modeling provides additional information in infants with monotonic VL decay dynamics, allowing us to identify factors driving the TTS.

A Low Baseline VL and a High Baseline CD4% Lead to a Short TTS

The slope parameter γ has the strongest correlation with TTS (see Figure S5, Supplemental Digital Content, <http://links.lww.com/QAI/B428>) and is a phenomenological param-

eter describing the “quality” of suppression. We used linear regression analysis to assess which biological parameters (age, VL, or CD4%) were the best predictors of the slope parameter γ . Although a univariable model confirmed the Spearman correlations from Figure S5, Supplemental Digital Content, <http://links.lww.com/QAI/B428>, the results of a multivariable model highlighted a significant effect of the estimated baseline VL on the slope parameter γ (Table 1). This significance further increased in a subsequent stepwise model. Here, the factor age dropped out, whereas the estimated baseline VL and measured baseline CD4% remained as biological factors explaining the slope parameter γ . We then performed a similar linear regression analysis for the estimated TTS (Table 1). Again age, baseline VL, and baseline CD4% were significantly associated with TTS in a univariable model. In a multivariable model, baseline VL was the only remaining significant factor. This remained the case in a subsequent stepwise model, in which the CD4% was also retained. Thus, TTS depends mainly on the baseline VL and CD4%, and the effect of age might be indirect as it affects baseline VL and baseline CD4%.

Perinatal HIV Infection Progresses Rapidly in Untreated Infants

To study how age affects VL and CD4%, we considered the natural course of HIV infection before the onset of treatment. We used all pretreatment measurements of infants from the EPPICC cohorts to form a cross-sectional time-course of VL and CD4% (Fig. 3). In general, infants have very low or undetectable VLs at birth. As they grow older, their HIV infection progresses in the absence of treatment. VL measurements increase within a few weeks to reach a plateau around $6 \log_{10}(\text{VL})$ after approximately 50 days (Fig. 3A). In contrast to adults, infants maintain this high setpoint VL for at least half a year.^{8,16} The measured CD4% then exhibits a constant decline as HIV infection progresses (Fig. 3B). We used the raw baseline CD4% measurements and did not normalize them to correct for the natural CD4% decline because the natural CD4% barely shows a decline during the first 6 months of life (see dashed band in Fig. 3B).¹⁷ The observed decline in CD4% is much steeper than the natural

TABLE 1. Baseline VL and Baseline CD4% Determine TTS

	Univariable		Multivariable		Stepwise	
	β	<i>P</i>	β	<i>P</i>	β	<i>P</i>
Slope (γ)						
Age	-5.31×10^{-5}	<0.001	-4.11×10^{-6}	0.805	—	—
V_0	-4.82×10^{-3}	<0.001	-3.63×10^{-3}	0.002	-3.72×10^{-3}	<0.001
CD4%	1.07×10^{-4}	0.022	5.81×10^{-5}	0.227	6.28×10^{-5}	0.154
TTS						
Age	0.44	<0.001	0.08	0.554	—	—
V_0	46.25	<0.001	35.02	<0.001	36.96	<0.001
CD4%	-1.05	0.01	-0.52	0.204	-0.62	0.101

Linear regression models for slope parameter γ and for TTS. The independent variables are age at start of ART, estimated VL (V_0), and observed baseline CD4%. The regression coefficients β and the *P*-values are given for a univariable, multivariable model, and the final model resulting from a stepwise approach. Significant *P*-values are marked in bold.

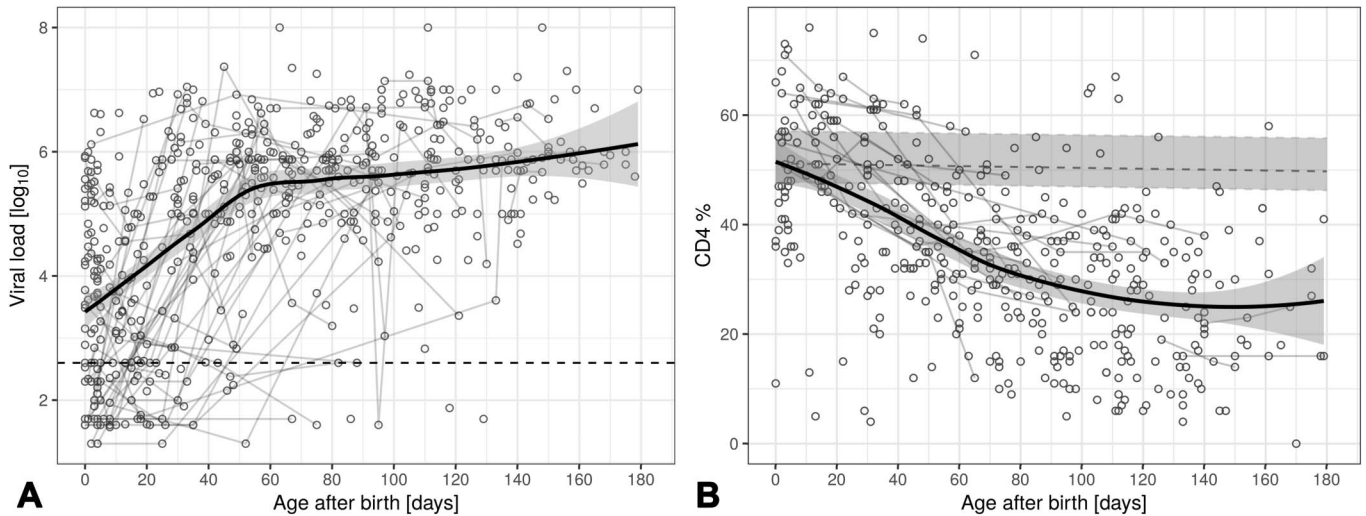


FIGURE 3. VL increases and CD4% decreases as HIV infection progresses. Cross-sectional time course of HIV infection before treatment. A, From 360 infants in the EPPICC database, we obtained 580 VL measurements before any treatment was applied. These VL measurements are plotted against age on a logarithmic scale. The dashed line represents the threshold VL of 400 copies/mL. B, From 309 infants in the EPPICC database, we obtained 427 CD4% measurements before treatment. These CD4% measurements are plotted against age. Reference values and 95% confidence intervals of natural CD4% decline in healthy children are represented by the dashed band (data retrieved from Huenecke et al¹⁷). LOESS regression with 95% confidence interval is shown in black. Infants with more than 3 follow-up measurements are represented by connected grey lines.

decline in infants (Fig. 3B). Thus, age markedly affects the VL and CD4%, as infants starting treatment late are expected to have a high VL and a low CD4%. Consequently, these infants are expected to have a long TTS (Table 1). Note that the effect of age on the VL vanishes after 6–8 weeks, when most infants have approached a plateau level, while the effect of CD4% persists (Fig. 3). Summarizing, we find that baseline VL and CD4% are the main drivers of TTS and that age operates indirectly by increasing VL and decreasing CD4%. Although at an early stage of an HIV infection age increases TTS by increasing VL and decreasing CD4%, at a later stage, the effect of the VL stabilizes and the deteriorated immune

system reflected by declining CD4% determined prolonged TTS (see also Fig. 4A).

Baseline VL and Baseline CD4% Predict TTS

Finally, we checked how well the results of the stepwise linear model (Table 1), ie,

$$TTS = 36.96 \times V_0 - 0.62 \times CD4_0 - 82.81 \quad (2)$$

could be used to predict TTS. This model (Equation 2), including an intercept ($P = 0.085$), predicts the TTS estimated

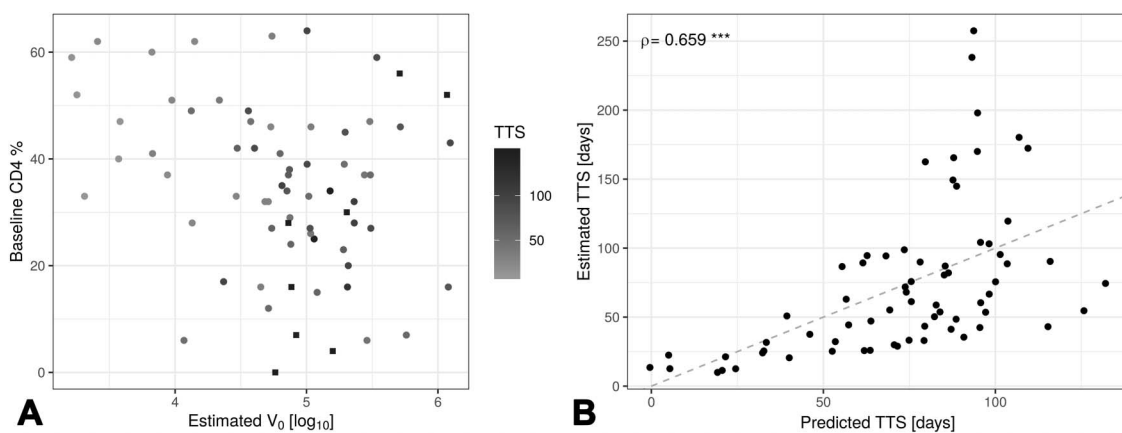


FIGURE 4. Baseline VL and baseline CD4% predict TTS. A, Estimated TTS in dependence of estimated VL and baseline CD4% measurements ($n = 69$). Increasing intensity of the grey scale represents longer TTS. TTS greater than 150 days are marked by squares. B, Correlation plot between predicted and estimated TTS. TTS was estimated by fitting data, and TTS was predicted with model of Equation 2. The dashed line represents the diagonal. Spearman’s correlation coefficient ρ is given with a P -value < 0.001 (***)

by the model of Equation 1 reasonably well (Fig. 4B, $P < 2.2 \times 10^{-16}$), using the estimated baseline VL (V_0 , $P < 7 \times 10^{-5}$) and the measured baseline CD4% ($CD4_0$, $P = 0.101$) as input values. We decided for the estimated VL baseline values as we had more data available, but the results with the measured VL baseline values performed equally good. The TTS could be predicted well with Equation 2, if the baseline VL was within the increasing VL phase of a natural HIV infection course (Fig. 3A) because the baseline VL is the major predictor (Table 1). Once the VL plateau is reached, the model (Equation 2) underestimates the TTS. These skewed model predictions for late times to suppression suggest that the effect of CD4% is nonlinear and might play a more dominant role in predicting TTS at a later stage of an HIV infection. This prediction, therefore, provides a lower bound for the expected TTS at the start of treatment. Equation 2 could be used as a diagnostic tool for early treated infants that are responding poorly to their treatment.

DISCUSSION

We studied “clean” VL decay dynamics of early treated perinatally HIV-infected infants from a heterogeneous database (EPPICC) to identify the major factors underlying TTS during the first treatment. We identified low baseline VL and high baseline CD4% as the major factors contributing to fast TTS. Because VL might be regarded as a trivial factor influencing TTS, the contribution of CD4% at start of ART is our most interesting and surprising result, although the association with TTS is not significant at the 5% level (Table 1). In addition, high CD4 levels are likely to be protective because the CD4% was also significantly lower in infants experiencing an erratic response. The exact mechanisms by which CD4 T-cell levels affect viral suppression remain elusive, but our results suggest that the status of the deteriorating immune system at the onset of treatment influences the TTS. Further immunological studies, considering different immune compartments, eg, levels of CD8 T cells or innate immune cells, are required to investigate and clarify the contribution of the immune system to viral suppression.

In contrast to a previous analysis of a somewhat larger subset of the same EPPICC cohort,¹² we cannot confirm an independent effect of age on TTS. A major difference is that we selected for a subset of infants with “clean” VL decay dynamics to ensure that our analysis was not confounded by inefficient or ineffective treatment. In the previous studies,^{11,12} the TTS need not be the response to the initial treatment and could also be the response to a later initiated treatment because of subsequent changes in treatment. Note that we have not generated an age bias by excluding infants with an “erratic” VL decay dynamics (see Table S1, Supplemental Digital Content, <http://links.lww.com/QAI/B428>). Thus, we find that age describes the TTS in an indirect manner (via baseline VL and CD4%).

The age at start of treatment need not reflect the time period of progression of an HIV infection. With improving diagnosis and treatment of pregnant women, increasing proportions of infants are in-utero infected as opposed to

intrapartum infected, and although infants usually start ART soon after birth, they might have progressed already in their HIV infection. In-utero infections might, therefore, confound the age effect on TTS, and baseline VL and baseline CD4% are more appropriate covariates in this setting. Unfortunately, we lacked any information about time of infection (in-utero or intrapartum). To determine whether a separate age contribution affects TTS, comparisons between in-utero versus intrapartum-infected infants are necessary, which will require more detailed studies addressing virological and immunological dynamics in young infants (eg, the EARTH study by EPIICAL).

We based our analyses on a simple phenomenological mathematical model describing the decay in the VL during treatment. Typically, a mechanistic model is used to capture the multiphase decay dynamic of HIV,¹⁵ but the sparseness of virological measurements did not allow proper identification of all parameters of this mechanistic model. The EPPICC database is a collection of observational studies and was not specifically designed to investigate TTS. Available virological and immunological measurements were sparse, and baseline measurements, and regular measurements during the initial VL decay, were often missing. The sparse datum is a generic problem in studies with infants. According to the WHO recommendations, only a limited volume of blood can be extracted from infants, which reduces the variety of medical tests that can be performed. To counter these issues, we used an exponential fit to the log-transformed VL data to predict missing baseline VLs and to estimate decay rates reflecting multiphase VL decay dynamics phenomenologically. In addition, we used a nonlinear mixed-effect model approach to compensate for missing data with a population mean. By combining phenomenological and mixed-effect approaches, we dealt with the sparseness of the data and nevertheless obtained individual estimates for infants in a heterogeneous data set allowing us to precisely determine individual TTS.

In conclusion, the data presented here support the fact that age at start of ART is less helpful than markers of infection such as VL and CD4% to determine TTS. Thus, an early ART initiation not only prevents disease progression but also shortens TTS. In addition to a low VL and a high CD4% at start of treatment, a “clean” VL decay pattern might predict an initially successful treatment responses resulting in fast TTS. Early ART initiation combined with a fast viral suppression may lead to the reduction of the early established latent viral reservoir by shortening the viral exposure time, which may allow for temporarily sustained VL suppression during treatment interruptions as seen in cases such as the Mississippi baby¹⁸ and the VISCONTI child.¹⁹ The opportunity to schedule treatment interruptions for perinatally HIV-infected infants has been investigated in the CHER trial.^{20,21} However, only a minority of early treated children achieved a prolonged viral suppression without ART,^{21,22} and novel therapeutic strategies are needed to obtain viral remission.²³ Testing novel immunotherapeutic approaches in the early treated HIV-infected children is the challenge of the EPIICAL network, of which we are part of.⁹ Potential candidates for such treatment interruptions studies should be chosen on an

individual basis. Our results provide a first step for such selection criteria, namely early ART initiation, low baseline VL, high baseline CD4%, and a “clean” VL decay pattern to suppression. As a next step, the effect of these criteria on sustained VL suppression has to be investigated.

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